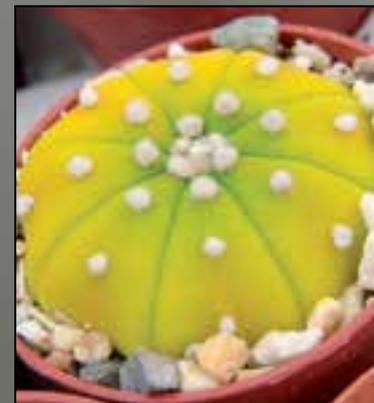


# Plant Cell and Tissue Culture

## Phytopathology

## Biochemicals



**D**uchefa



Catalogue 2010-2012

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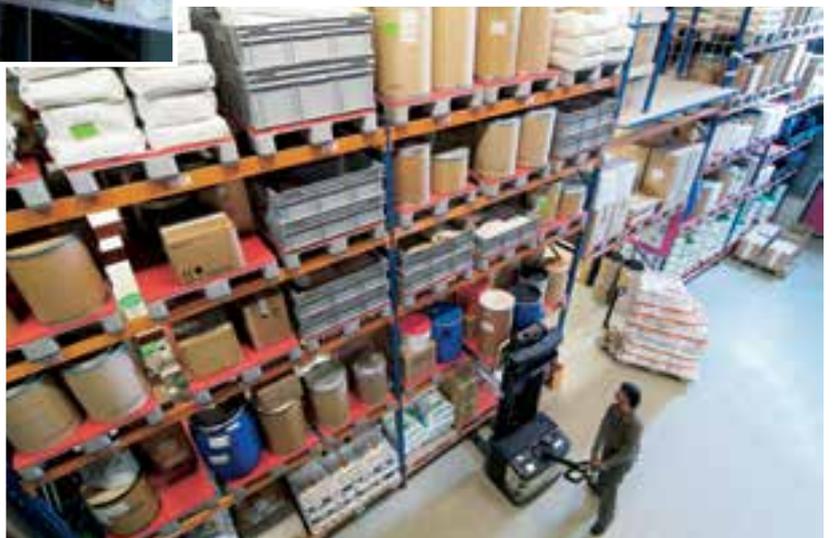
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# Duchefa



Dear customer,

After turning the cover we symbolically invite you to enter Duchefa Biochemie's warehouse filled with products aiming at the world of Plant Cell Tissue Culture and Biochemicals. At the same time it gives us great pleasure to introduce the edition 2010 – 2012 of our catalogue to you. In the tradition we have experienced over the years many of you were kind enough to send us illustrative photomaterial. Some of you even allowed us to pay a visit with our professional photographer to capture what Plant Cell and Tissue Culture is all about. This kind of reception has made it possible for us to make a catalogue once again to the best of our tradition. For this support and for your continuous interest in our products we honestly thank you very much.

Although the range of products we offer for all your needs arising from your professional activities in biochemistry has developed over the years to allow in contemporary terminology "one stop shopping", some minor additions and changes were made in our productportfolio.

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Sincerely yours,  
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drs C.M. Teves  
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**Flammable:** liquids with a flash point of 21°C or more and below or equal to 55°C.



**Oxidizing:** substances which produce highly exothermic reactions in contact with other substances.



**Very toxic:** substances which can cause extremely serious acute or chronic effects.



**Toxic:** substances which can cause serious, acute or chronic effects.



**Harmful:** substances which can have limited effects on health.



**Irritant:** substances which can cause inflammation.



**Corrosive:** substances which can destroy living tissue.



**Dangerous for the environment**



# PLANT TISSUE CULTURE MEDIA

Media used in plant tissue culture are composed of several components: salts, vitamins, amino acids, growth regulators, sugars, agar or Gelrite™ and water. All these compounds fulfil one or more functions in the *in vitro* growth of plants.

The minerals present in plant tissue culture media can be used by the plant cell as building blocks for the synthesis of organic molecules, or as catalysators in enzymatic reactions. The ions of the dissolved salts play an important role as counterion in the transport of ionized molecules by the plant, in the osmotic regulation, and in maintaining the electrochemical potential of the plant.

Nitrogen, sulphur and phosphorus are components of proteins and nucleic acids. Magnesium and many micro-elements form essential parts of enzymes and cell organelles, and are therefore important in the catalyzation of various reactions. Calcium and boric acid are mainly found in the cell wall and especially calcium has an important task in the stabilization of biomembranes. Potassium and chloride, on the contrary, are important in the osmotic regulation, for maintenance of the electrochemical potential, and for the activation of a large number of enzymes.

## Micro- and macro-elements

The salts in media can be divided into micro- and macro-elements. Fe, Cu, Mn, Co, Mo, B, I, Ni, Cl and Al are considered as micro-elements and Mg, Ca, P, S, N and K as macro-elements. This subdivision in micro- and macro-elements is mainly based on the needs of the plant for these elements. The need for micro-elements is small, reflected by the low concentrations of these elements in the medium. Most micro-elements are present in micromolar quantities. The need for macro-elements is much larger and therefore present in millimolar concentrations.

The smaller need for micro-elements is certainly not a guide-line for the importance of these elements for the plant. As for macro-elements, an iron deficiency can have catastrophic effects for the growth and development of the plant cell as well. However, in practice, a shortage of micro-elements in media is easier replenished by, for example, pollutions that are naturally present in agars, salts and water.

The necessity of some micro-elements as a medium component is not yet clear. Cobalt, aluminum and nickel might be useful for the plant, but are probably not essential.

In fact, of most micro-elements only the mineral part of its related salt is of importance to the plant. The anion is mostly not essential. The main function of copper sulphate is exerted by  $\text{Cu}^{2+}$ . The  $\text{SO}_4^{2-}$  ion is abundantly present in media and mainly derived from magnesium or potassium sulphate.

It is hard to recommend the minimal required amount of minerals to be added to a medium. In the Murashige and Skoog medium, developed for *Nicotiana tabacum*, the concentrations of Fe, B, Mn and Zn are significantly increased as compared to the starting medium. These increased concentrations result in a higher yield for growth. Litvay also used higher concentrations of micro-elements for suspension cultures of *Daucus carotus*. Eriksson, on the contrary, reports an increase in the yield of growth if the concentration of micro-elements present in the MS medium is reduced to one tenth of the initial concentration.

The need of a plant for macro-elements is much greater. In general, from the macro-elements both anions and cations are important for the plant cell. For example, of potassium nitrate, both  $\text{K}^+$  and  $\text{NO}_3^-$  are essential. Obviously, the macro-elements have the highest concentrations in the media used for plant cell and tissue culture. Within the group of macro-elements, the nitrogen containing salts, mainly in the form of potassium, ammonium or calcium nitrate, are used most.

The concentration of ammonium that can be supplied without harmful consequences for the plant is sometimes sharply defined. This is particularly well demonstrated by the description of the medium developed by Chu *et al.*



Cactaceae: *Pelecyphora asseli formis*,  
Succulent Tissue Culture, The Netherlands

## Vitamins

Vitamins are added to the plants in several forms and concentrations. Certainly, these compounds are essential for many biochemical reactions. In almost all media for plant cell and tissue culture, Thiamine (vitamin B1) is included. Linsmaier and Skoog assert, after a thorough revision of the vitamins present in the MS medium, that this vitamin is essential for growth. The importance of the role of Thiamine is stressed by other authors as well. Inositol is often mentioned as a vitamin that significantly stimulates the growth and development of plants. However, the vitamin is not essential for growth. Concerning other vitamins, it is hard to judge their virtual importance. The effect of vitamins on the development of the cell *in vitro* differs from species to species or might even be harmful.

Duchefa Biochemie B.V. produces custom-made media for prominent laboratories, institutes and companies which are all very active in the field of plant- and tissue culture. This production is performed under guaranteed secrecy and therefore these media are not described in this work. It is clear that, taking the considerable production of these uniquely composed media into consideration, with the help of the nutrients present in the medium still a large area has to be explored on the development and stimulation of growth under *in vitro* circumstances.

## MICRO ELEMENTS

Boron, Chloride, Iron, Cobalt, Copper, Manganese, Molybdenum, Zinc.

### BORON (B)

Of all elements necessary for the growth of plants, the need for boron is least understood. Boron is taken up by the roots and transported via the xylem to other parts of the plant. In the cell membrane it is mainly present as a borate ester. There are no enzymes known that contain boron or that are activated by boron. However, there are indications that cis-diol borate complexes can be formed with components present in or on membranes. The formation of these complexes might influence the activity of membrane-bound enzymes. The functions of boron are mainly extracellular. The element is involved in the lignification of the cell wall and differentiation of the xylem.

#### Cell wall

Boric acid is capable of forming stable mono- and diesters with cis-diols, present in molecules with many OH groups (polyhydroxyl compounds). A number of sugars like mannitol and polymannuronic acid have a similar configuration, making the formation of boric esters possible. These sugar-borate esters are part of the hemicellulose fraction of cell walls. Most of the boron present in the plant is in the form of an ester localized in the cell wall of the plant. The higher demand for boron by dicotyledons in comparison with monocotyledons is most probably due to higher concentrations of components with cis-diol configurations in the cell wall of the former. It is assumed that boron, like calcium, has a regulating role in the synthesis of the cell wall as well as in the stabilisation of constituents of the cell wall and cell membrane.

A deficiency of boron immediately results in inhibition of the length growth of primary and secondary roots. IAA oxidase activity strongly increases. Furthermore, boron participates in the regulation of the phenol metabolism and the synthesis of lignins by forming a stable borate ester between boric acid and phenolic acids.

### CHLORINE (Cl)

The concentrations of chlorine present in the plant vary from 70 to 700 mM per kilogram dry weight (2000 to 20000 mg/kg dry weight). Chlorine is taken up as  $\text{Cl}^-$  and is very mobile in the plant. The main functions of the ion are osmoregulation and compensation of charges.

#### Chloroplast

Chloride most probably plays a role in photosystem II during the Hill reaction, when  $\text{H}_2\text{O}$  is split into  $\text{O}_2$  and  $2\text{H}^+$ . It is assumed that chloride functions as a cofactor in the oxygen generating manganese complex. The chloroplasts of spinach and sugar beet contain chloride in a concentration of approximately 100 mM. In the leaves, less than 10 mM is present, showing a clear preference of chloride to accumulate in the chloroplasts.

#### Osmotic potential

The chloride ion regulates the opening and closing of stomata.  $\text{Cl}^-$  compensates the  $\text{K}^+$  influx during opening of the guard cells. In onions, which

lack chloride, opening of the stomata is prevented. At the closing time of the stomata, an efflux of  $\text{K}^+$  and accompanying anions, mainly  $\text{Cl}^-$ , out of the guard cells takes place. During shortage of chloride the stomata remain open, which might result in a severe loss of water.

Chloride is very important in the regulation of the osmotic potential of vacuoles and to turgor related processes.

#### ATPase

Monovalent cations, like  $\text{K}^+$ , highly stimulate Mg-ATPases located in the cell membrane in generating an  $\text{H}^+$  efflux. There are indications that a second type of  $\text{H}^+$  transporting ATPase exists in membranes of cell organelles in the cytoplasm. This ATPase is not activated by monovalent cations, but by  $\text{Cl}^-$  ions. Protons and chloride ions are simultaneously transported over the tonoplast, thereby creating a pH gradient between cytoplasm ( $\text{pH} > 7$ ) and vacuole ( $\text{pH} < 6$ ).

#### Nitrogen metabolism

Chloride activates asparagine synthetase, an enzyme important in nitrogen metabolism. This enzyme converts glutamine into asparagine and glutamic acid. In the presence of  $\text{Cl}^-$ , the reaction speed is increased seven-fold. Therefore, in plant species that use asparagine as the main carrier of nitrogen over longer distances, chloride fulfils an important function in nitrogen metabolism.

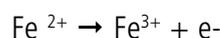


Willemsen en Bourgondiën B.V., The Netherlands

### IRON (Fe)

In plants iron is mainly bound to chelators and complex compounds. Free  $\text{Fe}^{2+}$ ,  $\text{Fe}^{3+}$  levels are extremely low ( $10^{-10}$  mM). Most plants only absorb  $\text{Fe}^{2+}$ . Therefore,  $\text{Fe}^{3+}$  has to be reduced to  $\text{Fe}^{2+}$  at the root surface before it is transported to the cytoplasm (only grasses mainly take up iron in the form of  $\text{Fe}^{3+}$ ).

During transport over longer distances, through the xylem of plants, iron is mainly transported as an iron-carbohydrate complex. Generally, this occurs as  $\text{Fe}^{3+}$ -citrate or as iron-peptide complex. The major function of iron in the plant is to form iron chelates. The element functions as a reversible oxydation-reduction system, according to:



## Hemoproteins

The iron containing proteins can be separated into hemoproteins and ironsulphur proteins.

The most well known hemoproteins are the cytochromes, which contain an iron-porphyrin complex as prosthetic group. Cytochromes form an integral part of the redox system of the electron transporting chain in chloroplasts and mitochondria of plant cells (see magnesium).

Cytochromes function as intermediates for electrons, required for the reduction of nitrate to nitrite by the enzyme nitrate reductase (see nitrogen) in the nitrogen assimilation.

In nitrogen fixation in legumes, cytochromes are intermediates of the electron transport chain along which electrons are transported to finally reduce  $N_2$  into  $NH_3$ .

Catalases and peroxidases are also heme-iron containing enzymes. Catalases participate in the photorespiration, glycolysis and the dismutation of hydrogen peroxide, according to the following equation:



Hydrogen peroxide is formed by superoxide dismutase in order to neutralize superoxide radicals. Hydrogen peroxide, in its turn, is neutralized by catalase. Peroxidases are abundantly present in plant cells. Cell wall bound peroxidases catalyse the polymerization of phenols to lignins. Roots contain high levels of peroxides and play a role in the iron uptake of the plant. An excess of phenols, which occurs in iron deficiency, will be excreted externally.

## Iron-sulphur proteins

The second group of iron binding proteins are the iron-sulphur proteins. The iron is bound to a thiol group (-SH) of cysteine and/or inorganic sulphur. Ferridoxin is the most common iron-sulphur protein and functions as carrier in the electron transport of reactions catalyzed by nitrite reductase, sulphate reductase, the synthesis of  $NADP^+$  during photosynthesis and nitrogen reduction executed by the nitrogenase complex. Three different iron-sulphur proteins, lying in serial order, are involved in the electron transport chain of the nitrogenase complex.

Besides these two groups of iron containing proteins, the plant has a number of other enzymes that contain iron. The element is necessary for redox reactions and the stabilisation of enzyme substrate complexes.

Iron is important in the biosynthesis of chlorophyll. In young leaves, iron deficiency is immediately followed by a reduction in the concentration of chlorophyll, because the protein synthesis is blocked. The number of ribosomes in the cells is also drastically reduced.

Iron deficiency in the roots is manifested by morphological changes. The elongation of the roots decreases, but the diameter and amount of root hairs increase. In green leaves 80% of the iron is located in the chloroplast. During a shortage of iron, all will be located in the chloroplast.

## COPPER (Cu)

Copper is a divalent cation and is taken up by the plant as  $Cu^{2+}$  or as a copper chelate complex. If equimolar concentrations of  $Cu^{2+}$  and complexed copper are present, the plant seems to have a preference for the free copper ion. In the xylem and phloem, copper is almost exclusively transported as a copper complex, mostly an amino acid-copper complex. Within the cell, copper is mostly part of enzyme complexes and important in redox reactions  $[(Cu^{2+})/(Cu^+)]$  executed by these enzymes. A shortage of copper immediately results in a decrease of the activity of many copper containing enzymes.



Willemsen en Bourgondiën B.V., The Netherlands

## Photosynthesis

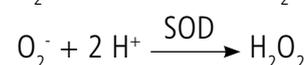
About 50% of the copper present in chloroplasts is bound to plastocyanin. This intermediate of the electron transport chain between photosystem I and II, contains one copper atom per molecule.

In copper deficiency, the concentration of plastocyanins is decreased. Like plastocyanins, plastoquinones play an important role in the transfer of electrons between photosystem I and II. When copper is deficient, the membrane of the chloroplast lacks two proteins which influence the mobility of plastoquinones. For the synthesis of plastoquinones the presence of the enzyme laccase is required. Laccase is a copper containing enzyme of which the activity is immediately reduced in copper deficiency. Therefore, a shortage of copper is followed very quickly by a decrease in the photosynthesis.

## Super Oxide Dismutase

Copper is, in addition to zinc, part of the enzyme Super Oxide Dismutase (Cu-Zn.SOD), which plays an important role in the neutralization of the highly reactive superoxide anion radical  $O_2^-$ , which is formed during photorespiration. Beside the Cu-Zn.SOD a manganese containing SOD is present in the cell as well.

SOD detoxifies the reactive  $O_2^-$  radical into  $H_2O_2$  and  $O_2$ , thereby protecting the cell for the destructive capacity of this radical. SOD is, together with catalase, involved in the following reactions:



Superoxide is neutralized by SOD and the  $H_2O_2$  is subsequently detoxified into oxygen and water by catalase.

The copper-zinc containing SOD enzymes are mainly found in the stroma of chloroplasts. Most  $O_2^-$  and  $H_2O_2$  is formed in the chloroplast. In young leaves, 90% of the SOD is located in the chloroplasts and only 4-5% in the mitochondria.

If copper is deficient, changes in the structure of chloroplasts occur, clearly showing the protective function of copper.

Copper also plays an important role in the mitochondrial electron transport chain. The terminal cytochrome oxidase contains two copper and two iron atoms in a heme configuration.

## COBALT (Co)

The function of cobalt in the plant is not known. On the other hand, cobalt is important in nitrogen fixation, like in root tubers of legumes of *Rhizobium* species.

Cobalt is an essential component of the cobalamin enzyme. Co(III) is the metal component situated between four nitrogen atoms in a porphyrin structure. Three enzyme systems of *Rhizobium* bacteria are known to contain cobalamin. A relation is found between the cobalt concentration, nitrogen fixation and root tuber development.

Cobalt is required for bacterial methionine synthesis, ribonucleotide synthesis and synthesis of methylmalonyl-coenzyme A mutase. Methylmalonyl-coenzyme A mutase is necessary for the synthesis of leghemoglobin.

Leghemoglobin is of great importance in the protection of nitrogenase against oxygen, which is able to irreversibly damage the enzyme.

It is not clear if cobalt has a function in higher plants. Only one cobalamin dependent enzyme is known, leucine-2,3-aminomutase in potatoes. For lower plants, cobalt is essential and present in several subcellular fractions and the thylakoids of chloroplasts.



## MANGANESE (Mn)

Manganese is taken up by the plant as bivalent, unbound  $Mn^{2+}$  ion and transported in this form from the roots via the xylem to other parts of the plant.

The element is strongly bound to several metalloproteins, either as structural part of the binding site of the enzyme or as part of the [Mn(II)/Mn(III)] redox system.

### Hill reaction

Manganese has two important functions in the plant. The ion is involved in the so-called Hill reaction of photosystem II, in which water is split into oxygen and protons, according to:



It is assumed that the four manganese atoms are a part of a protein, which catalyzes the hydrolysis of water. The released electrons are subsequently transferred to magnesium containing pigment 680, the center of photosystem II.

### Super Oxide Dismutase

Until now only a few manganese containing enzymes have been isolated. The most important manganese containing enzyme is manganese Super Oxide Dismutase (Mn-SOD). (See copper for more information about SOD).

Like for copper, if manganese is deficient, changes in the structure of the chloroplasts occur, clearly showing the protective role of manganese.

## MOLYBDENUM (Mo)

Molybdenum is in aqueous solutions mainly present as  $MoO_4^{2-}$ . In a weak acidic environment, the molybdate ion can, depending on the acidity, accept one or two protons, according to:

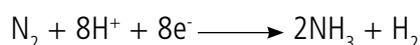


Polyanions like tri- and hexamolybdate can be formed as well. Molybdenum has limited mobility in plants and is probably transported through the xylem and phloem as  $MoO_4^{2-}$  ion.

### Nitrogenase

A few enzymes are known to use Mo as a co-factor. The two most described molybdenum containing enzymes are nitrogenase and nitrate reductase.

Nitrogenase is involved in nitrogen fixation in root tubers of leguminoses by *Rhizobium* bacteria:

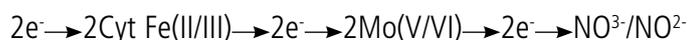


Molybdenum is directly involved in the reduction of  $N_2$ . The nitrogen molecule is bound to the molybdenum atom in the nitrogenase complex. Each nitrogen molecule is bound to two molybdenum atoms, which in turn are part of an iron-molybdenum protein. After activation of the nitrogenase complex using ATP, the iron-molybdenum complex changes its structure. Due to this conformational change, reduction of  $N_2$  occurs. The electrons required for this reduction by the iron-molybdenum protein are supplied by an iron-sulphur protein of the nitrogenase complex.

## Nitrate reductase

Nitrate reductase reduces nitrate into nitrite in the nitrogen assimilation process of the plant cell (for further information see the paragraph about nitrogen).

Nitrate reductase contains a heme-iron molecule and two molybdenum atoms. The enzyme catalyzes the reduction of nitrate in nitrite as follows:



FAD, cytochromes (Fe(II)/Fe(III)) and molybdenum (Mo(V)/(VI)) are functional parts of the nitrate reductase complex and the electron transport chain. Electrons derived from NADPH are used to reduce nitrate to nitrite. The activity of nitrate reductase is strongly reduced during molybdenum deficiency, but can be restored quickly by adding molybdenum.

## ZINC (Zn)

Zinc is taken up by the root system as  $Zn^{2+}$ . It is transported in the xylem as a free  $Zn^{2+}$  ion or as zinc-salt of an organic acid. Zinc is neither oxidized nor reduced in the plant. The element easily forms a tetrahedral complex and is in this way the metal component of a number of enzymes. It can be the structural as well as the regulatory cofactor of the enzyme complex.

## Enzymes

The plant has a number of zinc containing enzymes, including alcohol dehydrogenase in the meristem zone of the plant.

In Super Oxide Dismutase (SOD) Zn is complexed with Cu by means of a nitrogen atom from histidine (see copper for more information about SOD). The enzyme carbonic anhydrase binds  $CO_2$ , according to the following equation:



This reaction makes it possible for the plant to reversibly store  $CO_2$  as  $HCO_3^-$ . After conversion into  $CO_2$ ,  $HCO_3^-$  can be used as substrate for Ribulose Biphosphate Carboxylase. This enzyme consists of six subunits to each of which a zinc atom is attached and can be found in the chloroplast and in the cytoplasm.

## Protein synthesis

Zinc is very important for protein synthesis. A shortage of zinc results in considerable reduction of protein synthesis. Desintegration of ribosomes and accumulation of protein precursors, like amino-acids and amides, might occur.

Zn is essential for the activity of RNA polymerase. Under normal conditions, RNA polymerase contains two Zn atoms that determine the



Cactaceae: *Epithelantha micromeris*,  
Succulent Tissue Culture, The Netherlands

proper structure of the enzyme. Furthermore, an inversely proportional relation between the Zn concentration and the activity of RNAses exists. A low zinc concentration results in increased RNase activity.

## IAA synthesis

A shortage of zinc also disturbs the synthesis of Indol Acetic Acid by the plant.



Zinc plays a role in the synthesis of thryptophan, a precursor of IAA. For example, zinc deficiency in maize can be compensated by addition of tryptophan.

# MACRO ELEMENTS

Calcium, Phosphor, Potassium, Magnesium, Nitrogen, Sulfur

## CALCIUM (Ca)

In contrast to the other macro-nutrients, calcium is largely bound to the cell wall and cell membrane. This unique distribution is caused by the large number of  $\text{Ca}^{2+}$  binding places on the cell wall and the limited mobility of calcium through the membrane into the cytoplasm. Between two cell walls  $\text{Ca}^{2+}$  mainly binds to R-COO groups of polygalacturonic acids under formation of pectates. In apple, 90% of the total amount of calcium in the cell can be stored as pectate. The high concentration of calcium in the cell wall and cell membrane mainly serves to strengthen the cell wall and the regulation of the cell membrane structure. Transport of  $\text{Ca}^{2+}$  through the phloem as well as that from cell to cell is very limited.



### Cell wall

Pectin is broken down by the enzyme polygalacturonase. However, calcium strongly inhibits the activity of polygalacturonase. A high enzyme activity is observed in absence of calcium, causing degradation of the cell wall. The result is a softening of the plant tissue. If sufficient calcium is available, most pectin will be in the form of calcium pectate. In this way, the cell wall is highly resistant to the destructive activity of polygalacturonase. The presence of  $\text{Ca}^{2+}$  is also important for the resistance against fungal infections.

### Cell membrane

The stability of the cell membrane is highly influenced by  $\text{Ca}^{2+}$ . A shortage of  $\text{Ca}^{2+}$  results in an increased leakage of low-molecular compounds out of the membrane. A severe  $\text{Ca}^{2+}$  deficiency causes total disintegration of the membrane.  $\text{Ca}^{2+}$  stabilizes the membrane by interactions with phosphates, carboxylate groups of phospholipids, and proteins present in the membrane.

### Enzymes

Contrary to Magnesium, which is involved in the activation of many enzymes, calcium activates only a few enzymes like  $\alpha$ -amylase and ATPases. Calcium mainly stimulates membrane bound enzymes of which the activity is regulated by the structure of the membrane. However,  $\text{Ca}^{2+}$  also inhibits some cytoplasmatic enzymes.

The calcium binding protein calmodulin is important for the regulation of many enzymes in human and animal cells. Increasing evidence exists that this protein plays a role in the regulation of intracellular  $\text{Ca}^{2+}$  and enzymes in plants as well. Calmodulin in the cell is able to activate enzymes like phospholipases by forming  $\text{Ca}^{2+}$ -calmodulin complexes with these enzymes. Furthermore, it is assumed that calmodulin plays a role in the transport of  $\text{Ca}^{2+}$  to vacuoles.

### Location

Free  $\text{Ca}^{2+}$  is present in the cell in very low concentrations, approximately  $1 \mu\text{M}$ . This small amount prevents precipitation of  $\text{P}_i$ . Due to the low calcium level in the cell, competition with  $\text{Mg}^{2+}$  for cation binding sites is prevented, and inactivation or uncontrolled activation of enzymes is avoided. The cell membrane is a good barrier against influx of  $\text{Ca}^{2+}$  and since  $\text{Ca}^{2+}$  efflux is easy, a low intracellular calcium concentration is guaranteed.

Especially in leaf cells with vacuoles, a large amount of bound calcium is present. Calcium is necessary for the cation-anion balance by counteracting organic and inorganic anions. Most  $\text{Ca}^{2+}$  is bound to oxalate. Although in this form it is poorly soluble, it keeps the calcium concentration in cytoplasm and chloroplasts low. Calcium oxalate also has a function in the osmoregulation of the cell.

Calcium is important in cell and root multiplication. Furthermore, development of the pollen tube is  $\text{Ca}^{2+}$  dependent and is chemotrophically led by extracellular calcium. IAA is involved in the transport of calcium. Auxin inhibitors like TIBA, inhibit the  $\text{Ca}^{2+}$  distribution in the plant causing the appearance of calcium deficiency features.

## PHOSPHORUS (P)

Phosphorus is taken up as  $\text{H}_2\text{PO}_4^{2-}$  by the roots of the plant and is, contrary to nitrate and sulphate, not reduced. It can be present in the plant as inorganic phosphate (Pi) or esterified via an OH group to a C atom (C-O-P). The highly energetic pyrophosphate bond of phosphorus when bound to another P atom, as in ATP, is very important for the energy metabolism of the cell.

### Nucleic acids

Phosphorus is an essential element in DNA and RNA to connect the individual ribonucleic acids to form macro molecules.

### Phospholipids

Phospholipids in biomembranes also contain a large amount of phosphorus. In these phospholipids phosphorus makes, via a phosphate-ester bond, a connection between a diglyceride and an amino acid, amine or alcohol. Phospholipids consist of a hydrophobic tail, the diglyceride, and a hydrophilic head containing  $\text{PO}_4$ . Both have an important function in the stabilization of membranes. Membranes consist of two monolayers of phospholipids, together referred to as a lipid bilayer.

The hydrophilic parts of the phospholipids point outward towards the water, while the hydrophobic ends are orientated toward the inside of the membrane and interact with each other.

### Energy metabolism

Phosphorus is very important for the energy metabolism of the plant in forming energy rich phosphate esters (C-P), like in glucose-6-phosphate. These phosphate esters are important for the metabolism and the biosynthesis of the plant.

More important in the energy metabolism of the cell is the highly energetic pyrophosphate bond between two P atoms ( $\text{P}<\text{P}$ , 30 kJ), as in AdenosineTriPhosphate (ATP). The energy released during the glycolysis, oxidative phosphorylation or photosynthesis is used to synthesize ATP

and this energy is liberated during the hydrolysis of ATP in ADP and Pi. ATP is unstable and therefore has a high turnover. A single gram actively metabolizing root tips of maize can synthesize 5 gram ATP each day with an average turnover time of 30 seconds.

### Phosphate pool

Cells with vacuoles contain two different phosphate fractions. The metabolic pool, mainly in the form of phosphate esters, is present in cytoplasm and mitochondria. The non-metabolic pool, mainly in the form of Pi, is present in the vacuole. If phosphorus is sufficiently available, 85 to 95% of the total amount of Pi will be localized in the vacuoles. If the phosphorus supply to the plant is stopped, the Pi concentration in the vacuole immediately reduces, while reduction in the metabolic pool occurs much slower.

A sufficient supply of phosphorus results in an increase in all the phosphorus-containing organelles in the cell. However, above a certain level only Pi in the vacuoles increases. Therefore, an overdose P is stored as Pi.

### Enzymes

Pi also has a strong regulatory function in many metabolic processes in the plant. Therefore, compartmentation of phosphorus is essential for a good regulation of the metabolism of the cell.

In tomatoes, Pi, released from the vacuoles in the cytoplasm, stimulates the phosphofructokinase activity. This enzyme is important in the substrate influx in the glycolysis and induces an increase in cell respiration during ripening. At the same time, a shortage of phosphorus can cause a delay in the ripening process of tomatoes.

Phosphorus is also important in the regulation of starch production in chloroplasts. Only a low Pi concentration already causes inhibition of the synthesis of starch. ADP-glucose-pyrophosphorylase, the most important enzyme in the synthesis of starch, is inhibited by Pi and stimulated by triosephosphates. Consequently, the balance between both phosphorus containing compounds is very important in the regulation of starch synthesis in the chloroplast.

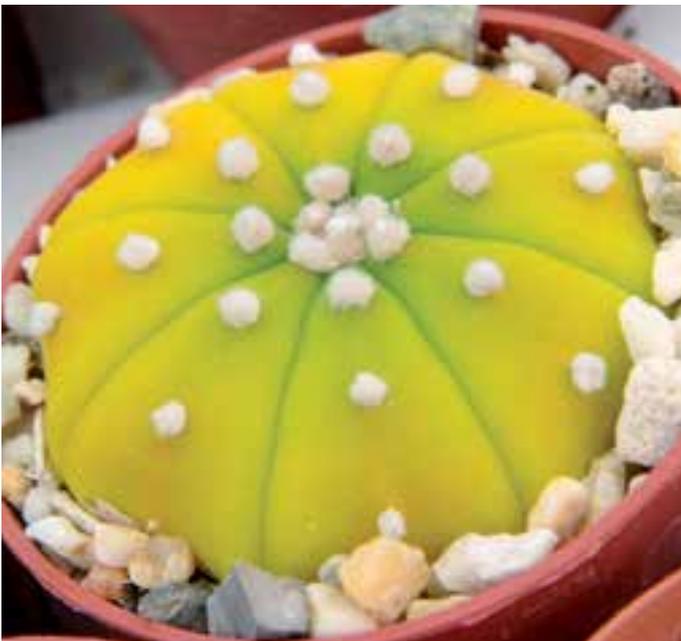
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Pi regulates starch synthesis in the chloroplast in another way as well. A phosphate carrier in the membrane brings Pi inside the cell and the triosephosphates outside. In this way, the Pi concentration in the chloroplast increases and that of the triosephosphates decreases. This, in turn, influences starch synthesis in the chloroplast, which is regulated by the mechanism described above.

Ribulose biphosphate (RuBP) is important in the carbon fixation as acceptor of CO<sub>2</sub>. Triosephosphates are required for the regeneration of RuBP. A high concentration Pi stimulates the export of these compounds out of the chloroplasts inducing a shortage of triosephosphates and thereby inhibiting CO<sub>2</sub> fixation. Phosphorus is important in the regulation of many other enzymes as well.

For optimal growth, 0.3 to 0.5 gram phosphorus per gram dry weight is required. Phosphorus deficiency results in delayed growth and a darkgreen color of the leaves. This is because during a shortage of phosphorus, leaf development is slower than chlorophyll synthesis, resulting in higher chlorophyll concentrations in the leaves.



Cactaceae: *Astrophytum asterias*,  
Succulent Tissue Culture, The Netherlands

## POTASSIUM (K)

Potassium is a monovalent cation with a high mobility in the plant, both at the cellular level as in the transport over longer distances in the xylem and phloem. Of all elements, the potassium ion is present in the highest concentration, in the cytoplasm between 100 and 200 mM and in the chloroplasts between 20 and 200 mM. Potassium salts have an important function in the osmotic regulation of the cell. In cell extension and other processes regulated by the turgor, the K<sup>+</sup> ion serves as counteracting ion for soluble (in)organic ions and to maintain a pH between 7 and 8, the ideal acidity for most enzymes. The osmotic pressure of the cytoplasm is also mainly regulated by the potassium ion.

## Enzymes

K<sup>+</sup> is essential for the activation of many enzymes. More than 50 enzymes in the plant depend on, or are stimulated by potassium. The binding of K<sup>+</sup> induces conformational changes in the structure of many enzymes, thereby increasing the V<sub>max</sub> and substrate affinity. During a K<sup>+</sup> shortage, an increase in the concentration of soluble carbohydrates and nitrogen containing compounds together with a decrease in the concentration of starch in the plant is observed.

This change in the carbohydrate metabolism is due to the strong need of K<sup>+</sup> for some regulatory enzymes in the carbon metabolism. K<sup>+</sup> is important in the activation of membrane bound ATPases. At first, these enzymes are activated by magnesium, but they need further stimulation by potassium ions. In higher plants, K<sup>+</sup> is needed for protein synthesis. K<sup>+</sup> is probably required for the translation and binding of tRNA to the ribosomes. The synthesis of Ribulose Biphosphate Carboxylase is also strongly dependent on the K<sup>+</sup> concentration. The ion is important for both the activation and synthesis of the nitrate reductase.

The role of K<sup>+</sup> in the photosynthesis is, besides the activation of many enzymes, to regulate the ion balance and pH of the chloroplasts. K<sup>+</sup> is the most important counteracting ion for the light induced H<sup>+</sup> flux over the thylakoid membrane. The ion is also involved in the induction of a transmembrane pH gradient, necessary for the synthesis of ATP. An increase in the K<sup>+</sup> concentration is related to an increase of the photosynthesis, the respiration and the Ribulose Biphosphate Carboxylase activity.

## Cell extension

The development of a large central vacuole in the cell is an important process in the cell extension. To create this vacuole, first a sufficient enlargement of the cell wall should be possible. Secondly, the osmotic potential of the vacuole has to increase. This can be achieved by accumulation of K<sup>+</sup>, causing a strong increase in the volume of the vacuole because of osmosis. GA<sub>3</sub> and K<sup>+</sup> apparently work synergistically in increasing the stalk length.

## Ion balance

K<sup>+</sup> is important in the maintenance of the ion balance. It neutralizes non-mobile anions in the cytoplasm and many mobile anions in xylem, phloem and vacuoles. In the nitrate metabolism K<sup>+</sup> functions mostly as counterion for NO<sub>3</sub><sup>-</sup> in the transport over longer distances in the xylem and for the storage in vacuoles.

For nitrate reduction in the leaves the remaining K<sup>+</sup> should be used for stoichiometric synthesis of organic acids to neutralize the K<sup>+</sup> ions. Potassium salts of organic acids, e.g. potassium malate, are transported to the roots. Then, the potassium ion can serve either as counterion of the nitrate present in the root cells or for the transport of nitrate through the xylem.

## MAGNESIUM (Mg)

Mg<sup>2+</sup> ions are very mobile and able to form a complex with strong nucleophilic ligands like phosphoryl groups. Magnesium is essential for many enzymatic reactions in providing the correct stereometric structure between enzyme and substrate. Magnesium is very important for the photosynthesis. Most Mg<sup>2+</sup> ions present are involved in the regulation of the intracellular pH and right cation-anion balance.

## Photosynthesis

Magnesium is the central atom in chlorophyll molecules of photosystem I and II, which are parts of the photosynthesis. In chlorophyll, absorbed photons cause an electron current thereby generating ATP and NADPH and resulting in fixation of CO<sub>2</sub>. If magnesium is optimally available, 10 to 20% of the Mg<sup>2+</sup> ions in the leaves will be localized in the chloroplasts. High concentrations Mg<sup>2+</sup> and K<sup>+</sup> ions in the chloroplast are necessary to maintain a pH between 6.5 and 7.5 in chloroplast and cytoplasm. This is in contrast to a pH between 5.0 and 6.0 in the vacuoles of the cell. The pH determines the structure of proteins and enzymes to a great extent and therefore has influence on the function of chloroplasts and on protein synthesis.



Echeveria, Succulent Tissue Culture, The Netherlands

## Enzymes

Magnesium is essential for the tertiary structure of many enzyme-substrate complexes, because it creates the proper stereometric conformation between enzyme and substrate.

In protein synthesis, Mg<sup>2+</sup> is involved at different levels. Magnesium forms a bridge between both ribosome subunits. In magnesium deficiency, the subunits will dissociate and protein synthesis stagnates. Magnesium is required for the activity of RNA polymerases, enzymes involved in the synthesis of RNA. A shortage of Mg<sup>2+</sup> will block RNA synthesis. In the leaves, 25% of the total proteins is localized in chloroplasts. Consequently, if insufficient magnesium is present the structure and function of the chloroplasts will be immediately affected.

Magnesium is also important for Ribulose Biphosphate Carboxylase activity. This CO<sub>2</sub> binding enzyme is highly pH and Mg<sup>2+</sup> dependent. Binding of magnesium to the enzyme increases the substrate affinity for CO<sub>2</sub> and the V<sub>max</sub>.

## Energy metabolism

Magnesium is indispensable for the energy metabolism of the plant because of its importance in the synthesis of ATP (ADP + Pi → ATP). The element builds a bridge between the enzyme and ADP. Especially the synthesis of ATP in the chloroplast is strongly stimulated by magnesium. Furthermore, magnesium is able to form a complex with ATP. ATPases, in their turn, transfer the highly energetic phosphoryl group to a protein or a sugar.

Even though magnesium has many regulatory functions, most of the time magnesium is stored in the vacuoles to serve as counterion for inorganic and organic anions in the cation-anion balance.

## NITROGEN (N), NITRATE (NO<sub>3</sub><sup>-</sup>) AND AMMONIUM (NH<sub>4</sub><sup>+</sup>)

The major component of almost all media is inorganic nitrogen in the form of nitrate or ammonium. The salts that are mostly used are potassium nitrate (KNO<sub>3</sub>), ammonium nitrate (NH<sub>4</sub>NO<sub>3</sub>) and calcium nitrate (Ca(NO<sub>3</sub>)<sub>2</sub>·4H<sub>2</sub>O). These compounds provide the plant with inorganic nitrogen to synthesize complex organic molecules.

Ammonium is mainly stored in the roots as organic nitrogen. Nitrate can be transported via the xylem to other parts of the plant, where it participates in the nitrogen assimilation. Nitrate can be stored in the vacuoles of the cell and fulfill an important function in the osmoregulation and anion-cation balance of the plant.

## Nitrate reductase

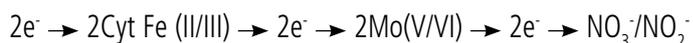
Nitrate cannot simply be used to synthesize organic molecules but has to be reduced to ammonia first. It is reduced according to the following reaction:



This reaction is executed in two steps by the enzymes nitrate- and nitrite reductase.

First, nitrate is converted into nitrite by nitrate reductase.

Secondly, nitrite is reduced into ammonia by nitrite reductase. The conversion of nitrate into nitrite occurs in the cytoplasm according to:



Nitrate reductase consists of FAD, cytochromes (Fe(II)/Fe(III)) and molybdenum (Mo(V)/(VI)). These components form integral parts of the electron transport chain through which electrons derived from NADPH are supplied to reduce nitrate to nitrite. During molybdenum deficiency, the activity of nitrate reductase significantly decreases. In most plants, nitrate reduction can occur in both leaves and shoots.

To which extent reduction can take place, strongly depends on factors like plant species, age of the plant and the presence of nitrate. Particularly woody species have a high nitrate reducing capacity. In low nitrate

concentrations, most is reduced in the roots. Conversely, if high nitrate concentrations are available, it is also reduced in the leaves. The complementary cation of nitrate is important for its uptake. If  $K^+$  is the cation, nitrate reductase activity in the roots is low and nitrate will be transported to the shoots of the plant. With  $Ca^{2+}$  as a cation, nitrate reductase activity of the roots is higher.

## Nitrite reductase

The reduction of  $NO_2^-$  to  $NH_3$  by nitrite reductase is carried out in the leaves. Reduced ferredoxin supplies the electrons for the reduction of nitrite. Ferredoxin, reduced by electrons generated in photosystem I, supplies the electrons for the reduction of nitrite.

## Reduced nitrogen containing compounds

Ammonium and ammonia ( $NH_3 \rightarrow NH_4^+ + OH^-$ ) are toxic for plants, even in low concentrations. Therefore, they should be converted quickly into nontoxic low-molecular nitrogen containing compounds like glutamine, asparagine, arginine, allantoin and betain. Glutamine synthetase and glutamate synthase, both present in roots and shoots, are key enzymes in the conversion of ammonium (see also, phosphinothricin *P 0159*).

Besides detoxification of ammonia and ammonium, low-molecular nitrogen compounds have several other functions. The most important function is the supply of organically bound N and  $NH_2$ , which is taken up by the plant as inorganic nitrogen, for the synthesis of amino acids and proteins. The low-molecular-weight compounds are also used as carrier of some cations, e.g. manganese and copper, over long distances in the plant.

Furthermore, these small nitrogen containing molecules serve as a storage place for an excess of nitrogen. Contrary to humans and animals, plants are not able to excrete organically bound nitrogen, as urea for example, but this mechanism enables them to store an excess of nitrogen.

## SULFUR (S)

Sulfur is taken up as  $SO_4^{2-}$  in the roots of the plant at a relatively low speed. Like nitrate, sulphate has to be reduced first before it can be used for the synthesis of reduced sulfur containing compounds like amino acids, proteins and enzymes. In the nonreduced form sulfur is incorporated in sulpholipids and polysaccharides.

## Sulfur assimilation

The first step in the sulfur assimilation is activation of  $SO_4^{2-}$  by the enzyme ATP sulfurylase, under use of ATP. This reaction yields adenosine phosphosulphate (APS) and pyrophosphate (Pi). Then, two different chemical routes can be followed. In one route, sulfur is not reduced but incorporated in polysaccharides present in sulpholipids. In another route, sulfur is reduced to a -SH group (thiol group) and the sulfuryl group of APS is transferred to glutathione (Glut-SH). Subsequently, the -SH group is transferred to acetylserine and broken down into acetate and cysteine. Cysteine is the first stable product in the assimilatory reduction and the precursor of all organic compounds in plants that contain reduced sulfur, like proteins, coenzymes, secondary metabolites etc. Sulfur assimilation mainly takes place in the chloroplast. During sulfur deficiency, protein synthesis is inhibited and the amount of chlorophyll in the leaves decreases.



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## Proteins

In proteins sulfur is present in cysteine and methionine. Both amino acids are precursors of all reduced sulfur-containing compounds in the plant. Sulfur has, as constituent of several coenzymes and prosthetic groups, an important function in various redox reactions, according to:



R can be a cysteine residue, but also the tripeptide glutathione. Glutathione is soluble in water, and therefore important as redox system in the chloroplast and cytosol of plants. Sulfur bridges between two cysteine residues are very important for the tertiary structure of proteins and the activity of enzymes. -SH groups, in APS sulphotransferase mentioned above and in coenzyme A (Krebs cycle) forms part of the functional group of the enzyme.

## Metallothioneins

Low-molecular sulfur containing compounds, the metallothioneins, are frequently found in plants. Most of these compounds contain cysteine. Especially metals like copper, cadmium and zinc are bound by metallothioneins. Most probably, these small proteins are involved in the elimination of an excess of these metals, before they are irreversibly bound to functional SH groups of enzymes.

## Nonreduced sulfur

In the nonreduced form sulfur is a component of sulpholipids, which form a structural constituent of membranes. Sulfur is present as a sulphate ester of sulphate and a C<sub>6</sub> sugar, for example glucose. Sulpholipids are abundantly present in thylakoid membranes of chloroplasts. Sulpholipids probably play a role in the transport of ions across other membranes as well. Further, the presence of sulpholipids in the membrane is positively related to the salt tolerance of plants.

The characteristic odor of species like onions and garlic is mainly due to the presence of volatile sulfur containing compounds.



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# PLANT HORMONES

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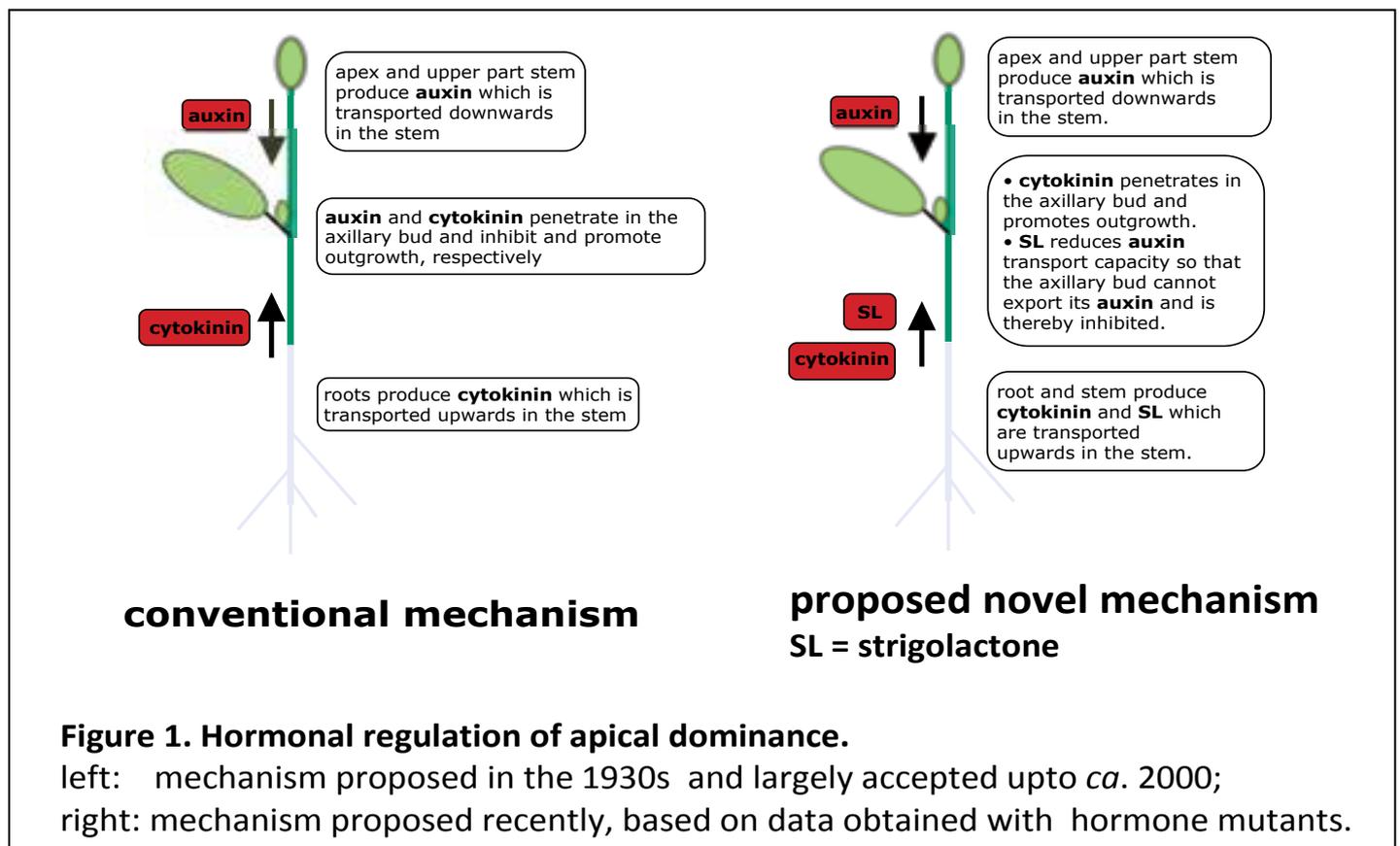
The ingredients of plant tissue culture media include plant hormones, inorganic nutrients, organic nutrients and vitamins. Plant hormones are added to regulate growth. In tissue culture, they are mainly used to stimulate adventitious regeneration of roots, shoots and embryos, outgrowth of axillary buds, and formation of callus. Moreover, cytokinin and auxin are often required to achieve quantitative growth (increase of cell number and volume). In tissue culture, usually only cytokinin and auxin are added. Plant hormones are typically added within the range 0.1–10  $\mu\text{M}$  (0.02–2 mg.l<sup>-1</sup>). A major part of the research efforts in plant tissue culture concern modification of the concentrations and types of plant hormones. The dose-response curves of plant hormones are generally bell-shaped. At a too low concentration there is no effect, and at a too high concentration the added hormone is inhibitory. The promotive effect only occurs at intermediate concentrations. To detect these concentrations, usually first a broad range is taken (0, 0.1, 1, 10, 100  $\mu\text{M}$ ), and after that a narrow one. It should be remembered that hormones act in a logarithmic way.

## General backgrounds

In animal physiology, hormones denote substances that are synthesized in very low amounts in one part of an organism and are transported to target tissues in other parts where they exert an effect. In plants, such chemical messengers have also been found. A classical example occurs in germinating barley seeds: gibberellin synthesized and released by the embryo diffuses into the aleurone layer where it induces synthesis and secretion of hydrolytic enzymes. These enzymes degrade macromolecular reserves to small fragments that are used by the embryo for initial growth. Another

notable example is the inhibition of the outgrowth of axillary buds by auxin synthesized in the apex and transported downwards in the stem (Fig. 1). In contrast to animal hormones, though, the synthesis of a plant hormone is often not restricted to a specific tissue, but may occur in many different tissues. Furthermore, plant hormones may be transported to distant tissues, but often they act at the site of synthesis. Another property of plant hormones is their lack of specificity: each influences a wide range of processes. Auxin, for example, has been found to promote cell elongation, cell division, formation of primary vascular tissue, adventitious root formation, senescence, fruit growth, outgrowth of axillary buds and sex expression. Because of the differences between animal and plant hormones, many researchers deny that the latter are genuine hormones and prefer to use phrases like 'plant growth substance' or 'plant growth regulator'. Nevertheless, the term 'plant hormone' is widely used.

In animals, hormones are to distant target tissues via the cardiovascular system. In plants growing *ex vitro*, almost all long-distance transport occurs via water flow in xylem and phloem (the notable exception is polar auxin transport). In this context, it should be noted that long distance transport via diffusion is very slow, taking ca. one week (!) for a distance of 2 cm (<http://4e.plantphys.net/article.php?ch=t&id=26>). Knowledge about water flow in vascular tissues of tissue-cultured plants is virtually absent. Flow in the xylem is most likely decimated by the lack of transpiration brought about by the very high humidity in the headspace. Applied plant hormones increase the hormone level within the target tissues, but how much depends on the rate of transport from the source. In addition, most increase is transient because plant hormones are rapidly inactivated after uptake. Inactivation can be permanent (by oxidation) or reversible (by conjugation to sugar or amino acid molecules). Ethylene is an exception but this gaseous compound can be rapidly released from the plant into the air. Usually very small amounts of the applied hormones remain in the free form. It has been shown for auxins, that an equilibrium exists between the free and the conjugated form, less than 1% being present in the free form.



**Table 1. Main auxins and cytokinins.**

auxins	indole-3-acetic acid - IAA	cytokinins	zeatin - Z
	indole-3-butyric acid - IBA		zeatinriboside - ZR
	1-naphthaleneacetic acid - NAA		isopentenyladenine - iP
	phenylacetic acid - PAA		6-bezylaminopurine - BAP
	2,4-dichlorophenoxyacetic acid - 2,4-D		6-furfurylaminopurine - kinetin
	2,4,5-trichlorophenoxyacetic acid - 2,4,5-T		N <sup>6</sup> -( <i>meta</i> -hydroxybenzyl)adenine - topolin
	picloram		thidiazuron - TDZ
	dicamba		forchlorfenuron - CPPU or 4PU-30
	p-chlorophenoxyacetic acid - CPA		

The effect of hormones depends also on the stability in the medium and in the tissue, and on the sensitivity of the target tissue: Cells in a certain tissue or at a certain developmental stage may not recognize the hormonal signal, or they may be incapable of carrying out the desirable response. Applied hormones influence synthesis and degradation of endogenous hormones belonging to the same class as the applied hormone or to other classes. A notable example is the induction of ethylene synthesis by auxin. All this results in a very complex situation and it is often difficult to discover how the observed effect has been brought about.

Most knowledge about the role of plant hormones originates from studies in which hormones have been applied to plant tissues. Instead of the hormones themselves, compounds that affect their metabolism, transport or action may be added. Experimentation *in vitro* has many advantages: tissue culture facilitates application of hormones via the cut surfaces of the explants, avoids microbial degradation of applied hormones and allows to study of the effect of hormones on isolated plant organs. At the same time, effects of the specific tissue culture conditions should be kept in mind. Recently, a vast amount of insight has been obtained from hormone mutants, in particular in *Arabidopsis*. Researchers also use plants transformed with cytokinin or auxin biosynthetic genes from *Agrobacterium tumefaciens* or with *rol*-genes from *A. rhizogenes* (the latter influence among others the signal transduction pathway).

## Auxins

Naturally occurring auxins include: IAA, IBA, 4-Cl-IAA, PAA and conjugates of these auxins. In addition, many chemical analogues have been synthesized: NAA, 2,4-D, 2,4,5-T, dicamba and 4-CPA (Table 1). Auxins were discovered in the 1920s by the Dutch plant physiologist F.W. Went. He observed that auxins produced in the tip of an *Avena* coleoptile influence the curvature of the coleoptile just below the tip. Shortly after, the root-inducing capability of IAA was discovered, the role of auxin in inhibiting outgrowth of axillary buds was observed, and NAA and IBA were chemically synthesized.

## Effects of auxin

The major roles of auxin in tissue culture were established by Skoog and Miller in 1957. They observed that pith tissues excised from tobacco stems form shoots at high cytokinin and low auxin concentration, roots at low cytokinin and high auxin concentration, or callus at intermediate concentrations of both plant hormones. The formation of roots from pith fragments corresponds with the effect of auxin on rooting of cuttings, and the reduction of shoot formation with the inhibition of the outgrowth of

axillary buds by auxin. A few years after the classical Skoog and Miller experiment, the formation of somatic embryos was observed after treatment with 2,4-D.

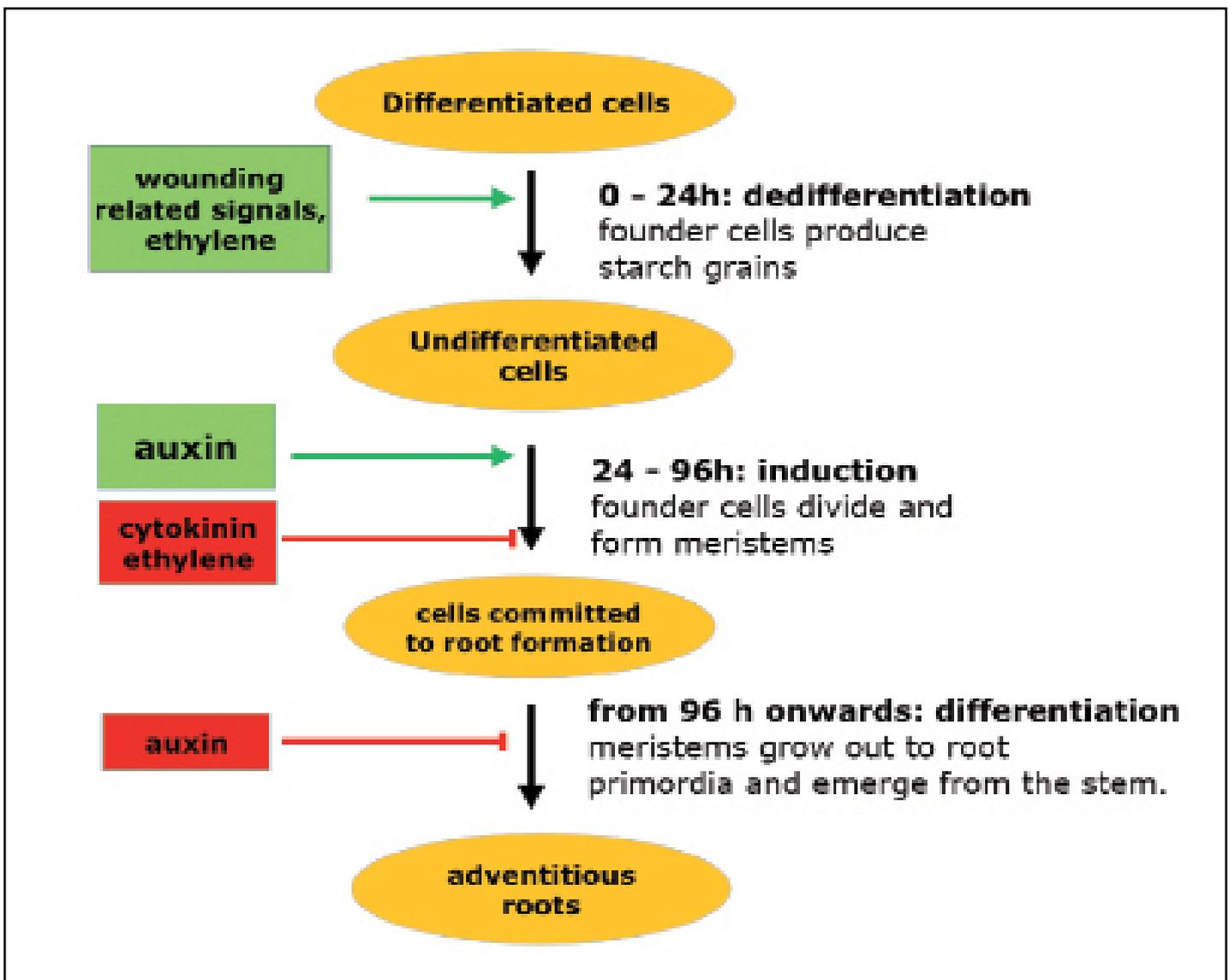
It should be noted that auxins are only required during the initial phases of adventitious root formation and somatic embryogenesis. After that, they become inhibitory and block the outgrowth of the root initials and embryos. Figure 2 shows the effect of various hormones in the successive stages of rooting of apple microcuttings. The effect of hormones is restricted both to a specific period of time during the development and to specific tissues/cells. The rhizogenic action of auxins in apple microcuttings is 24h – 96h after start of the rooting treatment and is restricted to specific cells near the interfascicular cambium adjacent to the vascular bundles.

2,4-D is often referred to as a strong auxin but this only applies to the formation of callus and somatic embryos: 2,4-D is a weak auxin with respect to the formation of adventitious root primordia or the inhibition of axillary buds. In contrast, IAA or IBA are not very effective in the formation of callus and somatic embryos, but show a high performance with respect to adventitious root formation and inhibition of axillary buds.

## Transport, uptake, and metabolism

In plants, auxin is synthesized predominantly in the apical region and transported downwards. The underlying mechanism of this transport has been examined extensively. Uptake of auxin into cells occurs by diffusion and by active uptake via an influx carrier termed AUX1. The rate of uptake via diffusion depends on the dissociation of the molecule. Auxin is more protonated outside the plasmalemma than inside the cell (in the cell wall the pH is ca. 5.5 but the cytoplasm has a pH of ca. 7; IAA is a weak acid with a pKa of 4.7). The undissociated lipophilic auxin diffuses through the plasmalemma into the cell. In the cytoplasm the anionic form prevails, so auxin cannot easily diffuse out through the plasmalemma and is 'trapped' within the cells. Auxin is actively transported out of the cells by efflux carriers, the PIN-proteins. Because the efflux carriers are located predominantly at the basal side of a cell, auxin is transported from cell to cell in a basipetal direction, i.e., from apical to basal regions. Inside the cells, auxin moves from the apical to the basal side by diffusion. The rate of auxin transport is ca. one cm.h<sup>-1</sup>. The active auxin transport occurs mainly in xylem parenchyma. Polarity itself is likely a major morphogenetic factor. In addition to directional transport, auxin can also move via water flow in the phloem.

When explants are cultured on medium with auxin, it is rapidly taken up probably via the same mechanism as described above (anion-trapping). This results in depletion of the medium. When plant tissues are cultured in



**Figure 2. Successive steps in adventitious root formation.** Similar schemes can be made for adventitious shoot formation and somatic embryogenesis, but of course the hormonal players and the durations are very different. Green indicates promotion, red inhibition.

liquid medium, most of the auxin may have disappeared from the medium within a few days. In solid medium only local exhaustion occurs because of the slowness of diffusion over large distances (see before). From the crucial medium components, auxin seems to be the only one that is so very rapidly depleted. The epidermis of plants is relatively impermeable to auxin and most uptake by explants occurs via the cut surface. How auxin reaches target tissues in the explant has not been studied. Roots are formed from founder cells close to the cut ends so auxin may reach these cells by diffusion.

Plant tissues inactivate auxins by conjugation or (enzymatic) oxidation. All auxins can be conjugated. It is believed that conjugated auxin is inactive. However, conjugation is reversible and the free, active form may be released. It has been suggested that in the plants an equilibrium exists between the free and conjugated forms. Experimental data show that 2,4-D is slower conjugated than IAA, IBA or NAA. IAA is rapidly oxidized by plant tissues, in particular by wounded tissues. IBA is also oxidized but slower. The various auxins have different chemical stabilities in the tissue culture medium. When exposed to light, IAA is very rapidly oxidized. MS-salts accelerate the rate of IAA oxidation. When using IAA, the rapid photooxidation of IAA should be kept in mind. IAA is also unstable during autoclaving, but bioassays and chemical determinations show a loss less than 20%. IBA is slower photooxidized than IAA, whereas other auxins,

e.g. NAA, are not or only very little photooxidized. Riboflavin may be added to medium to enhance photooxidation of IBA. The photooxidation of IAA and of IBA in the presence of riboflavin may be turned to advantage. For example, in adventitious root formation cultures with IAA may be left in the dark until the root meristemoids have been formed by the rhizogenic action of auxin (see Fig. 2). After that, when auxins have become inhibitory, the cultures are transferred to the light to degrade the auxin. It should be noted that for the choice of auxin, chemical stability is only one of the factors to consider. The efficiency with respect to the developmental process that should be promoted, is another major factor. The endogenous level of auxin and auxin action can be manipulated in various ways. In plant tissues, auxin is actively transported in a polar way (see above). TIBA (triiodobenzoic acid) and NPA (N-1-naphthylphthalamic acid) block this transport, because these compounds bind to the efflux carrier. The endogenous level of auxin can be increased by transforming plants with the auxin biosynthetic genes of *Agrobacterium tumefaciens*. The transformed plants show expected changes in their phenotype. Phenolic compounds (e.g., ferulic acid or phloroglucinol) may inhibit oxidation of applied auxin. This is not specific inhibition of enzymatic oxidation, photooxidation is also inhibited by adding phenolic compounds to the medium. PCIB is a genuine anti-auxin and competes with auxin for the auxin binding site at the auxin receptor.

## Cytokinins

Cytokinins are a complex class of plant hormones. The naturally occurring cytokinins include Z, iP, and DHZ and their ribosides ZR, iPA and DHZR (Table 2). In addition, conjugated (non-active) and phosphorylated (active) cytokinins have been isolated from plant tissues. For a long time, BAP has been considered to be a synthetic cytokinin, but has been recently shown a naturally occurring one. In addition to these cytokinins that are all of the purine-type, nonpurine cytokinins have been reported such as thidiazuron (TDZ) and CPPU (4-PU-30). These compounds have a very high cytokinin activity and are particularly successful in woody plants. TDZ is used commercially as a cotton defoliant. In this case, it acts by inducing ethylene synthesis. Meta-topolin is a highly active aromatic cytokinin that was first isolated from *Populus*. In tissue culture, BAP and the synthetic cytokinins kinetin and TDZ are most frequently used.

## Effects of cytokinins

The discovery of cytokinins is closely linked to tissue culture. In the starting period of plant tissue culture, it was observed that malt, coconut and yeast extracts promote both the growth and initiation of buds *in vitro*. Because these preparations all contain purines, nucleic acids were tested. It was observed that autoclaving of nucleic acids strongly enhanced their effect. The active compound formed by autoclaving appeared to be kinetin, a hitherto unknown purine. In 1964, Letham isolated zeatin from immature corn.

Cytokinins promote cell division, but they likely influence another step in the cell cycle than auxins. Thus, addition of cytokinins is usually required to obtain callus growth. In micropropagation, cytokinins are applied to promote axillary branching. High concentrations of cytokinin lead to extreme bushiness. This may result in undesirable bushiness long after transfer of micropropagated plantlets to soil. Transformation of plants with the cytokinin biosynthetic gene of *A. tumefaciens* may result in plants with reduced apical dominance. Other applications of cytokinin in tissue culture are promotion of adventitious shoot formation, prevention of senescence, reversion of the deteriorating effect of auxin on shoots, and, occasionally, inhibition of excessive root formation (e.g., in germinating somatic embryos). Cytokinins inhibit root formation and are therefore omitted from rooting media. Cytokinins may have other undesirable side-effects such as hyperhydricity and loss of the chimeric structure.

## Transport, uptake and metabolism

Roots are considered as the main site of cytokinin synthesis and cytokinin is transported to the shoot via the water flow in the xylem. Xylem exudates contain high levels of cytokinins. Recently, evidence has been found for active transport via carriers.

When plant tissues are cultured on medium with cytokinins, they are rapidly taken up, although at a much smaller rate than auxin (3 to 10 times slower). It is not known how cytokinins reach target tissues like axillary buds (to break apical dominance) and leaves (to reduce senescence) which both are at relatively large distance from the source but probably cytokinins are transported via water flow in the vascular tissues. Z, ZR, iP and iPA are conjugated and/or oxidized by plant tissues. Oxidation involves oxidative side chain cleavage. DHZ, DHZR and BAP are conjugated, but not oxidized. Cytokinins can be N-glucosylated on the purine ring or O-glucosylated on the N6-substituted side-chain. The N-glucosides are biologically inactive and stable. The O-glucosides, that are formed from Z and DHZ may have a storage function. Just as with other plant hormones, after uptake only a very small percentage of cytokinin remains in the free form. TDZ is an exception and is conjugated only at a very low rate: after long periods (12 to 33 days) of culture of *Phaseolus* callus on medium with radioactive labelled TDZ, 60% of the TDZ taken up from the medium was in the free, nonconjugated form. BAP is a chemically stable cytokinin in tissue culture medium, whereas most other purine-type cytokinins are

considered to be to some extent chemically unstable. The nonpurine type cytokinins CPPU and TDZ are chemically stable.

Compounds that influence cytokinin oxidation (phenolic compounds), conjugation and action, have been studied occasionally. They have hardly been used in tissue culture. The synthesis of cytokinins is inhibited by lovastatin or simvastatin. In human medicine statins are used to lower cholesterol.

## Ethylene

In contrast to other hormones, ethylene is a gas and a very 'simple' molecule (Fig. 3). The synthesis of ethylene increases during senescence and ripening. In tissue culture systems, wounding and auxins increase ethylene synthesis. Ethylene promotes senescence of flowers and leaves, and ripening of fruits. Because of promotion of senescence, ethylene is usually undesirable in tissue culture. Ethylene may accumulate in the headspace of tissue-culture containers when they are too tightly closed and this accumulation may be detrimental to the plant tissues. Ethylene may also accumulate in submerged tissues because of the low diffusion of gases in water (10,000 times lower than in air!). Apart from ethylene, other (toxic) gasses may also accumulate. In adventitious regeneration, ethylene may enhance the sensitivity to organogenic stimuli.

There are various ways to reduce the effect of ethylene produced by the plant. Ethylene may be removed from air by a KMnO<sub>4</sub> solution, and by purafil or power-pellets (trade names), bead-like porous material coated with KMnO<sub>4</sub>. The synthesis of ethylene is inhibited by AVG. This compound blocks the synthesis of ACC. STS blocks the action of ethylene. Often AgNO<sub>3</sub> is used, but this compound is not well transported in plants whereas STS is. Addition of ethylene as a gas is inconvenient and therefore ethephon (ethrel, 2-chloroethylphosphonic acid) is usually added. This compound is stable at pH 4 or less but decomposes at higher pH to produce ethylene. So, when ethrel diffuses in the cell, it will release ethylene in the cytoplasm (the pH of the cytoplasm is ca. 7). In plants, the ethylene-precursor ACC is transported over long distance in the xylem. The conversion of ACC to ethylene by ACC-oxidase is not a rate-limiting step in ethylene synthesis and usually applied ACC is rapidly converted to ethylene. Therefore, addition of ACC to tissue-culture medium is a convenient method to increase ethylene levels in plant tissues. Ethylene may be metabolized by plants but this unlikely plays a major role in regulating ethylene action.

## Abscisic acid, strigolactone and gibberellins

ABA has been isolated from plants thirty years ago. It plays a role in dormancy development in embryos, buds and bulbs, and in leaf abscission. When present in tissue culture media, ABA inhibits growth of shoots and germination of embryos. Another major effect of ABA is closure of stomata. In line with this, ABA has been found to accumulate under drought stress. When taken up, ABA is just like other hormones conjugated. In addition, it is irreversibly metabolized to phaseic acid. ABA-synthesis may be inhibited by fluridone. As this inhibitor acts by blocking one of the steps in the synthesis of carotenoids, the tissues bleach. Thus, tissues formed in the presence of fluridone have low ABA levels and are white. Fluridone may be used in tissue culture to prevent plants from entering dormancy. Just as ABA, strigolactones are carotenoid-derived. They trigger germination of parasitic plant seeds, for example of striga (witchweed, family Orobanchaceae) from which they gained their name. In plants strigolactones have been recently implicated in inhibition of shoot branching. This is an essential correction of the traditional theory (Fig. 1) and may have a major impact on propagation via axillary branching. In tissue culture, gibberellins are only used incidentally. They promote flowering, influence phase change (transition the juvenile and adult states)

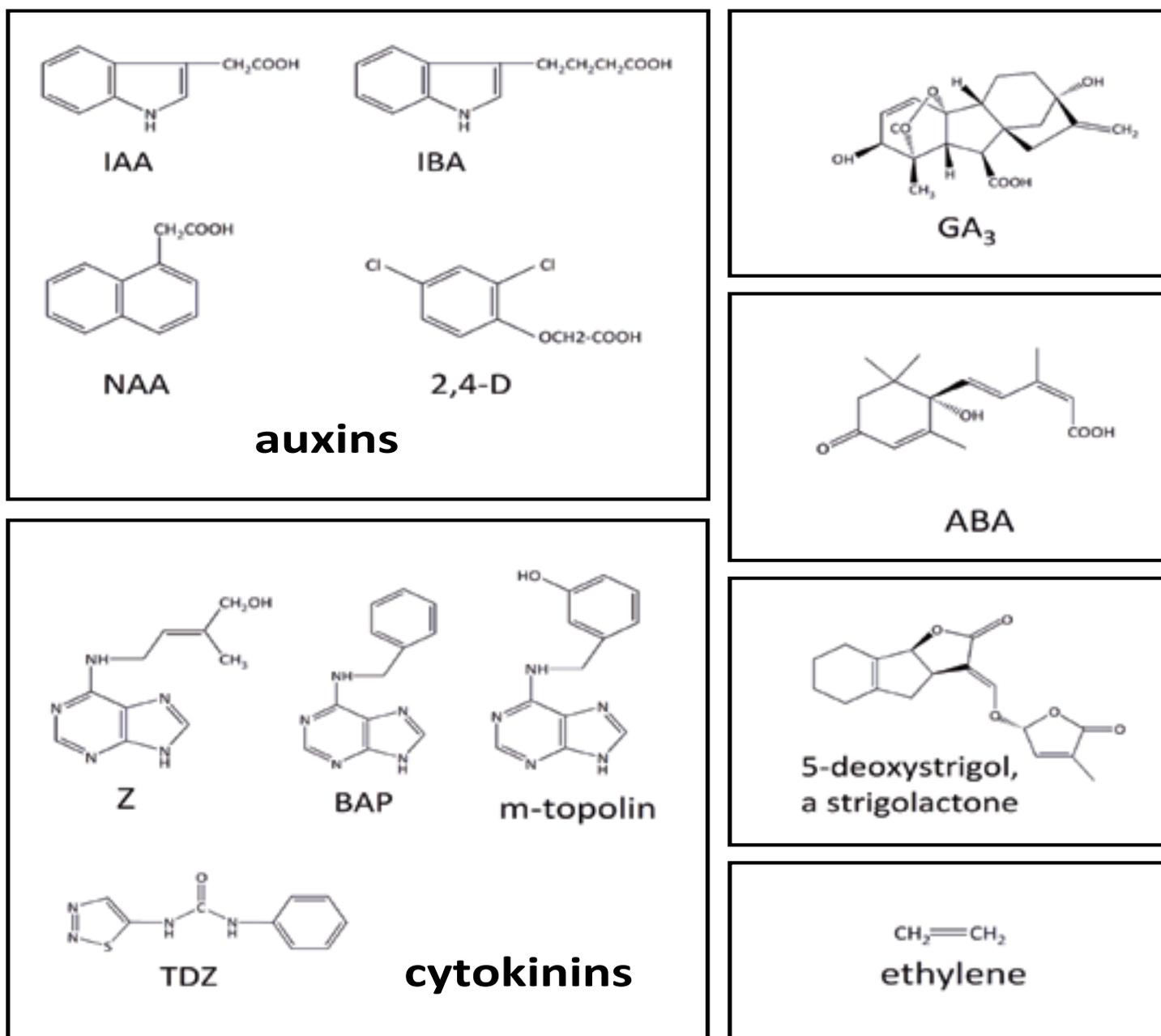
**Table 2. Overview of the five 'classical' plant hormones.**

	Effects in tissue culture	modulators of metabolism, action or transport
auxin	<ul style="list-style-type: none"> <li>• Formation of meristems of adventitious roots.</li> <li>• Induction of somatic embryos (in particular 2,4-D).</li> <li>• Cell division.</li> <li>• Callus formation and growth.</li> <li>• Inhibition of outgrowth of axillary buds.</li> <li>• Inhibition of root growth.</li> </ul>	<ul style="list-style-type: none"> <li>• 2,3,4-Triiodobenzoic acid (TIBA) and 1-N-naphthylphthalamic acid (NPA) inhibit polar auxin transport.</li> <li>• p-Chlorophenoxyisobutyric acid (PCIB) inhibits auxin action as a genuine anti-auxin by binding to the auxin receptor.</li> <li>• Phenolic compounds (e.g. ferulic acid or phloroglucinol) inhibit auxin oxidation.</li> <li>• Riboflavin strongly promotes photooxidation of IBA and IAA.</li> </ul>
cytokinin	<ul style="list-style-type: none"> <li>• Adventitious shoot formation .</li> <li>• Inhibition of adventitious root formation.</li> <li>• Cell division.</li> <li>• Callus formation and growth.</li> <li>• Stimulation of outgrowth of axillary buds.</li> <li>• Inhibition of shoot elongation.</li> <li>• Inhibition of leaf senescence.</li> </ul>	Compounds have been reported that inhibit cytokinin synthesis (lovastatin), degradation and action. The various effects are, however, not yet well studied or ambiguous.
gibberellin	<ul style="list-style-type: none"> <li>• Shoot elongation</li> <li>• Release from dormancy in seeds, somatic embryos, apical buds and bulbs.</li> <li>• Inhibition of adventitious root formation.</li> <li>• Synthesis-inhibitors promote root formation.</li> <li>• Synthesis-inhibitors promote tuber, corm and bulb</li> <li>• Synthesis-inhibitors inhibit shoot elongation</li> <li>• Synthesis-inhibitors facilitate acclimatization.</li> </ul>	There are various gibberellin synthesis inhibitors, among others paclobutrazole, ancymidol and flurprimidol.
ethylene	<ul style="list-style-type: none"> <li>• Senescence of leaves.</li> <li>• Ripening of fruits.</li> <li>• Promotion or inhibition of adventitious regeneration (depending on the time of application or on the genotype?).</li> </ul>	<ul style="list-style-type: none"> <li>• 1-Aminocyclopropane-1-carboxylic acid (ACC) is a precursor of ethylene and is metabolized by plant tissues to ethylene.</li> <li>• Aminoethoxyvinylglycine (AVG) inhibits ethylene synthesis. <math>\text{Co}^{2+}</math>, <math>\alpha</math>-aminoxy-acetic acid and <math>\alpha</math>-aminoisobutyric acid also inhibit ethylene synthesis but have a lower efficiency.</li> <li>• Silver ions inhibit ethylene action. Silver is applied as silverthiosulphate (STS) or <math>\text{AgNO}_3</math>.</li> <li>• <math>\text{KMnO}_4</math>, coated on porous grains effectively oxidizes ethylene.</li> </ul>
abscisic acid	<ul style="list-style-type: none"> <li>• Maturation of somatic embryos.</li> <li>• Facilitation of acclimatization.</li> <li>• Bulb and tuber formation.</li> <li>• Promotion of the development of dormancy.</li> </ul>	Fluridone inhibits ABA synthesis. As it acts by inhibiting an early step in carotenoid synthesis, plants bleach. However, fluridone does not seem to be toxic. Paclobutrazol also inhibits ABA synthesis.

in both directions depending on the species, break dormancy of seeds, buds, corms and bulbs, promote degradation of reserves in seeds, and cause stem elongation. There are over one hundred gibberellins known. GA3 (gibberellic acid), GA1, GA4 and GA7 are mostly used. Once taken up, gibberellins are conjugated. The synthesis of gibberellins is inhibited by compounds like paclobutrazol, flurprimidol and ancymidol. In tissue culture, these inhibitors are used more frequently than gibberellins themselves: they may promote bulb and corm formation and embryo maturation,

enhance rootability of shoots, block shoot elongation and may ease acclimatization. It should be noted that some inhibitors of GA-synthesis also block ABA synthesis.

Together, auxins, cytokinins, ethylene, gibberellins and abscisic acid are often denoted as the "five classical plant hormones". An overview of their actions and the various ways to influence transport, catabolism and action is in Table 2. The structural formulas are in Fig. 3.



**Figure 3.** Structural formulas of major plant hormones.

### Other hormones and hormone-like compounds

When plants are wounded, synthesis of jasmonates occurs by degradation of lipids in the membranes. Jasmonates activate the synthesis of stress proteins. Commercially, jasmonic acid and its volatile methylester (MeJa) can be purchased. Jasmonates promote leaf senescence, fruit ripening, tuber and bulb formation. They play a role in dormancy development and breaking. It has been observed that jasmonates promote regeneration of shoots and roots.

A large number of compounds has been found to influence developmental processes in plants. These include peptides, brassinosteroids, fusicoccin, NO (nitric oxide), phenolic compounds (such as salicylic acid), uridine, elicitors and lipochitooligosaccharides (LCOs). These compounds may become major tools, for example, in achieving adventitious regeneration of shoots, roots or embryos.

### Literature

Useful general information on plant hormones is given by P.J. Davies (ed.) 'Plant Hormones, Physiology, Biochemistry and Molecular Biology', Kluwer Academic Publishers, Dordrecht, Boston, London, 1995. The 2004 edition is less physiologically oriented.

The textbook Plant Physiology contains excellent chapters on plant hormones ('Plant Physiology' by Lincoln Taiz and Eduardo Zeiger, Sinauer Associates Inc, Sunderland, 2006).

Most aspects of the use of plant hormones in tissue culture are discussed in E.F. George 'Plant Propagation by Tissue Culture. Part 1, The Technology, 2<sup>nd</sup> edition; Part 2, In Practice', Exegetics Ltd., Edington, 1993, 1995. Update: 'Plant Propagation by Tissue Culture: Volume 1. The Background' by E.F. George, M.A. Hall, and G.J. De Klerk (eds), 2008. Springer, Dordrecht.

## Plant nutrition in tissue culture

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Plants require carbohydrates and inorganic compounds to sustain growth. Carbohydrates are used as building blocks for macromolecules, starting material in many biosynthetic reactions, energy source, and also as driving force of phloem transport. Under natural conditions, carbohydrates are synthesized during photosynthesis. In tissue culture the need for carbohydrates is met by sugar added in the nutrient medium but photosynthesis also occurs. Inorganic compounds have numerous functions in plants (Table 1). Under natural conditions inorganic compounds are supplied by the soil and in tissue culture by the nutrient medium.

### 1. Inorganic nutrition

Under natural conditions, plants need to take up from the soil:

- Large amounts of ions of some inorganic elements (macronutrients), viz. nitrogen (N), potassium (K), calcium (Ca), phosphorus (P), magnesium (Mg) and sulphur (S); and
- Small quantities of ions of other elements (micronutrients), viz. iron (Fe), nickel (Ni), chlorine (Cl), manganese (Mn), zinc (Zn), boron (B), copper (Cu), and molybdenum (Mo).

the nutrient formulation by dose-response studies is very time-consuming because of the large number of elements and the interactions between elements. A shortcut is the use of the composition of a well-growing plant: supposedly, each species has its own characteristic elementary composition which can be used to adapt the medium formulation. Such media result frequently but not always in improved growth.

Nutrients, especially micronutrients, are also added via impurities in particular via agar. Table 3 shows major inorganic impurities of various agar brands and their relative contribution to MS. Gelrite also contains inorganic contaminations at high concentrations. In addition to inorganic impurities, agar contains many organic impurities that may determine the performance of plants *in vitro*.

### 2. Uptake and transport of inorganics

Whole plants (with roots) absorb inorganic nutrients from soil almost entirely as ions. An ion is an atom, or a group of atoms, which has gained a positive charge (a cation) or a negative charge (an anion). Inorganic nutrients are added to plant culture media as salts. In aqueous solutions salts dissociate into cations and anions. The ions are taken up by the roots passively, or through active mechanisms involving the expenditure of energy. Both systems are influenced by the concentration of other elements, pH, temperature, and the biochemical or physiological status of the plant tissues. These factors can in turn be controlled by the solution presented to the roots, or they may dictate the ionic balance of an ideal solution. For

Table 1. Summary of the functions of the essential inorganic compounds.

Group 1. Nutrients that are part of carbon compounds, e.g. amino acids and nucleic acids	N, S
Group 2. Nutrients that are important in energy storage (ATP) or structural integrity (contribute e.g. to cell wall properties)	P, Si, B
Group 3. Nutrients that remain in ionic form function, e.g. as cofactors of enzymes and in establishing cell turgor	K, Ca, Mg, Cl, Mn, Na
Group 4. Nutrients that are involved in redox reactions, e.g. constituents of cytochromes, alcohol dehydrogenase etc.	Fe, Zn, Cu, Ni, Mo

Together with carbon (C), oxygen (O) and hydrogen (H), these elements constitute the 17 essential elements. Certain other elements, such as cobalt (Co), aluminium (Al), sodium (Na) and iodine (I), are essential or beneficial for some species but their widespread essentiality has not been established. The need for microelements has only been discovered over the past 50-60 years. Since plants in tissue culture entirely depend on added nutrients, discovery of the essentiality of microelements was crucial for successful growth *in vitro*.

The most commonly used formulation for inorganic nutrition in tissue culture is the one of Murashige and Skoog ('MS'). This medium was developed in 1962 to obtain optimal growth of tobacco callus. Table 2 shows the composition of MS compared to the composition of well-growing plants and to modified Hoagland, a modern formulation for a nutrient solution. Major differences between the compositions of MS and plants are the high levels of Cl and Mo and the low levels of Cu, Ca, P and Mg in MS. Interestingly, Hoagland is more similar to plants. MS is used for a very wide range of crops. Experimentation to improve for each crop

example,  $Mg^{2+}$  competes with other cations for uptake. High  $K^+$  or  $Ca^{2+}$  concentrations may lead to Mg deficiency, and vice versa. No studies have been made how uptake of nutrients occurs in shoot cultures. In tissue culture, uptake is generally proportional to the medium concentration up to a concentration of twice MS. For the plant hormone IAA, it has been shown that most uptake is via the cut surface and that only a small fraction is taken up via the epidermis. The same likely holds for minerals. It should be noted, though, that in tissue culture the stomata are always open. Thus, in tissue culture uptake via the stomata may be more prominent.

There are two ways of movement of compounds in water, (1) via diffusion and (2) via water flow. The former is very slow over large distances (according Fick's law, one meter diffusion takes 32 years; 2 cm takes ca. one week). Consequently, in plants transport over large distances occurs via water flow in the vascular bundles. Accordingly, once ions are taken up long-distance transport occurs in the water flow of the xylem. Water flow in the xylem is driven by transpiration. *In vitro* the atmosphere is very humid so transpiration is most likely much reduced and it is not known whether

Table 2. The levels of elements in shoots taken from well growing plants, in MS and in a modified Hoagland formulation used in horticulture. The major differences between MS and 'plants' are indicated.

	In tissue (mmol kgDW <sup>-1</sup> )	InMS (mmol l <sup>-1</sup> )	modified Hoagland (mmol l <sup>-1</sup> )	In tissue (mol%)	In MS (mol%)	modified Hoagland (mol%)
N	1000	60	16.0	64.4	64.4	53.0
K	250	20	6.0	16.1	21.3	19.9
Ca	125	3	4.0	8.0	3.2	13.3
Mg	80	1.5	1.0	5.1	1.6	3.3
P	60	1.25	2.0	3.9	1.3	6.6
S	30	1.5	1.0	1.9	1.6	3.3
Cl	3	6	0.05	0.19	6.4	0.17
Fe	2	0.1	0.05	0.13	0.11	0.17
Mn	1	0.1	0.002	0.06	0.11	0.007
B	2	0.1	0.025	0.13	0.11	0.08
Zn	0.3	0.03	0.002	0.02	0.03	0.007
Cu	0.1	0.0001	0.0005	0.0060	0.0001	0.002
Mo	0.001	0.001	0.0005	0.0001	0.0011	0.002
Ni	0.001	0	0.0005	0.0001	0.000	0.002
Na		0.1	0.05		0.11	0.17
total	15.5	93.7	30.2	100	100	100

the water flow is sufficient to provide growing tissues with sufficient nutrients (The rate of transpiration in vitro has not yet been examined). In liquid medium, almost all  $\text{PO}_4^{3-}$ ,  $\text{NH}_4^+$  and  $\text{NO}_3^-$  are taken up in the first two weeks of culture (Fig. 1).

### 3. Inorganic macronutrients

#### Nitrogen

Nitrogen (N) is essential to plant life. It is a constituent of proteins, nucleic acids and chlorophyll. Most animals cannot assimilate inorganic N and also cannot synthesize many of the amino acids unless assisted by bacteria (e.g. in the rumen of cattle). From the inorganic nutrients in tissue culture media, N has by far the highest concentration. It is usually added both as  $\text{NO}_3^-$  and as  $\text{NH}_4^+$  and nearly all published media provide the majority of their available nitrogen as  $\text{NO}_3^-$ .  $\text{NO}_3^-$  is often the only source of N for plants growing under natural conditions. Once within the cell,  $\text{NO}_3^-$  is reduced to  $\text{NH}_4^+$  before being utilised.  $\text{NO}_3^-$  is first converted to  $\text{NO}_2^-$  by nitrate reductase.  $\text{NO}_2^-$  is reduced to  $\text{NH}_4^+$  by nitrite reductase. Unlike  $\text{NH}_4^+$ ,  $\text{NO}_3^-$  is not toxic but  $\text{NO}_2^-$  can become toxic should it accumulate within plant tissues or in the medium, for example when growth conditions are

not favourable to high nitrite reductase activity and when nitrate is the only nitrogen source.

In the natural and agricultural environments, plant roots usually encounter little reduced nitrogen, because bacteria rapidly oxidize available sources. An exception is forest soils in mountainous regions of the northern hemisphere where  $\text{NO}_3^-$  is usually not available. If  $\text{NH}_4^+$  and other reduced nitrogen compounds are available -and this is particularly the case in the in vitro environment-, they can be taken up and effectively utilized by plants. Why not simply supply nitrogen as  $\text{NH}_4^+$  and avoid the use of  $\text{NO}_3^-$  altogether? The reason lies in the latent toxicity of  $\text{NH}_4^+$  at high concentration, and in the need to control the pH of the medium. Shoots grown on medium containing a high proportion of ammonium ions may become stunted or hyperhydric.

These effects can sometimes be

reversed by transfer to a medium containing a high proportion of  $\text{NO}_3^-$  or to one where  $\text{NO}_3^-$  is the only N source. Hyperhydricity is the in vitro formation of abnormal organs, which are brittle and have a water-soaked appearance. Plant culture media are usually started at pH 5.4-5.8. When both  $\text{NO}_3^-$  and  $\text{NH}_4^+$  are added, a rapid uptake of  $\text{NH}_4^+$  into plant tissue causes the pH to fall to ca. 4.2-4.6.

Reduced nitrogen may also be added as amino acids. For most tissue culture purposes, the addition of amino acids may be unnecessary, providing media contain adequate amounts of  $\text{NO}_3^-$  and  $\text{NH}_4^+$ . When media contain suboptimal amounts, a casein hydrolysate (a mixture of amino acids) may substantially increase growth, whereas only marginal increases in yield are achieved when optimal amounts of inorganic N occurs. In literature, many examples can be found of improvement of growth of cell cultures, shoot cultures and enhanced adventitious regeneration of shoots, roots and embryos by amino acid mixtures and by individual amino acids. It should be noted that in plants the natural transport vehicles of reduced N are asparagine and glutamine.

Nitrogen is available in the atmosphere as  $\text{N}_2$  but only legumes have the capacity to utilize this nitrogen using Rhizobium bacteria in the root nodules.

Table 3. Increase of the content of Na, S and Cu relative to MS brought about by agar (0.6%) obtained from various companies (1-8) or gelrite (0.2%). Increases are shown as percentages. The proportional increase in other elements is maximally 20% .

	Agar 1	Agar 2	Agar 3	Agar 4	Agar 5	Agar 6	Agar 7	Agar 8	gelrite
Na	1212	336	3312	1980	2562	3804	684	313	591
S	69	29	87	111	77	98	25	69	0.8
Cu	90	204	108	144	24	96	nd	28	91

## Phosphorus

Phosphorus (P) is a vital element in plant biochemistry. It occurs in numerous macromolecules such as nucleic acids, phospholipids and co-enzymes. It functions in energy transfer via the pyrophosphate bond in ATP. Phosphate groups attached to different sugars provide energy in respiration and photosynthesis and phosphate bound to proteins regulates their activity. P is absorbed by roots in the form of the anions  $\text{H}_2\text{PO}_4^-$  and  $\text{HPO}_4^{2-}$  by an active process. In contrast to  $\text{NO}_3^-$  and  $\text{SO}_4^{2-}$ , phosphate is not reduced, but remains in the highly oxidized form and is used as  $\text{PO}_4^{3-}$ . In culture media the element is provided as soluble  $\text{H}_2\text{PO}_4^-$  and  $\text{HPO}_4^{2-}$ .  $\text{H}_2\text{PO}_4^-$  predominates at pH values below 7, characteristic of most tissue culture media. Phosphate is usually taken up most rapidly (Fig. 1). At the same time, movement of phosphate in solidified medium by diffusion seems to be much slower than movement of other inorganic nutrients.

## Potassium

Potassium (K) is the major cation (positive ion) within plants, reaching in the cytoplasm and chloroplasts concentrations of 100 – 200 mM.  $\text{K}^+$  is not metabolised. Unlike  $\text{NH}_4^+$ ,  $\text{NO}_3^-$ ,  $\text{SO}_4^{2-}$ , and  $\text{H}_2\text{PO}_4^-$ , it is not incorporated into organic molecules. It contributes significantly to the osmotic potential of cells, functions in cell extension through the regulation of turgor and has a major role in stomatal movements.  $\text{K}^+$  counterbalances the negative charge of inorganic and organic anions, and functions in long-distance nutrient flow. In intact plants,  $\text{K}^+$  ions are thought to cycle moving up- and downwards in the vascular bundles. Many proteins show a high specificity for  $\text{K}^+$  which, acting as a cofactor, alters their configuration so that they become active enzymes.  $\text{K}^+$ -ions also neutralise organic anions produced in the cytoplasm, and so stabilise the pH and osmotic potential of the cell. In whole plants, deficiency of  $\text{K}^+$  results in loss of cell turgor, limp tissues and an increased susceptibility to drought, salinity, frost damage and fungal attack.  $\text{K}^+$ -deficiency in plant culture media is said to lead to hyperhydricity, and a decrease in absorption of phosphate. Murashige and Skoog medium contains 20 mM  $\text{K}^+$ .

## Sodium

Sodium ions ( $\text{Na}^+$ ) are taken up into plants, but in most cases they are not required for growth and development and many plants actively secrete them from their roots to maintain a low internal concentration. In some plants, though,  $\text{Na}^+$  does appear to have a beneficial nutritional effect and is therefore considered as a functional element. In wheat, oats, cotton and cauliflower  $\text{Na}^+$  can partially replace  $\text{K}^+$ , but is not essential. The element can function as an osmotic stabilizer in halophytic plants. Most nutrient formulations do not contain any  $\text{Na}^+$  with the exception of NaFeEDTA. Agar and gelrite contain high levels of  $\text{Na}^+$ .

## Magnesium

Magnesium (Mg) is an essential component of chlorophyll, and is required for the activity of many enzymes, especially those involved in the transfer of phosphate. Magnesium is the central atom in the porphyrin structure of the chlorophyll molecule. ATP synthesis has an absolute requirement for  $\text{Mg}^{2+}$  and it is a bridging element in the aggregation of ribosome subunits. Within plants,  $\text{Mg}^{2+}$  is mobile and diffuses freely and serves like  $\text{K}^+$  as a cation balancing and neutralising anions and organic acids. Plant culture media invariably contain relatively low concentrations of  $\text{Mg}^{2+}$  (MS only 1.5 mM). Very often  $\text{MgSO}_4$  is used as the unique source of both  $\text{Mg}^{2+}$  and  $\text{SO}_4^{2-}$ .

## Sulphur

Under natural conditions sulphur (S) is mainly absorbed as  $\text{SO}_4^{2-}$ , which is also the usual source of the element in nutrient media. However, that which is incorporated into organic compounds occurs mainly as reduced -SH, -S- or -S-S- groups. The sulphur-containing amino acids cysteine and methionine are incorporated into proteins. Between or within polypeptides, two cysteine amino acids can form disulfide (S-S) bridges. Sulphur is used by plants in lipid synthesis and acts as a ligand joining ions of iron, zinc and copper to metalloproteins and enzymes.

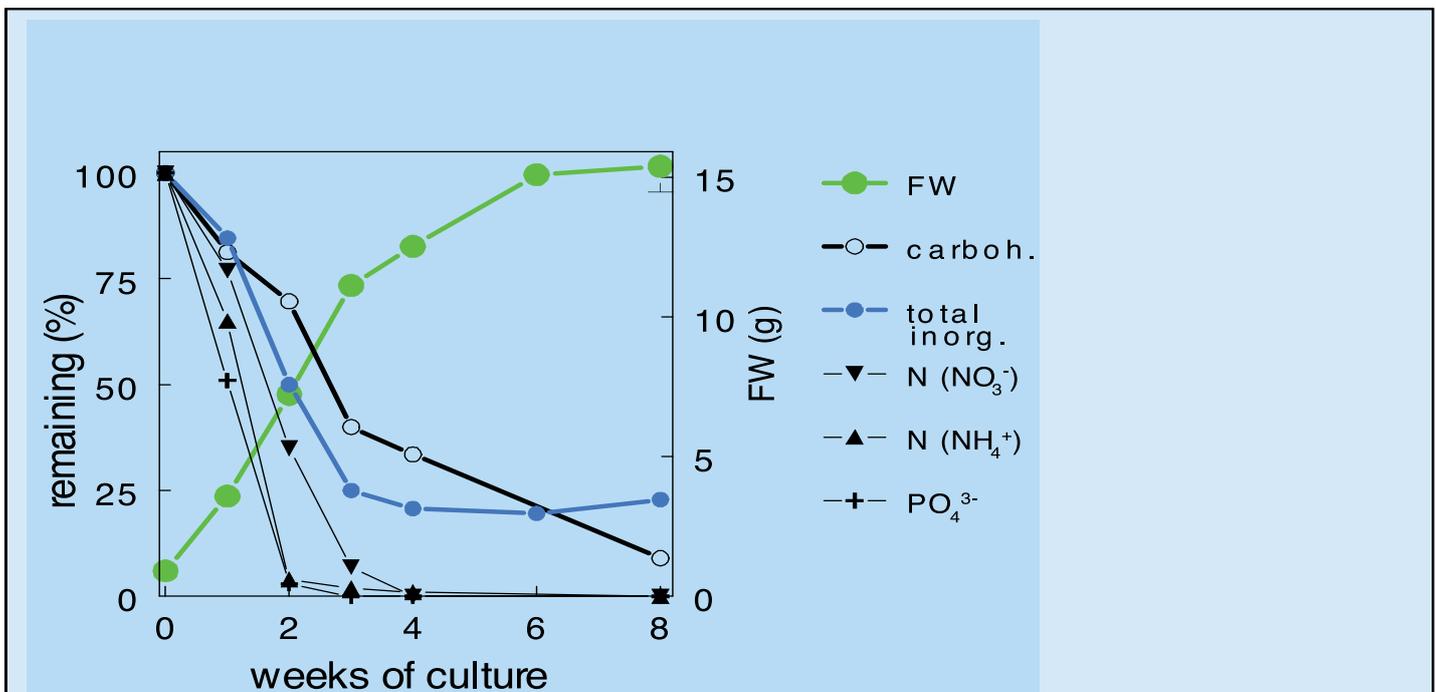


Figure 1. Exhaustion of medium components during culture of dahlia shoots in liquid medium (50 ml with 5 explants). On solidified medium, exhaustion and growth were much slower (not shown). Total inorganics was measured with an EC-meter and total carbohydrates with a brix-meter. Both determinations were shown to be accurate in separate experiments.

## Calcium

As a major cation, calcium ions ( $\text{Ca}^{2+}$ ) helps to balance anions within the plant, but unlike  $\text{K}^+$  and  $\text{Mg}^{2+}$  it is not readily mobile. Because of its capacity to link biological molecules together with coordinate bonds, the element is involved in the structure and physiological properties of cell membranes and the middle lamella of cell walls. The enzyme  $\beta$ -(1 $\rightarrow$ 3)-glucan synthase depends on  $\text{Ca}^{2+}$ , and cellulose synthesis by cultured cells does not occur unless there are at least  $\mu\text{M}$  concentrations of  $\text{Ca}^{2+}$  in the medium. Many other plant enzymes are  $\text{Ca}^{2+}$ -dependent and  $\text{Ca}^{2+}$  is a cofactor in the enzymes responsible for the hydrolysis of ATP. Although  $\text{Ca}^{2+}$  can be present in mM concentrations within the plant as a whole,  $\text{Ca}^{2+}$ -ions are pumped out of the cytoplasm of cells. The active removal of  $\text{Ca}^{2+}$  is necessary to prevent the precipitation of phosphate and interference with the function of  $\text{Mg}^{2+}$ . The uniquely low intra-cellular concentration of  $\text{Ca}^{2+}$  allows plants to use calcium as a chemical 'second messenger' in hormonal signalling. Regulatory mechanisms are initiated when  $\text{Ca}^{2+}$  binds with the protein calmodulin, which is thus enabled to modify enzyme activities.  $\text{Ca}^{2+}$ -deficiency in plants may result in a cessation of growth and in death of the shoot tip. Tip necrosis has been especially observed in shoot cultures and often occurs after several subcultures have been accomplished. In *Plectranthus*,  $\text{Ca}^{2+}$  reduces necrosis. Tip necrosis occurs in *Psidium guajava* shoot cultures if shoots are allowed to grow longer than 3 cm, and is common in rapidly growing cultures. It occurs in *Sequoiadendron giganteum* shoots when they are grown on relatively dilute media. Elemental analysis of necrotic apices has shown them to be deficient in  $\text{Ca}^{2+}$  and a shortage of this element has been associated with tip necrosis in *Amelanchier*, *Betula*, *Populus*, *Sequoia*, *Ulmus*, *Cydonia* and other woody plants. As  $\text{Ca}^{2+}$  is not or only little remobilised within plant tissues, actively growing shoots need a constant fresh supply of ions in the transpiration stream. An inadequate supply of  $\text{Ca}^{2+}$  can result from limited uptake and from inadequate transport, the latter being caused by the absence of transpiration due to the high humidity in the culture vessel. A remedy can sometimes be obtained by reducing the culture temperature so that the rate of shoot growth matches

*Haworthia micropropagation*,  
Succulent Tissue Culture, The Netherlands



$\text{Ca}^{2+}$  supply, using vessels which promote better gas exchange (thereby increasing the transpiration and xylem transport), or by increasing the concentration of  $\text{Ca}^{2+}$  in the medium. There is a limit to the concentration of  $\text{Ca}^{2+}$ , which can be employed in tissue culture media because many Ca-salts have limited solubility.

## Chloride

The chloride ion ( $\text{Cl}^-$ ) has been found to be essential for plant growth, but seems to be involved in few biological reactions and only very small quantities are really necessary.  $\text{Cl}^-$  is required for the water-splitting protein complex of photosystem II, and it can function in osmoregulation in particular in stomatal guard cells.  $\text{Cl}^-$  is freely transported and many plants can tolerate the presence of high concentrations without showing toxicity. The chief role of  $\text{Cl}^-$  seems to be in the maintenance of turgor and in balancing rapid changes in the level of free cations such as  $\text{K}^+$ ,  $\text{Mg}^{2+}$  and  $\text{Na}^+$ .

The concentration of  $\text{Cl}^-$  in MS is 6 mM. Agar (a product obtained from seaweed) also contains  $\text{Cl}^-$  and may increase the concentration by 1 mM. A too high concentration may lead in woody species to yellow leaves and weak stems: sometimes tissues collapse and die. An excess of  $\text{Cl}^-$  has been thought to be one of the causes of hyperhydricity, and omission of  $\text{Cl}^-$  seems to prevent hyperhydricity in *Prunus*.

## 4. Micronutrients

The essential micronutrients Fe, Mn, Zn, B, Cu, Co and Mo are components of proteins or have metabolic and physiological importance. At least five of these elements are, for instance, necessary for chlorophyll synthesis and chloroplast function. Micronutrients have roles in the functioning of the genetic apparatus and several are involved with the activity of growth substances.

*Manganese* (Mn) has been included in the majority of plant tissue culture media. It is generally added in concentrations between 25-150  $\mu\text{M}$ .



Cactaceae: *Solisiapectinifera*,  
Succulent Tissue Culture, The Netherlands

The most probable role for Mn is in definition of the structure of metalloproteins involved in respiration and photosynthesis. It is known to be required for the activity of several enzymes, among others decarboxylases, dehydrogenases, kinases and oxidases and superoxide dismutase enzymes. Mn is necessary for the maintenance of chloroplast ultrastructure. Because Mn(II) can be oxidized to Mn(IV), Mn plays an important role in redox reactions. The evolution of oxygen during photosystem II is dependent on an Mn-containing enzyme and is proportional to Mn content. Mn is toxic at high concentration.

**Zinc** (Zn) is a component of stable metallo-enzymes with many diverse functions. Zn is required in more than 300 enzymes including alcohol dehydrogenase, carbonic anhydrase, superoxide dismutase and RNA-polymerase. Zn is involved in chlorophyll synthesis.

**Boron** (B) is involved in plasma membrane integrity and functioning, probably by influencing membrane proteins, and cell wall intactness. The element is required for the metabolism of phenolic acids, and for lignin biosynthesis. It is probably a component, or co-factor of the enzyme which converts p-coumaric acid to caffeate and 5-hydroxyferulate. B is necessary for the maintenance of meristematic activity and is thought to be involved in the maintenance of membrane structure and function, possibly by stabilizing natural metal chelates which are important in wall and membrane structure and function. B is concerned with regulating the activities of phenolase enzymes; these bring about the biosynthesis of phenylpropane compounds, which are polymerized to form lignin. Lignin biosynthesis does not take place in the absence of B. B also mediates the action of phytochrome and the response of plants to gravity. B has

no effect during the induction of somatic embryogenesis from cultured carrot petiole explants, but strongly influences the development of somatic embryos: at low B development of roots is promoted with simultaneous retardation of shoot development, and at high B shoot development is favoured at the expense of the root system.

**Copper** (Cu) is an essential micronutrient, even though plants normally contain only a very low level of the element. Two kinds of copper ions exist, the monovalent ion  $\text{Cu}^+$  and the divalent ion  $\text{Cu}^{2+}$ . The former is easily oxidized to the latter and the latter is easily reduced. The element becomes attached to enzymes, many of which bind to and react with oxygen. They include the cytochrome oxidase enzyme system, responsible for oxidative respiration, and superoxide dismutase (an enzyme which contains both copper and zinc atoms). Detrimental superoxide radicals, which are formed from molecular oxygen during electron transfer reactions, are reacted by superoxide dismutase and thereby converted to water. Cu atoms occur in plastocyanin, a pigment participating in electron transfer. High concentrations of Cu can be toxic. Most culture media include ca. 0.1-1.0  $\mu\text{M}$   $\text{Cu}^{2+}$ , usually added through  $\text{CuSO}_4$ . The concentration of Cu in tissue culture media is very small relative to the level in plants (Table 2). It is therefore not surprising that a number of authors report strong increases of growth when Cu is added at 1- 5  $\mu\text{M}$ .

**Molybdenum** (Mo) is absorbed by plants as the molybdate ion ( $\text{MoO}_4^{2-}$ ). This is normally added to culture media as  $\text{Na}_2\text{MoO}_4$  at concentrations up to 1  $\mu\text{M}$ . Considerably higher levels have occasionally been introduced apparently without adverse effect. Mo is a component of several plant enzymes, e.g., nitrate reductase and nitrogenase. It is therefore essential for nitrogen utilisation. Tissues and organs presented with  $\text{NO}_3^-$  in a Mo-deficient medium can show symptoms of nitrate toxicity because the ion is not reduced to ammonia.

**Iron** (Fe) is an essential micronutrient for plant tissue culture media and can be provided from either ferrous or ferric salts. It functions in electron transfer as a component of cytochromes. To keep Fe in solution, chelating compounds are essential. Chelates are organic compounds capable of forming complexes with metal cations, in which the metal is held with fairly tight chemical bonds. In this way, metal ions are held in solution under conditions where free ions would react with anions to form insoluble compounds. Despite tight bonding, there is always an equilibrium between chelate complexes and ions in solution. For a chelated metal ion to be utilised by a plant there must be some mechanism whereby the complex can be broken permanently. This could occur if it is absorbed directly and the ion displaced by another more avid binding agent, or if the complex is biochemically denatured. Metals in very stable complexes can be unavailable to plants, copper in EDTA chelates may be an example. Within the plant very many constituents such as proteins, peptides, porphyrins, carboxylic acids and amino acids act as chelating agents. Plants secrete chelating agents to assist the uptake of iron. Divalent organic acids such as citric, maleic, malic and malonic acid are found in the xylem sap of plants, where together with amino acids they can complex with metal ions and assist their transport. These acids can be secreted from cultured tissues into the nutrient medium and will contribute to the conditioning effect. Malic and citric acids, released into the medium by rice cells, are able to make unchelated ferric iron available, so correcting an iron deficiency.

**Cobalt** (Co) is not regarded as an essential element. Nevertheless, Murashige and Skoog (1962) included Co in their medium because it had been shown to be required by lower plants and it was thought that it might have a role in regulating morphogenesis in higher plants. However, no stimulatory effect on the growth of tobacco callus was observed by adding  $\text{CoCl}_2$  to the medium at several concentrations from 0.1  $\mu\text{M}$  and above, and at 80.0 and 160  $\mu\text{M}$  the compound was toxic.

**Other micronutrients.** Several workers have included aluminium (Al) and nickel (Ni) in their micronutrient formulations. However, the general

**Table 4. Hydrolysis of sucrose to fructose and glucose during autoclaving, depending upon the pH.**

pH	hydrolysis (%)
3.0	100
3.4	75
3.8	40
4.2	25
4.7	12.5
5.0	10
6.0	0

benefit of adding the former metal does not seem to have been adequately demonstrated. It has been reported that the lack of Ni and the inclusion of Co leads to reduced urease activity in plants grown on MS medium. Iodine is not recognised as an essential element for the nutrition of plants, although it may be necessary for the growth of some algae. The iodide ion has been added to many tissue culture media. Silicon (Si) is the second most abundant element on the surface of the earth. Si has been demonstrated to be beneficial for the growth of plants and to alleviate biotic and abiotic stress. The silicate ion is not normally added to tissue culture media, although it is likely to be present in low concentrations. Deliberate addition to the medium might, however, improve the growth of some plants.

## 5. Organic Nutrition: Sucrose

In plants, carbohydrates have various essential functions. They are substrates for respiration, play a role in the synthetic pathways of many compounds, are building blocks of macromolecules (starch and cellulose) and are a major driving force of water flow in the phloem. Carbohydrates influence many developmental processes. Sucrose plays a role in dormancy development, storage organ formation and maturation of somatic embryos. Recent findings suggest a regulatory role of sugar levels in the transition to flowering. Starch synthesized from sucrose taken up from the nutrient medium accumulates especially in cells from which adventitious shoot or root primordia are being formed. How sugars act as regulating molecules remains to be elucidated.

In tissue culture, sucrose is usually added as the carbohydrate source. Sucrose has almost invariably been found to be the best carbohydrate. Glucose is generally found to support growth well, and in a few plants it may result in better in vitro growth than sucrose, or promote organogenesis where sucrose will not. But being more expensive than sucrose, glucose will only be preferred for micropropagation where it produces clearly advantageous results. Sucrose is the most common carbohydrate in the phloem sap of angiosperms. In sieve element sap sucrose can reach concentrations of 0.3 to 0.9 M. In tissue culture, concentrations range from 2% to 9% (20 – 90 g.l<sup>-1</sup>; 58 – 263 mM). The high concentrations are used when storage organs like bulbs should develop. The common concentration is 3%. Invertases that are released by the explant into the medium, split sucrose into glucose and fructose. Thus, explants are usually exposed to a mixture of sucrose, glucose and fructose. In in-vitro cultures, carbohydrates play also an important role as osmotic agent. The presence of sucrose in tissue culture media specifically inhibits chlorophyll formation. A hydrolysis of sucrose takes place during autoclaving of media depending on pH (Table 4).

In higher plants growing under natural conditions, sucrose is the major product of photosynthesis and is transported to various sink tissues via the phloem. Sucrose synthesized in mesophyll cells is loaded into the sieve element-companion cell complex of the phloem. Long-distance transport in the phloem uses the water flow that is brought about by a hydrostatic pressure gradient. In sink tissue, phloem unloading appears to depend on the sink strength.

## 6. Undefined supplements

Many undefined supplements were employed in early tissue culture media. Their use has slowly declined. Nevertheless several supplements of uncertain and variable composition are still in common use. The first successful cultures of plant tissue involved the use of yeast extract. Other undefined additions made to plant tissue culture media have been include meat extract, potato extract, malt extract, banana homogenate and coconut milk.

## Literature

Chapter 3 of 'Plant Propagation by Tissue Culture: Volume 1. The Background' by E.F. George, M.A. Hall, and G.J. De Klerk (eds), 2008. Springer, Dordrecht. The textbook Plant Physiology contains excellent chapters on mineral nutrition and solute transport ('Plant Physiology' by Lincoln Taiz and Eduardo Zeiger, Sinauer Associates Inc, Sunderland, 2006).

**Cactaceae: Gymnocactus  
Succulent Tissue Culture, The Netherlands**



## ANTIBIOTICS

Duchefa Biochemie B.V. is a supplier of a wide range of antibiotics.

Application of these antibiotics are

- Suppressing bacterial, fungal and mould growth in cell cultures.
- Selective agents in combination with marker genes.

Antibiotics can be produced by various species of micro-organisms or are chemically synthesized. All have the capacity of inhibiting growth of micro-organisms.

Most of our antibiotics have been tested for use in cell cultures and have no cytotoxic effects. Some antibiotics have been specially tested for use in plant cell and tissue cultures.

If you might have any questions regarding the use of antibiotics, please don't hesitate to contact us. Since our company has pharmaceutical, biochemical and microbiological knowledge available, we will be able to give you an answer in most cases.

**All antibiotics are for laboratory use only.**

**Not for drug, household or other uses**



There are many antibiotics known and at least as many different modes of antimicrobial action active against more or less definite spectra of bacteria. In their turn bacteria have developed numerous types of resistance mechanisms against all kinds of antibiotics.

Antibiotics can be grouped in several classes such as their molecular mode of action.

In biotechnology the most often used groups are Inhibitors of Bacterial Cell Wall Synthesis and Inhibitors of Protein Synthesis. The first group is mostly used to eliminate bacteria for instance *Agrobacterium* after transformation. The second group called Inhibitors of Protein Synthesis, such as Kanamycin, is most often used as a selective agent in combination with marker genes.

## Inhibitors of Bacterial Cell Wall Synthesis

This group of antibiotics focuses on the synthesis of the bacterial cell wall. By application of these antibiotics, several bacterial key enzymes and cell wall binding blocks are knocked out. As a result, build up of the bacterial cell wall is ceased and lysis of the cell as a result of osmotic shock will occur.

The bacterial cell wall, also called peptidoglycan, encases the cell membrane as a continuous, highly cross linked molecule, preventing rupturing of the cell membrane in a hypotonic milieu. The build up of the bacterial cell is a continuous process of synthesis and degrading.

Synthesis takes part in three steps. In the first step, production of basic building blocks takes place inside the cell. Cycloserine, because of its similarity to certain substrates of key enzymes involved in this process, inhibits major reactions in this process. As a result no final buildings blocks are made.

In the second step, ready made building blocks are transported across the cell membrane and covalently linked to the already existing cell wall. This results in long linear polymers of building blocks attached to the already existing cell wall. Because these polymers are not cross linked yet they do not provide any strength to the bacterial cell wall. Bacitracin and Vancomycin act inhibitory in this sequence of reactions.

Within the third and final stage, all linear polymers are cross linked to form the rigid network which is the backbone of the bacterial cell wall or peptidoglycan. Transpeptidase is the key enzyme involved in this cross linking step and is inhibited by Penicillins and Cephalosporins like Carbenicillin, Cefotaxim, Ampicillin etc.

Blocking one of these three steps causes inhibition in the build up of the cell wall, finally resulting in nicks in the peptidoglycan by which the membrane protrudes into the hypotonic medium and ultimately last ruptures.

Bacteria can develop resistance against Penicillins and Cephalosporins by producing Beta-Lactamase. Both Penicillins and Cephalosporins have a Beta-Lactam ring in their center. A major part of this ring structure is a C-N bond which is an absolute requirement for antimicrobial activity. This C-N bond is also the substrate site of Beta-Lactamase, which is capable of hydrolyzing the binding between the carbon and nitrogen atom. Once broken, there is no antimicrobial activity left due to a structural change in the penicillin or cephalosporin molecule.

In Cefotaxim and to a lesser degree in Carbenicillin this Achilles heel is protected by molecular side chains preventing beta-lactamase to unite with its substrate site.

Another way of protecting Amoxicillin or Ticarcillin against inactivation by Beta-Lactamase is the addition of Clavulanic acid. This small molecule is a look-alike structure of the C-N bond present in the Lactam ring. Due to an irreversible binding between Clavulanic acid and the substrate site of Beta-Lactamase, hydrolysis of the C-N bond is prevented.

The two major groups within the family of Bacterial Cell Wall inhibitors are Penicillins and Cephalosporins.

Bacterial Cell Wall Inhibitors		
Penicillins	Cephalosporins	Others
Ampicillin	Cefalexin	Bacitracin
Amoxycillin	Cefotaxim	Cycloserin
Carbenicillin		Vancomycin
Penicillin G		
Ticarcillin		

### Bactericide Inhibitors of Protein Synthesis

The main group within this family is the group of Aminoglycosides. The collection of Aminoglycosides represents a large set of structurally related polycationic molecules containing two or more sugars connected by glycosidic linkage to a hexose core. Aminoglycosides have a strong antibacterial effect. Bacteria exposed to these antibiotics undergo a wide variety of metabolic changes, including changes in cell permeability and transport, inhibition of protein synthesis and misreading of the genetic code.

Aminoglycosides can both bind to prokaryotic and eukaryotic ribosomes and are capable of contacting ribosome binding sites at both subunits. By attachment of these antibiotics to their respective binding sites, protein translating at various stages can be inhibited.

Aminoglycosides are often used in biotechnology as selective agents in combination with certain antibiotic resistance marker genes. Antibiotics used are Kanamycin, G418, Hygromycin B, Paromomycin etc.

The most frequently used marker gene is based on phosphorylation by O-Phosphotransferase which is coded via NPT II (APH 3' gene). The enzyme phosphorylates the 3'OH group present on Kanamycin or G418. Due to the attachment of a strong electronegative phosphate group at the sugar part, the stereometric structure of the aminoglycoside molecule changes in such a way that the antibiotic does not fit anymore at its ribosome's binding site.

Bactericide Inhibitors of Protein Synthesis	
Aminoglycosides	
Gentamycin	Paromycin
Hygromycin B	Streptomycin
Kanamycin	Tobramycin
Neomycin	G-418

### Bacteriostatic Inhibitors of Protein Synthesis

This set of antibiotics includes various groups of antibiotics with the capability to bind at the 30S ribosomal subunit. As a result, protein synthesis may be inhibited at several stages during the translation process. In contrast to Aminoglycosides binding of bacteriostatic inhibitors of protein synthesis is reversible. Protein synthesis and finally

bacterial growth will start again after exposed bacteria are transferred to media without antibiotics.

Bacteriostatic inhibitors, amongst many others, are Tetracyclines, Chloramphenicol, Spectinomycin and Erythromycin. All have their respective binding sites at the 30S ribosomal subunit. All antibiotics are capable of inhibiting protein synthesis, but their modes of action differ.

Bacteriostatic Inhibitors of Protein Synthesis	
Chloramphenicol	Lincomycin
Chlortetracycline	Oxytetracyclin
Clindamycin	Spectinomycin
Doxycyclin	Tetracyclin
Erythromycin	

### Antifungal Agents

Amphotericin B and Nystatin are two commonly applied antifungal antibiotics in biotechnology. Both affect cell membrane permeability of moulds and fungi due to interactions with sterols present in the membrane. As a results small trans membrane channels are formed leaking valuable ions and causing cell death.

Antifungal Agents	
Amphotericin B	Miconazole
Nystatin	Cycloheximide

Besides the above mentioned families of antibiotics there are many more. Each differs in mode of action, spectrum, resistance patterns etc.

Inhibitors of Nucleic Acid Metabolism	
Amsacrine	Mitomycin C
Doxorubicin	Nalidixic acid
Rifampicin	

Antimetabolites	
Methotrexate	Trimethoprim
Metronidazole	Sulphametoxazole
Miconazole	

Nucleic Acid Analogues	
5-Fluorouracil	6-Mercaptopurine

# PLANT CELL AND TISSUE CULTURE MEDIA



# CUSTOM MADE MEDIUM FORM

## 1. ADDRESS INFORMATION

---

Name of company, university, institute, customer i.d. etc.:

---

Name of contact person :

---

Telephone No:

Fax No:

---

Shipment Address:

---

Billing Address:

---

Purchase Order Number:

VAT No:

---

## 2. NAME or NUMBER of the medium:

---

## 3. FORMULATION (mg/l and/or molarity)

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## 4. Quantity Required:

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(the minimal weight of the ordered quantity of medium should be one kilogram of powdered medium)

---

## 5. Delivery Schedule:

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Date:

Quantity:

---

## 6. Undersigned declaration of discretion YES or NO

---

## 7. Customer place, date, signature:

---

Please, fax this form to:  
+31- (0)23 - 531 80 27  
E-mail:order@duchefa.nl

# ANDERSON'S RHODODENDRON MEDIUM

A two-fold increase in shoot multiplication of red raspberry on Anderson's medium was achieved as compared to the Murashige and Skoog formulation. A comparison of the inorganics of both formulations showed a reduction to approximately 1/4 strength on  $\text{NH}_4\text{NO}_3$  and  $\text{KNO}_3$  in Anderson inorganics. The optimal concentrations of growth regulators for shoot multiplication of red and black raspberry were 0.1-2.5  $\mu\text{M}$  IBA and 4.5-9.0  $\mu\text{M}$  BA. In vitro rooting of black and red raspberries were successful using the basal media that included Anderson's Inorganics, 5 $\mu\text{M}$  IBA and 600 mg/l activated charcoal. The latter is essential for high rooting percentages.

Anderson W.C., Tissue culture propagation of Red and Black Raspberries, *Rubus Idaeus* and *R. Occidentalis* Act. Hort., 112, 13 (1980).

## A 0201 ANDERSON'S RHODODENDRON

### Micro and Macro elements

A 0201.0001	1 l	(1.8 g)
A 0201.0010	10 l	(18.3 g)
A 0201.0050	50 l	(91.4 g)

## A 0202 ANDERSON'S RHODODENDRON

### Micro and Macro elements including Vitamins

A 0202.0001	1 l	(2.0 g)
A 0202.0010	10 l	(20.1 g)
A 0202.0050	50 l	(100.5 g)

## MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	73.40	200.00
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.30	1.81
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
$\text{KNO}_3$	480.00	4.75
$\text{MgSO}_4$	180.54	1.50
$\text{NaH}_2\text{PO}_4$	330.60	2.75
$\text{NH}_4\text{NO}_3$	400.00	5.00

Total concentration Micro and Macro elements: 1828.86 mg/l

## VITAMINS

	mg/l	$\mu\text{M}$
Adenine sulphate	80.00	197.87
myo-Inositol	100.00	554.94
Thiamine HCl	0.40	1.19

Total concentration Micro and Macro elements including vitamins: 2009.26 mg/l

Willemsen en Bourgondiën B.V., The Netherlands



# CHÉE AND POOL (C2D) VITIS MEDIUM

In the medium defined by Chée and Pool the original Murashige and Skoog concentration of Chlorine, Iodine and Manganese is decreased, resulting in an improved shoot multiplication of Vitis. Substituting calcium nitrate for calcium chloride improved the quality of grapevine shoots produced in culture. Shoot multiplication was dramatically improved by omitting Iodine and lowering the concentration of Manganese. This might be the result of the involvement of both ions in auxin metabolism and transport.

Chée, R., and Pool, R.M.,  
Improved Inorganic Media Constituents for In Vitro Shoot Multiplication of Vitis, *Scientia Horticulturae*, 32 (1987) 85-95.

## C 0248 CHÉE & POOL BASAL SALT MEDIUM

Micro and Macro elements

C 0248.0010                      10 l      (44.5 g)

## C 0249 CHÉE & POOL BASAL SALT MEDIUM

Micro and Macro elements including Vitamins

C 0249.0010                      10 l      (44.6 g)

## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
MnSO <sub>4</sub> ·H <sub>2</sub> O	0.85	5.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
Ca(NO <sub>3</sub> ) <sub>2</sub>	492.30	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

Total concentration Micro and Macro elements: 4445.49 mg/l

## VITAMINS

	mg/l	µM
myo-Inositol	10.00	55.5
Nicotinic Acid	1.00	8.12
Pyridoxine HCl	1.00	5.00
Thiamine HCl	1.00	3.00

Total concentration Micro and Macro elements including vitamins:  
4458.49 mg/l

Willemsen en Bourgondiën B.V., The Netherlands



# CHU (N<sub>6</sub>) MEDIUM

Chu (N<sub>6</sub>) medium is defined to improve the formation, growth and differentiation of pollen callus in rice. The concentration of ammonium proved to be crucial for the development of callus. The optimum concentration NH<sub>4</sub><sup>+</sup> is 7.0 mM (equal to 3.5 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>). Higher concentrations of ammonium drastically inhibited the growth and differentiation of the rice pollen. The concentration of KNO<sub>3</sub> and the other medium components did not affect the development of the callus.

Chu C.C., The N<sub>6</sub> medium and its application to anther culture of cereal crops, Proc. Symp. Plant Tissue Cult., Peking, 43 (1978).

Chu C.C. et al., Establishment of an efficient medium for anther culture of rice through comparative experiments on the nitrogen sources. Scientia Sinica, 18, 659 (1975).

## C 0203 CHU (N<sub>6</sub>) MEDIUM

### Micro and Macro elements

C 0203.0001	1 l	(4.0 g)
C 0203.0010	10 l	(39.5 g)
C 0203.0050	50 l	(197.6 g)

## C 0204 CHU (N<sub>6</sub>) MEDIUM

### Micro and Macro elements including Vitamins

C 0204.0001	1 l	(4.0 g)
C 0204.0010	10 l	(39.6 g)
C 0204.0050	50 l	(197.8 g)

## C 0401 CHU VITAMIN MIXTURE

Package contains 0.4 g or 1.0 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre Chu (N<sub>6</sub>) medium of the proper final vitamin concentration.

C 0401.0100

Package to prepare 100 ml 1000 X stock solution

C 0401.0250

Package to prepare 250 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	μM
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	1.60	25.88
KI	0.80	4.81
MnSO <sub>4</sub> ·H <sub>2</sub> O	3.33	19.70
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	1.50	5.22

## MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	125.33	1.13
KH <sub>2</sub> PO <sub>4</sub>	400.00	2.94
KNO <sub>3</sub>	2830.00	27.99
MgSO <sub>4</sub>	90.27	0.75
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	463.00	3.50

Total concentration Micro and Macro elements: 3952.53 mg/l

## VITAMINS

	mg/l	μM
Glycine	2.00	26.64
Thiamine HCl	1.00	2.96
Pyridoxine HCl	0.50	2.43
Nicotinic acid	0.50	4.06

Total concentration Micro and Macro elements including vitamins: 3956.53 mg/l



Haworthia callus regeneration,  
Succulent Tissue Culture, The Netherlands

# CLC/IPOMOEA BASAL MEDIUM

## For Embryogenic Callus Growth (CP) and Embryo Development (EP)

Sweetpotato somatic embryo production is accomplished in two stages. Embryogenic callus is continuously proliferated by subculture on media containing 10  $\mu\text{M}$  2,4-D and 1  $\mu\text{M}$  6-BAP. Increasing the  $\text{K}^+$  concentration to 40-60 mM doubled the production of embryogenic callus, while the production of non embryogenic callus was reduced by 40%.

The development of embryos, triggered by the removal of 2,4-D and 6-BAP, was enhanced by decreasing ammonium ( $\text{NH}_4^+$ ) from 20 to 10 mM.

Cheé R. et al., Optimizing Embryogenic Callus and Embryo Growth of a synthetic seed system for Sweetpotato by varying media nutrient concentrations. J. Am. Soc. Hort. Sci. 117, 663 (1992).

### C 0228

CLC / Ipomoea

Embryogenic Callus Growth (CP medium)

Including Vitamins

C 0228.0001	1 l	(6.7 g)
C 0228.0010	10 l	(67.1 g)
C 0228.0050	50 l	(335.3 g)

### C 0229

CLC / Ipomoea

Embryo Development (EP) medium

Including Vitamins

C 0229.0001	1 l	(3.5 g)
C 0229.0010	10 l	(35.5 g)
C 0229.0050	50 l	(177.3 g)

### MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

### MACRO ELEMENTS, Embryo Development (EP)

	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	1900.00	18.79
$\text{MgSO}_4$	180.54	1.50
$\text{NH}_4\text{NO}_3$	800.40	10.00

Total concentration Micro and Macro elements (EP): 3452.49 mg/l

### MACRO ELEMENTS, Embryogenic Callus Growth (CP)

	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
KCl	2237.00	30.00
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	2022.00	20.00
$\text{MgSO}_4$	180.54	1.50
$\text{NH}_4\text{NO}_3$	1601.00	20.00

Total concentration Micro and Macro elements (CP): 6612.09 mg/l

### VITAMINS

	mg/l	$\mu\text{M}$
myo-Inositol	90.10	500.00
Nicotinic acid	1.23	10.00
Pyridoxine HCl	1.03	5.00
Thiamine HCl	1.69	5.00

Total concentration Micro and Macro elements (CP) including vitamins: 6706.14 mg/l

Total concentration Micro and Macro elements (EP) including vitamins: 3546.54 mg/l

# DE GREEF & JACOBS MEDIUM

Callus derived from leaf pieces of sugarbeet, exposed to a cold period of 3-9 weeks, could be regenerated into a normal plant after being returned to normal temperature. On media free of growth regulators, a regenerating callus could be formed with a high regeneration capacity.

De Greef W. and Jacobs M. In vitro culture of the sugarbeet: Description of a cell line with high regeneration capacity., Plant Science Letters., 17, 55-61 (1979).

## D 0205 DE GREEF AND JACOBS MEDIUM

### Micro and Macro elements

D 0205.0005	5 l	(18.9 g)
D 0205.0050	50 l	(188.5 g)

## D 0206 DE GREEF AND JACOBS MEDIUM

### Micro and Macro elements including Vitamins

D 0206.0005	5 l	(19.4 g)
D 0206.0050	50 l	(194.1 g)

## G 0403 DE GREEF & JACOBS VITAMIN MIXTURE

Package contains 11.20 g or 28.00 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre medium of the proper final vitamin concentration.

G 0403.0100	Package to prepare 100 ml 1000 X stock solution
G 0403.0250	Package to prepare 250 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.0025	0.01
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.0025	0.01
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	10.62	171.76
KI	1.58	9.54
MnSO <sub>4</sub> ·H <sub>2</sub> O	1.68	9.94
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.0025	0.01
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	1.06	3.69

## MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	226.50	2.04
KCl	600.00	8.05
KNO <sub>3</sub>	2000.00	19.78
MgSO <sub>4</sub>	244.33	2.03
NaH <sub>2</sub> PO <sub>4</sub>	250.00	2.08
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	400.00	3.03

Total concentration Micro and Macro elements: 3770.44 mg/l

## VITAMINS

	mg/l	µM
myo-Inositol	100.00	554.94
Nicotinic acid	1.00	8.12
Pyridoxine HCl	1.00	4.86
Thiamine HCl	10.00	29.65

Total concentration Micro and Macro elements including vitamins: 3882.44 mg/l



Potato tuberisation,  
SBW International BV, The Netherlands

# DKW/JUGLANS MEDIUM

The DKW medium has been defined for in vitro propagation of Paradox Walnut Rootstock (*Juglans hindsii* x *J. regia*) via nodal explants. The explants were placed on medium without growth-regulators for one week and subsequently on medium containing 6-BAP and IBA. Optimum shoot development was supported under 4.5  $\mu\text{M}$  6-BAP and 5 nM IBA. The basal ends of the tissue culture derived shoots were dipped in 5 mM IBA solution and subsequently rooted within 10 to 14 days in the greenhouse.

Driver, J.A., Kuniyuki, A.H. In Vitro Propagation of Paradox walnut Rootstock, Hort. Science, 19(4), August 1984.

## D 0246 DKW/JUGLANS MEDIUM

### Micro and Macro elements

D 0246.0001	1 l	(5.5 g)
D 0246.0005	5 l	(27.4 g)
D 0246.0010	10 l	(54.8 g)
D 0246.0025	25 l	(137.0 g)
D 0246.0050	50 l	(274.0 g)

## D 0247 DKW/JUGLANS MEDIUM

### Micro and Macro elements including Vitamins

D 0247.0001	1 l	(5.6 g)
D 0247.0005	5 l	(27.9 g)
D 0247.0010	10 l	(55.8 g)
D 0247.0025	25 l	(139.6 g)
D 0247.0050	50 l	(279.2 g)

## D 0414 DKW/JUGLANS VITAMIN MIXTURE

Package contains 10.50 g or 26.25 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre DKW medium of the proper final vitamin concentration.

D 0414.0100

Package to prepare 100 ml 1000 X stock solution

D 0414.0250

Package to prepare 250 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.25	1.00
FeNaEDTA	44.63	121.61
$\text{H}_3\text{BO}_3$	4.80	77.63
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	33.80	200.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.39	1.61
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	17.00	72.19

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	112.50	1.01
$\text{Ca}(\text{NO}_3)_2 \cdot 2\text{H}_2\text{O}$	1664.64	8.30
$\text{KH}_2\text{PO}_4$	265.00	1.95
$\text{K}_2\text{SO}_4$	1559.00	8.95
$\text{MgSO}_4$	361.49	3.00
$\text{NH}_4\text{NO}_3$	1416.00	17.70

Total concentration Micro and Macro elements: 5479.50 mg/l

## VITAMINS

	mg/l	$\mu\text{M}$
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	1.00	8.12
Thiamine HCl	2.00	5.93

Total concentration Micro and Macro elements including vitamins: 5584.50 mg/l



# ERIKSSON (ER) MEDIUM

The Eriksson medium was developed for cell suspension cultures of *Haplopappus gracilis*. An increase in the growth of cell suspensions, especially with small inocula, was achieved by a 10% reduction of the concentration MS microelements, except for Fe and Zn. Equimolar replacement of  $ZnSO_4 \cdot 7H_2O$  by  $ZnNa_2EDTA$  improved cell growth as well. MnEDTA and CoEDTA did not improve the growth of the cell culture. A reduction of Murashige and Skoog  $NH_4NO_3$  concentration of 1650 to 1200 mg/l and an increase in phosphate to 2.5 mM also stimulated cell growth.

Eriksson T., *Physiol. Plant*, 18, 976 (1965).

## E 0207 ERIKSSON (ER) MEDIUM

### Micro and Macro elements

E 0207.0001	1 l	(4.0 g)
E 0207.0010	10 l	(40.1 g)
E 0207.0050	50 l	(200.3 g)

## E 0208 ERIKSSON (ER) MEDIUM

### Micro and Macro elements including Vitamins

E 0208.0001	1 l	(4.0 g)
E 0208.0010	10 l	(40.1 g)
E 0208.0050	50 l	(200.5 g)

## E 0402 ERIKSSON (ER) VITAMIN MIXTURE

Package contains 0.35 g to prepare 100 ml of a 1000 X vitamin stock solution. Use 1 ml vitamin stock solution to prepare 1 litre Eriksson medium of the proper final vitamin concentration.

E 0402.0100

Package to prepare 100 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	μM
$CoCl_2 \cdot 6H_2O$	0.0025	0.01
$CuSO_4 \cdot 5H_2O$	0.0025	0.01
FeNaEDTA	36.70	100.00
$H_3BO_3$	0.63	10.19
$MnSO_4 \cdot H_2O$	1.69	10.00
$Na_2MoO_4 \cdot 2H_2O$	0.025	0.10
$ZnSO_4 \cdot 7H_2O$	9.15	31.80

## MACRO ELEMENTS

	mg/l	mM
$CaCl_2$	332.02	2.99
$KH_2PO_4$	340.00	2.50
$KNO_3$	1900.00	18.79
$MgSO_4$	180.54	1.50
$NH_4NO_3$	1200.00	14.99

Total concentration Micro and Macro elements: 4000.92 mg/l

## VITAMINS

	mg/l	μM
Glycine	2.00	26.64
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.50	1.48

Total concentration Micro and Macro elements including vitamins: 4004.42 mg/l



# GAMBORG B5 MEDIUM

The B5 medium has been defined for the growth of cell suspensions of soybean root cells in the presence of 2,4 D. Nitrate was required in a concentration of 20-30 mM. An addition of 2 mM ammonium sulphate led to an increase in cell growth.  $\text{NH}_4^+$  when added as the sole source of nitrogen, did not support growth. Similar results were obtained when  $\text{NH}_4\text{NO}_3$  was substituted for  $(\text{NH}_4)_2\text{SO}_4$ . However, ammonium ions depressed growth when the concentration exceeded 2 mM. Variations in the concentrations of phosphate, calcium and magnesium resulted in relatively minor changes in growth rate. Thiamine is known to be an essential nutrient for cell growth and is increased in concentration up to 10 mg/l.

Gamborg O.L., Miller R.A., Ojima K., Nutrient requirement of suspensions cultures of soybean root cells. *Exp. Cell Res.*, 50, 151 (1968).

## G 0209 GAMBORG B5 MEDIUM

### Micro and Macro elements

G 0209.0001	1 l	(3.1 g)
G 0209.0005	5 l	(15.3 g)
G 0209.0010	10 l	(30.5 g)
G 0209.0025	25 l	(76.3 g)
G 0209.0050	50 l	(152.6 g)

## G 0210 GAMBORG B5 MEDIUM

### Micro and Macro elements including Vitamins

G 0210.0001	1 l	(3.2 g)
G 0210.0005	5 l	(15.8 g)
G 0210.0010	10 l	(31.6 g)
G 0210.0025	25 l	(79.1 g)
G 0210.0050	50 l	(158.2 g)

## G 0415 GAMBORG B5 VITAMIN MIXTURE

Package contains 11.20 g or 28.00 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre medium of the final vitamin concentration.

G 0415.0100

Package to prepare 100 ml 1000 X stock solution

G 0415.0250

Package to prepare 250 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	3.00	48.52
KI	0.75	4.52
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	10.00	59.16
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	2.00	6.96

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	113.23	1.02
$\text{KNO}_3$	2500.00	24.73
$\text{MgSO}_4$	121.56	1.01
$\text{NaH}_2\text{PO}_4$	130.44	1.09
$(\text{NH}_4)_2\text{SO}_4$	134.00	1.01

Total concentration Micro and Macro elements: 3051.98 mg/l

## VITAMINS

	mg/l	$\mu\text{M}$
myo-Inositol	100.00	554.94
Nicotinic acid	1.00	8.12
Pyridoxine HCl	1.00	4.86
Thiamine HCl	10.00	29.65

Total concentration Micro and Macro elements including vitamins: 3163.98 mg/l



# GRESSHOFF & DOY (DBM2) MEDIUM

The medium defined by Gresshoff and Doy is developed for growth of haploid callus and plants of *Arabidopsis thaliana* cultured from the diploid anthers. The anthers were removed during the late prophase of meiosis, selecting a genotype favouring callus formation from dividing sporocytes on a high auxin - low kinetin concentration in a fully defined medium. Further differentiation was induced by transfer to a low auxin - high kinetin medium with a light-dark cycle. Haploid callus cultures of tomato, barley and *Vitis vinifera* have been cultured as well using this method.

Gresshoff P.M. et al., Haploid *Arabidopsis thaliana* callus and plants from anther culture. *Aust. J. Biol. Sci.*, 25, 259 (1972).

Gresshoff P.M. et al., Derivation of a haploid cell line from *Vitis vinifera* and the importance of the stage of meiotic development of anthers for haploid culture of this and other genera, *Z. Pflanzenphysiol.* 73, 132-141, (1974).

## G 0211 GRESSHOFF & DOY MEDIUM

### Micro and Macro elements

G 0211.0001	1 l	(2.6 g)
G 0211.0010	10 l	(26.3 g)
G 0211.0050	50 l	(131.5 g)

## G 0212 GRESSHOFF & DOY MEDIUM

### Micro and Macro elements including Vitamins

G 0212.0001	1 l	(2.7 g)
G 0212.0010	10 l	(27.5 g)
G 0212.0050	50 l	(137.3 g)

## G 0404 GRESSHOFF & DOY (DBM2) VITAMIN MIXTURE

Package contains 11.6 g or 29.0 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre Gresshoff & Doy medium of the proper final vitamin concentration.

G 0404.0100

Package to prepare 100 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	µM
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	0.30	4.85
KI	0.80	4.82
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	1.00	5.92
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.025	0.10
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	0.30	1.04

## MACRO ELEMENTS

	mg/l	mM
$\text{Ca}(\text{NO}_3)_2 \cdot 2\text{H}_2\text{O}$	208.81	1.04
KCl	65.00	0.87
$\text{KH}_2\text{PO}_4$	300.00	2.20
$\text{KNO}_3$	1000.00	9.89
$\text{MgSO}_4$	17.09	0.14
$\text{NH}_4\text{NO}_3$	1000.00	12.49

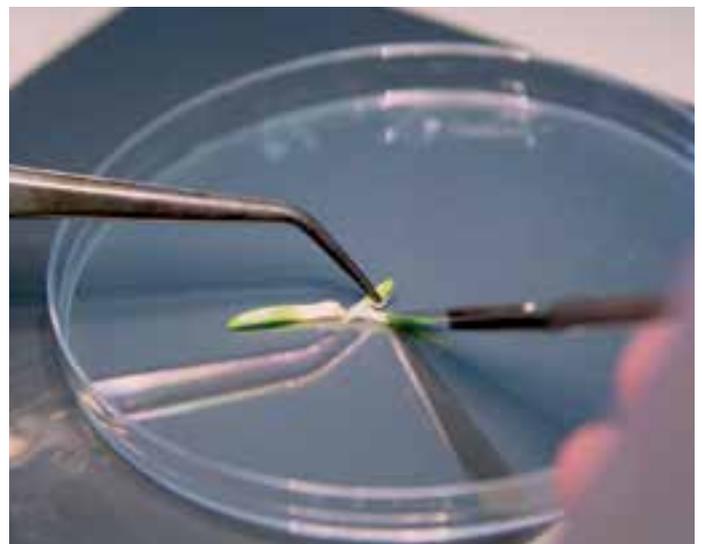
Total concentration Micro and Macro elements: 2630.10 mg/l

## VITAMINS

	mg/l	µM
Glycine	4.00	53.28
myo-Inositol	100.00	554.94
Nicotinic acid	1.00	8.12
Pyridoxine HCl	1.00	4.86
Thiamine HCl	10.00	29.65

Total concentration Micro and Macro elements including vitamins: 2746.10 mg/l

Preparation of material before the real meristem is isolated.  
Iribov BV the Netherlands,



# HELLER MEDIUM

Heller R., Ann. Sci. Nat. Bot. Biol. Veg. 11th Ser., 14, 1 (1953).

## H 0213 Heller medium

### Micro and Macro elements

H 0213.0001	1 l	(1.6 g)
H 0213.0005	5 l	(8.2 g)
H 0213.0025	25 l	(41.1 g)

## MICRO ELEMENTS

	mg/l	µM
$\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$	0.054	0.22
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.03	0.12
$\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$	1.00	3.70
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.015	0.09
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	0.08	0.47
$\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.14
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	1.00	3.48

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	56.62	0.51
KCl	750.00	10.06
$\text{MgSO}_4$	121.56	1.01
$\text{NaNO}_3$	600.00	7.06
$\text{NaH}_2\text{PO}_4$	108.70	0.91

Total concentration Micro and Macro elements: 1645.29 mg/l



### Virus elimination of plants.

Typically meristems of 0,1-0,2 mm are isolated. Sometimes pretreatment is given (temperature treatment or application chemicals for virus suppression. Development of culture of micro-explants is in most cases the critical factor.

Iribov B.V.,  
Middenweg 591b  
1704 BH Heerhugowaard  
The Netherlands

 **Iribov**  
breeding support laboratory

# KAO & MICHAYLUK MEDIUM

The medium defined by Kao and Michayluk was designed to grow cells and protoplasts of *Vicia hajastana* at a very low population density in liquid media. The inability of the plant cells to grow at a very low population density may be caused by excessive diffusion of metabolic intermediates into the medium, resulting in their dilution in the cell to a level below that required for survival. *Vicia* cells were able to grow at an initial population density of 1-2 cells/ml when the mineral salt medium was enriched with organic acids, sugars, sugar alcohols, growth regulators, amino acids and other organic compounds. The percentage of cell division could be increased by raising the concentration of  $\text{CaCl}_2$  from 1 mM, as in Gamborg B5, to 5 mM. Calcium may play an important role in the process of cell division because of its ability to preserve the structural and functional integrity of plant cell membranes.

Kao K.N., O.L. Gamborg et al., The effects of sugars and inorganic salts on cell regeneration and sustained division in plant protoplasts. Colloques internationaux C.N.R.S., 212, Protoplastes et fusion de cellules somatiques végétales.

Kao K.N. and Michayluk M.R., Nutritional requirements for growth of *Vicia hajastana* cells and protoplasts at a very low population density in liquid media. *Planta (Berl.)*, 126, 105 (1975).

## K 0214 KAO & MICHAYLUK MEDIUM

### Micro and Macro elements

K 0214.0001	1 l	(3.6 g)
K 0214.0005	5 l	(18.1 g)
K 0214.0010	10 l	(36.2 g)

### MICRO ELEMENTS

	mg/l	μM
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	3.00	48.52
KI	0.75	4.52
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	10.00	59.17
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	2.00	6.96

### MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	453.00	4.08
KCl	300.00	4.02
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	1900.00	18.79
$\text{MgSO}_4$	146.84	1.22
$\text{NH}_4\text{NO}_3$	600.00	7.50

Total concentration Micro and Macro elements: 3622.59 mg/l

Willemsen en Bourgondiën B.V., The Netherlands



# KNUDSON C ORCHID MEDIUM, MOREL MODIFICATION

Morel, G.M., Cymb. Soc. News, 20, (1965).

## K 0215 KNUDSON C ORCHID MEDIUM, MOREL MODIFICATION

### Micro and Macro elements

K 0215.0001	1 l	(1.9 g)
K 0215.0005	5 l	(9.5 g)
K 0215.0010	10 l	(18.9 g)
K 0215.0025	25 l	(47.4 g)
K 0215.0050	50 l	(94.7 g)

Willemsen en Bourgondiën B.V., The Netherlands

### MICRO ELEMENTS

	mg/l	µM
$\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$	25.00	89.92
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	5.68	33.61

### MACRO ELEMENTS

	mg/l	mM
$\text{Ca}(\text{NO}_3)_2$	241.30	1.43
KCl	250.00	3.35
$\text{KH}_2\text{PO}_4$	250.00	1.84
$\text{MgSO}_4$	122.15	1.02
$\text{NH}_4\text{NO}_3$	500.00	6.25
$(\text{NH}_4)_2\text{SO}_4$	500.00	3.78

Total concentration Micro and Macro elements: 1894.13 mg/l



# LINDEMANN ORCHID MEDIUM

Lindemann E.G.P., Amercan. Orch. Bull 39, 1002 (1970).

## L 0216 LINDEMANN ORCHID MEDIUM

### Micro and Macro elements

L 0216.0001	1 l	(2.6 g)
L 0216.0010	10 l	(26.0 g)
L 0216.0050	50 l	(129.9 g)

### MICRO ELEMENTS

	mg/l	µM
$\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$	0.56	2.32
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.02	0.08
FeCitrate	4.40	
$\text{H}_3\text{BO}_3$	1.01	16.34
KI	0.10	0.60
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	0.05	0.31
$\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$	0.03	0.13
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	0.57	1.98

### MACRO ELEMENTS

	mg/l	mM
$\text{Ca}(\text{NO}_3)_2$	347.20	2.12
$\text{KH}_2\text{PO}_4$	135.00	0.99
KCl	1050.00	14.08
$\text{MgSO}_4$	58.98	0.49
$(\text{NH}_4)_2\text{SO}_4$	1000.00	7.57

Total concentration Micro and Macro elements: 2597.92 mg/l

Willemsen en Bourgondiën B.V., The Netherlands



# LINSMAIER & SKOOG MEDIUM

Linsmaier and Skoog have made a systematic study of the organic requirements of Tobacco cultures in addition to the studies of mineral requirements done by Murashige and Skoog. It was found that of all MS vitamins only Thiamine and Inositol are essential. The optimum concentration for Thiamine HCl was 0.4 mg/l (MS 0.1 mg/l). At a lower concentration growth decreased and the cells became necrotic after 4 weeks. Inositol also had a very stimulatory effect on the cell growth but was not as essential as Thiamine. All other Murashige & Skoog vitamins were not required for cell growth and could be omitted without any disadvantageous effect. Folic acid, p-Aminobenzoic acid, l-Glutamic acid and Ascorbic acid also had a positive influence on cell growth of *Nicotiana tabacum*, however the effect was much less than that of Thiamine and Inositol.

Linsmaier E.M. and Skoog F., *Physiol. Plantarum*, 18, 100, (1965).

## L 0230 LINSMAIER & SKOOG MEDIUM

### Micro and Macro elements including Vitamins

L 0230.0001	1 l	(4.4 g)
L 0230.0005	5 l	(22.0 g)
L 0230.0010	10 l	(44.0 g)
L 0230.0025	25 l	(110.1 g)
L 0230.0050	50 l	(220.1 g)

## L 0406 LINSMAIER & SKOOG VITAMIN MIXTURE

Package contains 10.04 g or 25.10 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre Linsmaier & Skoog medium of the proper final vitamin concentration.

L 0406.0100

Package to prepare 100 ml 1000 X stock solution

L 0406.0250

Package to prepare 250 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	332.02	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

Total concentration Micro and Macro elements: 4302.09 mg/l

## VITAMINS

	mg/l	µM
myo-Inositol	100.00	554.94
Thiamine HCl	0.40	1.19

Total concentration Micro and Macro elements including vitamins: 4402.49 mg/l

Willemsen en Bourgondiën B.V., The Netherlands



# LITVAY MEDIUM

Litvay's medium is composed for the in vitro culture of cell suspension of *Daucus carotus* and finally for *Pinus taeda* L. An increase of the phosphate concentration from 0.5 mM to 2.5 mM was essential for improved cell growth and embryogenesis. Increasing the magnesium concentration from 0.75 mM to 7.5 mM and decreasing the calcium concentration from 1.5 mM to 0.15 mM was also of positive influence. However, these alterations are not as drastical as the improvement by the enrichment of the medium by additional phosphate.

Litvay J.D., Verma D.C., Morris A.J., *Plant Cell Rep.*, 4, 325 (1985).

## L 0217 LITVAY MEDIUM

### Micro and Macro elements

L 0217.0001	1 l	(5.0 g)
L 0217.0010	10 l	(49.5 g)
L 0217.0050	50 l	(247.4 g)

## L 0218 LITVAY MEDIUM

### Micro and Macro elements including Vitamins

L 0218.0001	1 l	(5.1 g)
L 0218.0010	10 l	(50.5 g)
L 0218.0050	50 l	(252.4 g)

## L 0407 LITVAY VITAMIN MIXTURE

Package contains 10.07 g or 25.18 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre Litvay medium of the proper final vitamin concentration.

L 0407.0100	Package to prepare 100 ml 1000 X stock solution
L 0407.0250	Package to prepare 250 ml 1000 X stock solution

Astilbe propagation,  
SBW International BV, The Netherlands

## MICRO ELEMENTS

	mg/l	µM
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.125	0.53
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.50	2.00
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	31.00	501.37
KI	4.15	25.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	21.00	124.25
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	1.25	5.17
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	43.00	149.54

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	16.61	0.15
$\text{KH}_2\text{PO}_4$	340.00	2.50
$\text{KNO}_3$	1900.00	18.79
$\text{MgSO}_4$	903.38	7.51
$\text{NH}_4\text{NO}_3$	1650.00	20.61

Total concentration Micro and Macro elements: 4947.72 mg/l

## VITAMINS

	mg/l	µM
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.10	0.49
Thiamine HCl	0.10	0.30

Total concentration Micro and Macro elements including vitamins: 5048.42 mg/l



# McCOWN WOODY PLANT MEDIUM

Lloyd G. and McCown. Commercially-feasible micropropagation of mountain laurel, *Kalmia latifolia*, by use of shoot-tip culture. *B., Int. Plant Prop. Soc. Proc.* 30, 421 (1980).

## M 0219 McCOWN WOODY PLANT MEDIUM

### Micro and Macro elements

M 0219.0001	1 l	(2.4 g)
M 0219.0005	5 l	(11.8 g)
M 0219.0010	10 l	(23.6 g)
M 0219.0025	25 l	(59.0 g)
M 0219.0050	50 l	(117.9 g)

## M 0220 McCOWN WOODY PLANT MEDIUM

### Micro and Macro elements including Vitamins

M 0220.0001	1 l	(2.5 g)
M 0220.0005	5 l	(12.3 g)
M 0220.0010	10 l	(24.6 g)
M 0220.0025	25 l	(61.6 g)
M 0220.0050	50 l	(123.13 g)

## M 0408 McCOWN WOODY PLANT VITAMIN MIXTURE / MURASHIGE & SKOOG MODIFIED VITAMIN MIXTURE

Package contains 10.4 g or 26.0 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre McCown Woody Plant medium of the proper final vitamin concentration.

M 0408.0100

Package to prepare 100 ml 1000 X stock solution

M 0408.0250

Package to prepare 250 ml 1000 X stock solution

Anthurium propagation,  
SBW International BV, The Netherlands

## MICRO ELEMENTS

	mg/l	µM
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.25	1.00
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	6.20	100.27
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	22.30	131.94
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	72.50	0.65
$\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$	471.26	2.35
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{K}_2\text{SO}_4$	990.00	5.68
$\text{MgSO}_4$	180.54	1.50
$\text{NH}_4\text{NO}_3$	400.00	5.00

Total concentration Micro and Macro elements: 2358.60 mg/l

## VITAMINS

	mg/l	µM
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	1.00	2.96

Total concentration Micro and Macro elements including vitamins:  
2462.60 mg/l



# MURASHIGE & SKOOG MEDIUM

MS medium is the most used tissue culture medium, of which many variations have been developed. The medium is derived from White's medium and originally developed for the cultivation of *Nicotiana tabacum* calli. Compared to the White medium, the concentration of all ingredients is increased. An increase to 50-60 mM nitrogen stimulated the growth of *Nicotiana* cells significantly, however a concentration of 80 mM and higher was clearly disadvantageous to the cells. The increase of all other elements, especially the macro elements, also stimulated the growth of the calli. Due to the high concentration of minerals, MS medium is a very rich and saline medium and can be too salty to certain plant species. To avoid this problem, MS is often used with the micro elements in full concentration, but with the macro elements in respectively half or threequarter of the concentration as originally described by the authors. Sometimes the original MS vitamins are replaced by the vitamins of Linsmaier and Gamborg B5 medium regarding the higher concentration of Thiamine in relation to the requirement of this vitamin by plants.

Murashige T. and Skoog F., *Physiol. Plant*, 15, 473 (1962).

## M 0221 MURASHIGE & SKOOG MEDIUM

### Micro and Macro elements

M 0221.0001	1 l	(4.3 g)
M 0221.0005	5 l	(21.5 g)
M 0221.0010	10 l	(43.0 g)
M 0221.0025	25 l	(107.6 g)
M 0221.0050	50 l	(215.1 g)

## M 0222 MURASHIGE & SKOOG MEDIUM

### Micro and Macro elements including Vitamins

M 0222.0001	1 l	(4.4 g)
M 0222.0005	5 l	(22.0 g)
M 0222.0010	10 l	(44.1 g)
M 0222.0025	25 l	(110.1 g)
M 0222.0050	50 l	(220.3 g)

## M 0409 MURASHIGE & SKOOG VITAMIN MIXTURE

Package contains 10.31 g or 25.80 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 liter MS medium of the proper final vitamin concentration.

M 0409.0100

Package to prepare 100 ml 1000 X stock solution

M 0409.0250

Package to prepare 250 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	μM
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

## MACRO ELEMENTS

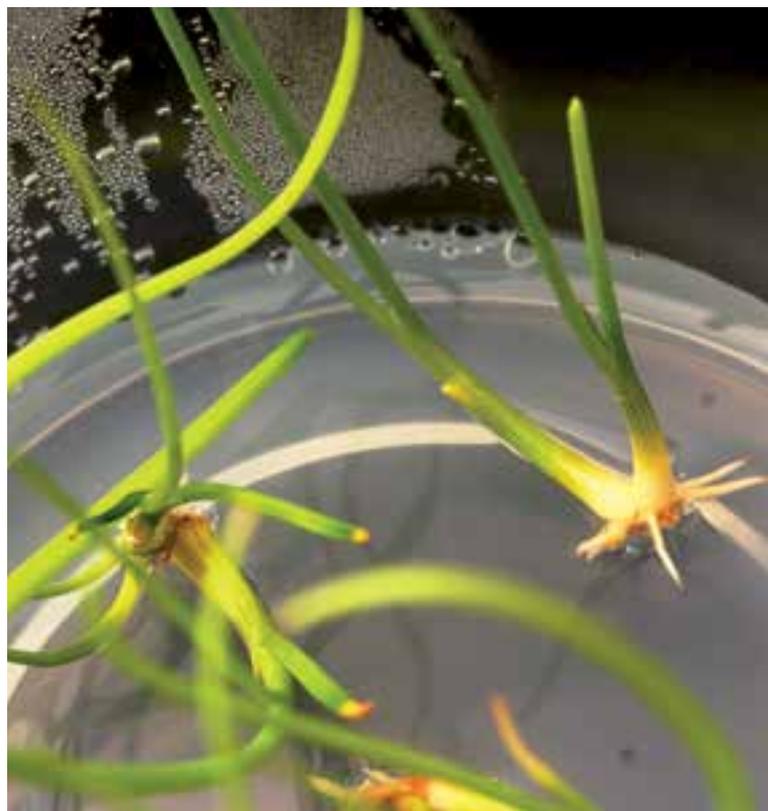
	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	1900.00	18.79
$\text{MgSO}_4$	180.54	1.50
$\text{NH}_4\text{NO}_3$	1650.00	20.61

Total concentration Micro and Macro elements: 4302.09 mg/l

## VITAMINS

	mg/l	μM
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.10	0.30

Total concentration Micro and Macro elements including vitamins: 4405.19 mg/l



Onion micropropagation.

Ing. Bernadette van Kronenberg and Dr. Olga Scholten,  
Wageningen UR Plant Breeding

# MURASHIGE & SKOOG MEDIUM

## including Modified Vitamins

### M 0245 MURASHIGE & SKOOG MEDIUM

#### Micro and Macro elements including Modified Vitamins

M 0245.0001	1 l	(4.4 g)
M 0245.0010	10 l	(44.1 g)
M 0245.0050	50 l	(220.3 g)

### M 0408 MURASHIGE & SKOOG MODIFIED VITAMIN MIXTURE/ McCOWN WOODY PLANT VITAMIN MIXTURE

Package contains 10.40 g or 26.00 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre MS medium of the proper final vitamin concentration.

M 0408.0100

Package to prepare 100 ml 1000 X stock solution

M 0408.0250

Package to prepare 250 ml 1000 X stock solution

Apple at the start of a subculture cycle.

Dr. Geert-Jan de Klerk,

Wageningen UR Plant Breeding

## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

## MACRO ELEMENTS

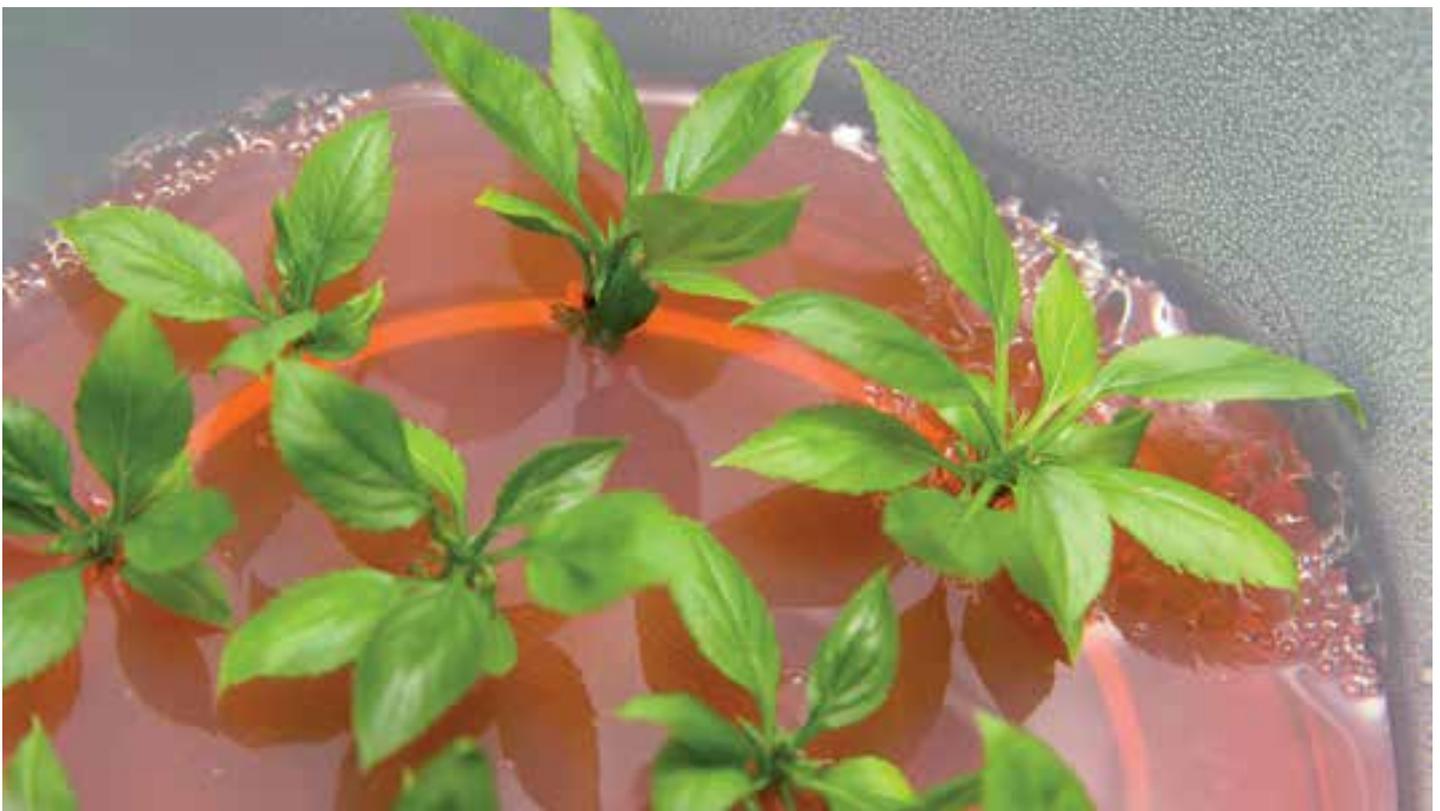
	mg/l	mM
CaCl <sub>2</sub>	332.02	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

Total concentration Micro and Macro elements: 4302,09 mg/l

## VITAMINS, 10x concentration of Thiamine HCl

	mg/l	µM
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	1.00	2.96

Total concentration Micro and Macro elements including vitamins: 4406.09 mg/l



# MURASHIGE & SKOOG MEDIUM

including Gamborg B5 vitamins

## M 0231 MURASHIGE & SKOOG MEDIUM

Micro and Macro elements including  
Gamborg B5 Vitamins

M 0231.0001	1 l	(4.4 g)
M 0231.0005	5 l	(22.1 g)
M 0231.0010	10 l	(44.1 g)
M 0231.0025	25 l	(110.4 g)
M 0231.0050	50 l	(220.7 g)

## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	332.02	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

Total concentration Micro and Macro elements: 4302.09 mg/l

## VITAMINS, Gamborg B5

	mg/l	µM
myo-Inositol	100.00	554.94
Nicotinic acid	1.00	8.12
Pyridoxine HCl	1.00	4.86
Thiamine HCl	10.00	29.65

Total concentration Micro and Macro elements including vitamins:  
4414.09 mg/l



Transgenic apple scion grafted on a non-transgenic rootstock.

Dr. Frans Krens.  
Wageningen UR Plant Breeding

# MURASHIGE & SKOOG MEDIUM

## including Nitsch vitamins

To improve the growth of Geranium species in tissue culture the original vitamins as described by Murashige and Skoog in 1962 are replaced by the vitamins as described by Nitsch et al in 1969.

Nitsch J.P. and Nitsch C., Science 169, 85 (1969).

### M 0256 MURASHIGE & SKOOG MEDIUM

Micro and Macro elements including Nitsch vitamins

M 0256.0001	1 l	(4.4 g)
M 0256.0010	10 l	(44.1 g)
M 0256.0050	50 l	(220.5 g)



### MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

### MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	332.02	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

Total concentration Micro and Macro elements: 4302,09 mg/ml

### VITAMINS, Nitsch

	mg/l	µM
Biotin	0.05	0.21
Folic acid	0.50	1.13
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	5.00	40.62
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.50	1.48

Total concentration Micro and Macro elements including vitamins:  
4410.64 mg/l

Multiple transgenic apple scions grafted on non-transgenic rootstocks ready for a greenhouse scab-resistance assay.

Dr. Frans Krens.  
Wageningen UR Plant Breeding

# MURASHIGE & SKOOG MEDIUM

## including MES Buffer

To prevent acidification of the medium during cultivation in this formulation of Murashige and Skoog medium MES (2-MorpholinoEthaneSulfonic acid (cat. no. M 1501)) has been added in a concentration of 500 mg/l. Applied as a buffer in Plant Tissue Culture media, MES is non toxic for plant tissue and plant cells and makes almost no interactions with inorganic cations present in the medium. MES is an excellent buffer for use in Plant Tissue Culture media, because of high buffer capacity and its pH range of 5.5 - 6.7.

### M 0254 MURASHIGE & SKOOG MEDIUM

#### Micro and Macro elements including MES Buffer

M 0254.0001	1 l	(4.8 g)
M 0254.0010	10 l	(48.0 g)
M 0254.0050	50 l	(240.1 g)

### M 0255 MURASHIGE & SKOOG MEDIUM

#### Micro and Macro elements incl. Vitamins and MES Buffer

M 0255.0001	1 l	(4.9 g)
M 0255.0010	10 l	(49.1 g)
M 0255.0050	50 l	(245.3 g)



## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	332.02	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

## BUFFER

	mg/l	mM
MES	500.00	2.35

Total concentration Micro and Macro elements including MES buffer:  
4802.09 mg/l

## VITAMINS

	mg/l	µM
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxin HCl	0.50	2.43
Thiamine HCl	0.10	0.30

Total concentration Micro and Macro elements including MES buffer  
and vitamins: 4905.19 mg/l

Flowers of transgenic *Crambe abyssinica* (fam. Cruciferae) plants.

Dr. Frans Krens.  
Wageningen UR Plant Breeding

# MURASHIGE & SKOOG MEDIUM

## MODIFICATION No. 1: 1/2 CONCENTRATION MACRO ELEMENTS

### M 0232 MURASHIGE & SKOOG MEDIUM MODIFICATION

#### No. 1 A

##### Micro and 1/2 concentration Macro elements

M 0232.0001	1 l	(2.2 g)
M 0232.0010	10 l	(21.8 g)
M 0232.0050	50 l	(109.2 g)

### M 0233 MURASHIGE & SKOOG MEDIUM MODIFICATION

#### No. 1 B

##### Micro and 1/2 concentration Macro elements

##### including Vitamins

M 0233.0001	1 l	(2.3 g)
M 0233.0010	10 l	(22.9 g)
M 0233.0050	50 l	(114.3 g)

Seeds set after selfing on transgenic *Crambe abyssinica* plants.

Dr. Frans Krens.  
Wageningen UR Plant Breeding

### MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

### MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	166.00	1.50
KH <sub>2</sub> PO <sub>4</sub>	85.00	0.63
KNO <sub>3</sub>	950.00	9.40
MgSO <sub>4</sub>	87.86	0.73
NH <sub>4</sub> NO <sub>3</sub>	825.00	10.30

Total concentration Micro and Macro elements: 2183.39 mg/l

### VITAMINS

	mg/l	µM
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.10	0.30

Total concentration Micro and Macro elements including vitamins:  
2286.49 mg/l



# MURASHIGE & SKOOG MEDIUM

## MODIFICATION No. 2: 3/4 CONCENTRATION MACRO ELEMENTS

### M 0234 MURASHIGE & SKOOG MEDIUM MODIFICATION

No. 2 A

Micro and 3/4 concentration Macro elements

M 0234.0001	1 l	(3.2 g)
M 0234.0010	10 l	(32.4 g)
M 0234.0050	50 l	(162.2 g)

### M 0235 MURASHIGE & SKOOG MEDIUM MODIFICATION

No. 2 B

Micro and 3/4 concentration Macro elements including Vitamins

M 0235.0001	1 l	(3.3 g)
M 0235.0010	10 l	(33.5 g)
M 0235.0050	50 l	(167.4 g)

### MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

### MACRO ELEMENTS, 3/4 concentration

	mg/l	mM
CaCl <sub>2</sub>	249.02	2.24
KH <sub>2</sub> PO <sub>4</sub>	127.50	0.94
KNO <sub>3</sub>	1425.00	14.09
MgSO <sub>4</sub>	136.01	1.13
NH <sub>4</sub> NO <sub>3</sub>	1237.50	15.46

Total concentration Micro and Macro elements: 3244.56 mg/l

### VITAMINS

	mg/l	µM
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.10	0.30

Total concentration Micro and Macro elements including vitamins: 3347.66 mg/l

Echeveria micropropagation,  
Succulent Tissue Culture, The Netherlands



# MURASHIGE & SKOOG MEDIUM

## MODIFICATION No. 3: 1/2 CONCENTRATION $\text{NH}_4\text{NO}_3$ and $\text{KNO}_3$

### M 0236 MURASHIGE & SKOOG MEDIUM MODIFICATION

#### No. 3 A

##### Micro and Macro elements

##### 1/2 concentration $\text{NH}_4\text{NO}_3$ and $\text{KNO}_3$

M 0236.0001	1 l	(2.5 g)
M 0236.0010	10 l	(25.3 g)
M 0236.0050	50 l	(126.4 g)

### M 0237 MURASHIGE & SKOOG MEDIUM MODIFICATION

#### No. 3 B

##### Micro and Macro elements

##### 1/2 concentration $\text{NH}_4\text{NO}_3$ and $\text{KNO}_3$ including Vitamins

M 0237.0001	1 l	(2.6 g)
M 0237.0010	10 l	(26.3 g)
M 0237.0050	50 l	(131.5 g)

Variegated *Haworthia* micropropagation,  
Succulent Tissue Culture, The Netherlands

### MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

### MACRO ELEMENTS, 1/2 concentration $\text{NH}_4\text{NO}_3$ and $\text{KNO}_3$

	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	950.00	9.40
$\text{MgSO}_4$	180.54	1.50
$\text{NH}_4\text{NO}_3$	825.00	10.30

Total concentration Micro and Macro elements: 2527.09 mg/l

### VITAMINS

	mg/l	$\mu\text{M}$
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.10	0.30

Total concentration Micro and Macro elements including vitamins:  
2630.19 mg/l



# MURASHIGE & SKOOG MEDIUM

## MODIFICATION No. 4: $\text{NH}_4\text{NO}_3$ Free

### M 0238 MURASHIGE & SKOOG MODIFICATION No. 4

#### $\text{NH}_4\text{NO}_3$ Free

M 0238.0001	1 l	(2.7 g)
M 0238.0010	10 l	(26.5 g)
M 0238.0050	50 l	(132.6 g)

### MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

### MACRO ELEMENTS, $\text{NH}_4\text{NO}_3$ Free

	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	1900.00	18.79
$\text{MgSO}_4$	180.54	1.50

Total concentration Micro and Macro elements: 2652.09 mg/l

# MURASHIGE & SKOOG MEDIUM

## MODIFICATION No. 5: $\text{NH}_4\text{NO}_3$ replaced by $\text{NaNO}_3$

### M 0239 MURASHIGE & SKOOG MEDIUM MODIFICATION

#### No. 5 $\text{NH}_4\text{NO}_3$ replaced by $\text{NaNO}_3$

M 0239.0001	1 l	(4.4 g)
M 0239.0010	10 l	(44.0 g)
M 0239.0050	50 l	(220.2 g)

### MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

### MACRO ELEMENTS, $\text{NH}_4\text{NO}_3$ replaced by $\text{NaNO}_3$

	mg/l	mM
$\text{CaCl}_2$	332.00	2.99
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	1900.00	18.79
$\text{MgSO}_4$	180.54	1.50
$\text{NaNO}_3$	1751.00	20.60

Total concentration Micro and Macro elements: 4403.07 mg/l

# MURASHIGE & SKOOG MEDIUM

## FINER & NAGASAWA MODIFICATION (1988):

(1.6 x concentration of  $\text{KNO}_3$  / 0.5 x concentration of  $\text{NH}_4\text{NO}_3$ )

A rapidly growing, maintainable, embryogenic suspension culture of *Glycine max.* could be generated in a revised version of MS medium. Highly embryogenic callus was cultivated in MS medium with Gamborg B5 vitamins and 5 mg/l 2,4-D. Substitution of MS nitrogen with 10 mM  $\text{NH}_4\text{NO}_3$  and 30 mM  $\text{KNO}_3$  plus 15 mM Glutamine or 5 mM Asparagine improved the growth of the calli suspension.

Finer J.J., and Nagasawa A, Development of an embryogenic suspension culture of soybean (*Glycine max.* Merrill). *Plant Cell, Tissue and Organ Culture*, 15, 125, (1988).

### M 0240 MURASHIGE & SKOOG MEDIUM FINER & NAGASAWA MODIFICATION

#### Micro and Macro elements

M 0240.0001	1 l	(4.6 g)
M 0240.0010	10 l	(46.1 g)
M 0240.0050	50 l	(230.4 g)

#### MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	100.00
FeNaEDTA	36.70	100.27
$\text{H}_3\text{BO}_3$	6.20	0.10
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

#### MACRO ELEMENTS, (1.6 x concentration of $\text{KNO}_3$ / 0.5 x concentration of $\text{NH}_4\text{NO}_3$ )

	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	3030.00	29.97
$\text{MgSO}_4$	180.54	1.50
$\text{NH}_4\text{NO}_3$	825.00	10.30

Total concentration Micro and Macro elements: 4607.09 mg/l

Willemsen en Bourgondiën B.V., The Netherlands



# MURASHIGE & SKOOG MEDIUM

## van der SALM MODIFICATION (1994)

### FeNaEDTA replaced by FeEDDHA as iron source

Fe-EDDHA is a highly stable chelate providing a source of iron that is easily absorbed by plants. In vitro propagation of the rose rootstock 'Moneyway', on Murashige & Skoog and Quirin & LePoivre medium resulted, despite good growth, after three weeks in chlorosis of newly formed leaves and was correlated with a lower chlorophyll content of shoots. Replacement of FeNaEDTA by FeEDDHA resulted in the development of green shoots for more than three months.

L. Moneyway, van der Salm T.M.P. et al., Importance of the iron chelate formula for micropropagation of *Rosa hybrida* Plant Cell Tiss. and Organ Cult, 37: 73-77, 1994

### M 0241 MURASHIGE & SKOOG MEDIUM van der SALM MODIFICATION

#### Micro and Macro elements

M 0241.0001	1 l	(4.4 g)
M 0241.0010	10 l	(43.6 g)
M 0241.0050	50 l	(218.1 g)

### M 0242 MURASHIGE & SKOOG MEDIUM van der SALM MODIFICATION

#### Micro and Macro elements including Vitamins

M 0242.0001	1 l	(4.5 g)
M 0242.0010	10 l	(44.6 g)
M 0242.0050	50 l	(223.2 g)

*Echeveria* micropropagation,  
Succulent Tissue Culture, The Netherlands

## MICRO ELEMENTS

	mg/l	µM
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeEDDHA	96.00	
$\text{H}_3\text{BO}_3$	6.20	100.27
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	332.02	2.99
$\text{KH}_2\text{PO}_4$	170.00	1.25
$\text{KNO}_3$	1900.00	18.79
$\text{MgSO}_4$	180.54	1.50
$\text{NH}_4\text{NO}_3$	1650.00	20.61

Total concentration Micro and Macro elements: 4361.39 mg/l

## VITAMINS

	mg/l	µM
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	0.50	4.06
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.10	0.30

Total concentration Micro and Macro elements including vitamins: 4464.49 mg/l



# MURASHIGE & SKOOG MEDIUM

## SYNGONIUM STAGE I & II

A procedure for clonal multiplication of *Cordyline terminalis*, *Dracena godseffian*, *Scindapsus aureus* and *Syngonium podophyllum* was established using MS minerals, LS vitamins and 3% sucrose. The optimum for 2-iP, kinetin and IAA was determined for each plant species. Addition of Phosphate increased the multiplication rate significantly. Adenine sulphate had a repressive action on shoot multiplication of *Syngonium* and was omitted from the medium.

Murashige T. and Miller L.R., *In Vitro*, 12, 796, (1976).

### M 0243 MURASHIGE & MILLER MEDIUM SYNGONIUM STAGE I & II

M 0243.0001	1 l	(4.7 g)
M 0243.0010	10 l	(47.0 g)
M 0243.0050	50 l	(234.9 g)

Hardening of TC plants. Compartment with first fase after tissue culture. Humidity controlled with fog system.

Cosmo Plant, joint hardening facility of Iribov, Allplant and Maatschap Holtmaat.

### MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

### MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	332.02	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NaH <sub>2</sub> PO <sub>4</sub> .anhydrous	295.41	2.15
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

Total concentration Micro and Macro elements: 4597.50mg/l

### VITAMINS

	mg/l	µM
myo-Inositol	100.00	554.94
Thiamine HCl	0.40	1.19

Total concentration Micro and Macro elements including vitamins: 4697.90 mg/l



# MURASHIGE & MILLER MEDIUM

## SHOOT MULTIPLICATION MEDIUM B

Huang L.C. and Murashige T., TCA Manual, 3 (1), 539 (1976).

### M 0244 MURASHIGE & SKOOG MEDIUM SHOOT MULTIPLICATION B

M 0244.0001	1 l	(4.5 g)
M 0244.0010	10 l	(45.3 g)
M 0244.0050	50 l	(226.6 g)

### MICRO ELEMENTS

	mg/l	μM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.83	5.00
MnSO <sub>4</sub> ·H <sub>2</sub> O	16.90	100.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

### MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	332.02	2.99
KH <sub>2</sub> PO <sub>4</sub>	170.00	1.25
KNO <sub>3</sub>	1900.00	18.79
MgSO <sub>4</sub>	180.54	1.50
NaH <sub>2</sub> PO <sub>4</sub> ·anhydrous	128.40	1.07
NH <sub>4</sub> NO <sub>3</sub>	1650.00	20.61

Total concentration Micro and Macro elements: 4430.49 mg/l

### VITAMINS

	mg/l	μM
myo-Inositol	100.00	554.94
Thiamine HCl	0.40	1.19

Total concentration Micro and Macro elements including vitamins: 4530.89 mg/l

Agavaceae micropropagation,  
Succulent Tissue Culture, The Netherlands



# NITSCH MEDIUM

The medium defined by Nitsch was used in the production of haploid plants of various species of *Nicotiana* raised from pollen grains. In this procedure, pollen that were still uninucleate were isolated and then cultured in vitro. Some pollen grains proliferate into embryo-like structures that develop in stages similar to those of zygotic embryos. The plantlets matured and flowered profusely, but did not set seed.

Nitsch J.P. and Nitsch C., Haploid plants from pollen grains, *Science* 169, 85 (1969).

Nitsch J.P., Experimental androgenesis in *Nicotiana*, *Phytomorphology* 19, 389 (1969).

## N 0223 NITSCH MEDIUM

### Micro and Macro elements

N 0223.0001	1 l	(2.1 g)
N 0223.0005	5 l	(10.4 g)
N 0223.0010	10 l	(20.7 g)
N 0223.0025	25 l	(51.8 g)
N 0223.0050	50 l	(103.5 g)

## N 0224 NITSCH MEDIUM

### Micro and Macro elements including Vitamins

N 0224.0001	1 l	(2.2 g)
N 0224.0005	5 l	(10.9 g)
N 0224.0010	10 l	(21.8 g)
N 0224.0025	25 l	(54.5 g)
N 0224.0050	50 l	(108.9 g)

## N 0410 NITSCH VITAMIN MIXTURE

Package contains 10.85 or 27.13 g vitamins to prepare 100 ml or 250 ml of a 1000 X vitamin stock solution.

Use 1 ml vitamin stock solution to prepare 1 litre Nitsch medium of the proper final vitamin concentration.

N 0410.0100

Package to prepare 100 ml 1000 X stock solution

N 0410.0250

Package to prepare 250 ml 1000 X stock solution

## MICRO ELEMENTS

	mg/l	µM
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	10.00	161.73
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	18.94	111.94
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	10.00	34.78

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	166.00	1.50
$\text{KH}_2\text{PO}_4$	68.00	0.50
$\text{KNO}_3$	950.00	9.40
$\text{MgSO}_4$	90.27	0.75
$\text{NH}_4\text{NO}_3$	720.00	9.00

Total concentration Micro and Macro elements: 2070.19 mg/l

## VITAMINS

	mg/l	µM
Biotin	0.05	0.21
Folic acid	0.50	1.13
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	5.00	40.62
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.50	1.48

Total concentration Micro and Macro elements including vitamins: 2178.74 mg/l

# NLN MEDIUM

The composition of the NLN medium originated from the medium described by Nitsch. The medium was developed for anthercultures of *Brassica Napus* in liquid medium and the induction of haploid plants from isolated pollen. NLN medium is provided free of Calcium nitrate. In the original medium  $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$  is present in a concentration of 500 mg/l.

To prepare the proper NLN medium formulation 500 mg/l  $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$  has to be added extra to the already dissolved powdered medium.

Lichter, R., Z. Planzephiol., 103, 229-237, 1981

Lichter, R., Z. Planzephiol., 105, 427-434, 1982

## N 0252 NLN MEDIUM

### Micro and Macro elements

N 0252.0001	1 l	(0.4 g)
N 0252.0010	10 l	(3.9 g)
N 0252.0050	50 l	(19.3 g)

## N 0253 NLN MEDIUM

### Vitamin mixture

N 0253.0001	1 l	(1.0 g)
N 0253.0010	10 l	(10.4 g)
N 0253.0050	50 l	(51.9 g)



## MICRO ELEMENTS

	mg/l	μM
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025	0.10
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	10.00	161.73
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	18.95	111.94
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	10.00	34.78

## MACRO ELEMENTS, $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ Free

	mg/l	mM
$\text{KH}_2\text{PO}_4$	125.00	0.92
$\text{KNO}_3$	125.00	1.24
$\text{MgSO}_4$	61.00	0.51

Total concentration Micro and Macro elements: 386.95 mg/l

## VITAMINS

	mg/l	μM
D(+)-Biotine	0.05	0.21
Folic Acid	0.50	1.13
L-Glutamine	800.00	5473.83
Gluthatione (reduced)	30.00	97.61
Glycine	2.00	26.64
Myo-Inositol	100.00	554.94
Nicotinic Acid	5.00	40.62
Pyridoxine HCl	0.50	2.43
L-Serine	100.00	951.57
Thiamine HCl	0.50	1.48

Total concentration vitamins: 1038.55 mg/l

# ORCHIMAX

## Orchid maintenance medium

Orchimax medium is a nutritious and well buffered medium for the cultivation of orchid species. Besides sucrose and the required inorganics and vitamins, the medium is enriched by trypton to provide an additional source of reduced organic nitrogen, vitamins and nutritional agents. To prevent acidification during the cultivation of the plants, 1 gram of MES (Morpholino Ethane Sulfonic acid) is present in the medium.

Applied as a buffer in Plant Tissue Culture media MES is non-toxic for plant tissue and plant cells and makes almost no interactions with inorganic cations as being present in the medium. MES is an excellent buffer for use in Plant Tissue Culture media because of its high buffer capacity its pH range of 5.5 - 6.7.

### O 0257 ORCHIMAX without activated charcoal

O 0257.0001	1 l	25.3 g
O 0257.0010	10 l	252.8 g
O 0257.0016	16 l	404.5 g

### O 0262 ORCHIMAX including activated charcoal

O 0262.0001	1 l	27.3 g
O 0262.0010	10 l	272.8 g
O 0262.0016	16 l	436.5 g



### MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.0125	0.05
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.0125	0.05
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	3.10	50.16
KI	0.415	2.50
MnSO <sub>4</sub> ·H <sub>2</sub> O	8.45	50.00
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.125	0.52
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	5.30	18.42

### MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	166.00	1.50
KH <sub>2</sub> PO <sub>4</sub>	85.00	0.62
KNO <sub>3</sub>	950.00	9.40
MgSO <sub>4</sub>	90.35	0.75
NH <sub>4</sub> NO <sub>3</sub>	825.00	10.31

Total concentration Micro and Macro elements: 2170.47 mg/l

### VITAMINS

	mg/l	µM
myo-Inositol	100.00	554.94
Nicotinic acid	1.00	8.12
Pyridoxin HCl	1.00	4.86
Thiamine HCl	10.00	29.65

Total concentration Micro and Macro elements including MES buffer and vitamins : 2292.47mg/l

### BUFFER

	mg/l	mM
MES	1000.00	4.69

Total concentration Micro and Macro elements including MES buffer: 3170.47mg/l

### ORGANICS

	g/l	mM
Sucrose	20.0	58.43
Tryptone	2.0	
Activated charcoal	2.0	

Total concentration Micro and Macro elements including MES buffer, vitamins and organics: 27.28 g/l

Willemsen en Bourgondiën B.V., The Netherlands

# QUIRIN & LEPOIVRE MEDIUM

Prunus species plantlets could be regenerated from root callus on a medium defined by Quirin and Lepoivre. The calli were formed on the roots of plantlets derived from meristem culture containing 6-benzylaminopurine and Gibberellic acid. Micropropagation of Rosa hybrida L. cultivars is also described on this medium.

Quirin & Lepoivre medium has several differences in comparison to Murashige & Skoog. The ammonium ion concentration is strongly reduced, the calcium ion concentration is increased and the chlorine ions are almost eliminated. This formulation avoids vitrification problems.

Druart. P., Sci. Hort., 12, 339-342, (1980).

Quirin M. and Lepoivre P., Acta Hort, 78, 437, (1977).

Valles. M., Boxus, Ph., Acta Hort., 212, (1987).

## Q 0250 QUIRIN & LEPOIVRE MEDIUM

### Micro and Macro elements

Q 0250.0001	1 l	(3.3 g)
Q 0250.0010	10 l	(32.8 g)
Q 0250.0050	50 l	(163.9 g)

## Q 0251 QUIRIN & LEPOIVRE MEDIUM

### Micro and Macro elements including Vitamins

Q 0251.0001	1 l	(3.4 g)
Q 0251.0010	10 l	(33.8 g)
Q 0251.0050	50 l	(168.9 g)

## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.025	0.10
FeNaEDTA	36.70	100.00
H <sub>3</sub> BO <sub>3</sub>	6.20	100.27
KI	0.08	0.48
MnSO <sub>4</sub> ·H <sub>2</sub> O	0.76	4.50
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
Ca(NO <sub>3</sub> ) <sub>2</sub> .anhydrous	578.92	3.53
KH <sub>2</sub> PO <sub>4</sub>	270.00	1.99
KNO <sub>3</sub>	1800.00	17.82
MgSO <sub>4</sub>	175.79	1.46
NH <sub>4</sub> NO <sub>3</sub>	400.00	5.00

Total concentration Micro and Macro elements: 3278.00 mg/l

## VITAMINS

	mg/l	µM
myo-Inositol	100.00	554.94
Thiamine HCl	0.40	1.19

Total concentration Micro and Macro elements including vitamins: 3378.40 mg/l

Geranium propagation, SBW International BV



# RUGINI OLIVE MEDIUM

The Olive (*Olea europaea sativa* L.) plays an important role in the economies of countries in the Mediterranean area. The in vitro culture of Olive, a particular difficult species to propagate in vitro, required the development of a specific medium formulation. Rugini medium is dedicated to the proliferation of the olive shoots. The medium has an enriched composition compared to MS. Olive tissues are characterized by a high content of Ca, Mg, S, Cu and Zn. The best nitrogen source is a combination of  $\text{NO}_3^-$  and  $\text{NH}_4^+$  supplemented with glutamine 2,19 mg/l. The better carbon source is mannitol (30-36 gr/l) compared to sucrose. A better cytokinin to be used is zeatin: 1 mg/l if filter sterilized, 3-4 mg/l when autoclaved. TDZ and 2iP are less effective. Shoots grow more rapidly compared to other media. The proliferation rate increases and more tender, sturdier shoots with less basal callus are obtained.

Rugini E., In vitro propagation of some olive cultivars, *Scientia Horticulturae* 24, 123 (1984)

Jacoboni A., Luppino M., Rugini E., Role of basal shoot darkening *Scientia Horticulturae*, 53:63 (1993)

## R 0258 RUGINI OLIVE MEDIUM

R 0258.0001	1 l	(4.02 g)
R 0258.0010	10 l	(40.24 g)
R 0258.0050	50 l	(201.18 g)

## MICRO ELEMENTS

	mg/l	$\mu\text{M}$
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025	0.11
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.25	1.00
FeNaEDTA	36.70	100.00
$\text{H}_3\text{BO}_3$	12.40	200.55
KI	0.83	5.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90	100.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25	1.03
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	14.30	49.75

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	332.16	2.99
$\text{Ca}(\text{NO}_3)_2$	416.92	2.54
KCl	500.00	6.71
$\text{KH}_2\text{PO}_4$	340.00	2.50
$\text{KNO}_3$	1100.00	10.88
$\text{MgSO}_4$	732.60	6.09
$\text{NH}_4\text{NO}_3$	412.00	5.15

Total concentration Micro and Macro elements: 3915.34 mg/l

## VITAMINS

	mg/l	$\mu\text{M}$
Biotin	0.05	0.20
Folic acid	0.50	1.13
Glycine	2.00	26.64
myo-Inositol	100.00	554.94
Nicotinic acid	5.00	40.62
Pyridoxine HCl	0.50	2.43
Thiamine HCl	0.50	1.48

Total concentration Micro and Macro elements including vitamins: 4023.89 mg/l

Heuchera propagation, SBW International BV



# SCHENK & HILDEBRANDT MEDIUM

Schenk en Hildebrandt medium has been developed for growth of both monocotyle and dicotyle cell suspensions. A high level of auxin-type growth regulators, 2,4-D (0.5 mg/l) and 4-CPA (2.0 mg/l), generally favoured monocotyledonous cell cultures, while low levels of cytokin, kinetin (0.1 mg/l), were essential for most dicotyledonous cell cultures.

Schenk R.U. and Hildebrandt A.C., Medium and techniques for induction and growth of monocotyledonous and dicotyledonous plant cell cultures. Can. J. Bot. 50, 199 (1972).

## S 0225 SCHENK & HILDEBRANDT MEDIUM

### Micro and Macro elements

S 0225.0001	1 l	(3.2 g)
S 0225.0010	10 l	(31.8 g)
S 0225.0050	50 l	(159.2 g)

## S 0411 SCHENK & HILDEBRANDT VITAMIN MIXTURE

Package contains 10.1 g or 25.3 g vitamins to prepare 100 ml or 250 ml of a 100 X vitamin stock solution.

Use 10 ml vitamin stock solution to prepare 1 litre Schenk & Hildebrandt medium of the proper final vitamin concentration.

S 0411.0100

Package to prepare 100 ml 100 X stock solution

S 0411.0250

Package to prepare 250 ml 100 X stock solution

## MICRO ELEMENTS

	mg/l	µM
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.10	0.42
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.20	0.80
FeNaEDTA	19.80	53.94
$\text{H}_3\text{BO}_3$	5.00	80.87
KI	1.00	6.02
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	10.00	59.16
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.10	0.41
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	1.00	3.48

## MACRO ELEMENTS

	mg/l	mM
$\text{CaCl}_2$	151.00	1.36
$\text{KNO}_3$	2500.00	24.73
$\text{MgSO}_4$	195.05	1.62
$(\text{NH}_4)_2\text{H}_2\text{PO}_4$	300.00	2.61

Total concentration Micro and Macro elements: 3183.25 mg/l

## VITAMINS

	mg/l	µM
myo-Inositol	1000.0	5549.39
Nicotinic acid	5.0	40.61
Pyridoxine HCl	0.5	2.43
Thiamine HCl	5.0	14.82

Total concentration Micro and Macro elements including vitamins: 4193.75mg/l

Echinaceae propagation, SBW International BV



# S - MEDIUM Milieu S Milieu de Bouturage

Bourgoin J.P., Chupeau Y., Missonnier C., *Physiol Plant*, 45, 288-292, 1979

Chupeau et al., *Biotechnology*, 7, 503-507, 1989

## S 0261 S-Medium

Micro and Macro elements including vitamins, buffer and organics

S 0261.0001	1 l	(13.0 g)
S 0261.0010	10 l	(129.7 g)

## MICRO ELEMENTS, Heller medium

	mg/l	µM
AlCl <sub>3</sub> ·6H <sub>2</sub> O	0.05	0.21
CuSO <sub>4</sub> ·5H <sub>2</sub> O	0.03	0.12
Ferric Ammonium Citrate	50.00	160.00
H <sub>3</sub> BO <sub>3</sub>	1.00	16.17
KI	0.01	0.06
MnSO <sub>4</sub> ·H <sub>2</sub> O	0.10	0.59
NiCl <sub>2</sub> ·6H <sub>2</sub> O	0.03	0.13
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	1.00	0.48

## MACRO ELEMENTS, 1/2 concentration MS medium

	mg/l	mM
CaCl <sub>2</sub>	166.12	1.50
KH <sub>2</sub> PO <sub>4</sub>	85.00	0.62
KNO <sub>3</sub>	950.00	9.40
MgSO <sub>4</sub>	90.30	0.75
NH <sub>4</sub> NO <sub>3</sub>	825.00	10.31

## Vitamins, Morel and Wetmore medium

	mg/l	µM
Biotine	0.01	0.04
myo-Inositol	100.00	554.94
Nicotinic Acid	1.00	8.12
Pantothenate Ca.salt	1.00	2.10
Pyridoxine HCl	1.00	4.86
Thiamine HCl	1.00	2.96

## Buffer, Organics

	mg/l	mM
MES	700.00	3.59
Sucrose	10,000.00	29.21

Total concentration: 12,972.65 mg/l

Willemsen en Bourgondiën B.V., The Netherlands



# WESTVACO WV5 MEDIUM

A significant improvement in the initiation of embryogenic cultures of loblolly pine from immature seeds was achieved on Westvaco's WV5 medium defined by Coke with the addition of 30 g/l sucrose, 3 mg/l 2,4-D, 0.5 mg/l BA, 500 mg/l casein hydrolysate, and 1.25 mg/l Gelrite™. Up to a threefold increase in embryogenic culture initiation was seen with WV5 medium over other published media. WV5 medium was also found suitable for embryo development.

Shoot cultures of loblolly pine have also been established and micro-propagated using Westvaco's WV5 medium. Seedling shoots cultured on WV5 medium with 20 g/l sucrose, 5 g/l activated charcoal, and 8 g/l agar showed improved survival and shoot growth compared to that seen with other published media. Shoot quality was excellent and rooting response was good.

Coke J.E, Basal nutrient medium for in vitro cultures of loblolly pine. United States Patent#5,534,433. July 9, 1996.

## W 0260 WESTVACO WV5 MEDIUM

### Micro and Macro elements including Vitamins

W 0260.0001	1 l	(5.2 g)
W 0260.0010	10 l	(52.2 g)
W 0260.0050	50 l	(261.1 g)

## MICRO ELEMENTS

	mg/l	µM
CoCl <sub>2</sub> · 6H <sub>2</sub> O	0.025	0.11
CuSO <sub>4</sub> · 5H <sub>2</sub> O	0.25	1.00
FeNaEDTA	36.71	100.00
H <sub>3</sub> BO <sub>3</sub>	31.00	501.37
KI	0.83	5.00
MnSO <sub>4</sub> · H <sub>2</sub> O	15.16	89.69
Na <sub>2</sub> MoO <sub>4</sub> · 2H <sub>2</sub> O	0.25	1.03
ZnSO <sub>4</sub> · 7H <sub>2</sub> O	8.60	29.91

## MACRO ELEMENTS

	mg/l	mM
CaCl <sub>2</sub>	452.88	4.08
KCl	718.67	9.64
KH <sub>2</sub> PO <sub>4</sub>	270.00	1.98
KNO <sub>3</sub>	1084.06	10.72
MgSO <sub>4</sub>	903.79	7.51
NH <sub>4</sub> NO <sub>3</sub>	700.00	8.74

Total concentration Micro and Macro elements: 4222.23 mg/l

## VITAMINS

	mg/l	µM
myo-Inositol	1000.00	5549.39
Thiamine HCl	0.40	1.19

Total concentration Micro and Macro elements including vitamins: 5222.63 mg/l

Succulent Tissue Culture, The Netherlands



# VACIN & WENT MEDIUM

Vacin E.F. and Went E.W., Bot. Gaz. 110, 605 (1949).

## V 0226 VACIN & WENT MEDIUM

### Micro and Macro elements

V 0226.0001	1 l	(1.6 g)
V 0226.0010	10 l	(16.3 g)
V 0226.0050	50 l	(81.3 g)

## MICRO ELEMENTS

	mg/l	μM
$\text{Fe}_2(\text{C}_4\text{H}_4\text{O}_6)_3$	23.13	32.49
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	5.68	33.61

## MACRO ELEMENTS

	mg/l	mM
$\text{Ca}_3(\text{PO}_4)_2$	200.00	0.64
$\text{KH}_2\text{PO}_4$	250.00	1.84
$\text{KNO}_3$	525.00	5.19
$\text{MgSO}_4$	122.00	1.01
$(\text{NH}_4)_2\text{SO}_4$	500.00	3.78

Total concentration Micro and Macro elements: 1625.81 mg/l

Succulent Tissue Culture, The Netherlands



# WHITE MEDIUM

White P.R., The cultivation of Animal and Plant Cells, Ronald Press, New York (1963).

## W 0227 WHITE MEDIUM

### Micro and Macro elements

W 0227.0001	1 l	(0.96 g)
W 0227.0010	10 l	(9.64 g)
W 0227.0050	50 l	(48.2 g)

## MICRO ELEMENTS

	mg/l	μM
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.001	$4.0 \times 10^{-3}$
$\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$	3.47	12.48
$\text{H}_3\text{BO}_3$	1.50	24.26
KI	0.75	4.52
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	5.31	31.42
$\text{MoO}_3$	0.0001	$0.69 \times 10^{-3}$
$\text{Na}_2\text{SO}_4$	200.00	1400.05
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	2.67	9.29

## MACRO ELEMENTS

	mg/l	mM
$\text{Ca}(\text{NO}_3)_2$ anhydrous	208.47	1.27
KCl	65.00	0.87
$\text{KNO}_3$	80.00	0.79
$\text{MgSO}_4$	351.60	2.92
$\text{NaH}_2\text{PO}_4$	16.80	0.14

Total concentration Micro and Macro elements: 963.39 mg/l

Genetically modified strawberries with a changed antioxidant composition.

Dr. Jan Schaart  
Wageningen UR Plant Breeding



## MICRO-MACRO MEDIA

Using ready-made mineral mixtures, the creation of variations in the concentration of the various components is difficult. The addition of some minerals is feasible, but decreasing the concentration of specific minerals is not possible. In practice this may prove to be a disadvantage. In order to counterbalance this drawback, Duchefa

Biochemie B.V. has created micro and macro mixtures. The medium is divided into micro and macro components and ammonium or potassium nitrate, so the concentration of media components can be varied as needed. The composition of the various micro- and macro media is described on the following pages.

## MICRO-MACRO GAMBORG'S B5 MEDIUM

To obtain the proper concentration of Gamborg's B5 medium add to 1 litre demi water:

- 1.00 g micro-salt mixture
- 1.00 g macro-salt mixture
- 1.05 g (1051.98 mg) potassium nitrate

### M 0302 MICRO-SALT MIXTURE B5

M 0302.0025                      25 l      (25.00 g)

### M 0304 MACRO-SALT MIXTURE B5

M 0304.0025                      25 l      (25.00 g)

### MICRO-SALT MIXTURE

	mg/l
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025
FeNaEDTA	36.70
$\text{H}_3\text{BO}_3$	3.00
KI	0.75
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	10.00
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	2.00
$\text{KNO}_3$	947.25

Total concentration Micro-salt mixture 1000.00 mg/l

### MACRO-SALT MIXTURE

	mg/l
$\text{CaCl}_2$	113.23
$\text{NaH}_2\text{PO}_4$	130.44
$(\text{NH}_4)_2\text{SO}_4$	134.00
$\text{MgSO}_4$	121.56
$\text{KNO}_3$	500.77

Total concentration Macro-salt mixture 1000.00 mg/l

### POTASSIUM NITRATE

	mg/l
$\text{KNO}_3$	1051.98



Genetically modified strawberries with a changed antioxidant composition.

Dr. Jan Schaart  
Wageningen UR Plant Breeding

## MICRO-MACRO MURASHIGE & SKOOG MEDIUM

To obtain the proper concentration of MS medium add to 1 litre demi water:

- 1.00 g micro-salt mixture
- 1.65 g (1652.09 mg) macro-salt mixture
- 1.65 g ammonium nitrate

### M 0301 MICRO-SALT MIXTURE MS

M 0301.0025	25 l	(25.00 g)
M 0301.0050	50 l	(50.00 g)

### M 0305 MACRO-SALT MIXTURE MS

M 0305.0025	25 l	(41.30 g)
M 0305.0050	50 l	(82.60 g)

### MICRO-SALT MIXTURE

	mg/l
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.025
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025
FeNaEDTA	36.70
$\text{H}_3\text{BO}_3$	6.20
KI	0.83
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	16.90
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	8.60
$\text{KNO}_3$	930.47

Total concentration Micro-salt mixture 1000.00 mg/l

### MACRO-SALT MIXTURE

	mg/l
$\text{CaCl}_2$	332.02
$\text{KH}_2\text{PO}_4$	170.00
$\text{KNO}_3$	969.53
$\text{MgSO}_4$	180.54

Total concentration Macro-salt mixture 1652.09 mg/l

## MICRO-MACRO NITSCH MEDIUM

To obtain the proper concentration of Nitsch medium add to 1 litre demi water:

- 0.50 g micro-salt mixture
- 0.85 g (850.19 mg) macro-salt mixture
- 0.72 g ammonium nitrate

### M 0303 MICRO-SALT MIXTURE NITSCH

M 0303.0025	25 l	(12.50 g)
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### M 0306 MACRO-SALT MIXTURE NITSCH

M 0306.0025	25 l	(21.25 g)
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### MICRO-SALT MIXTURE

	mg/l
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.025
FeNaEDTA	36.70
$\text{H}_3\text{BO}_3$	10.00
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	18.94
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	0.25
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	10.00
$\text{KNO}_3$	424.85

Total concentration Micro-salt mixture 500.00 mg/l

### MACRO-SALT MIXTURE

	mg/l
$\text{CaCl}_2$	166.00
$\text{KH}_2\text{PO}_4$	68.00
$\text{MgSO}_4$	90.27
$\text{KNO}_3$	525.92

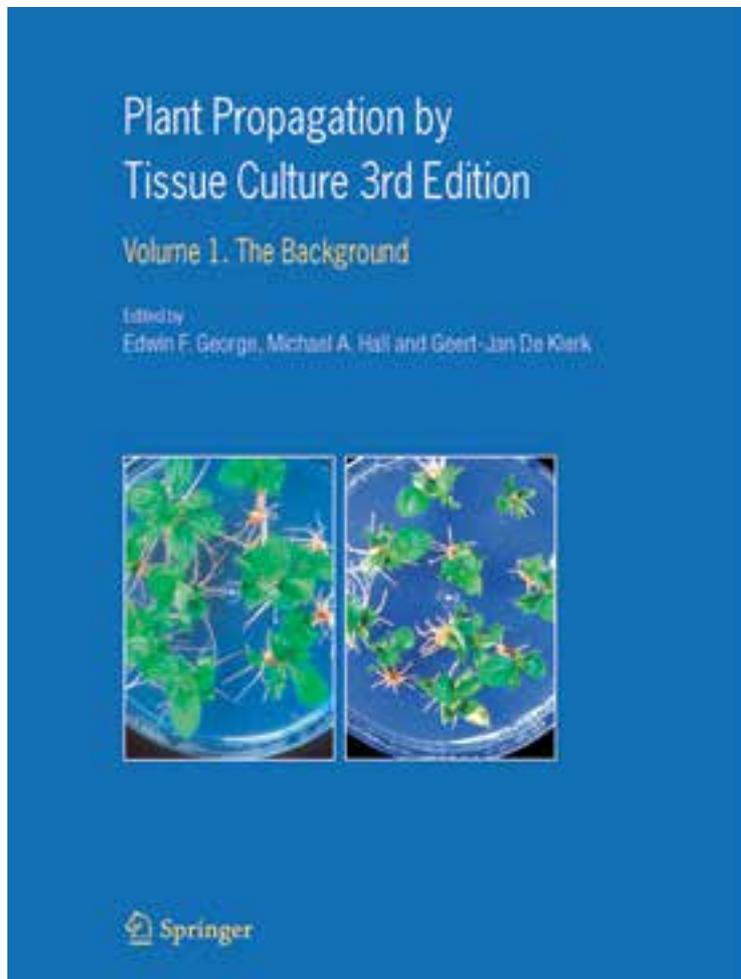
Total concentration Macro-salt mixture 850.19 mg/l

### AMMONIUM NITRATE

	mg/l
$\text{NH}_4\text{NO}_3$	720.00

# Plant Propagation by Tissue Culture

EF George, MA Hall & G-J de Klerk



Procedures for plant tissue culture have been developing from ca. 1930 onwards and are now essential in many domains of science and teaching. The use of these techniques for plant propagation only began to emerge some 40 years later.

The first edition of *Plant Propagation by Tissue Culture* by Edwin F. George appeared in 1986. A second edition consisting of two volumes appeared in 1993 and 1996. For researchers and students, George's books have become the standard works on in vitro plant propagation.

These volumes also contain a wealth of information crucial for researchers and companies working in related areas; particularly plant breeding, genetic engineering, phytopathology, production of secondary metabolites and conservation.

Scientific knowledge has expanded rapidly since the second edition and it would now be a daunting task for a single author to cover all aspects adequately. Therefore, in this third edition, topics are being covered by a number of specialists in the field. However, this edition still maintains the integration that was characteristic of the previous editions.

The first volume of the new edition highlights the scientific background of in vitro propagation. The second volume, which is in preparation, will cover the practice of micropropagation and describe its various applications.

P 5001.0001

## Chapter

- 1 Introduction to tissue culture
- 2 Micropropagation: uses and methods
- 3 The components of plant tissue culture media (1): Macro- and micronutrients
- 4 The components of plant tissue culture media (2): Organic supplements, organic acids, osmotic and pH effects, support systems
- 5 Plant growth regulators (1): Auxins, their analogues and inhibitors
- 6 Plant growth regulators (2): Introduction; cytokinins, their analogues and antagonists
- 7 Plant growth regulators (3): Gibberellins, ethylene, abscisic acid, their analogues and inhibitors; miscellaneous compounds
- 8 Plant developmental biology
- 9 Somatic embryogenesis
- 10 Adventitious regeneration
- 11 Effects of endogenous biological factors
- 12 Effects of the physical environment
- 13 Morphology of tissue cultured plants

## Contributor

- EF George  
 EF George & PC Debergh  
 EF George and G-J de Klerk
- T Thorpe, C Stasolla, EC Yeung, G-J de Klerk, A Roberts & EF George  
 I Machakova, E Zazimalova & EF George  
 J van Staden, E. Zazimalova & EF George
- IE Moshkov, GV Novikova, MA Hall & EF George
- D Chriqui  
 S Von Arnold  
 PB Gahan & EF George  
 J Preece  
 EF George & W Davies  
 M Ziv & Jianxin Chen



A 0941

**(+)-CIS, TRANS-ABSCISIC ACID (S-ABA)** $C_{15}H_{20}O_4 = 264.3$ 

Assay (HPLC) : &gt; 98 %

 $[\alpha]^{20}_D = +425^\circ$  (c= 0.052, MeOH)

- store between -25°C and -15°C
- protect from light
- S: 22-24/25
- CAS 21293-29-8

A 0941.0100	100 mg
A 0941.0250	250 mg
A 0941.1000	1 g

A 1366

**ACETYSALICYLIC ACID** $C_9H_8O_4 = 180.2$ 

Assay: &gt; 99.5%

- store at room temperature
- soluble in water (20 °C / 3.3 g/l)
- R: 22
- CAS 50-78-2

A 1366.0100	100 g
A 1366.0250	250 g

A 1334

**ADENOSINE**

9-β-Ribofuranosyladenine

 $C_{10}H_{13}N_5O_4 = 267.2$ 

Assay (HPLC) : &gt; 98%

Loss on drying : &lt; 0.5%

- store at 2-8°C
- soluble in water
- S: 22-24/25
- CAS 58-61-7

A 1334.0005	5 g
A 1334.0025	25 g

A 0183

**ACYCLOVIR** $C_8H_{11}N_5O_3 = 225.2$ 

Acyclovir inhibits viral DNA synthesis by selective interaction with two distinct viral proteins. Cellular uptake and initial phosphorylation are facilitated by thymidine kinase. Cellular enzymes convert the monophosphate to acyclovir triphosphate and compete for endogenous deoxyguanosine triphosphate (dGTP). Acyclovir triphosphate competitively inhibits viral DNA polymerases and, to a much smaller extent, cellular DNA polymerases. Acyclovir triphosphate is also incorporated into viral DNA, where it acts as a chain terminator because of the lack of 3'-hydroxyl group. By a mechanism termed suicide inactivation, the terminated DNA template containing acyclovir binds the enzyme and leads to irreversible inactivation of the DNA polymerase.

- store at room temperature
- soluble in dilute aqueous solutions of alkali hydroxides and mineral acid.
- R: 20/21/22
- S: 26-36
- CAS No. 59277-89-3

A 0183.1000	1 g
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A 0908

**ADENINE HEMISULPHATE DIHYDRATE**

6-Aminopurine sulphate dihydrate

 $C_{10}H_{12}N_{10}O_4S \cdot 2H_2O = 404.3$  $(C_5H_5N_5)_2 : H_2SO_4 : 2H_2O$ 

Cytokinin growth regulator

Assay: &gt; 99%

- soluble in water
- powder storage at room temperature
- liquid storage at 2-8°C
- sterilization: autoclavable
- concentration 50-250mg/l
- R: 22
- S: 22-24/25
- CAS 321-30-2

A 0908.0005	5 g
A 0908.0025	25 g
A 0908.0100	100 g
A 0908.0250	250 g
A 0908.0500	500 g

A 1335

**ADENOSINE-5-TRIPHOSPHATE**

ATP disodiumsalt

 $C_{10}H_{14}N_5O_{13}P_3Na_2 \cdot xH_2O = 551.1 \cdot x 18.0$ 

Assay (calculated on dry weight)	: > 96%
Dry weight	: > 90%
Heavy metals	: < 0.002%
White crystalline powder	

- store dry at 2-8°C
- soluble in water (20°C / 50 mg/ml)
- CAS 987-65-5

A 1335.0001	1 g
A 1335.0005	5 g
A 1335.0010	10 g

**AGAR**

Agar is a natural product that is obtained from various types of seaweeds. All qualities have been extensively analysed for the remaining mineral grade, limpidity, gel strength, ash content and humidity.

- store at room temperature
- CAS 9002-18-0

P 1001

**PLANT AGAR**

Plant Agar is applied in plant cell and tissue culture as a general purpose agar that combines a good quality with a favourable price.

Plant Agar can be used in a minimal concentration of 5.5 g/l to obtain a solid gel.

**General Characteristics**

Gel strength	: min. 1100 g/cm <sup>2</sup>
Crude ash: < 3%	
Ash, acid insoluble (1.5% conc. in boiling water)	: < 0.5%

P 1001.1000	1 kg
P 1001.5000	5 kg
P 1001.9025	25 kg
	2 x 25 kg
	bulk

D 1004

**DAISHIN AGAR**

Daishin Agar is a well known agar brand in Plant Tissue Culture and is tested for the micropropagation of numerous plants.

D 1004.1000	1 kg
D 1004.5000	5 kg

M 1002

**MICRO AGAR**

Micro Agar is a purified agar with a high gel strength and excellent properties for use in plant cell and tissue culture as well as microbiological work.

Micro Agar can be used in a minimal concentration of 5.0 g/l to obtain a solid gel.

**General Characteristics**

Gel strength	: > 900 g/cm <sup>2</sup>
Sulphated ash	: < 6%
Calcium	: < 2000 ppm
Ash, acid insoluble	: < 0.5%

M 1002.1000	1 kg
M 1002.5000	5 kg
M 1002.9025	25 kg
	2 x 25 kg
	bulk

P 1003

**PHYTO AGAR**

Phyto Agar is a specially selected plant tissue culture tested agar with a high gel strength.

Phyto Agar can be used in a minimal concentration of 5.0 g/l to obtain a solid gel.

**General Characteristics**

Gel strength	: 950-1050 g/cm <sup>2</sup>
Moisture	: < 18%
Ash content	: < 3.5%

P 1003.1000	1 kg
P 1003.5000	5 kg

A 1203

## AGAROSE SPI

Agarose is a highly purified linear galactan hydrocolloid isolated from *Gelidium* species of seaweed. The gelmatrix formed by agarose is almost ideal for diffusion and electrokinetic movement of biopolymers like DNA and RNA.

Duchefa Biochemie **AGAROSE SPI** is ideally suited for electrophoresis of nucleic acids > 1000 bp.

**AGAROSE SPI** is recommended for preparative, as well as analytical nucleic acid electrophoresis. It provides very firm gels at low concentrations.

**AGAROSE SPI** is quality assured specifically to meet the stringent requirement of nucleic acid applications.

**AGAROSE SPI** is manufactured under very stringent conditions and quality controlled to assure conformance to the demanding requirements of nucleic acids and applications.

## Specifications:

- **Gel strength:**  
The force that must be applied to a gel to cause it to fracture.
- **Gelling temperature:**  
The temperature at which an aqueous agarose solution forms a gel as it cools. The gelpoint of an agarose solution is not the same as its melting temperature
- **Sulphate content:**  
May be used as an indicator of purity since sulphate is the major ionic group present.
- **Electroendosmosis (EEO)**  
The movement of liquid through the gel towards the cathode. Because of the electric movement of nucleic acids in the direction of the anode, cathodal EEO can disrupt separations by internal convection. The EEO phenomenon is caused by the migration of dissociable cations and their hydration spheres towards the cathode. The anionic groups in an agarose gel are affixed to the matrix and thus restrained from such movement.

Gel strength, 1%	: > 1200 g/cm <sup>2</sup>
Gel strength, 1.5%	: > 2500 g/cm <sup>2</sup>
Gelling temperature	: 34.5-37.5°C
Melting temperature	: 86.5-89.5°C
Sulphate	: < 0.2%
Electroendosmosis	: 0.09-0.13
Residue on ignition	: < 0.5%
Loss on drying	: < 7%
DNA Binding	: None Detected
DNase and RNase activity	: None Detected

- store at room temperature
- soluble in water
- CAS 9012-36-6

A 1203.0100	100 g
A 1203.0500	500 g
A 1203.1000	1 kg

L 1204

## LOW MELTING AGAROSE PPC

### Specifically selected for Protoplast Cultures

Low Melting Agarose PPC is specifically selected for use in cloning lines where the low gelling temperature obviates the risk of exposing the cell to damaging temperatures. The low gelling temperature of 24-30°C allows the culturist to manipulate cells within the sol at 37°C without having to be concerned about premature gelation. Cooling the agarose to < 26°C immobilizes cells for clonal growth or other experiments.

Gel strength, 1,5%	: > 1000 g/cm <sup>2</sup>
Gelling temperature, 1,5%	: 24-30°C
Melting temperature	: < 65°C
Electroendosmosis	: < 0.12
Moisture	: < 5%
Sulphate	: < 0.12%

- store at room temperature
- soluble in water
- CAS 9012-36-6

L 1204.0100	100 g
L 1204.0250	250 g

S 1202

## SEAPLAQUE™ AGAROSE

Seaplaque™ agarose is particularly useful in cloning lines where the low gelling temperature obviates the risk of exposing the cell to damaging temperatures.

Gel strength, 1,0% gel.	: > 200 g/cm <sup>2</sup>
Gelling temperature, 1,0% sol	: 26-30°C
Melting temperature, 1,0% sol	: < 65°C
Electroendosmosis	: < 0.10
Moisture	: < 10%
Sulphate	: < 0.10%

- store at room temperature
- soluble in water
- CAS 9012-36-6

S 1202.0100	100 g
S 1202.0250	250 g



A 0703

**L-ALANINE** $C_3H_7NO_2 = 89.1$ 

Assay \_\_\_\_\_ : &gt; 98.5%

- store at room temperature
- soluble in water (25°C / 166.5 g/l)
- CAS 56-41-7

A 0703.0025 \_\_\_\_\_ 25 g

A 0703.0100 \_\_\_\_\_ 100 g

A 0532

**ALUMINIUM CHLORIDE  
HEXAHYDRATE** $AlCl_3 \cdot 6H_2O = 241.4$ 

Assay \_\_\_\_\_ : &gt; 98%

- soluble in water (20°C / 1330g/l)
- R: 36/38
- S: 26
- CAS 7784-13-6

A 0532.0025 \_\_\_\_\_ 25 g

A 0532.0100 \_\_\_\_\_ 100 g

A 0601

**p-AMINO BENZOIC ACID**

4-Aminobenzoic Acid, Vitamin H', PABA

 $C_7H_7NO_2 = 137.1$ 

Assay \_\_\_\_\_ : &gt; 99%

White crystalline powder

- slightly soluble in water (4.7 g/l)
- store at room temperature
- R: 22-36/37/38-43
- S: 26-36
- CAS 150-13-0

A 0601.0025 \_\_\_\_\_ 25 g

A 0601.0100 \_\_\_\_\_ 100 g

A 0185

**Amiprofos Methyl** $C_{11}H_{17}N_2O_4PS = 304.3$ 

Used as antimicrotubule herbicide for the production of doubled haploid plants from anther-derived maize callus.  
Theor. Appl. Genet. 81: 205-211, 1991

Assay \_\_\_\_\_ : &gt; 98%

- store at 2-8°C
- R : 22
- S: 36
- CAS 36001-88-4

A 0185.0250 \_\_\_\_\_ 250 mg

A 0185.1000 \_\_\_\_\_ 1 g

A 0528

**AMMONIUM CHLORIDE** $NH_4Cl = 53.5$ 

Assay \_\_\_\_\_ : &gt; 99%

- store at room temperature
- soluble in water (20°C / 370 g/l)
- R: 22-36
- S: 22
- CAS 12125-02-9

A 0528.1000 \_\_\_\_\_ 1 kg

A 1338

**AMMONIUM DIHYDROGEN  
PHOSPHATE**

Ammonium phosphate monobasic  
( $NH_4$ ) $H_2PO_4 = 115.0$

Assay \_\_\_\_\_ : &gt; 99%

- store at room temperature
- soluble in water (20°C / 370 g/l)
- R: 36/37
- S: 26-37/39
- CAS 7722-76-1

A 1338.1000 \_\_\_\_\_ 1 kg

A 0501

## AMMONIUM NITRATE

 $\text{NH}_4\text{NO}_3 = 80.0$ 

Assay \_\_\_\_\_ : &gt; 97.5%

- store at room temperature
- soluble in water (20°C / 1183 g/l)
- hygroscopic
- R: 8-9
- S: 15-16-41
- UN 1942
- CAS 6484-52-2

A 0501.1000	1 kg
A 0501.5000	5 kg
A 0501.9025	25 kg

A 0502

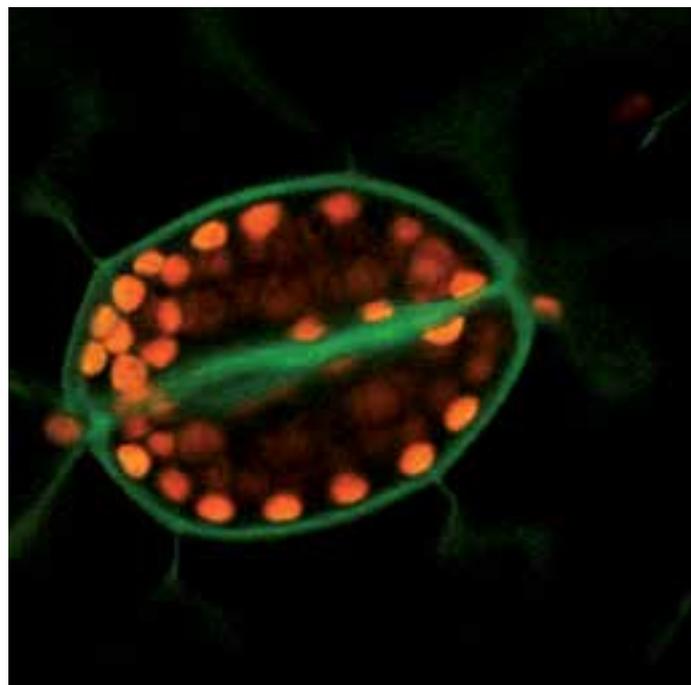
## AMMONIUM SULPHATE

 $(\text{NH}_4)_2\text{SO}_4 = 132.1$ 

Assay \_\_\_\_\_ : &gt; 99%

- store at room temperature
- soluble in water (20°C / 760 g/l)
- CAS 7783-20-2

A 0502.1000	1 kg
A 0502.5000	5 kg



A 0101

## AMOXICILLIN TRIHYDRATE

 $\text{C}_{16}\text{H}_{19}\text{N}_3\text{O}_5\text{S}\cdot 3\text{H}_2\text{O} = 419.5$ 

Assay \_\_\_\_\_ : &gt; 95%

Inhibitor of bacterial cell wall synthesis.

Amoxicillin inhibits the crosslinking of peptidoglycan by binding and inactivating of transpeptidases. High activity against gram-negative bacteria like Agrobacterium species. Sensitive to  $\beta$ -lactamase.

- store at room temperature
- soluble in water
- R: 42/43
- S: 22-24/25-36
- CAS 61336-70-7

A 0101.0010	2x5 g
A 0101.0025	25 g

A 0189

AMOXICILLIN SODIUM /  
CLAVULANATE POTASSIUM

Amoxicillin sodium and clavulanate potassium mixed in a ratio of 5:1

Amoxicillin is an inhibitor of bacterial cell wall synthesis. It inhibits the crosslinking of peptidoglycan by binding and inactivating of transpeptidases. High activity against gram-negative bacteria like Agrobacterium species.  $\beta$ -lactamase sensitive.Clavulanic acid is a specific inhibitor of  $\beta$ -lactamase and protects amoxicillin against inactivation by  $\beta$ -lactamase.

- store dry at 2-8°C
- soluble in water
- R: 42/43
- S: 22-36/37

A 0189.0002	2 g
A 0189.0010	10 g
A 0189.0025	25 g

Stomata cell of transgenic tobacco expressing GFP- overlay, confocal laser microscopy, Leica Germany

(Dr. J. Imani, Institute of Phytopathology &amp; Applied Zoology, Justus-Liebig-University-Giessen, Germany, Prof. R. Hueckelhoven, Centre of Life and Food Sciences Weihenstephan, Germany)

A 0103

## AMPHOTERICIN B

 $C_{47}H_{73}NO_{17} = 924.1$ 

Amphotericin B is a polyene antifungal antibiotic produced by *Streptomyces nodosus*. It appears mainly by interfering with the permeability of the cell membrane of sensitive fungi and yeasts by binding to sterols.

Assay \_\_\_\_\_ : &gt; 750 µg/mg \_\_\_\_\_

- store at 2-8°C
- soluble in DMSO
- R: 20/21/22
- S: 36/37/39-45
- CAS 1397-89-3

A 0103.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

A 0103.0010 \_\_\_\_\_ 10 g \_\_\_\_\_

A 0192

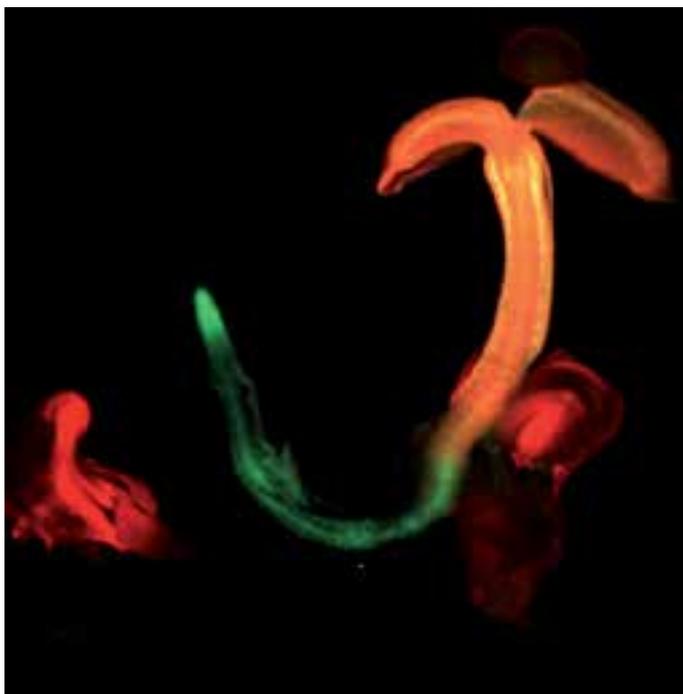
## AMPHOTERICIN B SUSPENSION

Aqueous suspension of 100 mg/ml Amphotericin B

 $C_{47}H_{73}NO_{17} = 924.1$ 

- store at room temperature.

A 0192.0040 \_\_\_\_\_ 40 ml \_\_\_\_\_



A 0104

## AMPICILLIN SODIUM

 $C_{16}H_{18}N_3O_4SNa = 371.4$ 

Ampicillin is an inhibitor of bacterial cell wall synthesis. It inhibits the crosslinking of peptidoglycan by binding and inactivating of transpeptidases. High activity against gram-negative bacteria.  $\beta$ -lactamase sensitive. Ampicillin is used as a selective agent for the transformation of plasmids encoding for  $\beta$ -lactamase production such as pBR322 or pUC (AMP<sup>R</sup>).

Assay \_\_\_\_\_ : &gt; 91% \_\_\_\_\_

- store dry at 2-8°C
- soluble in water
- hygroscopic, protect from moisture
- R: 36/37/38-42/43
- S: 22-26-36/37
- CAS 69-52-3

A 0104.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

A 0104.0010 \_\_\_\_\_ 10 g \_\_\_\_\_

A 0104.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

A 0164

## APRAMYCIN SULPHATE



Nebramycin II

 $C_{21}H_{41}N_5O_{11} \cdot nH_2SO_4 = 539.6 + 98n$  (n=2-2.5)

Apramycin is an aminoglycoside antibiotic and has a bactericidal action against many gram-negative bacteria. Apramycin is a structurally unique antibiotic that contains a bicyclic sugar moiety and a monosubstituted deoxystreptamine. Apramycin can only be acetylated by AAC(3)IV and as a consequence of this enzymatic modification, the antibiotic is unable to enter the cell to bind to its target, the ribosome.

Antimicrobial Agents and chemotherapy, July 1978, p.69-72

Assay \_\_\_\_\_ : &gt; 50% (base) \_\_\_\_\_

- store at 2-8°C
- soluble in water
- R: 20/21/22-61
- S: 22-36/37/39-45
- CAS 65710-07-8

A 0164.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

A 0164.0010 \_\_\_\_\_ 10 g \_\_\_\_\_

A 0164.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

GFP expressing *A. thaliana* plantlet -GFP2 filter

(Dr. J. Imani, Institute of Phytopathology &amp; Applied Zoology, Justus-Liebig-University Giessen, Germany)

A 0704

## L-ARGININE

 $C_6H_{14}N_4O_2 = 174.2$ 

Assay \_\_\_\_\_ : > 98.5%  
 Foreign amino acids \_\_\_\_\_ : < 0.3%

- store at room temperature
- soluble in water (20°C / 150 g/l)
- R: 36
- S: 26
- CAS 74-79-3

A 0704.0025 \_\_\_\_\_ 25 g  
 A 0704.0100 \_\_\_\_\_ 100 g  
 A 0704.0500 \_\_\_\_\_ 500 g  
 A 0704.1000 \_\_\_\_\_ 1 kg

A 0602

## L-ASCORBIC ACID

Vitamin C

 $C_6H_8O_6 = 176.1$ 

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water (20°C / 333 g/l)
- CAS 50-81-7

A 0602.0100 \_\_\_\_\_ 100 g  
 A 0602.0250 \_\_\_\_\_ 250 g  
 A 0602.1000 \_\_\_\_\_ 1 kg



A 0725

## L-ASPARAGINE MONOHYDRATE

 $C_4H_8N_2O_3 \cdot H_2O = 150.1$ 

Assay \_\_\_\_\_ : > 98%

- store at room temperature
- soluble in water (20°C / 30 g/l)
- CAS 5794-13-8

A 0725.0025 \_\_\_\_\_ 25 g  
 A 0725.0100 \_\_\_\_\_ 100 g  
 A 0725.1000 \_\_\_\_\_ 1 kg

A 0705

## L-ASPARTIC ACID

 $C_4H_7NO_4 = 133.1$ 

Assay \_\_\_\_\_ : > 98.5%

- store at room temperature
- soluble in water (25°C / 5 g/l)
- R: 36
- S: 26
- CAS 56-84-8

A 0705.0100 \_\_\_\_\_ 100 g  
 A 0705.0500 \_\_\_\_\_ 500 g

A 0156

## ATRAZINE

 $C_8H_{14}ClN_5 = 215.7$ 

Atrazine is an inhibitor of photosynthesis by blocking the electron transport due to binding of the Qb protein in the thylakoid membrane.

Assay \_\_\_\_\_ : > 97%

- store at room temperature
- soluble in chloroform
- R: 43-48/22-50/53 S: 36/37-60-61
- UN 2811
- CAS 1912-24-9

A 0156.0250 \_\_\_\_\_ 250 mg

GFP expressing *A. thaliana* plantlet -GFP3 filter

(Dr. J. Imani, Institute of Phytopathology & Applied Zoology, Justus-Liebig-University Giessen, Germany)

B 0106

**BACITRACIN** $C_{66}H_{103}N_{17}O_{16}S = 1421.6$ 

Bacitracin is active against gram-positive bacteria. Most gram-negative bacteria are resistant. It interferes with bacterial cell wall synthesis by blocking the function of the lipid carrier molecule that transfers cell wall subunits across the cell membrane. Toxic to plant cells.

Potency \_\_\_\_\_ &gt; 60 IU/mg \_\_\_\_\_

- soluble in ethanol and methanol
- slightly soluble in water
- store at 2-8°C
- hygroscopic, protect from moisture
- S: 22-24/25
- CAS 1405-87-4

B 0106.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

B 0106.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

B 1304

**BANANA POWDER**

Produced by freeze drying banana-puree without additives. 100 grams banana powder is equivalent to approximately 420 gram fresh fruit.

Light brownish powder. \_\_\_\_\_

Moisture content \_\_\_\_\_ : &lt; 5% \_\_\_\_\_

- Store dry at room temperature

B 1304.0500 \_\_\_\_\_ 500 g \_\_\_\_\_

B 1304.1000 \_\_\_\_\_ 1 kg \_\_\_\_\_

B 1304.5000 \_\_\_\_\_ 5 kg \_\_\_\_\_



B 0904

**6-BENZYLAMINOPURINE**6-BAP, N<sup>6</sup>-Benzyladenine $C_{12}H_{11}N_5 = 225.2$ 

Cytokinin growth regulator

Assay \_\_\_\_\_ : &gt; 99% \_\_\_\_\_

- soluble in 1N NaOH
- store powder at room temperature
- store liquid at 2-8°C
- sterilization : autoclavable or filtration
- concentration : 0.01-5.0 mg/l
- R: 22-36/37/38
- S:24/25-26-36
- CAS 1214-39-7

B 0904.0001 \_\_\_\_\_ 1 g \_\_\_\_\_

B 0904.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

B 0904.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

B 0930

**6-BENZYLAMINOPURINE RIBOSIDE**N<sup>6</sup>-Benzyladenosine $C_{17}H_{19}N_5O_4 = 357.4$ 

Cytokinin growth regulator

Assay \_\_\_\_\_ : &gt; 99.5% \_\_\_\_\_

- soluble in 1N NaOH
- store powder at 2-8°C
- store liquid at 2-8°C
- sterilization : filtration
- concentration : 0.01-5.0 mg/l
- R: 22-36/37/38
- S:26-36
- CAS 4294-16-0

B 0930.0250 \_\_\_\_\_ 250 mg \_\_\_\_\_

B 0930.1000 \_\_\_\_\_ 1 g \_\_\_\_\_

LED-Light cultivation,  
Succulent Tissue Culture, The Netherlands

B 0932

**N-BENZYL-9-(2-TETRAHYDROPYRANYL)-ADENINE**

BPA, PBA

6-Benzylamino-9-[2-tetrahydropyranyl]-9H-purine

 $C_{17}H_{19}N_5O = 309.4$ 

BPA is a highly mobile synthetic cytokinin. Foliar spray of BPA increased branching in carnation, chrysanthemum, pointsettia, petunia and fuchsia. In no instance did BPA reduce plant height. Application of BPA to flower buds at an early stage increased both the diameter and the fresh weight of carnation flowers or chrysanthemum inflorescences (Jeffcoat, B. J. of Hort. Sc. 52:143-153 (1977).

In *Lilium longiflorum*, spraying with BPA resulted in delayed anthesis and increased dry matter accumulation in flowers under high photosynthetic photon flux. Application of BPA induced the formation of numerous bulbils in the leaf axils (Wang YT, Hort Sc. 31 (6) 976-977 (1996).

BPA can be used as cytokinin for haploid plant regeneration from cultured anthers of strawberry (Owen H.R. and Miller AR. PCR 15: 905-909 (1996).

Assay \_\_\_\_\_ : &gt; 98.5%

White crystalline powder

- store at 2-8°C
- soluble in ethanol
- powder storage 2-8°C
- liquid storage 2-8°C
- sterilization : filtration or autoclave
- R : 22 S : 36
- CAS 2312-73-4

B 0932.0100 \_\_\_\_\_ 100 mg

B 0932.0500 \_\_\_\_\_ 500 mg

B 1514

**BES**

(N,N-bis[2-Hydroxyethyl]-2-aminoethanesulfonic acid)

 $C_6H_{15}NO_5S = 213.2$ 

pKa (20°C) \_\_\_\_\_ : 6.9-7.3

pH range \_\_\_\_\_ : 6.4 – 7.8

Assay \_\_\_\_\_ : &gt; 99%

Moisture \_\_\_\_\_ : &lt; 1%

UV Absorbance (1 M aq. sol., 1 cm cell, 260 nm) : &lt; 0.1

A 5% solution in water is clear and colourless

- store at room temperature
- R: 36/37/38
- S: 26-36
- CAS 10191-18-1

B 1514.0025 \_\_\_\_\_ 25 g

B 1514.0250 \_\_\_\_\_ 250 g

B 1514.1000 \_\_\_\_\_ 1 kg

LED-Light cultivation,  
Succulent Tissue Culture, The Netherlands

B 0603

**D(+)-BIOTIN**

Vitamin H (Coenzyme R)

 $C_{10}H_{16}N_2O_3S = 244.31$ 

Assay \_\_\_\_\_ : &gt; 97.5%

White crystalline powder

- store at 2-8°C
- soluble in warm water
- soluble in slightly alkaline and acid solutions
- CAS 58-85-5

B 0603.0500 \_\_\_\_\_ 500 mg

B 0603.1000 \_\_\_\_\_ 1 g

B 1516

**Bis-Tris buffer grade**

Bis-(2-hydroxyethyl)-imino-tris-(hydroxyl-methyl)-methane

 $C_8H_{19}NO_5 = 209,2$ 

Assay \_\_\_\_\_ : &gt; 99%

pH range \_\_\_\_\_ : 5.8 – 7.2

- store at room temperature
- soluble in water
- R: 36/37/38
- S: 26-36
- CAS 6976-37-0

B 1516.0100 \_\_\_\_\_ 100 g

B 1516.0500 \_\_\_\_\_ 500 g

B 0107

**BLEOMYCIN SULPHATE**

MW = approximately 1400  
1 unit per mg solid

The sulphates of bleomycin are a mixture of basic antineoplastic glycopeptide antibiotics produced by *Streptomyces verticillus*. Bleomycin binds to DNA and causes strand scissions.

- store at 2-8°C
- soluble in water (20°C / 20 g/l)
- R: 39/23/25-42/43-40-45-46-61 S: 13-22-36/37/39-45-53
- CAS 9041-93-4

B 0107.0015

15 mg

B 0503

**BORIC ACID**H<sub>3</sub>BO<sub>3</sub> = 61.8

Assay : &gt; 99%

- store at room temperature
- soluble in water (20°C / 50 g/l)
- R: 62-63-36/37/38 S: 26-36
- CAS 10043-35-3

B 0503.1000

1 kg

B 0503.5000

5 kg

X 1402

**5-BROMO-4-CHLORO-3-INDOLYL-β-D-GALACTOPYRANOSIDE**C<sub>14</sub>H<sub>15</sub>BrClNO<sub>6</sub> = 408.6

X-Gal is a chromogenic substrate of β-galactosidase. X-Gal is used in conjunction with Isopropyl-b-D-1-thiogalactoside (IPTG) (I1401) for the detection of β-galactosidase activity in bacterial colonies in a colorimetric assay in order to distinguish recombinants (white) from non-recombinants (blue).

X-gal is cleaved at the β1-4 bond between galactose and the 5-Bromo-4-chloro-3-indolyl part of X-Gal by β-galactosidase via hydrolysis. The enzymatic cleavage of X-Gal results in the production of a water insoluble blue dichloro-dibromo-indigo precipitate. In cloning strategies with vectors like Lambda-11, M13mp18 and 19, pUC18 and 19, pUR222 the *E. coli lacZ* gene is transformed to lac<sup>c</sup> cells. After transformation, the cells show β-galactosidase activity in the presence of IPTG and X-Gal containing media. The insertion of a DNA fragment into the cloning sites of the lacZ gene results in the disruption of β-galactosidase activity leading to the appearance of white colonies on X-Gal and IPTG containing media. Non recombinant cells produce a blue indigo dye on these media.

Assay : &gt; 98%

- store dry at 2-8°C or below
- allow to warm to room temperature before opening
- protect from light and moisture
- soluble in DMSO and DMF
- S: 22-24/25
- CAS 7240-90-6

X 1402.0100

100 mg

X 1402.1000

1 g

X 1402.5000

5 g

GUS expression in carrot leaves under control of 35sCaMV promoter (Dr. J. Imani, Institute of Phytopathology & Applied Zoology, Justus-Liebig-University-Giessen, Germany, Prof. R. Hueckelhoven, Centre of Life and Food Sciences Weihenstephan, Germany)



## X 1405

### 5-BROMO-4-CHLORO-3-INDOLYL- $\beta$ -D-GLUCURONIC ACID CYCLOHEXYLAMMONIUM SALT

#### X-GlcA, Cyclohexylammonium salt

$C_{14}H_{13}BrClNO_7 \cdot C_6H_{13}N = 521.8$

**X-GlcA**, 5-Bromo-4-chloro-3-indolyl- $\beta$ -D-glucuronic acid is a substrate for  $\beta$ -D-Glucuronidase (GUS) encoded by the *gusA* gene. The substrate is used as a qualitative histochemical marker of specific GUS expressions in cells and tissue. X-GlcA is cleaved by GUS at the  $\beta$ 1 glucuronic bond between glucuronic acid and the 5-Bromo-4-chloro-3-indolyl part of X-GlcA via hydrolysis. The enzymatic cleavage of X-GlcA results in the precipitation of a water insoluble blue dichloro-dibromo-indigo precipitate. Color formation requires three separate reactions. After enzymatic turnover, the released indoxyl derivative dimerises and is subsequently oxidized to the final indigo dye.

Assay	: > 98%
Specific Opt. Rotation	: -87.5° +/- 2°
(a <sub>20</sub> <sup>o</sup> /D; c=1 in H <sub>2</sub> O : DMF = 1:1)	

- store dry at 2-8°C
- allow to warm to room temperature before opening
- hygroscopic, protect from light and moisture
- soluble in DMSO and DMF
- S: 22-24/25
- CAS 114162-64-0

X 1405.0100	100 mg
X 1405.1000	1 g
	5 x 1 g
	10 x 1 g

## X 1406

### 5-BROMO-4-CHLORO-3-INDOLYL- $\beta$ -D-GLUCURONIC ACID SODIUM SALT TRIHYDRATE

#### X-GlcA, Sodium salt trihydrate

$C_{14}H_{12}BrClNO_7 \cdot Na \cdot 3H_2O = 498.7$

Assay	: > 98.5%
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- store dry at 2-8°C
- allow to warm to room temperature before opening
- protect from light and moisture
- soluble in DMSO and DMF
- S: 22-24/25
- CAS 129541-41-9

X 1406.0100	100 mg
X 1406.1000	1 g
	5x1 g
	10x1 g

## M 1412

### 5-BROMO-6-CHLORO-3-INDOLYL- $\beta$ -D-GLUCURONIC ACID CYCLOHEXYLAMMONIUM SALT

#### Magenta-GlcA, Cyclohexylammonium salt

$C_{14}H_{13}BrClNO_7 \cdot C_6H_{13}N = 521.8$

An alternative for X-GlcA producing a magenta colour.

Assay	: > 98%
Water	: < 1.0%
Specific rotation	: -68.0° +/- 3°
(a <sub>20</sub> <sup>o</sup> /D; c =1 in H <sub>2</sub> O : DMF = 1:1)	

- store dry at 2-8°C
- allow to warm to room temperature before opening
- protect from light and moisture
- soluble in DMSO and DMF
- CAS 144110-43-0

M 1412.0010	100 mg
M 1412.0100	1 g
	10 x 1 g

Please inquire for annual bulk discounts.

## X 1410

### 5-BROMO-4-CHLORO-3-INDOLYL-PHOSPHATE DISODIUM SALT



#### X-Phos disodium salt

$C_8H_4BrClNO_4 \cdot P \cdot Na_2 = 370.4$

X-Phos is a colorimetric substrate for detection of alkaline phosphatase activity in blotting immunohistochemical and cytochemistry techniques. In conjunction with nitro blue tetrazolium (NBT) (N1411), a purple insoluble precipitate is formed that can be read visually.

Assay	: > 99%
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- store between -25°C and -15°C
- allow to warm to room temperature before opening
- protect from light
- soluble in water
- R: 36/37/38
- S: 22
- CAS 102185-33-1

X 1410.0100	100 mg
X 1410.1000	1 g

X 1413

### 5-BROMO-4-CHLORO-3-INDOLYL-PHOSPHATE p-TOLUIDINE SALT



BCIP p-Toluidine salt, X-Phos p-Toluidine salt

$C_8H_6BrClNO_4 \cdot P \cdot C_7H_9N = 433.64$

X-Phos is a colorimetric substrate for detection of alkaline phosphatase activity in blotting immunohistochemical and cytochemistry techniques. In conjunction with nitro blue tetrazolium (NBT) (N1411), a purple insoluble precipitate is formed that can be read visually.

Assay \_\_\_\_\_ : > 99%

- store between -25°C and -15°C
- allow to warm to room temperature before opening
- protect from light
- soluble in DMSO and DMF
- R: 20/21/22-36/37/38-40 S: 22-24/25-36/37
- CAS 6578-06-9

X 1413.0100 \_\_\_\_\_ 100 mg

X 1413.1000 \_\_\_\_\_ 1 g

B 1414

### 5-BROMO-INDOLYL-β-D-GALACTOPYRANOSIDE

Blue-Gal An alternative to X-Gal producing a darker blue color.

$C_{14}H_{16}BrNO_6 = 374.2$

Assay (TLC) \_\_\_\_\_ : > 98%

Spec. Opt. Rot. \_\_\_\_\_ : -34° ± 2°

(α 20°/D; c = 1 in 1:1 H<sub>2</sub>O: DMFO)

Water \_\_\_\_\_ : < 1.0%

- store dry between -25°C and -15°C
- allow to warm to room temperature, before opening
- protect from light and moisture
- soluble in DMSO and DMF
- S: 22-24/25
- CAS 97753-82-7

B 1414.0100 \_\_\_\_\_ 100 mg

B 0157

### BROMOXYNIL



3,5-Dibromo-4-hydroxy-benzonitril

$Br_2C_6H_2(CN)OH = 267.9$

Bromoxynil inhibits photosynthesis in plants by binding to electron-transport components of photosystem II in the thylakoid membrane.

- store at room temperature
- very slightly soluble in water
- soluble in tetrahydrofuran
- R: 25-26-43-50/53-63 S: 27/28-36/37-45-60-61-63
- UN 2588
- CAS 1689-84-5

B 0157.0250 \_\_\_\_\_ 250 mg

C 0529

### CALCIUM CARBONATE



$CaCO_3 = 100.1$

Assay \_\_\_\_\_ : > 98.5%

- store at room temperature
- insoluble in water
- R: 37/38-41 S: 26-39
- CAS 471-34-1

C 0529.1000 \_\_\_\_\_ 1 kg

C 0504

### CALCIUM CHLORIDE DIHYDRATE



$CaCl_2 \cdot 2H_2O = 147.0$

Assay \_\_\_\_\_ : > 97%

- store at room temperature
- soluble in water
- hygroscopic
- R: 36 S: 22-24
- CAS 10035-04-8

C 0504.1000 \_\_\_\_\_ 1 kg

C 0504.5000 \_\_\_\_\_ 5 kg

GUS expression in carrot flower under the control of mannopine synthase (mas) promoter (Dr. J. Imani, Institute of Phytopathology & Applied Zoology, Justus-Liebig-University Giessen, Germany)



C 0530

**CALCIUM CITRATE TETRAHYDRATE**

tri-Calcium-di-citrate tetrahydrate  
 $\text{Ca}_3(\text{C}_6\text{H}_5\text{O}_7)_2 \cdot 4\text{H}_2\text{O} = 570.5$

Assay \_\_\_\_\_ : > 98%

- store at room temperature
- soluble in water (23° C / 0.96 g/l)
- CAS 5785-44-4

C 0530.1000 \_\_\_\_\_ 1 kg

C 0531

**CALCIUM GLUCONATE MONOHYDRATE**

$\text{C}_{12}\text{H}_{22}\text{CaO}_{14} \cdot \text{H}_2\text{O} = 448.4$

Assay \_\_\_\_\_ : > 98.5%

Additional Calcium ( $\text{Ca}^{2+}$ ) source in Plant Tissue Culture media.

- store at room temperature
- soluble in water (20° C / 30 g/l)
- CAS 299-28-5

C 0531.0250 \_\_\_\_\_ 250 g

C 0531.1000 \_\_\_\_\_ 1 kg

C 0505

**CALCIUM NITRATE TETRAHYDRATE**

$\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O} = 236.2$

Assay \_\_\_\_\_ : > 98.5%

Crystalline powder

- store at room temperature
- soluble in water (20°C / 2600 g/l)
- hygroscopic
- R: 8-36/38
- S: 26-17
- UN 1454
- CAS 13477-34-4

C 0505.1000 \_\_\_\_\_ 1 kg

C 0505.5000 \_\_\_\_\_ 5 kg

C 0506

**CALCIUM PHOSPHATE TRIBASIC**

$\text{Ca}_3(\text{PO}_4)_2 = 310.2$

Assay ( $\text{Ca}^{2+}$ ) \_\_\_\_\_ : > 35-40%

- store at room temperature
- insoluble in water, soluble in diluted acids
- CAS 7758-87-4

C 0506.1000 \_\_\_\_\_ 1 kg

C 1006

**CARRAGEENAN, Iota type**

Carrageenan is a naturally-occurring family of polysaccharides extracted from red seaweed. Upon cooling and in the presence of appropriate cations, ( $\text{K}^+$ ,  $\text{Ca}^{2+}$ ), carrageenan polymers align themselves to form double helices.

Iota carrageenan binds water and forms dry, elastic gels in the presence of calcium salts.  $\text{Ca}^{2+}$  ions make bonds between the carrageenan molecules to form helices. The negative charges associated with the 2-sulphate groups on the Iota carrageenan molecules do not allow the helices to aggregate to the same extent as Kappa carrageenan.

- store at room temperature
- soluble in water (60 °C / 5g/l)

- CAS 9062-07-1

C 1006.0100 \_\_\_\_\_ 100 g

G 1007

**GELCARIN GP- 812**

Gelcarin GP-812 is a well tested source of carrageenan for use in Plant Tissue Culture. It forms a clear, palebrown firm gel. Gelcarin should be dispersed in cold water and then heated above its solubility temperature to obtain maximum functionality. Upon cooling and in the presence of appropriate cations ( $\text{K}^+$ ,  $\text{Ca}^{2+}$ ) carrageenan polymers align themselves to form double helices. These helices associate with divalent cations, i.e. calcium, to form a gel matrix.

- CAS 9000-07-1

G 1007.0250 \_\_\_\_\_ 250 g

G 1007.1000 \_\_\_\_\_ 1 kg

G 1007.5000 \_\_\_\_\_ 5 kg

C 0109

## CARBENICILLIN DISODIUM

 $C_{17}H_{16}N_2Na_2O_6S = 422.4$ 

Assay	: > 90%
Water	: < 5.5%

Carbenicillin is an inhibitor of bacterial cell wall synthesis. It inhibits the crosslinking of peptidoglycan by binding and inactivation of transpeptidases. High activity against gram-negative bacteria. Commonly used for the elimination of *Agrobacterium* species after inoculation. Sensitive to  $\beta$ -lactamase. Non toxic to plant cells.

- store dry at 2-8°C
- soluble in water
- hygroscopic
- protect from moisture
- R: 42/43
- S: 36/37/39
- CAS 4800-94-6

C 0109.0005	5 g
C 0109.0025	25 g

C 0160

## CARBOXIN

 $C_{12}H_{13}NO_2S = 235.3$ 

Carboxin is a fungicide and inhibits the oxydation of succinate in sensitive yeasts and fungi.

- store at room temperature
- soluble in ethanol
- R: 21/22
- S: 36
- CAS 5234-68-4

C 0160.0250	250 mg
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C 1301

## CASEIN HYDROLYSATE

Pancreatic hydrolysate of casein.

Due to its low NaCl content this quality is well suited for Plant Tissue Culture.

Total nitrogen (TN)	: 12.5%-13.5%
Amino nitrogen (AN)	: 3.0%-4.0%
NaCl	: < 6.0%

- store dry at room temperature
- soluble in water
- CAS 9000-71-9

C 1301.0250	250 g
C 1301.0500	500 g
C 1301.1000	1 kg

C 0110

## CEPHALEXIN MONOHYDRATE

 $C_{16}H_{17}N_3O_4S \cdot H_2O = 365.4$ 

Cephalexin is an inhibitor of bacterial cell wall synthesis. The antibiotic inhibits the crosslinking of peptidoglycan by binding and inactivating of transpeptidases. Active against gram-positive bacteria and moderately active against gram-negative bacteria.  $\beta$ -lactamase sensitive.

Assay	: > 95.0%
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- store at 2-8°C
- soluble in water
- R: 20/21/22-36/37/38-42/43
- S: 26-36
- CAS 15686-71-2

C 0110.0005	5 g
C 0110.0010	10 g

C 0111

## CEFOTAXIME SODIUM



$C_{16}H_{16}N_5NaO_7S_2 = 477.4$   
plant cell culture tested

Cefotaxime is an inhibitor of bacterial cell wall synthesis. The antibiotic inhibits the crosslinking of peptidoglycan by binding and inactivating of transpeptidases. High activity against gram-negative bacteria. Very often used for elimination of *Agrobacterium* species after inoculation. Cefotaxime has high resistance against  $\beta$ -lactamase activity. Non toxic to plant cells.

Assay	: 916 - 964 $\mu$ g/mg
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- store dry at 2-8°C
- soluble in water
- R: 42/43
- S: 22-24/25
- CAS 64485-93-4

C 0111.0001	1 g
C 0111.0005	5 g
C 0111.0025	25 g

C 8001

## CELLULASE R-10

"Cellulase Onozuka R-10" from *Trichoderma Viride*.

1 unit (U) of Cellulase will release 1.0 µmole of glucose from carboxy-methyl cellulose. Routinely used for the isolation of protoplasts, for its ability to degrade cell walls. Cellulase "Onozuka R-10" is often used in combination with Macerozyme R-10 (cat. no. M 8002).

Beldman, G. et al., The cellulase of *Trichoderma Viride*, . J. Biochem., 146, 301-308, 1985.

Potrykus, J., et al., Protoplasts: Isolation, culture, plant regeneration, 118, 549-578, 1986.

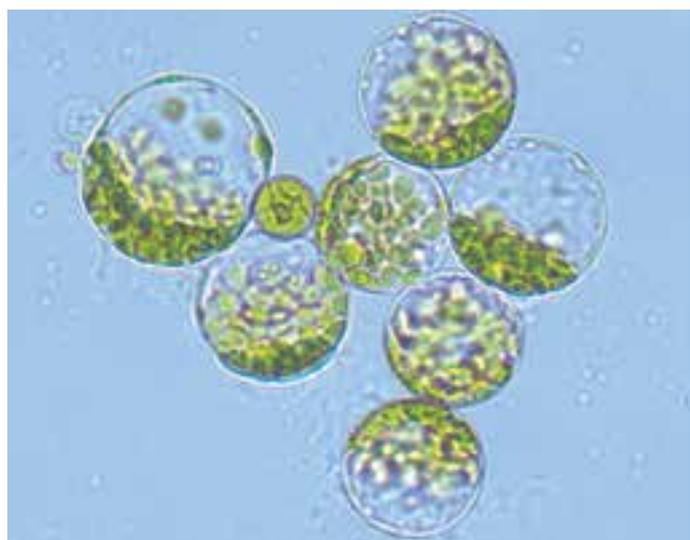
Tewes, A., et al., High yield isolation and rapid recovery of protoplasts from suspension cultures of tomato (*Lycopersicon esculentum*), 113, 141-150, 1984.

Evans, D.A. et al., Plant protoplast isolation and culture, Int. Rev. Cyt. Suppl, 16, 33-53, 1983.

Loss on drying	: < 10%
Enzyme activity	: >10,000 U/g
Beige lyophilisate	

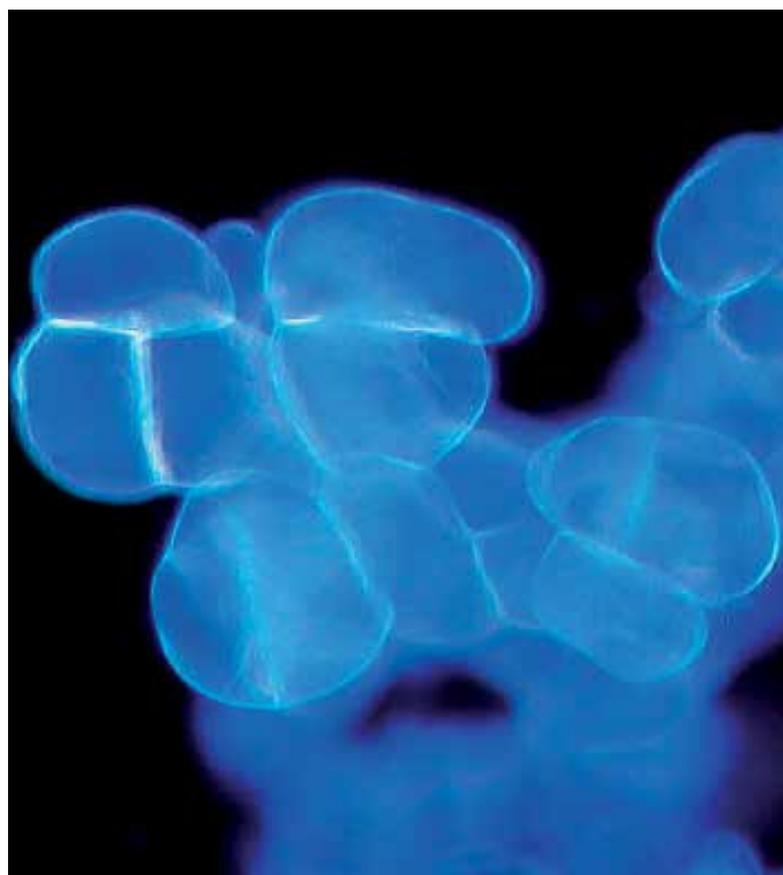
- optimum pH between 4 and 5
- store at 2-8°C
- CAS 9012-54-8

C 8001.0001	1 g
C 8001.0005	5 g
C 8001.0010	10 g



Protoplast from barley leaf

(Dr. J. Imani, Institute of Phytopathology & Applied Zoology, Justus-Liebig-University Giessen, Germany)



Cell wall staining in protoplasts, Iris Heidmann

C 8003

## CELLULASE RS

"Cellulase Onozuka RS"

Cellulase "Onozuka RS" is produced by a mutant *Trichoderma viride* that was derived from the parent strain for Cellulase "Onozuka R-10". Cellulase RS contains a very high activity of decomposing natural celluloses. This type of cellulase can be used to obtain protoplasts in a very short time and dissolves cell walls of a wider range of plants.

Loss on drying	: < 10%
Enzyme activity	: > 16,000 U/g
Off white dry powder	

- optimum pH : 4.0 – 5.0
- optimum temperature : 50 – 60°C
- Xylanase : Cellulase RS contains about three times as high xylanase activity as Cellulase R-10
- Activity : Cellulase RS contains more than 16,000 units per gram of filter decomposing activity.
- readily soluble in water
- store at 2-8°C
- CAS 9012-54-8

C 8003.0001	1 g
C 8003.0005	5 g
C 8003.0010	10 g

## C 1397

## N-TETRADECYL -N,N,N,-TRIMETHYL AMMONIUM BROMIDE



Cetrimide

 $C_{17}H_{38}NBr = 336.4$ 

Assay \_\_\_\_\_ : &gt; 96%

- soluble in water
- store at room temperature
- R: 20/21/22-34 S: 26-27-36/37/39
- UN 3077
- CAS 8044-71-1

C 1397.0050	50 g
C 1397.0100	100 g
C 1397.0500	500 g
C 1397.1000	1 kg

## C 1393

## N-CETYL-N,N,N, -TRIMETHYL AMMONIUM BROMIDE



Hexadecyltrimethylammonium Bromide, Cetrimonium Bromide, CTABr

 $C_{19}H_{42}NBr = 364.5$ 

Assay \_\_\_\_\_ : &gt; 96%

- soluble in water
- store at room temperature
- R: 22-36/38-50/53 S: 26-39-61
- UN 3077
- CAS 57-09-0

C 1393.0050	50 g
C 1393.0100	100 g
C 1393.0500	500 g
C 1393.1000	1 kg

## C 1302

## CHARCOAL

Steam activated

Assay \_\_\_\_\_ : 100%

pH (5% in water) \_\_\_\_\_ : 5-7

- store dry at room temperature
- water insoluble
- CAS 7440-44-0

C 1302.1000	1 kg
C 1302.5000	5 kg

## C 1374

## CHAPS

3-[(3-Cholamidopropyl)dimethyl-ammonio]-1-propanesulfonate  
 $C_{32}H_{58}N_2O_7S = 614.9$ 

CHAPS is a nondenaturing zwitterionic detergent suitable for use as a solubilizing agent for membrane proteins. Combines the useful properties of both sulfobetaine-type and the bile salt detergents. The low background absorption in the UV region is an attractive feature for use in the UV monitoring of membrane proteins. CHAPS can be easily removed by dialysis or gel filtration.

L.M. Hjelmeland, A nondenaturing zwitterionic detergent for membrane biochemistry, Proc. Nat. Acad. Sci. USA, 77, 6368 (1980).

Assay \_\_\_\_\_ : &gt; 97%

Water \_\_\_\_\_ : &lt; 3%

Absorption (280 nm) \_\_\_\_\_ : &lt; 0.22

- store at room temperature
- soluble in water
- hygroscopic
- R: 36/37/38
- S: 26/36
- CAS 75621-03-3

C 1374.0001	1 g
C 1374.0005	5 g
C 1374.0025	25 g
C 1374.0100	100 g

## C 0113

## CHLORAMPHENICOL

 $C_{11}H_{12}Cl_2N_2O_5 = 323.1$ 

Bacteriostatic agent against gram-negative and gram-positive bacteria. Enters sensitive cells by active transport. Within the cell, it binds to the 50S subunit of bacterial ribosomes and inhibits bacterial protein synthesis by preventing attachment of amino-acyl transfer RNA to its acceptor site on the ribosome, thus preventing peptide bond formation by peptidyl transferase.

Assay \_\_\_\_\_ : &gt; 98%

- store at room temperature
- slightly soluble in water (2.5 g/l)
- soluble in ethanol
- R: 42/43-45-46-63
- S: 36/37/39-45-53
- CAS 56-75-7

C 0113.0025	25 g
C 0113.0100	100 g

C 0114

**CHLORHEXIDINE  
DIGLUCONATE**

20% aqueous solution

 $C_{22}H_{30}Cl_2N_{10}.2(C_6H_{12}O_7) = 897.8$ 

Chlorhexidine is a bisbiguanide antiseptic and disinfectant that is bactericidal or bacteriostatic against a wide range of gram-positive and gram-negative bacteria. It inhibits mycobacteria, fungi and some viruses. Chlorhexidine is most active at a neutral or slightly acidic pH. It is used for disinfection of skin, clean instruments and hard surfaces in a concentration of 0.05 to 0.5% in water or 70% alcohol.

- store at room temperature
- soluble (miscible) in water
- R: 41-50 S: 26-37/39-61
- UN 3082
- CAS 18472-51-0

C 0114.0250	250 ml
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C 0114.1000	1 l
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C 0115

**CHLORHEXIDINE  
HYDROCHLORIDE** $C_{22}H_{30}Cl_2N_{10}.2HCl = 578.4$ 

- store at room temperature
- soluble in water
- R: 36/37/38-43 S: 22-24/25
- UN 3077
- CAS 3697-42-5

C 0115.0010	10 g
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C 0115.0025	25 g
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S 1403

**6-CHLORO-3-INDOLYL-β-D-GALACTO-  
PYRANOSIDE****Salmon-Gal** $C_{14}H_{16}ClNO_6 = 329.7$ 

**Salmon-GAL** is an alternative chromogenic substrate for β-D-Galactosidase. Salmon-Gal is used in conjunction with Isopropyl-b-D-1-thiogalactoside (IPTG) (I1401) for detection of β-galactosidase activity in bacterial colonies in a colorimetric assay, in order to distinguish recombinants (white) from non-recombinants. Salmon-Gal is cleaved at the β1-4 bond between galactose and the 5-Bromo-4-chloro-3-indolyl part of X-Gal by β-galactosidase via hydrolysis.

Assay	: > 98%
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- store dry at 2-8°C
- allow to warm to room temperature before opening
- protect from light and moisture
- soluble in DMSO and DMF
- S: 22-24/25
- CAS 138182-21-5

S 1403.0100	100 mg
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S 1403.1000	1 g
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S 1407

**6-CHLORO-3-INDOLYL-β-D-  
GLUCURONIC ACID,  
CYCLOHEXYLAMMONIUM SALT****Salmon-XGlcA cyclohexylammonium salt** $C_{14}H_{14}ClNO_7.C_6H_{13}N = 442.9$ 

**Salmon-XGlcA** is an alternative substrate for β-D-Glucuronidase (GUS) encoded by the **gusA** gene. Cleavage via hydrolysis of Salmon-Red-X-GlcA by GUS results in the precipitation of a water insoluble Salmon precipitate at the site of enzymatic cleavage. In conjunction with X-Gal, Salmon-X-GlcA is useful for simultaneous detection of GUS and Lac activities on the same plate. For more detailed information see X-GlcA.

Assay	: > 90%
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- store dry at 2-8 °C
- protect from light and moisture
- soluble in DMSO and DMF
- S: 22-24/25
- CAS 138182-20-4

S 1407.0100	100 mg
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Slugs?  
Iris Heidmann

C 0909

**p-CHLOROPHENOXYACETIC ACID**4-CPA; CPA  
 $C_8H_7ClO_3 = 186.6$ Assay \_\_\_\_\_ : > 97%  
off white to tan crystals

- soluble in ethanol
- liquid storage 2-8°C
- sterilization : autoclavable
- concentration : 0.1-10.0 mg/l
- R: 22 S:13-36-46
- UN 2811
- CAS 122-88-3

C 0909.0025 \_\_\_\_\_ 25 g  
C 0909.0100 \_\_\_\_\_ 100 g

D 0161

**CHLOROXYLENOL, 49 mg/ml**

Disinfectant for the skin.

- store at room temperature
- soluble in water
- CAS 88-04-0 (chloroxylenol)

D 0161.1000 \_\_\_\_\_ 1 litre

C 0177

**CHLORSULFURON** $C_{12}H_{12}ClN_5O_4S = 357.8$ 

Chlorsulfuron affects the biosynthesis of branched chain amino acids by inhibiting the enzyme acetolactate synthase (ALS). The crs1-1 gene from Arabidopsis thaliana confers resistance to chlorsulfuron (CS) by encoding an ALSS with a reduced affinity to Chlorsulfuron. Chlorsulfuron has been applied as a successful selective agent in the transformation of tobacco, maize and sugarbeet. Transgenic poplars and fertile rice plants have also been obtained by using the crs1-1 gene in combination with the CaMV 35S promoter.

Assay \_\_\_\_\_ : &gt;95%

- store at room temperature
- slightly soluble in methylene chloride
- soluble in water (25°C/150-300 ppm)
- R: 50/53 S: 60/61
- CAS 64902-72-3
- UN 3077

C 0177.0100 \_\_\_\_\_ 100 mg

C 0116

**CHLORTETRACYCLINE HYDROCHLORIDE** $C_{22}H_{23}ClN_2O_8.HCl = 515.3$ 

Bacteriostatic antibiotic with activity against gram-positive and gram-negative bacteria. Within the cell tetracyclines bind reversible to the 30S subunit of the ribosome, preventing the binding of aminoacyl transfer RNA and inhibiting protein synthesis and hence cell growth.

Assay \_\_\_\_\_ : > 89.5%  
pH \_\_\_\_\_ : 2.3-3.3  
Water \_\_\_\_\_ : < 2.0%  
Tetracycline \_\_\_\_\_ : > 94.5%

- store at 2-8°C
- soluble in water
- protect from light
- R: 20/21/22-63 S: 22-24/25-36/37-45
- CAS 64-72-2

C 0116.0025 \_\_\_\_\_ 25 g  
C 0116.0100 \_\_\_\_\_ 100 g

C 0605

**CHOLINE CHLORIDE** $C_5H_{14}NOCl = 139.6$ 

White crystals

- store at room temperature
- soluble in water
- hygroscopic
- R: 36/37/38 S: 26-36
- CAS 67-48-1

C 0605.0100 \_\_\_\_\_ 100 g

C 1303

**CITRIC ACID MONOHYDRATE** $C_6H_8O_7.H_2O = 210.1$ 

Assay \_\_\_\_\_ : &gt; 99.5%

- store at room temperature
- soluble in water
- R: 37/38-41
- S: 26-36/37/39
- CAS 5949-29-1

C 1303.1000 \_\_\_\_\_ 1 kg

C 0117

**CLINDAMYCIN  
HYDROCHLORIDE**

$C_{18}H_{33}ClN_2O_5 \cdot S \cdot HCl = 461.5$   
plant cell culture tested

Clindamycin is a lincosamide antibiotic with a primarily bacteriostatic action against gram-positive bacteria. It binds to the 50S subunit of the bacterial ribosome and inhibits the early stages of protein synthesis.

Assay \_\_\_\_\_ : > 84.% \_\_\_\_\_

- store at 2-8°C
- soluble in water (20°C / 50 g/l)
- R: 36/37/38 S: 26-36
- CAS 21462-39-5

C 0117.0001 \_\_\_\_\_ 1 g \_\_\_\_\_

C 0507

**COBALT CHLORIDE  
HEXAHYDRATE**

$CoCl_2 \cdot 6H_2O = 237.93$

Assay \_\_\_\_\_ : > 97% \_\_\_\_\_

- store at room temperature
- soluble in water (20°C / 76 g/l)
- R: 22-42/43-49-50/53 S: 22-45-53-60-61
- UN 3077
- CAS 7791-13-1

C 0507.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

C 0507.0100 \_\_\_\_\_ 100 g \_\_\_\_\_

C 1305

**COLCHICINE**

$C_{22}H_{25}NO_6 = 399.4$

Assay \_\_\_\_\_ : > 97% \_\_\_\_\_

- store at room temperature
- soluble in water and ethanol
- R: 26/28 13-36/37-45
- UN 1544
- CAS 64-86-8
- For colchicine an end user declaration is required

C 1305.0001 \_\_\_\_\_ 1 g \_\_\_\_\_

C 1305.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

C 1305.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

C 0118

**COLISTIN SULPHATE**

A mixture of the sulphates of polypeptides produced by certain strains of *Bacillus polymixa*. Colistin acts primarily by binding to membrane phospholipids and disrupting the bacterial cytoplasmic membrane. The antibiotic is active against gram-negative bacteria, especially *Pseudomonas* species.

Potency \_\_\_\_\_ : > 19.000 Units/mg \_\_\_\_\_

- store dry at 2-8°C
- soluble in water
- hygroscopic
- R: 25 S: 22-36/37-45
- CAS 1264-72-8

C 0118.0001 \_\_\_\_\_ 1 g \_\_\_\_\_

C 0118.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

C 0508

**CUPRIC SULPHATE  
PENTAHYDRATE**

$CuSO_4 \cdot 5H_2O = 249.7$

Assay \_\_\_\_\_ : > 99.5% \_\_\_\_\_

Crystalline

- store at room temperature
- soluble in water
- R: 22-36/38-50/53 S: 22-60-61
- UN 3077
- CAS: 7758-99-8

C 0508.0250 \_\_\_\_\_ 250 g \_\_\_\_\_

C 0508.0500 \_\_\_\_\_ 500 g \_\_\_\_\_

C0943

**4-CPPU**

N-(2-Chloro-4-pyridyl)-N'-phenylurea

$C_{12}H_{10}ClN_3O = 247.7$

Cytokinin plant growth regulator  
Takahashi, S. et al., *Phytochemistry* 17, 2101 (1978)

Assay \_\_\_\_\_ : > 98% \_\_\_\_\_

- store at room temperature
- soluble in DMSO or KOH 0.1 M
- R: 36/37 S: 26-36
- CAS 68157-60-8

C 0943.0250 \_\_\_\_\_ 250 mg \_\_\_\_\_

C 0726

## CYANOCOBALAMIN

Vitamin B12

 $C_{63}H_{88}CoN_{14}O_{14}P = 1355.4$ 

Assay \_\_\_\_\_ : &gt; 98%

- store at 2-8°C
- soluble in water (25°C / 12 g/l)
- S: 22-24/25
- CAS 68-19-9

C 0726.0100	100 mg
C 0726.1000	1 g

C 0176

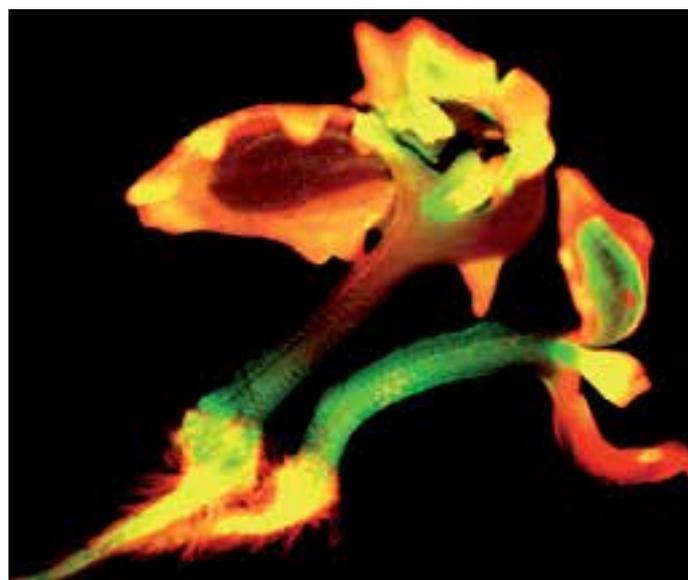
## CYCLOHEXIMIDE

 $C_{15}H_{23}NO_4 = 281.4$ 

Assay \_\_\_\_\_ : &gt;95%

- store at 2-8°C
- soluble in water
- R: 28-51/53-61-68
- S: 45-53-61
- CAS 66-81-9

C 0176.0001	1 g
C 0176.0005	5 g
C 0176.0025	25 g



Two seedlings, Iris Heidmann

C 0119

## D-CYCLOSERINE

 $C_3H_6N_2O_2 = 102.1$ 

Cycloserine interferes with the bacterial cell wall synthesis by competing with D-Alanine for incorporation into the cell wall. Cycloserine has some activity against gram-negative bacteria and is active against some mycobacteria.

Assay \_\_\_\_\_ : &gt; 900 µg/mg

- store at 2-8°C
- soluble in water
- CAS 68-41-7

C 0119.0005	5 g
C 0119.0025	5x5 g

C 0706

## L-CYSTEINE HYDROCHLORIDE MONOHYDRATE

 $C_3H_8NO_2SCL.H_2O = 175.6$ 

Assay \_\_\_\_\_ : &gt; 98.0%

- store at room temperature
- soluble in water
- R: 36/37/38
- S: 22-36
- CAS 7048-04-6

C 0706.0025	25 g
C 0706.0100	100 g
C 0706.0500	500 g
C 0706.1000	1 kg

D 1342

## DEXTRAN SULPHATE SODIUM

Produced from Dextran 500.000

Tested for suitability in nucleic acids hybridizations

Free sulphate	: < 0.2%
pH aqueous solution (1%)	: 5.0-7.5
Clarity (15% solution):	No suspended particles

- store at room temperature
- soluble in water
- CAS 9011-18-1

D 1342.0010	10 g
D 1342.0050	50 g
D 1342.0100	100 g

D 0920

## DICAMBA



3,6-Dichloro-o-Anisic Acid  
 $C_8H_6Cl_2O_3 = 221.0$

Auxin like growth regulator

Assay \_\_\_\_\_ : &gt; 89%

- store at room temperature
- liquid storage at 2-8°C
- sterilization : filtration
- concentration : 0.01-5.0 mg/l
- R: 22-41-52/53
- S: 26-61
- UN 3077
- CAS 1918-00-9

D 0920.0250 \_\_\_\_\_ 250 mg

D 0911

2,4-DICHLOROPHENOXYACETIC  
ACID

2,4 D  
 $C_8H_6Cl_2O_3 = 221.0$

Auxin growth regulator

Assay \_\_\_\_\_ : &gt; 96%

off white to tan crystals

- soluble in ethanol or 1N NaOH
- store powder at room temperature
- liquid storage at 2-8°C
- readily soluble in water
- sterilization : autoclavable
- concentration : 0.01-5.0 mg/l
- R: 22-37-41-43-52/53
- S: 24/25-26-36/37/39-46-61
- UN 3077
- CAS 94-75-7

D 0911.0100 \_\_\_\_\_ 100 g

D 0911.0250 \_\_\_\_\_ 250 g

D 0933

## DL-DIHYDROZEATIN

(diH)Z, DHZ, DZ  
 $C_{10}H_{15}N_5O = 221.3$

DL-Dihydrozeatin (DHZ) is a naturally occurring cytokinin that is generally very active. DHZ derivatives are commonly found in plant tissues and are frequent metabolites of applied zeatin. In a bioassay, DHZ and its conjugates are equally active as their zeatin analogues. In studies where DHZ has been externally supplied to plants it appears to be more 'stable' than zeatin. This may be because DHZ is not a substrate for cytokinin oxidase. DHZ may be important in the maintenance of cytokinin activity levels in an oxidative environment.

Assay \_\_\_\_\_ : &gt; 98%

white crystalline

- Zeatin < 0.1%
- soluble in ethanol
- powder storage 2-8°C
- liquid storage between -25°C and +5°C
- sterilization : filtration
- S: 22-36
- CAS 14894-18-9

D 0933.0025 \_\_\_\_\_ 25 mg

D 0933.0050 \_\_\_\_\_ 50 mg

D 0933.0100 \_\_\_\_\_ 100 mg

D 0933.0250 \_\_\_\_\_ 250 mg

D 0906

6-g-g-(DIMETHYLALLYLAMINO)-  
PURINE

2-iP; N<sup>6</sup>-[2-Isopentenyl]adenine  
 $C_{10}H_{13}N_5 = 203.2$

Cytokinin growth regulator

Assay \_\_\_\_\_ : &gt; 98%

Loss on drying \_\_\_\_\_ : &lt; 1.0%

White Crystalline

- soluble in 1N NaOH
- store powder between -25°C and -15°C
- liquid storage between -25°C and -15°C
- sterilization : autoclavable or filtration
- concentration : 1.0-30.0 mg/l
- S: 22-24/25
- CAS 2365-40-4

D 0906.0001 \_\_\_\_\_ 1 g

D 0906.0005 \_\_\_\_\_ 5 g

D 0906.0010 \_\_\_\_\_ 10 g

D 0934

## 6-(g-g-DIMETHYLALLYLAMINO) PURINE RIBOSIDE

2-iP-ribose, N6-[2-Isopentenyl]adenosine, N6-[g,g-, methylallyl]adenosine  
 $C_{15}H_{21}N_5O_4 = 335.4$

Assay \_\_\_\_\_ : > 97%  
 White crystalline (3 x recrystallized)

- store at 2-8°C
- soluble in water
- sterilization: filtration
- S: 22-24/25
- CAS 7724-76-7

D 0934.0100	100 mg
D 0934.0250	250 mg
D 0934.1000	1 g

D 1370

## DIMETHYL SULFOXIDE



DMSO, Methyl sulfoxide  
 $C_2H_6SO = 78.1$

Assay \_\_\_\_\_ : > 99.9%  
 $H_2O$  \_\_\_\_\_ : < 0.1%

- store at room temperature
- melting point 16-19°C
- soluble in water
- R: 36/38 S: 26
- CAS 67-68-5

D 1370.0100	100 ml
D 1370.0250	250 ml
D 1370.1000	1 l



Pineapple propagation in TIB, SBW International B.V., The Netherlands.

D 1308

## DITHIOERYTHREITOL, DTE


 $C_4H_{10}O_2S_2 = 154.2$ 

Assay \_\_\_\_\_ : > 98%  
 Melting Point \_\_\_\_\_ : 79-83°C

- store dry at 2-8°C
- soluble in water
- hygroscopic, protect from moisture
- R: 22-36/37/38 S: 22-24/25-28-36/37
- CAS 6892-68-8

D 1308.0005	5 g
D 1308.0010	10 g
D 1308.0025	25 g

D 1309

## DITHIOTHREITOL, DTT


 $C_4H_{10}O_2S_2 = 154.2$ 

Assay \_\_\_\_\_ : ≥ 99%  
 Melting Point \_\_\_\_\_ : 40-44°C

- store dry at 2-8°C
- soluble in water
- hygroscopic, protect from moisture
- R: 22 S: 22-24-36/37/39
- CAS 3483-12-3

D 1309 .0005	5 g
D 1309 .0010	10 g
D 1309 .0025	25 g

TIB propagation vessels, SBW International B.V., The Netherlands

D 0120

**DOXORUBICIN  
HYDROCHLORIDE** $C_{27}H_{29}NO_{11}.HCl = 580.0$ 

Doxorubicin is an antineoplastic antibiotic that may act by forming a stable complex with DNA and interfering with the synthesis of nucleic acids. It is a cell cycle nonspecific agent, but is most active against cells in S phase. Doxorubicin also acts on cell membranes.

- store at 2-8°C, protected from light
- R: 22-36/37/38-45 S: 36/37/39-45-53
- CAS 25316-40-9

D 0120.0010

5 ml

A 5 ml solution contains 10 mg doxorubicin hydrochloride dissolved in 0.9% NaCl

D 0121

**DOXYCYCLINE  
HYDROCHLORIDE** $C_{22}H_{24}N_2O_8.HCl = 480.9$ 

Doxycycline is a tetracycline with bacteriostatic properties against gram-positive and gram-negative bacteria. Within the cell, it binds reversibly to the 30S subunit of the ribosome, preventing the binding of aminoacyl transfer RNA and inhibiting protein synthesis and hence cell growth. Doxycycline is more active against most species than tetracycline.

- store at 2-8°C
- soluble in water
- protect from light
- R: 20/21/22-40 S: 22-36/37/39-45
- CAS 10592-13-9

D 0121.0010

10 g

D 0121.0025

25 g



Harvested TIB explant Pineapple, SBW International B.V., The Netherlands

E 0940

**24-EPIBRASSINOLIDE** $C_{28}H_{48}O_6 = 480.8$ 

Some 30 years ago, organic extracts of *Brassica napus* pollen were found to promote stem elongation and cell division in plants. The active components were identified as steroids and have therefore been named brassinosteroids. It is now recognized more and more that brassinosteroids are genuine plant hormones. In the nM to  $\mu$ M range, 24-epibrassinolide has been found to promote cell division of protoplasts and to cause hypocotyls elongation, but also to inhibit root extension. Evidence is mounting that it plays a role in vascular differentiation. Much research has been done on the ameliorative effect of brassinosteroids during stress. S.D. Clouse and J.M. Sasse: Brassinosteroids: essential regulators of plant growth and development. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* 49: 427-451 (1998)

Assay (HPLC) : &gt; 90%

22-epibrassinolide + 3- epibrassinolide : &lt; 10%

- store at 2-8°C
- soluble in DMSO
- R: 36 S: 26-36
- CAS 78821-43-9

E 0940.0010 10 mg

E 0940.0025 25 mg

Temporary Immersion Bioreactors (TIB), a PLC-operated system consisting of liquid medium storage units and plant reactor vessels, uses temporary submersion of plant parts which enables the culture period to be prolonged while propagation factors are maintained or even increased, compared to classical propagation methods.

SBW International BV has gained experience using large scale TIB systems and developed protocols for numerous ornamental crops like *Heliconia* and nutrition crops like banana and pineapple. The application of this technique provides high quality, homogenous starting material.

E 0122

## ERYTHROMYCIN

 $C_{37}H_{67}NO_{13} = 733.9$ 

Erythromycin is a macrolide antibiotic with a bacteriostatic action against primarily gram positive bacteria. It binds reversibly to the 50S subunit of the ribosome, resulting in blockage of the transpeptidation or translocation reactions, inhibition of protein synthesis and hence inhibition of cell growth.

Assay	: > 93%
Water	: < 6.5%

- store dry at room temperature
- soluble in ethanol
- R: 20/21/22-42/43 S: 36/37/39
- CAS 114-07-8

E 0122.0010	10 g
E 0122.0025	25 g

E 1343

## ESCULIN

 $C_{15}H_{16}O_9 \cdot 1\frac{1}{2} H_2O = 367.3$ 

Assay	: > 97.5%
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- store dry at room temperature
- soluble in water
- CAS 66778-17-4

E 1343.0005	5 g
E 1343.0025	25 g

F 0527

## ETHYLENEDIAMINE DI-2-HYDROXY-PHENYL ACETATE FERRIC



Fe-EDDHA, Red-Brown Microgranule. A highly stable chelate providing a source of iron easily absorbed by plants. Replacement for FeNaEDTA. T.P.M. van der Salm Plant Cell Tiss. and Organ Cult, 37: 73-77, 1994

Iron (Fe)	: > 5.7%
Chelating agent	: EDDHA

- store at room temperature
- soluble in water
- R: 22-36/37/38 S: 26-39
- CAS 16455-61-1

F 0527.0025	25 g
F 0527.0100	100 g
F 0527.0250	250 g

E 0509

## ETHYLENEDIAMINETETRA-ACETATE FERRIC SODIUM



FeNaEDTA

Ferric Sodium EDTA

 $C_{10}H_{12}N_2O_8FeNa = 367$ 

Assay	: > 99%
Iron (Fe)	: > 13.1%
pH: 1%	: 4-5.5

- store at room temperature
- soluble in water
- R: 22-36/37/38
- S: 26-39
- CAS 15708-41-5

E 0509.0100	100 g
E 0509.0250	250 g
E 0509.1000	1 kg

E 0511

## ETHYLENEDIAMINETETRA-ACETATE DISODIUM DIHYDRATE

Na<sub>2</sub>EDTA.2H<sub>2</sub>O $C_{10}H_{14}N_2O_8Na_2 \cdot 2H_2O = 372.2$ 

Assay	: > 99%
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- store at room temperature
- soluble in water ( 20°C/100 g/l)
- R: 36/37/38
- S: 26-36/37/39
- CAS 6381-92-6

E 0511.0250	250 g
E 0511.0500	500 g
E 0511.1000	1 kg

F 0512

## FERROUS SULPHATE HEPTAHYDRATE

 $FeSO_4 \cdot 7H_2O = 278.0$ 

Assay	: > 98%
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- store at room temperature
- soluble in water
- R: 22 S: 24/25
- CAS 7782-63-0

F 0512.0250	250 g
F 0512.1000	1 kg

F 0176

## 5-FLUORO OROTIC ACID



5-FOA

 $C_5H_3FN_2O_4 = 174.1$ 

Used in the selection of orotidine-5'-phosphate decarboxylase mutants of *S. cerevisiae*. Winstof, F. et al., Genetics, 107, 179 (1984).

Assay (NMR) \_\_\_\_\_ : > 98%

- store between -25°C and -15°C
- soluble in water/ethanol
- R: 20/21/22
- S: 26-36/37/39
- CAS 703-95-7
- UN 2783

F 0176.1000 \_\_\_\_\_ 1 g

F 0176.5000 \_\_\_\_\_ 5 g

F 0123

## 5-FLUOROURACIL



5-FU

 $C_4H_3FN_2O_2 = 130.1$ 

5-Fluorouracil, a pyrimidine analogue, is an antineoplastic agent that acts as an antimetabolite to uracil. After intracellular conversion to the active deoxynucleotide, it interferes with the synthesis of DNA by blocking the conversion of deoxyuridylic acid to thymidylic acid by the cellular enzyme thymidylate synthetase.

Assay \_\_\_\_\_ : > 98%

Loss on drying \_\_\_\_\_ : < 0.5%

- store at 2-8°C
- soluble in water (10 g/l)
- R: 20/21/22-45-60-61
- S: 7-13-22-26-27-36/37/39-45-53
- CAS 51-21-8

F 0123.0001 \_\_\_\_\_ 1 g

F 0123.0005 \_\_\_\_\_ 5 g

F 0919

## FLURIDON

 $C_{19}H_{14}F_3NO = 329.3$ 

Inhibitor of ABA-synthesis.

Kwang-Soo K., Davelaar E. and De Klerk G.J. Phys. Plantarum 90, 59-64 1994

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- slightly soluble in methanol and diethylether
- sterilization : filtration
- concentration : 0.01-0.05 mg/l
- R: 51/53
- S: 60
- CAS 59756-60-4

F 0919.0250 \_\_\_\_\_ 250 mg



F 0935

## FLURPRIMIDOL

 $C_{15}H_{15}F_3N_2O_2 = 312.3$ 

Flurprimidol is an alternative for Ancyimidol. Flurprimidol is two to four times as active as Ancyimidol and more stable. Both Ancyimidol and Flurprimidol are synthetic inhibitors of Gibberellic Acid biosynthesis and block the pathway during the oxidation of ent-kaurene to ent-kaurenoic acid. Flurprimidol is used in Tissue Culture to control internode elongation, especially in liquid cultures.

Assay \_\_\_\_\_ : > 99%

- store powder at 2-8°C
- store solution at 2-8°C
- soluble in DMSO
- sterilization: autoclavable or filtration
- concentration: 0.25-10.0 mg/l
- R: 52-21/22
- CAS 56425-91-3

F 0935.0025	25 mg
F 0935.0050	50 mg
F 0935.0100	100 mg

F 0608

## FOLIC ACID

 $C_{19}H_{19}N_7O_6 = 441.4$ 

Assay \_\_\_\_\_ : > 96%

Crystalline

- store at room temperature
- slightly soluble in water (25°C / 1.6 mg/l)
- S: 22-24/25
- CAS 59-30-3

F 0608.0025	25 g
F 0608.0100	100 g

F0619

FOLINATE CALCIUM  
PENTAHYDRATE $C_{20}H_{21}N_7O_7Ca \cdot 5H_2O = 601.5$ 

Assay \_\_\_\_\_ : > 97%

Yellow powder

- store at 2-8°C
- soluble in water
- R: 36/37/38-42/43 S: 26-36
- CAS 41927-89-3

F 0619.0001	1 g
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F 0801

## D-FRUCTOSE

 $C_6H_{12}O_6 = 180.2$ 

Assay \_\_\_\_\_ : > 99.5%

Water \_\_\_\_\_ : < 0.15%

White crystalline

- store at room temperature
- soluble in water (20°C / 500 g/l)
- CAS 57-48-7

F 0801.0500	500 g
F 0801.1000	1 kg
F 0801.5000	5 kg

G 0175

## G-418 DISULPHATE

 $C_{20}H_{40}N_4O_{10} \cdot 2H_2SO_4 = 692.7$ 

G-418 is an aminoglycoside antibiotic and is applied as a selective agent in transformation experiments. The antibiotic binds to the 30S subunit of the prokaryotic ribosome, thereby inhibiting protein synthesis as well as generating errors in the transcription of the genetic code. Ribosomes of mitochondria and chloroplasts of higher plants are related to bacterial ribosomes and are also susceptible to aminoglycosides.

Being a derivative of gentamycin, the antibiotic contains a additional 3'OH that can be phosphorylated by NPT II. As a result of this phosphorylation, the charge and the steriomeric conformation of the G-418 molecule changes in such a way that the antibiotic is no longer capable of binding to the specific ribosome binding sites.

G-418 is used as an alternative for kanamycin in monocots, e.g., rice, Lolium, Graminea which are highly resistant to the latter. In all cases G-418 was shown to be more effective. This is most probably due to the better binding characteristics of the gentamycine shaped structure of G-418.

Activity \_\_\_\_\_ : > 650 µg/mg

- store at 2-8°C
- soluble in water
- R: 20/21-40-61
- S: 36/37/39-45-53
- CAS 108321-42-2
- UN 2811

G 0175.0001	1 g
G 0175.0005	5 g

G 0810

**D-GALACTOSE** $C_6H_{12}O_6=180.2$ 

Assay	: > 98%
Water	: < 1.0%

- store at room temperature
- soluble in water (25°C / 680 g/l)
- CAS 59-23-4

G 0810.0100	100 g
G 0810.0500	500 g
G 0810.1000	1 kg

G 1101

**GELRITE**

Gelrite is a naturally-derived gelling polymer that can be used in a variety of applications as a solidification agent instead of agar.

Produced by microbial fermentation, Gelrite is a highly purified natural anionic polysaccharide without the variations commonly associated with agar. Gelrite forms rigid, brittle, agar like gels at approximately half the use level of agar in presence of soluble salts like  $Mg^{2+}$  and  $Ca^{2+}$ . Gels prepared with Gelrite are remarkably clear in comparison to those formed with agar. Gelrite contains no contaminating matters (e.g., phenolic compounds) as found in agar that are toxic to certain sensitive organisms.

Li-Chun Huang, Toshio Murashige et al. Effects of common components on hardness of culture media prepared with Gelrite. *In Vitro Cell. Dev. Biol.* 31: 84-89, April 1995. Society for in Vitro Biology

Loss on drying	: <15%
Gel strength	: 400-700 g/cm <sup>2</sup>

- store at room temperature
- soluble in water
- It is advised to adjust Gelrite to the medium by means of a sieve to avoid lumping.
- CAS 71010-52-1

G 1101.0100	100 g
G 1101.0250	250 g
G 1101.0500	500 g
G 1101.1000	1 kg
G 1101.5000	5 kg
G 1101.9025	25 kg

Iris Heidmann, Arabidopsis flower

G 0124

**GENTAMICIN SULPHATE**

An aminoglycoside antibiotic with bactericidal activity against many gram-negative bacteria. Aminoglycosides are taken up into sensitive bacterial cells by an active transport process.

Within the cell, they bind to the 30S and to some extent to the 50S subunits of the bacterial ribosome, inhibiting protein synthesis and generating errors in the transcript of the genetic code.

Potency	: > 590 units/mg
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- store at room temperature
- soluble in water, (20°C / 100 g/l)
- R: 36/38-42/43
- S: 22-36/37/39-45
- CAS 1405-41-0

G 0124.0001	1 g
G 0124.0005	5 g
G 0124.0010	10 g
G 0124.0025	25 g



G 0907

## GIBBERELIC ACID 3



GA<sub>3</sub> Gibberellin A3  
C<sub>19</sub>H<sub>22</sub>O<sub>6</sub> = 346.4

GA<sub>3</sub> content \_\_\_\_\_ : > 90% of total gibberellins  
crystalline

- soluble in ethanol
- store powder at room temperature
- liquid storage at 2-8°C
- sterilization by filtration
- concentration : 0.01-5.0 mg/l
- R: 36 S: 26-36
- CAS 77-06-5

G 0907.0001 \_\_\_\_\_ 1 g  
G 0907.0005 \_\_\_\_\_ 5 g

G 0938

## GIBBERELIC ACID 4+7

(GA<sub>4+7</sub>), Gibberellin A4 + A7  
Mixture of GA<sub>4</sub>: GA<sub>7</sub> = 2:1

Assay (content A<sub>4</sub>+A<sub>7</sub>) \_\_\_\_\_ : > 90%

- soluble in ethanol
- store powder at room temperature
- liquid storage at 2-8°C
- sterilization by filtration
- concentration : 0.01-5.0 mg/l
- S: 22-24/25
- CAS GA<sub>4</sub>: 468-44-0
- CAS GA<sub>7</sub>: 510-75-8

G 0938.0250 \_\_\_\_\_ 250 mg  
G 0938.1000 \_\_\_\_\_ 1 g

G 0802

## D-GLUCOSE MONOHYDRATE

C<sub>6</sub>H<sub>12</sub>O<sub>6</sub>·H<sub>2</sub>O=198.2

Assay \_\_\_\_\_ : >99.5%

- store at room temperature
- soluble in water (25°C / 500 g/l)
- CAS 5996-10-1

G 0802.1000 \_\_\_\_\_ 1 kg  
G 0802.5000 \_\_\_\_\_ 5 kg

G 0707

## L-GLUTAMIC ACID

C<sub>5</sub>H<sub>9</sub>NO<sub>4</sub> = 147.1

Assay \_\_\_\_\_ : > 98.5%

- store at room temperature
- soluble in water (25°C / 11.1 g/l)
- CAS 56-86-0

G 0707.0500 \_\_\_\_\_ 500 g  
G 0707.1000 \_\_\_\_\_ 1 kg

G 0708

## L-GLUTAMINE

C<sub>5</sub>H<sub>10</sub>N<sub>2</sub>O<sub>3</sub> = 146.15

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water (18°C / 26 g/l)
- CAS 56-85-9

G 0708.0050 \_\_\_\_\_ 50 g  
G 0708.0100 \_\_\_\_\_ 100 g  
G 0708.0250 \_\_\_\_\_ 250 g  
G 0708.0500 \_\_\_\_\_ 500g



G 1346

## GLUTATHIONE REDUCED

 $C_{10}H_{17}N_3O_6S = 307.3$ 

Assay \_\_\_\_\_ : &gt; 98%

- store at 2-8°C
- soluble in water (20°C / ±100 g/l)
- CAS 70-18-8

G 1346.0005 \_\_\_\_\_ 5 g

G 1346.0025 \_\_\_\_\_ 25 g

G 1346.0100 \_\_\_\_\_ 100 g

G 1345

## GLYCEROL

 $C_3H_8O_3 = 92.1$ 

1 l = 1.26 kg

Assay \_\_\_\_\_ : &gt; 98.0%

Water \_\_\_\_\_ : &lt; 2%

- store at room temperature
- soluble in water
- CAS 56-81-5

G 1345.1000 \_\_\_\_\_ 1 l

G 1345.5000 \_\_\_\_\_ 5 l

G 0709

## GLYCINE

 $C_2H_5NO_2 = 75.1$ 

Cell culture tested

Assay \_\_\_\_\_ : &gt; 98.5%

- store at room temperature
- soluble in water (25°C / 250 g/l)
- CAS 56-40-6

G 0709.1000 \_\_\_\_\_ 1 kg

G 0709.5000 \_\_\_\_\_ 5 kg

*Urginea maritima*, a medicinal bulbous crop producing cardiac glycosides.

Heba Shanin MSc and Dr. Geert-Jan de Klerk,  
Wageningen UR Plant Breeding



G 0158

## GLYPHOSATE



N-(phosphonomethyl)glycine

 $C_3H_8NO_5P = 169.1$ 

Glyphosate inhibits the enzyme 5-enolpyruvyl-shikimate 3-phosphate synthetase (EPSPS) which is involved in the shikimate pathway. Inhibition of this enzyme results in an accumulation of shikimate, inhibition of synthesis of aromatic amino acids and secondary metabolites, and results in cell death.

## Enolpyruvylshikimate-phosphate synthase

Q

phosphoenolpyruvate + shikimate-3P R5-enolpyruvylshikimate-3-P

Q

Q

aromatic aminoacids

A. Wilmink and J.J.M. Dons

Plant Molecular Biology Reporter, Vol 11 (2) 1993

Assay \_\_\_\_\_ : &gt; 95%

- store at 2-8°C
- soluble in water
- R: 41-51/53
- S: 26-39-61
- CAS 1071-83-6

G 0158.0001 \_\_\_\_\_ 1 g

G 0158.0005 \_\_\_\_\_ 5 g

G 0167

## GRISEOFULVIN

 $C_{17}H_{17}ClO_6 = 352.8$ 

Griseofulvin is an antifungal agent that causes gross morphological changes in the fungus including the production of binucleate and multinucleate cells. Griseofulvin blocks microtubule assembly and may also affect microtubule function.

Assay \_\_\_\_\_ : &gt; 97%

- store at 2-8°C
- soluble in ethanol
- R: 40-43-60-61
- S: 22-28-36/37/39-45-53
- CAS 126-07-8

G 0167.0005 \_\_\_\_\_ 5 g

G 0167.0025 \_\_\_\_\_ 25 g

G 1375

## GUANIDINE HYDROCHLORIDE


 $\text{CH}_5\text{N}_3\text{HCl} = 95.5$ 

Being a so called chaotropic agent Guanidine HCl is used as a powerful protein denaturant in the purification of proteins and nucleic acids (DNA and RNA). Guanidine HCl is also applied as a solubilizing agent in electrophoresis and in molecular weight determination.

Assay	: > 99.7%
Melting Point	: 185-188
Moisture content	: < 0.2%

- store at room temperature
- soluble in water (30°C / 2280g/l)
- R: 22-36/38 S: 22
- CAS 50-01-1

G 1375.0100	100 g
G 1375.0250	250 g
G 1375.0500	500 g
G 1375.1000	1 kg

H 0710

## L-HISTIDINE

 $\text{C}_6\text{H}_9\text{N}_3\text{O}_2 = 155.2$ 

Assay	: > 99%
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- store at room temperature
- soluble in water (20°C / 41.6 g/l)
- CAS 71-00-1

H 0710.0100	100 g
H 0710.0500	500 g

Iris Heidmann, CMS in chicory.  
ENZA zaden Research and Development B.V.



H 1504

## HEPES



N-[2-Hydroxyethyl]piperazine-N'-[2-ethanesulfonic acid]  
 $\text{C}_8\text{H}_{18}\text{N}_2\text{O}_4\text{S} = 238.3$

Assay	: > 99%
pKa (25° C)	: 7.5
pH range	: 6.8 - 8.2
Moisture content	: < 0.5%

- store at room temperature
- soluble in water
- R: 36/37/38
- S: 26
- CAS 7365-45-9

H 1504.0025	25 g
H 1504.0100	100 g
H 1504.0250	250 g
H 1504.0500	500 g
H 1504.1000	1 kg

H 0168

## 8-HYDROXYQUINOLINE


 $\text{C}_9\text{H}_7\text{NO} = 145.2$ 

Assay	: > 99%
-------	---------

- store dark at room temperature
- insoluble in water
- R: 22-36/38 S: 22
- CAS 148-24-3

H 0168.0025	25 g
H 0168.0100	100 g

Iris Heidmann, Wild type chicory.  
ENZA zaden Research and Development B.V.



H 0192

## HYGROMYCIN B

 $C_{20}H_{37}N_3O_{13} = 527.0$ 

Toxic aminoglycoside produced by *Streptomyces hygrosopicus*. Hygromycin B interferes with the translation step of polypeptide synthesis of prokaryotes and eukaryotes. It inhibits peptide chain elongation by preventing elongation-factor EF-2 dependent translocation. Hygromycin B is used as a selective agent for the incorporation of the APH 4 gene in plant tissue.

- store at 2-8°C
- soluble in water
- R: 26/27/28-37/38-41
- S: 23-26-28-36/37/39-45
- CAS 31282-04-9
- UN 2810

H 0192.0001	1 x 10 <sup>6</sup> U
	5 x 10 <sup>6</sup> U

One vial of Hygromycin B solution contains 1 x 10<sup>6</sup> units and is approximately the equivalent of 1 gram Hygromycin B lyophilized powder.

I 0901

## INDOLE-3-ACETIC ACID

3-Indoleacetic acid, IAA, Heteroauxin

 $C_{10}H_9NO_2 = 175.2$ 

Auxin growth regulator

Assay	: > 99.0%
Melting point	: 166-169°C

- soluble in ethanol and 1N NaOH
- store powder between -20°C and 15°C
- store liquid between -25°C and 15°C
- sterilization : autoclavable or filtration
- concentration : 0.01-3.0 mg/l
- S: 22-24/25
- CAS 87-51-4

I 0901.0005	5 g
I 0901.0025	25 g
I 0901.0100	100 g

Rose after rooting treatment with auxin. Ethylene was removed from the headspace by porous grains coated with KMnO<sub>4</sub> (trade name "Power Pellets")

Dr. Geert-Jan de Klerk, Wageningen UR Plant Breeding

I 0902

## INDOLE-3-BUTYRIC ACID



3-Indolebutyric acid; IBA; 4-[3-indolyl]butyric acid

 $C_{12}H_{13}NO_2 = 203.2$ 

Auxin growth regulator

Assay	: > 98%
Melting point	: 122-124°C

- soluble in ethanol or 1N NaOH
- store powder at 2-8°C
- liquid storage at 2-8°C
- sterilization : autoclavable or filtration
- concentration : 0.01-3.0 mg/l
- R: 23/25-36/37/38
- S: 28-36/37/39-45
- CAS 133-32-4
- UN 2811

I 0902.0005	5 g
I 0902.0025	25 g

I 0609

## MYO-INOSITOL

i-inositol, meso-inositol

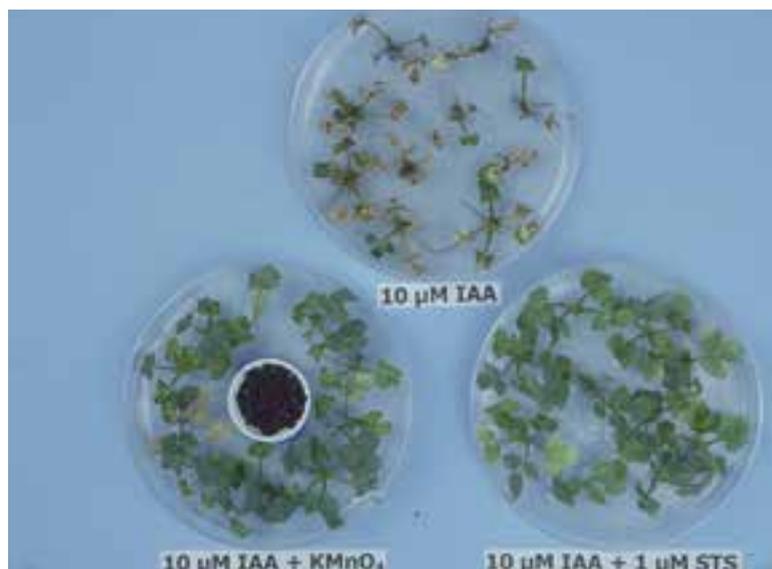
 $C_6H_{12}O_6 = 180.2$ 

Assay	: > 97%
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White powder

- store at room temperature
- soluble in water
- CAS 87-89-8

I 0609.0100	100 g
I 0609.0250	250 g
I 0609.0500	500 g
I 0609.1000	1 kg



I 0711

**L-ISOLEUCINE** $C_6H_{13}NO_2 = 131.2$ 

Assay : &gt; 98.5%

- store at room temperature
- soluble in water (20°C / 32.1 g/l)
- CAS 73-32-5

I 0711.0010 10 g

I 0711.0025 25 g

I 0711.0100 100 g

I 1401

**ISOPROPYL-β-D-1-THIOGALACTOSIDE**

IPTG, DIOXAN FREE

 $C_9H_{18}O_5S = 238.3$ 

Isopropyl-β-D-thiogalactoside, IPTG is a chemical analogue of galactose that can not be cleaved by β-galactosidase. Functioning as an analogue, IPTG binds and inhibits the powerful lac repressor, strongly inducing the production of β-galactosidase.

IPTG is used in conjunction with X-Gal for detection of lac<sup>+</sup> colonies or cells in a colorimetric assay, in order to distinguish recombinants (white) from non recombinants (blue) in cloning strategies using vectors like Lambda-11, M13mp18 and 19, pUC18 and 19, pUR222 containing the lacZ gene. For more detailed information see X-Gal.

Assay : &gt; 99%

Water : &lt; 1 %

Spec. Opt. Rot. : (-)31° - 33°

(α<sub>20</sub><sup>D</sup>; C=1 in H<sub>2</sub>O)

- store dry between -25°C and -15°C
- soluble in ethanol and water
- R: 20/21/22
- S: 22-24/25
- CAS 367-93-1

I 1401.0001 1 g

I 1401.0005 5 g

I 1401.0025 25 g

J 0936

**JASMONIC ACID**

([±]-1α,2β-3-Oxo-2-[cis-2-pentyl]cyclopentaneacetic acid)

 $C_{12}H_{18}O_3 = 210.3$ 

Ravnikar M., Vilhar B., Gogala N., J Plant Growth Regul (1992) 11:29-31

Ravnikar M., Gogala N., J Plant Growth Regul (1990) 9:233-236

Assay : &gt; 95%

- store at 2-8°C
- soluble in ethanol
- CAS 6894-38-8

J 0936.0250 250 mg

K 0126

**KANAMYCIN MONOSULPHATE MONOHYDRATE**

Kanamycin A Sulphate monohydrate

 $C_{18}H_{36}N_4O_{11} \cdot H_2SO_4 \cdot H_2O = 600.6$ 

plant cell culture tested

Kanamycin is an aminoglycoside antibiotic and has a bactericidal action against many gram-negative bacteria. Aminoglycosides are taken up into sensitive bacterial cells by an active transport process. Within the cell they bind to the 30S and to some extent to the 50S subunits of the bacterial ribosome, inhibiting protein synthesis and generating errors in the transcription of the genetic code. Kanamycin is used as a selective agent for the incorporation of the NPT II in 2005 en 2005 (APH3') gene in plant tissue.

Activity : &gt; 750 IU/mg

Kanamycin B : &lt; 4.0%

- store at room temperature
- soluble in water
- R: 61
- S: 45-53
- CAS 25389-94-0

K 0126.0001 1 g

K 0126.0005 5 g

K 0126.0010 10 g

K 0126.0025 25 g

K 0905

## KINETIN

6-Furfurylaminopurine  
 $C_{10}H_9N_5O = 215.2$

Assay : > 98%

Cytokinin growth regulator

- soluble in 1N NaOH
- store powder at 2-8°C
- liquid storage at between -25°C and -15°C
- sterilization : autoclavable or filtration
- concentration : 0.01-5.0 mg/l
- S: 22-24/25
- CAS 525-79-1

K 0905.0001	1 g
K 0905.0005	5 g
K 0905.0025	25 g

L 1372

## LACTOSE MONOHYDRATE

$C_{12}H_{22}O_{11} \cdot H_2O = 360.3$

- store at room temperature
- soluble in water
- CAS 10039-26-6

L 1372.1000	1 kg
L 1372.5000	5 kg

L 1705

## LB AGAR LOW SALT

Ingredients per litre

Tryptone	:	10 g
Sodium chloride	:	5 g
Yeast extract	:	5 g
Microbiological tested Agar	:	10 g

- store dry at room temperature
- dissolve 30 g in 1 l distilled water and adjust the pH to 7.2.
- sterilize by autoclaving at 121°C for 15 minutes.

L 1705.0100	100 g
L 1705.0500	500 g
L 1705.2500	2,5 kg

L 1706

## LB AGAR HIGH SALT

Ingredients per litre

Tryptone	:	10 g
Sodium chloride	:	10 g
Yeast extract	:	5 g
Microbiological tested Agar	:	10 g

- store dry at room temperature
- dissolve 35 g in 1 l distilled water and adjust the pH to 7.2.
- sterilize by autoclaving at 121°C for 15 minutes

L 1706.0100	100 g
L 1706.0500	500 g
L 1706.2500	2,5 kg

L 1703

## LB BROTH LOW SALT

Ingredients per litre

Tryptone	:	10 g
Sodium chloride	:	5 g
Yeast extract	:	5 g

- store dry at room temperature
- dissolve 20 g in 1 l distilled water and adjust the pH to 7.2.
- sterilize by autoclaving at 121°C for 15 minutes.

L 1703.0100	100 g
L 1703.0500	500 g
L 1703.2500	2.5 kg

L 1704

## LB BROTH HIGH SALT

Ingredients per litre

Tryptone	:	10 g
Sodium Chloride	:	10 g
Yeast extract	:	5 g

- store dry at room temperature
- dissolve 25 g in 1 l distilled water and adjust the pH to 7.2.
- sterilize by autoclaving at 121°C for 15 minutes.

H. Miller, Propagation and maintenance of E. coli for the preparation of phage and plasmid DNA., Meths. Enzymol. 152, 145 (1987)  
 S. Heber, B.E. Tropp, Biochim. Biophys. Acta 1129, 1 (1991)

L 1704.0100	100 g
L 1704.0500	500 g
L 1704.2500	2,5 kg

L 0712

**L-LEUCINE** $C_6H_{13}NO_2 = 131.18$ 

Assay \_\_\_\_\_ : &gt; 98.5%

- store at room temperature
- soluble in water (25°C / 25 g/l)
- CAS 61-90-5

L 0712.0100 \_\_\_\_\_ 100 g

L 0127

**LINCOMYCIN HYDROCHLORIDE MONOHYDRATE** $C_{18}H_{34}N_2O_6S \cdot HCl \cdot H_2O = 461.0$ 

Lincomycin is a lincosamide antibiotic with a primarily bacterio-static action against gram-positive bacteria. Lincomycin binds to the 50S subunit of the bacterial ribosome and inhibits the early stages of protein synthesis.

Assay \_\_\_\_\_ : &gt; 82.5% (dried substance)

- store at 2-8°C
- soluble in water
- S: 22-24/25
- CAS 7179-49-9

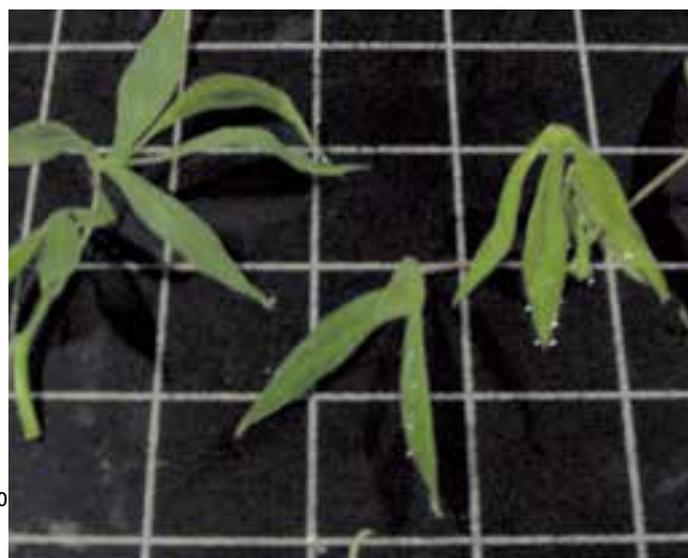
L 0127.0005 \_\_\_\_\_ 5 g

Luciferase-activity in genetically modified cassava-plants.

Left: no activity in leaves and stems when the luciferase-gene is driven by a tuber-specific promoter isolated from cassava.

Right: activity in leaves and stems when the luciferase-gene is driven by the constitutive 35S-promoter.

Ing. Herma Koehorst- van Putten,  
Wageningen UR Plant Breeding



L 1349

**D-LUCIFERIN**

(4,5-Dihydro-[6-hydroxy-2-benzothiazoyl]-4-thiazolecarboxylic acid)  
Free acid

 $C_{11}H_8O_3N_2S_2 = 280.3$ 

Used with firefly luciferase for the determination of ATP using bioluminescence. Firefly luciferase from *Photinus pyralis* catalyzes the adenosine triphosphate dependent oxidative decarboxylation of luciferin producing light emission at a wavelength of 562 nm.

Assay: \_\_\_\_\_

D-Luciferin, HPLC, chemical purity \_\_\_\_\_ : &gt; 99.4%

D-Luciferin HPLC, optical purity \_\_\_\_\_ : &gt; 99.3%

Contains 0.05% acetic acid as antistatic.

- store between -25°C and -15°C
- soluble in alkaline solutions
- protect from light and moisture
- S: 22-26
- CAS 2591-17-5

L 1349.0100 \_\_\_\_\_ 100 mg

L 1349.0250 \_\_\_\_\_ 250 mg

L 1349.0500 \_\_\_\_\_ 500 mg

L 1349.1000 \_\_\_\_\_ 1 g

L 0714

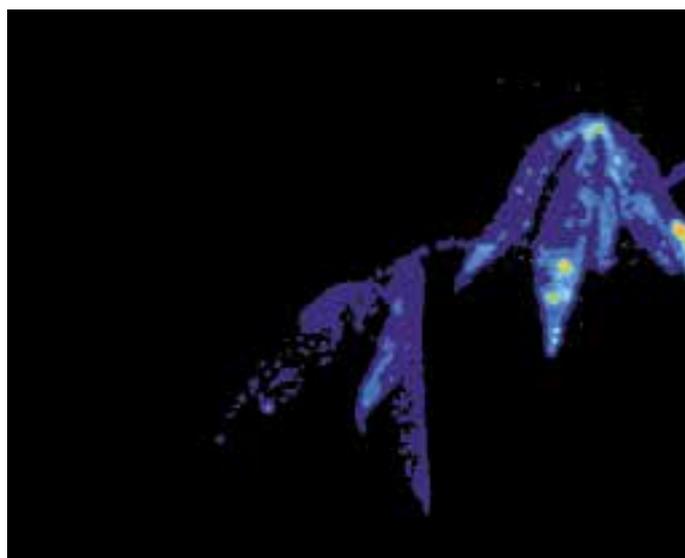
**L-LYSINE HYDROCHLORIDE** $C_6H_{15}ClN_2O_2 = 182.7$ 

Assay \_\_\_\_\_ : &gt; 98.5%

- store at room temperature
- soluble in water
- CAS 657-27-2

L 0714.0100 \_\_\_\_\_ 100 g

L 0714.0500 \_\_\_\_\_ 500 g



M 8002

## MACEROZYME R-10

Macerating Enzyme from *Rhizopus* sp. Macerozyme is well suited for the isolation of plant cells and is often used in combination with cellulase "Onozuka R-10" (Cat no. C 8001). A multi-component enzyme mixture containing the following enzyme activities:

Enzyme activity	: > 3,000 U/g
Pectinase	: 0.5 U/mg
Cellulase	: 0.1 U/mg
Hemicellulase	: 0.25 U/mg
Loss on drying	: < 10%

- solubility : 1 mg/ml 0.1 M Sodium acetate buffer pH 4.5
- store at 2-8°C
- pH optimum : 3.5 - 7.0
- CAS 9032-75-1

Yamada, Y et al., Agr. Biol. Chem. 36, 1055-1059, 1972  
 Barraclough, R. & Ellis, R.J., Eur. J. Biochem, 94, 165, 165-177  
 Okada, G., Methods Enzymol. Vol. 160, 259-263

M 8002.0001	1 g
M 8002.0005	5 g
M 8002.0010	10 g

M 0533

MAGNESIUM CHLORIDE  
HEXAHYDRATE
 $MgCl_2 \cdot 6H_2O = 203.3$ 

Assay \_\_\_\_\_ : > 98%

- store at room temperature
- soluble in water (20°C / 1670 g/l)
- CAS 7791-18-6

M 0533.1000	1 kg
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M 0513

MAGNESIUM SULPHATE  
HEPTAHYDRATE
 $MgSO_4 \cdot 7H_2O = 246.5$ 

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water (20°C / 710 g/l)
- CAS 10034-99-8

M 0513.1000	1 kg
M 0513.5000	5 kg



Shoots regeneration from somatic cell of barley immature scutellum (Dr. J. Imani, Institute of Phytopathology & Applied Zoology, Justus-Liebig-University Giessen, Germany)

M 0921

## MALEIC HYDRAZIDE


 $C_4H_4N_2O_2 = 112.1$ 

Assay \_\_\_\_\_ : > 98%

- soluble in 1N NaOH
- store powder at room temperature.
- liquid storage at 2-8°C
- sterilization : filtration
- concentration : 0.01-10.0 mg/l
- R: 36/37/38 S: 26-36
- CAS 123-33-1

M 0921.0100	100 g
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M 1315

## MALIC ACID-(DL)


 $C_4H_6O_5 = 134.1$ 

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water (20°C / ±530 g/l)
- R: 36 S: 26-36
- CAS 617-48-1

M 1315.1000	1 kg
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M 1327

## MALT EXTRACT

Prepared by extracting the soluble products from sprouted grain.

Assay	: > 60.0% maltose
Sodium chloride	: < 1.0%
pH (3% solution)	: 4.8-5.8

- store dry at room temperature
- soluble in water
- CAS 8002-48-0

M 1327.0100	100 g
M 1327.0500	500 g

M 0811

## MALTOSE MONOHYDRATE

 $C_{12}H_{22}O_{11} \cdot H_2O = 360.3$ 

Assay	: > 95%
Glucose	: < 3.0%

- store at room temperature
- soluble in water (25°C / 850 g/l)
- CAS 6363-53-7

M 0811.0250	250 g
M 0811.0500	500 g
M 0811.1000	1 kg
M 0811.5000	5 kg

Haworthia micropropagation  
Succulent Tissue Culture, The Netherlands



M 0514

MANGANESE SULPHATE  
MONOHYDRATE
 $MnSO_4 \cdot H_2O = 169.0$ 

Assay	: > 98%
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- store at room temperature
- soluble in water (20°C / 750 g/l)
- R: 48/20/22-51/53 S: 22-61
- CAS 10034-96-5
- UN 3077

M 0514.0250	250 g
M 0514.0500	500 g
M 0514.1000	1 kg

M 0803

## D-MANNITOL

 $C_6H_{14}O_6 = 182.2$ 

Assay	: > 98%
Sorbitol	: < 2%

- store at room temperature
- soluble in water (25°C/213 g/l)
- CAS 69-65-8

M 0803.1000	1 kg
M 0803.5000	5 kg

Micropropagation illuminated by LED-Light  
Succulent Tissue Culture, The Netherlands



M1392

**D-MANNOSE** $C_6H_{12}O_6 = 180.2$ 

Most plants are incapable of surviving on a synthetic medium containing mannose as energy source, because these plants miss the enzyme Phosphomannose isomerase (PMI). This leads to an accumulation of mannose 6-phosphate which depletes intracellular stores of inorganic phosphate.

In the presence of PMI, mannose 6-phosphate is converted into fructose 6-phosphate to enter the glycolytic pathway.

A new selection system has been developed by genetically transforming plant cells with the gene ManA, coding for PMI, as marker. Cells containing this gene are able to grow on mannose.

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water
- CAS 3458-28-4

M 1392.0100	100 g
M 1392.0500	500 g
M 1392.1000	1 kg

M 0129

**6-MERCAPTOPYRINE MONOHYDRATE** $C_5H_4N_4S \cdot H_2O = 170.2$ 

6-Mercaptopurine is an antineoplastic agent that acts as an antimetabolite. It is an analogue of the natural purines hypoxanthine and adenine. After the intracellular conversion of mercaptopurine to active nucleotides, it appears to exhibit a variety of actions including interference with nucleic acid synthesis.

Assay \_\_\_\_\_ : > 96.0%

- store at room temperature
- soluble in ethanol
- R: 23/25-40
- S: 22-28-53
- CAS 6112-76-1
- UN 2811

M 0129.0005	5 g
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M 1503

**MES MONOHYDRATE**

2-(N-morpholino)ethanesulfonic acid

 $C_6H_{13}NO_4S \cdot H_2O = 213.2$ 

A highly purified quality of MES with excellent properties for molecular biology and cell culture. MES is an excellent buffer for use in Plant Culture media because of its high buffer capacity and its pH range of 5.5-6.7.

Assay	: > 99%
pKa (20° C)	: 5.9 - 6.3
pH (0.5 M in water, 20°C)	: 2.5 - 4.0
pH range	: 5.5 - 6.7

- store at room temperature
- soluble in water (25°C / >100 g/l)
- R: 36/37/38 S: 26-36
- CAS 4432-31-9

M 1503.0025	25 g
M 1503.0100	100 g
M 1503.0250	250 g
M 1503.1000	1 kg

M 0715

**L-METHIONINE** $C_5H_{11}NO_2S = 149.2$ 

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water (20°C / 48 g/l)
- CAS 63-68-3

M 0715.0100	100 g
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M 0130

## METHOTREXATE



Amethopterin  
 $C_{20}H_{22}N_8O_5 = 454.4$

An antineoplastic agent that acts as an antimetabolite of folic acid. Within the cell folic acid is reduced to dihydrofolic - and tetrahydrofolic acid. Methotrexate competitively inhibits the enzyme dihydrofolate reductase and prevents the formation of tetrahydrofolate, which is necessary for purine and pyrimidine synthesis and consequently the formation of DNA and RNA. Most active against cells in the S phase.

- store dry at room temperature
- soluble in alkaline solutions
- protect from light
- R: 23/24/25-36/37/38-46-60-61
- S: 07-13-22-26-36/37/39-45-53
- CAS 59-05-2
- UN 2811

M 0130.0001 1 g

M 0918

## METHYL JASMONATE

$C_{13}H_{20}O_3 = 224.29$

Assay	:> 97%
Specific gravity	: 1028 mg/ml

- store at room temperature, dark and dry
- soluble in ethanol
- concentration: 0.01-5.0 mg/l
- CAS 39924-52-2

M 0918.0001 1 ml

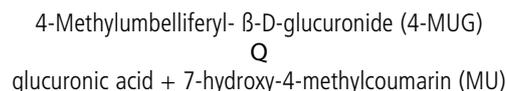
1 ml solution contains 100 ppm d,l-tocopherol

M 1404

4-METHYLUMBELLIFERYL- $\beta$ -D-GLUCURONIDE TRIHYDRATE

4-MUG trihydrate  
 $C_{16}H_{16}O_9 \cdot 3H_2O = 406.4$

4-Methylumbelliferyl- $\beta$ -D-glucuronide trihydrate (**4-MUG**) is a fluorescent substrate for  $\beta$ -D-glucuronidase (GUS) encoded by the *gusA* gene isolated originally from *E. coli*. Cleavage of the substrate 4-MUG by a  $\beta$ -glucuronidase activity leads to the generation of the fluorogenic product 4-MU, which can be visualized or detected by irradiation with UV light.



The fluorescence assay allows quantitation of GUS activity by means of a fluorimeter in protein extracts in conjunction with 4-MUG at a peak excitation of 365 nm (UV) and a peak emission of 455 nm (blue).

- store dry at 2-8°C
- soluble in DMF and DMSO
- S: 22-24/25
- CAS 6160-80-1

M 1404.0100 100 mg

M 1404.1000 1 g

M 0131

## METRONIDAZOLE

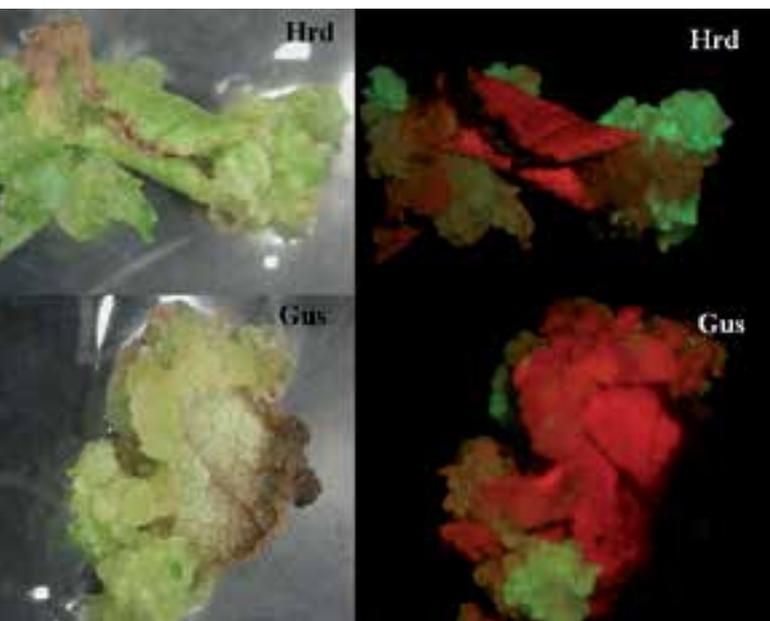


$C_6H_9N_3O_3 = 171.2$

- store at room temperature
- soluble in diluted acids and DMFO
- R: 20/21/22-33-40 S: 26-36/37/39
- CAS 443-48-1

M 0131.0025 25 g

M 0131.0100 100 g

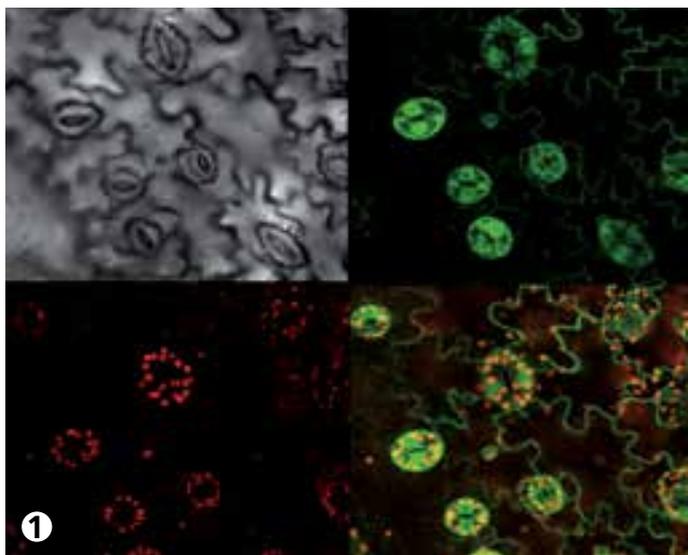


1. Stomata cell of transgenic barley expressing GFP. Overlay, confocal laser microscopy, Leica Germany.
2. Transgenic tobacco stomata cell expressing GFP- Confocal laser microscopy, Leica Germany

(Dr. J. Imani, Institute of Phytopathology & Applied Zoology, Justus-Liebig-University Giessen, Germany, Prof. R. Hueckelhoven, Centre of Life and Food Sciences Weihenstephan, Germany)

- ← GFP expression in strawberry, transformed with constructs containing the *gfp* gene in addition to other genes.  
 Left : normal light  
 Right: UV light

Ing. Aranka van der Burgh, Wageningen UR Plant Breeding



M 0132

### MICONAZOLE NITRATE



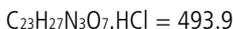
Miconazole as an imidazole antifungal agent interferes with ergosterol synthesis and therefore alters the permeability of the cell membrane of sensitive fungi and yeasts.

- store at room temperature
- soluble in propylene glycol
- R: 20/21/22-43 S: 36/37/39
- CAS 22832-87-7

M 0132.0001	1 g
M 0132.0005	5 g

M 0172

### MINOCYCLINE HYDROCHLORIDE



Minocycline is a bacteriostatic antibiotic with activity against gram-positive and gram-negative bacteria. Minocycline has a spectrum of activity like that of tetracycline but is more active against many species. Within the cell minocycline binds reversible to the 30S subunit of the ribosome, preventing the binding of aminoacyl transfer RNA and inhibiting protein synthesis and hence cell growth.

Assay	: > 96%
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- store at 2-8°C
- soluble in water
- protect from light
- R: 33-36/37/38-63-64 S: 26-36-45
- CAS 13614-98-7

M 0172.0001	1 g
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M 0133

### MITOMYCIN C



Mitomycin C is a toxic antibiotic with antineoplastic properties. It acts as an alkylating agent after activation and also suppresses the synthesis of nucleic acids. It is a cell-cycle non specific agent and is most active in the late G1 and early S phases.

- store at room temperature
- soluble in ethanol, slightly soluble in water
- R: 25-33-40-45 S: 22-28-36/37/39-45
- CAS 50-07-7, UN 2811

M 0133.0002	1 x 2 mg
	5 x 2 mg
	25 x 2 mg

Each vial contains 2 mg mitomycin C and 48 mg NaCl as recipient

M 1502

### MOPS



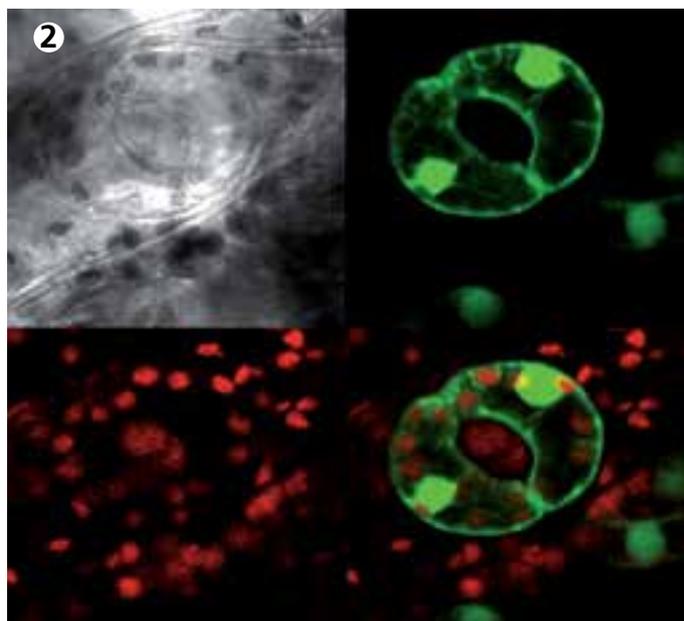
4-Morpholino propanesulfonic acid



Assay	: > 99.5%
pKa (25° C)	: 7.0 - 7.2
pH (10% in water)	: ca. 4.0
pH range	: 6,5 - 7,9

- store at room temperature
- soluble in water (20°C / > 100 g/l)
- R: 36/37/38 S: 26-36
- CAS 1132-61-2

M 1502.0025	25 g
M 1502.0100	100 g
M 1502.0250	250 g
M 1502.1000	1 kg



**M 1415**
**MTT**

Thiazolyl Blue Tetrazolium Bromide

 $C_{18}H_{16}N_5SBr = 414.3$ 

MTT is a water soluble salt of tetrazolium salt yielding a yellowish solution when prepared in media or salt solutions lacking phenol red. By cleavage of the tetrazolium ring by dehydrogenase enzymes, dissolved MTT is converted into insoluble purple formazan. This water insoluble formazan can be solubilized using isopropanol or other solvents and the dissolved material is measured spectrophotometrically yielding maximum absorbance at 565 nm as a function of concentration of converted dye.

Assay \_\_\_\_\_ : &gt; 98%

- store at 2-8°C
- soluble in water (20°C / > 10 g/l)
- S: 22-24/25
- CAS 298-93-1

M 1415.0001	1 g
M 1415.0005	5 g
M 1415.0025	25 g

Apple shoots after 3 weeks of rooting on medium with NAA and the ethylene inhibitor STS

Geert-Jan de Klerk, Wageningen UR Plant Breeding

**N 0134**
**NALIDIXIC ACID**

 $C_{12}H_{12}N_2O_3 = 232.2$ 

Nalidixic acid is active against gram-negative bacteria. The antibiotic is considered to act by interfering with the replication of bacterial DNA, probably by inhibiting DNA gyrase (topoisomerase) activity.

Assay \_\_\_\_\_ : &gt; 99.4%

- store at room temperature
- slightly soluble in water (23°C / 0.1 g/l)
- R: 40-42/43-63
- S: 22-24-36/37-45
- CAS 389-08-2

N 0134.0005	5 g
N 0134.0025	25 g

**N 0903**
**α-NAPHTHALENE ACETIC ACID**


NAA, 1-Naphthalene Acetic acid

 $C_{12}H_{10}O_2 = 186.2$ 

Auxin growth regulator

Assay \_\_\_\_\_ : &gt; 98%

- store at room temperature
- slightly soluble in water (20 °C / < 0,4 g/l), soluble in alcohol (20 °C / 30 g/l)
- liquid storage at 2-8°C
- sterilization : autoclavable
- R: 22 S: 13
- CAS 86-87-3

N 0903.0025	25 g
N 0903.0050	50 g
N 0903.0100	100 g



10 μM NAA

10 μM NAA + 10 μM STS

**N 0912**
**β-NAPHTHOXYACETIC ACID**


2-Naphthoxyacetic Acid

 $C_{12}H_{10}O_3 = 202.2$ 

Assay \_\_\_\_\_ : &gt; 97%

- Store powder at room temperature
- soluble in 1 N NaOH
- liquid storage at 2-8°C
- sterilization : autoclavable
- R: 36/37/38-20/21/22 S: 24/25
- CAS 120-23-0
- UN 2783

N 0912.0025	25 g
N 0912.0500	500 g

N 1350

**1-NAPHTHYLPHOSPHATE  
SODIUM MONOHYDRATE** $C_{10}H_8NaO_4P \cdot H_2O = 264.2$ 

Substrate for determination of phosphatase

Assay	: > 99 %
Free Phosphate (PO <sub>4</sub> )	: < 0.1%
Free Naphthyl	: < 0.1%
Water	: 5 - 10%

- store at 2-8°C
- soluble in water
- protect from moisture
- R: 36/37/38 S: 26-36
- CAS 81012-89-7

N 1350.0001	1 g
N 1350.0005	5 g

N 0926

**N-(1-NAPHTHYL)  
PHTHALAMIC ACID**Naptalam, NPA  
 $C_{18}H_{13}NO_3 = 291.3$ 

Non competitive transport inhibitor of auxin.

- R: 20 S: 22-24/25
- CAS 132-66-1, UN 2588

N 0926.0250	250 mg
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Willemsen en Bourgondiën B.V., The Netherlands



Willemsen en Bourgondiën B.V., The Netherlands

M 0135

**NEOMYCIN SULPHATE** $C_{23}H_{46}N_6O_{13} \cdot 3H_2SO_4 = 908.9$ 

Potency \_\_\_\_\_ : &gt; 680 µg/mg \_\_\_\_\_

Neomycin is an aminoglycoside with a bactericidal action against many gram-negative bacteria. Aminoglycosides are taken up into sensitive bacteria by an active transport process. In the cell they bind to 30S and to some extent to the 50S subunits of the bacterial ribosome, inhibiting protein synthesis and generating errors in the transcription of the genetic code. Neomycin is used as a selective agent for the incorporation of the NPT II (APH3') gene in plant tissue.

- store at 2-8°C
- soluble in water (20°C / 300 g/l)
- R: 36/37/38-42/43-63 S: 22-26-36/37/39
- CAS 1405-10-3

M 0135.0025	25 g
M 0135.0100	100 g

N 0610

**NICOTINAMIDE** $C_6H_6N_2O = 122.1$ 

Assay \_\_\_\_\_ : &gt; 99% \_\_\_\_\_

- store at room temperature
- soluble in water (25°C / 1000 g/l)
- R: 36/37/38 S: 26-36
- CAS 98-92-0

N 0610.0100	100 g
N 0610.0250	250 g

N 0611

## NICOTINIC ACID

 $C_6H_5NO_2 = 123.1$ 

Assay : &gt; 99%

- store at room temperature
- soluble in water (20°C / 18 g/l)
- R: 36 S: 22-26
- CAS 59-67-6

N 0611.0100 100 g

N 0611.0250 250 g

N 0611.0500 500 g

N 1411

## NITRO BLUE TETRAZOLIUM



Nitro Tetrazolium Blue, NBT

 $C_{40}H_{30}Cl_2N_{10}O_6 = 817.6$ 

NBT is used in conjunction with X-Phos for colorimetric detection of alkaline phosphatase activity in blotting, immunohistochemical and cytochemistry techniques.

Assay : &gt; 99%

Water : &lt; 1.5%

- store dry at 2-8°C
- soluble in methanol and water
- protect from light
- R: 20/21/22 S: 22-24/25-36
- CAS 38184-50-8

N 1411.0100 100 mg

N 1411.1000 1 g

O 1409

## 2-NITROPHENYL-β-D-GALACTOPYRANOSIDE

ONPG

 $C_{12}H_{15}NO_8 = 301.3$ 

ONPG is a colorimetric and spectrophotometric substrate for detection of β-galactosidase activity. ONPG is cleaved by β-galactosidase via hydrolysis at the β-1-4-glycosidic bond between 2-nitrophenol and galactose. The released 2-nitrophenol is measured spectrophotometrically at 405 nm. The absorbance intensity at this wavelength is directly related to the specific activity.

Assay : &gt; 90%

- store dry at 2-8°C
- soluble in DMSO, DMF and water
- protect from light and moisture
- S: 22-24/25
- CAS 369-07-3

O 1409.0005 5 g

O 1409.0025 25 g

N 1408

## p-NITROPHENYL-β-D-GLUCURONIDE

NPG

 $C_{12}H_{13}NO_9 = 315.2$ 

NPG is substrate for detection of β-glucuronidase activity. NPG is cleaved by GUS via hydrolysis at the β-1-glycosidic bond between 4-nitrophenol and glucuronic acid. The released 4-nitrophenol can be spectrophotometrically measured at 402-410 nm. The absorbance intensity at these wavelengths is directly related to the specific activity.

Assay : &gt; 99%

- store dry at between -25°C and -15°C
- soluble in DMSO, DMF and water
- protect from moisture
- S: 22-24/25
- CAS 10344-94-2

N 1408.0250 250 mg

N 1408.1000 1 g

In house developed system of growth chamber. Light armature integrated in construction. Light level adjustable between 500 - 10.000 lux. All shelves with water cooling. Setting of temperature variable between shelves in one growth room.

Iribov B.V., The Netherlands



N 0138

## NYSTATIN

 $C_{47}H_{75}NO_{17} = 926.1$ 

Nystatin is a polyene antifungal antibiotic produced by *Streptomyces noursei*. It acts mainly by interfering with the permeability of the cell membrane of sensitive fungi and yeasts by binding to sterols.

Potency \_\_\_\_\_ : > 5000 IU/mg \_\_\_\_\_

- store dry at 2-8°C
- soluble in DMSO
- S: 22-24/25
- CAS 1400-61-9

N 0138.0005	5 g
N 0138.0010	10 g
N 0138.0025	25 g

O 1351

## L-ORNITHINE HYDROCHLORIDE

 $C_5H_{12}N_2O_2 \cdot HCl = 168.6$ 

Polyamine growth regulator.

Assay \_\_\_\_\_ : > 99% \_\_\_\_\_

- store at room temperature
- soluble in water (25°C / 500 g/l)
- CAS 3184-13-2

O 1351.0025	25 g
O 1351.0100	100 g
O 1351.0500	500 g

O 1318

## ORYZALINE


 $C_{12}H_{18}N_4O_6S = 346.4$ 

Assay \_\_\_\_\_ : > 96% \_\_\_\_\_

- store at room temperature
- soluble in DMSO
- R: 51/53 S: 22-24/25-60
- CAS 19044-88-3

Verhoeven, H.A. et al. *Acta Bot. Neerl.*, 40(2) : 97 (1001) *Planta* 182 : 408 (1990) Van Tuyl J.M. et al. *Acta Horticultura* 325 : 625 (1992)

O 1318.1000 \_\_\_\_\_ 1 g \_\_\_\_\_

O 0140

## OXYTETRACYCLINE HYDROCHLORIDE


 $C_{22}H_{24}N_2O_9 \cdot HCl = 496.9$ 

Oxytetracycline is a bacteriostatic antibiotic with activity against gram-positive and gram-negative bacteria. Within the cell tetracyclines bind reversibly to the 30S subunit of the ribosome, preventing the binding of aminoacyl transfer RNA and inhibiting protein synthesis and hence cell growth.

- store at room temperature
- soluble in ethanol and water
- R: 63 S: 36/37/39-45-53
- CAS 2058-46-0

O 0140.0005	5 g
O 0140.0025	25 g

Succulent Tissue Culture, The Netherlands



P 0922

## PACLOBUTRAZOL



N-dimethylaminosuccinamic acid  
 $C_{15}H_{20}ClN_3O = 293.8$

G. Marino, The effect of Paclobutrazol on in vitro rooting, transplant establishment and growth of fruit plants. Plant Growth Reg. 7:237-246 (1981) Ziv, M. Ariel, Bud proliferation and plant regeneration in liquid-cultured Philodendron treated with Ancymidol and Paclobutrazol. Plant Growth Regulation 10 : 53-57, 1991.

- very slightly soluble in water (20 mg/l)
- store powder at room temperature
- liquid storage 2-8°C
- sterilization : filtration
- concentration : 0.25-0.5 mg/l
- R: 20/22-36 S: 36/37/39
- CAS 76738-62-0
- UN 1325

P 0922.0500	500 mg
P 0922.1000	1 g

C 0604

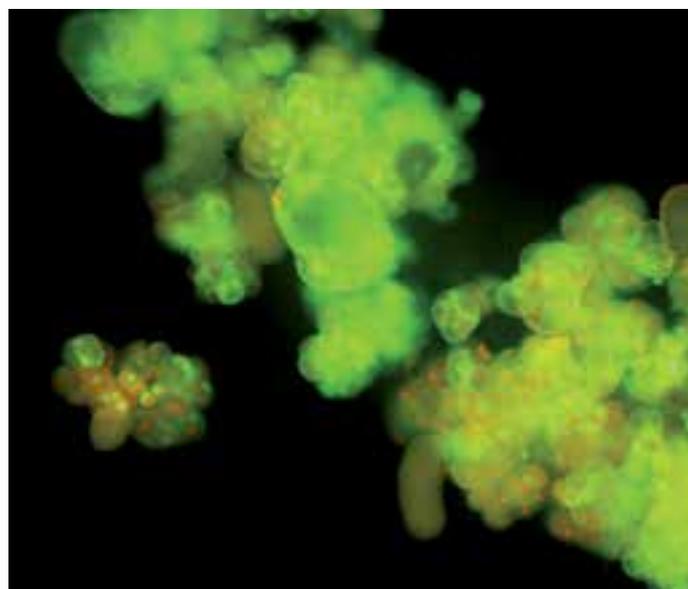
## D(+) PANTOTHENATE CALCIUM

$C_{18}H_{32}N_2O_{10}Ca = 476.5$

Assay \_\_\_\_\_ : > 98%

- store at room temperature
- soluble in water (330 g/l)
- CAS 137-08-6

C 0604.0100	100 g
C 0604.0500	500 g



P 0141

## PAROMOMYCIN SULPHATE



$C_{23}H_{45}N_5O_{14}.xH_2SO_4 = 615.6$  (base)

Paromomycin is an aminoglycoside antibiotic and has a mode of action similar to kanamycin and neomycin. It is used as a selective agent for the incorporation of the NPT II (APH3') gene in plant tissue. Because of the switch of the 3' NH<sub>2</sub> and 6' OH group in the 3-Amino-3-deoxyglucose ring of both antibiotics, paromomycin causes a higher misreading in plant cells and can be a better selective agent than kanamycin and neomycin.

Assay \_\_\_\_\_ : > 675 µg/mg

- store at room temperature
- soluble in water (20°C/ 250 g/l)
- R: 36/37/38-61
- S: 26-36-45
- CAS 1263-89-4

P 0141.0001	1 g
P 0141.0005	5 g
P 0141.0025	25 g

P 8004

## PECTOLYASE Y-23

Pectolyase Y-23 is a highly purified maceration enzyme from *Aspergillus japonicus*. It contains two types of pectinase such as endo-polygalacturonase and endo-pectin lyase in high activity.

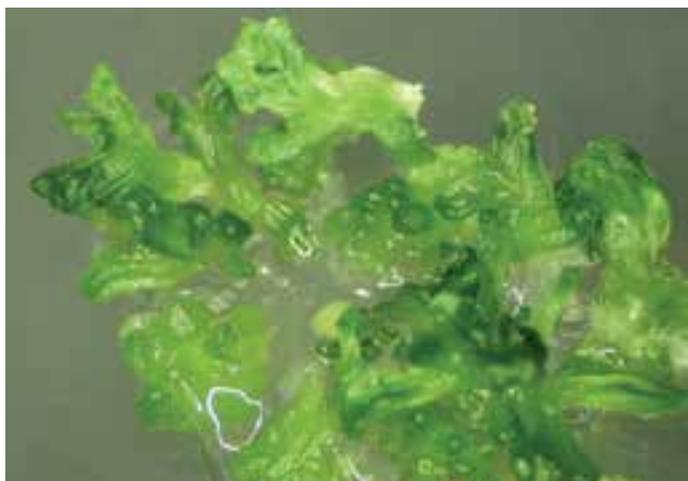
S. Ishii and T. Yokotsuka, Purification and properties of endo-polygalacturonase from (*aspergillus japonicus*), (Agric.Biol. Chem, 36, 1885 (1972)

Specific Activity \_\_\_\_\_ : approximately 1000 maceration units per gram

- store at 2-8°C
- CAS: 9033-35-6

P 8004.0001	1 g
P 8004.0005	5 g

Iris Heidmann,  
 Acridine orange staining on protoplasts.



Iris Heidmann,  
Arabidopsis regeneration from protoplasts.

P 0142

## PENICILLIN G SODIUM


 $C_{16}H_{17}N_2NaO_4S = 356.4$ 

Penicillin G is an inhibitor of bacterial cell wall synthesis. It inhibits the crosslinking of peptidoglycan by binding and inactivating of transpeptidases. Active against gram-positive and some gram-negative bacteria.  $\beta$ -lactamase sensitive.

Assay \_\_\_\_\_ : > 96%

- store at < 30°C protected from light
- soluble in water (100 g/l)
- R: 42 S: 22
- CAS 69-57-8

P 0142.0005	5 g
P 0142.0025	25 g
P 0142.0100	100 g

P 1328

## PEPTONE

Mix of peptides and free amino acids obtained by pancreatic hydrolysis of animal tissues. Due to its low NaCl content this quality is well suited for Plant Tissue Culture.

Sodium chloride	: < 7.0%
Total nitrogen (TN)	: 11.5-12.5%
Amino nitrogen (AN)	: 3.5-4.5%
AN/TN	: 0.28-0.39
Loss on drying	: < 6.0%

- store at room temperature
- soluble in water
- CAS 73049-73-7

P 1328.0100	100 g
P 1328.0500	500 g
P 1328.1000	1 kg

P 1707

## PEPTONE WATER

Ingredients per litre

Peptone	: 10 g
Sodium chloride	: 5 g

- store dry at room temperature
- dissolve 15 g in 1 l distilled water and adjust the pH to 7.2.

P 1707.0100	100 g
P 1707.0500	500 g
P 1707.2500	2,5 kg

B 1702

## BUFFERED PEPTONE WATER

Light Phosphate buffer

Ingredients per litre

Peptone	: 10 g
Phosphate buffer	: 5 g
Sodium chloride	: 5 g
Final pH 7.2 +/- 0.2 at 25°C	

- store dry at room temperature
- soluble in water

B 1702.0100	100 g
B 1702.0500	500 g

Inquire for bulk quantities.

P 0716

## L-PHENYLALANINE

 $C_9H_{11}NO_2 = 165.2$ 

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water (27 g/l / 20°C)
- CAS 63-91-2

P 0716.0100	100 g
P 0716.0500	500 g

P 0187

## PHLEOMYCIN



Phleomycin is produced by *Streptomyces verticillus* and part of the structurally related group of bleomycin/phleomycin type antibiotics. The antibiotic is applied as a selective agent in transformation experiments with mammalian cells, plant cells and yeast.

The cytotoxic action of the family of Bleomycin/Phleomycin related antibiotics results from their ability to cause fragmentation of DNA. The antibiotic binds to DNA through its amino-terminal peptide, and the activated complex generates free radicals that are responsible for scission of the DNA chain. Studies in vitro indicate that the antibiotic causes accumulation of the cells in the G2 phase of the cell cycle.

- store at 2-8°C
- soluble in water
- R: 22-40-42/43
- S: 24/25-36/37/39
- CAS 11006-33-0

P 0187.0100	100 mg
P 0187.0250	250 mg

P 0159

## DL-PHOSPHINOTHRICIN



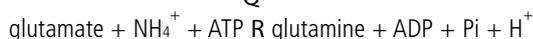
PPT  
 $C_5H_{15}N_2O_4P = 198.2$

DL-Phosphinothricin (PPT) is an analogue of glutamate and acts as a competitive inhibitor of glutamine synthetase.

This enzyme is involved in assimilation of ammonia and plays a key role in nitrogen assimilation.

## Glutamine synthetase

Q



A. Wilmink and J.J.M. Dons, Plant Molecular Biology Reporter, Vol 11 (2) 1993

- store at room temperature
- soluble in water
- R: 23/24/25
- S: 36/37/39-45
- CAS 77182-82-2

P 0159.0250	250 mg
P 0159.1000	1 g

P 1353

## PHLOROGLUCINOL



$C_6H_6O_3 = 126.1$

Assay \_\_\_\_\_ : > 98%

- store at room temperature
- soluble in water
- R: 36/37/38-41
- S: 26-36
- CAS 108-73-6

P 1353.0025	25 g
P 1353.0100	100 g

P 0914

## PICLORAM



4-Amino-3,5,6-tri-chloropicolinic acid  
 $C_6H_3Cl_3N_2O_2 = 241.5$

Collins, G.B., Use of 4-Amino-3,5,6-trichloropicolinic acid as an auxin source in plant tissue cultures Crop Science 18, 286 (1978)

- soluble in 1N NaOH
- store powder at room temperature
- liquid storage between -25°C and -15°C
- sterilization : autoclavable or filtration
- concentration : 0.01-3.0 mg/l
- R: 20/21/22-36-45
- S: 26-36/37/39-45
- CAS 1918-02-1

P 0914.0005	5 g
P 0914.0010	10 g
P 0914.0050	50 g



Herman Schreuder

P 1505

## PIPES

PIPERAZINE-N,N'-BIS-2-ETHANESULFONIC ACID

 $C_8H_{18}N_2O_6S_2 = 302.4$ 

Assay	: > 99%
pKa (25°C)	: 6.7 - 6.9
pH range	: 6.1 - 7.5

- store at room temperature
- slightly soluble in water, soluble in 0.2 N NaOH (20 °C / 30 g/l)
- S: 22-24/25
- CAS 5625-37-6

P 1505.0025	25 g
P 1505.0100	100 g
P 1505.0250	250 g
P 1505.0500	500 g

P 0813

## POLYETHYLENE GLYCOL 400

PEG 400

Average mol weight	: 380 - 420
Hydroxyl number	: 264 - 300
Viscosity	: 105 - 130 mPa.s

- store at room temperature
- soluble in water
- S: 24/25
- CAS 24322-68-3

P 0813.1000	1 kg
P 0813.5000	5 kg

P 0804

## POLYETHYLENE GLYCOL 4000

PEG 4000

Average mol weight	: 3700 - 4500
Hydroxyl number	: 25 - 32
Freezing point	: 53-59°C

- store at room temperature
- soluble in water
- CAS 25322-68-3

P 0804.1000	1 kg
P 0804.5000	5 kg

P 0805

## POLYETHYLENE GLYCOL 6000

PEG 6000

Average mol weight	: 5000 - 7000
Hydroxyl number	: 16 - 22
Freezing point	: 55 - 61°C

- store at room temperature
- soluble in water
- CAS 25322-68-3

P 0805.1000	1 kg
P 0805.5000	5 kg

P 0145

## POLYMXIN B SULPHATE

 $C_{55}H_{96}N_{16}O_{13} \cdot 2H_2SO_4 = 1385$ 

Polimixin B is a mixture of sulphates of polypeptides produced by certain strains of *Bacillus polymixa*. Polymixin acts primarily by binding to membrane phospholipids and disrupting the bacterial cytoplasmic membrane. It is active against gram-negative bacteria, especially *Pseudomonas* species.

Potency	: > 6500 units/mg
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- store at 2-8°C
- soluble in water
- R: 22 S: 36
- CAS 1405-20-5

P 0145.0001	1 g
P 0145.0005	5 g

P 1362

POLYOXYETHYLENESORBITAN  
MONOLAUATE

Tween 20, Polysorbate 20

 $C_{58}H_{114}O_{26} = 1227.7$ 

Fatty acid composition	: Lauric acid approximately 50%
Other fatty acids	: Myristic, palmitic and oleic acids
1 l =	1.08 – 1.12 kg

- store at room temperature
- soluble in water
- CAS 9005-64-5

P 1362.0500	500 ml
P 1362.1000	1l

P 1365

**POLYOXYETHYLENESORBITAN MONOOLEATE**

Tween 80, Polysorbate 80  
 $C_{64}H_{124}O_{26} = 1310$

Fatty acid composition : Oleic acid approximately 70%  
 Other fatty acids : Linoleic, palmitic and stearic acids  
 $l = 1.06 - 1.10 \text{ kg}$

- store at room temperature
- soluble in water
- CAS 9005-65-6

P 1365.0500 500 ml  
 P 1365.1000 1 l

P 1368

**POLYVINYL PYRROLIDONE**

PVP 10  
 Average mol weight 10,000

Absorbant for excreted phenolic substances

- store at room temperature
- soluble in water
- S: 22
- CAS 9003-39-8

P 1368.0100 100 g  
 P 1368.0500 500 g

P 0515

**POTASSIUM CHLORIDE**

KCl = 74.6

Assay : > 99%  
 crystalline

- store at room temperature
- soluble in water
- CAS 7447-40-7

P 0515.1000 1 kg

P 0574 (was P 0516)

**POTASSIUM DIHYDROGEN PHOSPHATE**

$KH_2PO_4 = 136.1$

Assay : > 98%  
 crystalline

- store at room temperature
- soluble in water (20°C / 222 g/l)
- CAS 7778-77-0

P 0574.1000 1 kg  
 P 0574.5000 5 kg

P 0573

**DI-POTASSIUM HYDROGEN PHOSPHATE**

$K_2HPO_4 = 174.2$

Assay : > 98%

- store at room temperature
- soluble in water
- S: 22-24/25
- CAS 7758-11-4

P 0573.1000 1 kg  
 P 0573.5000 5 kg

P 0517

**POTASSIUM HYDROXIDE**

KOH = 56.11

Assay : > 85%

- store dry at room temperature
- soluble in water (20°C / 1120 g/l)
- R: 22-35
- S: 26-36/37/39-45
- CAS 1310-58-3
- UN 1813

P 0517.0500 500 g  
 P 0517.1000 1 kg

P 0518

## POTASSIUM IODIDE

KI = 166.0

Assay \_\_\_\_\_ : &gt; 99% \_\_\_\_\_

- store at room temperature
- soluble at room temperature (1430 g/l / 20°C)
- CAS 7681-11-0

P 0518.0100 \_\_\_\_\_ 100 g \_\_\_\_\_

P 0519

## POTASSIUM NITRATE

KNO<sub>3</sub> = 101.1

Assay \_\_\_\_\_ : &gt; 99% \_\_\_\_\_

Crystalline

- store at room temperature
- soluble in water (20°C / 320 g/l)
- hygroscopic
- R: 8
- S: 16-41
- UN 1486
- CAS 7757-79-1

P 0519.1000 \_\_\_\_\_ 1 kg \_\_\_\_\_

P 0519.5000 \_\_\_\_\_ 5 kg \_\_\_\_\_

P 0519.9025 \_\_\_\_\_ 25 kg \_\_\_\_\_

P 0535

## POTASSIUM SULPHATE

K<sub>2</sub>SO<sub>4</sub> = 174.3

Assay \_\_\_\_\_ : &gt; 99% \_\_\_\_\_

- store at room temperature
- soluble in water (20°C / 110 g/l)
- CAS 7778-80-5

P 0535.1000 \_\_\_\_\_ 1 kg \_\_\_\_\_

P 0535.5000 \_\_\_\_\_ 5 kg \_\_\_\_\_

P 0717

## L-PROLINE

C<sub>5</sub>H<sub>9</sub>NO<sub>2</sub> = 115.1

Assay \_\_\_\_\_ : &gt; 99% \_\_\_\_\_

- store at room temperature
- soluble in water (25°C / 1623 g/l)
- CAS 147-85-3

P 0717.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

P 0717.0100 \_\_\_\_\_ 100 g \_\_\_\_\_

P 0717.0500 \_\_\_\_\_ 500 g \_\_\_\_\_

P 1391

## PROPYLENEGLYCOL

C<sub>3</sub>H<sub>8</sub>O<sub>2</sub> = 76.1

1 l = 1.04 kg

- store at room temperature
- soluble in water
- CAS 57-55-6

P 1391.1000 \_\_\_\_\_ 1 l \_\_\_\_\_

P 0927

PUTRESCINE  
DIHYDROCHLORIDE

1,4-Diaminobutane dihydrochloride

C<sub>4</sub>H<sub>12</sub>N<sub>2</sub>·2HCl = 161.1

Polyamine growth regulator

Polyamine growth regulator affecting the synthesis of macromolecules, the activity of macromolecules, membrane permeability and partial processes of mitosis and meiosis.

Assay \_\_\_\_\_ : &gt; 98% \_\_\_\_\_

- store at room temperature
- soluble in water
- R: 36/37/38
- S: 26-37/39
- CAS 333-93-7

P 0927.0001 \_\_\_\_\_ 1 g \_\_\_\_\_

P 0927.0005 \_\_\_\_\_ 5 g \_\_\_\_\_

P 0927.0025 \_\_\_\_\_ 25 g \_\_\_\_\_

P 0612

## PYRIDOXINE HYDROCHLORIDE



Vitamin B6

 $C_8H_{11}NO_3 \cdot HCl = 205.6$ 

Assay \_\_\_\_\_ : &gt; 99.0%

White crystalline powder

- store at room temperature
- soluble in water (25°C / 200 g/l)
- R: 36/37/38
- S: 26-36
- CAS 58-56-0

P 0612.0050 \_\_\_\_\_ 50 g

P 0612.0100 \_\_\_\_\_ 100 g

P 0612.0250 \_\_\_\_\_ 250 g

R 0182

## RIBAVIRIN

 $C_8H_{12}N_4O_5 = 244.2$ 

Ribavirin is a synthetic nucleoside analogue structurally related to guanine. Ribavirin inhibits the replication of a wide range of RNA and DNA viruses. The antiviral mechanism of action of Ribavirin is not fully defined, but relates to alteration of cellular nucleotide pools and inhibition of viral mRNA synthesis.

Intracellular phosphorylation of ribavirin into phosphate derivatives is mediated by host cell enzymes. Ribavirin monophosphates competitively inhibit cellular inosine-5'-phosphate dehydrogenase and interfere with the synthesis of guanosine triphosphate (GTP) and thus nucleic acid synthesis in general. Ribavirin triphosphate also competitively inhibits the GTP dependent 5'-capping of viral mRNA.

Assay \_\_\_\_\_ : &gt; 98%

- store at room temperature
- soluble in water
- R: 61
- S: 22-45-53
- CAS 36791-04-5

R 0182.0250 \_\_\_\_\_ 250 mg

R 0182.1000 \_\_\_\_\_ 1 g

R 0812

## RAFFINOSE PENTAHYDRATE

 $C_{18}H_{32}O_{16} \cdot 5H_2O = 594.5$ 

Assay \_\_\_\_\_ : 98%

- soluble in water
- store dry at room temperature
- CAS 17629-30-0

R 0812.0025 \_\_\_\_\_ 25 g

R 0812.0100 \_\_\_\_\_ 100 g

R 0613

## RIBOFLAVINE

 $C_{17}H_{20}N_4O_6 = 376.4$ 

Assay \_\_\_\_\_ : &gt; 97.0%

- store at room temperature
- soluble in alkaline solutions with decomposition
- CAS 83-88-5

R 0613.0025 \_\_\_\_\_ 25 g

R 0613.0100 \_\_\_\_\_ 100 g

R 0806

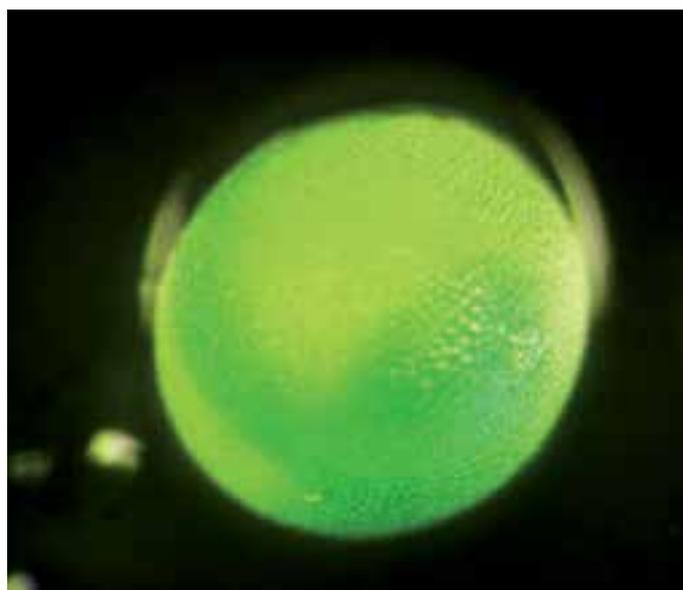
## D-RIBOSE

 $C_5H_{10}O_5 = 150.1$ 

- store at 2-8°C
- soluble in water
- CAS 50-69-1

R 0806.0025 \_\_\_\_\_ 25 g

Brassica embryo, Brenda de Lange



R 0146

## RIFAMPICIN



Rifampicin is active against gram-positive bacteria but less active against gram-negative bacteria. It interferes with the synthesis of nucleic acids by inhibiting DNA dependent RNA polymerase. Resistance to rifampicin can develop rapidly. The degree of resistance varies depending on the site of mutation in the RNA polymerase.

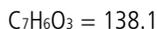
Assay \_\_\_\_\_ : > 97%

- store dry at 2-8°C
- soluble in dilute acid solution
- R: 22
- S: 36
- CAS 13292-46-1

R 0146.0001	1 g
R 0146.0005	5 g
R 0146.0025	25 g

S 1367

## SALICYLIC ACID



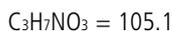
Assay \_\_\_\_\_ : > 99%

- store at room temperature
- slightly soluble in water (20°C / 1.8 g/l)
- R: 22-36/37/38-41
- S: 24-26-39
- CAS 69-72-7

S 1367.0100	100 g
S 1367.0500	500 g

S 0718

## L-SERINE



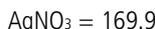
Assay \_\_\_\_\_ : > 900 ug/mg

- store at room temperature
- soluble in water (20°C / 250 g/l)
- CAS 56-45-1

S 0718.0025	25 g
S 0718.0100	100 g

S 0536

## SILVER NITRATE



Used with Sodium thiosulphate to produce a Silver thiosulphate solution (STS) containing the ethylene inhibitor  $[Ag(S_2O_3)_2]^{3-}$

Prepare a 0.1 M Sodium thiosulphate stock solution by dissolving 1.58 g of Sodium thiosulphate into 100 ml of water. Prepare a 0.1 M Silver nitrate stock solution by dissolving 1.7 g of Silver nitrate into 100 ml of water. Store the stock solutions in the dark until needed to prepare the Silver thiosulphate solution (STS).

In general the (STS) is prepared with a molar ratio between silver and thiosulphate of 1:4. Nearly all of the silver present in the solution is in the form of  $[Ag(S_2O_3)_2]^{3-}$ , the active complex for ethylene effect inhibition. Prepare a 0.02 M Silver thiosulphate solution (STS) by slowly pouring 20 ml of 0.1 M Silver nitrate stock solution into 80 ml of 0.1 M sodium thiosulphate stock solution. The Silver thiosulphate solution (STS) can be stored in the refrigerator for up to one month. However, preparation of the Silver thiosulphate solution (STS) just prior to use is recommended.

Assay \_\_\_\_\_ : > 99.8%

- store at room temperature
- soluble in water (20°C / 2150 g/l)
- protect from light
- R: 34-50/53 S: 26-36/37/39-45-60-61
- UN1493
- CAS 7761-88-8

S 0536.0005	5 g
S 0536.0025	25 g
	4 x 25 g

Iribov B.V., The Netherlands



S 1320

## SODIUM ALGINATE

Alginic acid sodiumsalt

Viscosity 1%, 100-200 mPa.s

A mixture of polyuronic acids composed of residues of D-mannuronic acid and L-guluronic acids extracted from algae belonging to the order *Phaeophyceae*. Alginates are used as suspending and thickening agents and in the preparation of water-miscible gels.

- store at room temperature
- soluble in water
- S: 22-24/25
- CAS 9005-38-3

S 1320.0250	250 g
S 1320.1000	1 kg

S 0520

SODIUM CHLORIDE 

NaCl = 58.4

Assay	: > 99%
Bromide (Br)	: < 0.005%
Sulphate (SO <sub>4</sub> )	: < 0.02%
Phosphate(PO <sub>4</sub> )	: < 0.0025%
Heavy metals	: < 0.0005%

- store at room temperature
- soluble in water (20°C / 310 g/l)
- R: 36/37/38 S: 22-24/25
- CAS 7647-14-5

S 0520.1000	1 kg
S 0520.5000	5 kg

S 0521

## TRI-SODIUM CITRATE DIHYDRATE

C<sub>6</sub>H<sub>5</sub>Na<sub>3</sub>O<sub>7</sub>·2H<sub>2</sub>O = 294.1

Assay	: > 99%
Crystalline	

- store at room temperature
- soluble in water (25°C / 720 g/l)
- CAS 6132-04-3

S 0521.1000	1 kg
S 0521.5000	5 kg

S 0522

## SODIUM DIHYDROGEN PHOSPHATE DIHYDRATE

NaH<sub>2</sub>PO<sub>4</sub>·2H<sub>2</sub>O = 156.0

Assay : &gt; 98%

- store at room temperature
- soluble in water (20°C / 850 g/l)
- CAS 13472-35-0

S 0522.1000	1 kg
S 0522.5000	5 kg

S 1377

1-SODIUM DODECYL SULPHATE SDS, Sodium Lauryl Sulphate  
C<sub>12</sub>H<sub>25</sub>O<sub>4</sub>SNa = 288.4

A twice recrystallized quality of SDS with excellent qualities for denaturing proteins before gel electrophoresis, molecular weight sieving and many other applications.  
Harewood K. and Wolff J.S., Anal. Biochem., 55, 573 (19730)

Assay : &gt; 99%

- store at room temperature
- soluble in water (20 °C/ >100 g/l)
- R: 20/22-36/37/38-41-42
- S: 22-26-36
- CAS 151-21-3

S 1377.0100	100 g
S 1377.0250	250 g
S 1377.0500	500 g
S 1377.1000	1 kg

S 0537

## DI-SODIUM HYDROGEN PHOSPHATE DIHYDRATE

Na<sub>2</sub>HPO<sub>4</sub>·2H<sub>2</sub>O = 178.0

Assay : &gt; 98%

- store dry at room temperature
- soluble in water (20°C / 779 g/l)
- CAS 10028-24-7

S 0537.1000	1 kg
S 0537.5000	5 kg

S 0523

## SODIUM HYDROXIDE



NaOH = 40.0

Caution, causes severe burns

Assay \_\_\_\_\_ : &gt; 98%

- store dry at room temperature
- soluble in water (20°C / 1090 g/l)
- R: 35
- S: 26-37/39-45
- CAS 1310-73-2
- UN 1823

S 0523.0500 \_\_\_\_\_ 500 g

S 0523.1000 \_\_\_\_\_ 1 kg

S 0525

## SODIUM MOLYBDATE DIHYDRATE

Na<sub>2</sub>MoO<sub>4</sub>·2H<sub>2</sub>O = 241.9

Assay \_\_\_\_\_ : &gt; 98.0%

Crystalline

- store at room temperature
- soluble in water (840 g/l / 20°C)
- S: 22-24/25
- CAS 10102-40-6

S 0525.0025 \_\_\_\_\_ 25 g

S 0525.0100 \_\_\_\_\_ 100 g

S 0524

## SODIUM NITRATE

NaNO<sub>3</sub> = 85.0

Assay \_\_\_\_\_ : &gt; 99%

- store dry at room temperature
- soluble in water (880 g/l / 20°C)
- R: 8-22-36
- S: 16-22-24-41
- CAS 7631-99-4
- UN 1498

S 0524.1000 \_\_\_\_\_ 1 kg

S 0538

## SODIUM THIOSULPHATE

Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> = 158.1Used with Silver nitrate to produce a Silver thiosulphate solution (STS) containing the ethylene inhibitor [Ag(S<sub>2</sub>O<sub>3</sub>)<sub>2</sub>]<sup>3-</sup> (see cat. no. S 0536).

Assay \_\_\_\_\_ : &gt; 98%

- soluble in water (20°C / 20 g/l)
- S: 22-24/25
- CAS 7772-98-7

S 0538.0250 \_\_\_\_\_ 250 g

S 0538.1000 \_\_\_\_\_ 1 kg

S 0807

## D-SORBITOL

C<sub>6</sub>H<sub>14</sub>O<sub>6</sub> = 182.2

Assay \_\_\_\_\_ : &gt; 97.0%

Water \_\_\_\_\_ : &lt; 1.0%

- store dry at room temperature
- soluble in water
- CAS 50-70-4

S 0807.1000 \_\_\_\_\_ 1 kg

S 0807.5000 \_\_\_\_\_ 5 kg

S 1330

## SOYA PEPTONE

From papaiic hydrolysis of soybean meal.

Typical analysis (% w/w):

total nitrogen (TN) \_\_\_\_\_ : approx. 9.0-10.5%

amino nitrogen (AN) \_\_\_\_\_ : approx. 2.5-3.5%

Sodium chloride \_\_\_\_\_ : approx. &lt; 1.0%

- store dry at room temperature
- soluble in water
- CAS 73049-73-7

S 1330.0100 \_\_\_\_\_ 100 g

S 1330.0500 \_\_\_\_\_ 500 g

S 0188

## SPECTINOMYCIN DIHYDROCHLORIDE PENTAHYDRATE


 $C_{14}H_{24}N_2O_7 \cdot 2HCl \cdot 5H_2O = 495.3$ 

Spectinomycin is an aminocyclitol antibiotic that acts by binding to the 30S subunit of the bacterial ribosome and inhibiting protein synthesis. Its activity is generally modest, particularly against gram-positive bacteria. Some gram-negative bacteria are sensitive. Resistance in vitro may develop by chromosomal mutation or may be plasmid located.

Assay \_\_\_\_\_ : > 95%

- store dry at 2-8°C
- soluble in water
- R: 36/37 S: 22-25-26
- CAS 22189-32-8

S 0188.0005 \_\_\_\_\_ 5 g  
S 0188.0025 \_\_\_\_\_ 25 g

S 1369

## SPERMIDINE


 $NH_2(CH_2)_7NH_2 = 145.2$ 

Assay \_\_\_\_\_ : >98%

- store at 2-8°C
- soluble in water
- R: 34 S: 26-36/37/39-45
- CAS 124-20-9
- UN 1760

S 1369.0001 \_\_\_\_\_ 1 g  
S 1369.0005 \_\_\_\_\_ 5 g  
S 1369.0025 \_\_\_\_\_ 25 g

*Echeveria*,  
Succulent Tissue Culture, The Netherlands



S 1511

## SSC-BUFFER

A homogeneous mixture of molecular grade Sodium chloride and Trisodium citrate to prepare SSC-buffer. Suitable for use in nucleic acid hybridisation.

NaCl	0.15 M	8.77 g/l
Trisodium citrate	0.015 M	4.41 g/l
		13.18 g/l

pH (water, 20°C): 8.3 ± 0.2

Sambrook, J., et al., Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory (1989), p.B.13.

- after dissolving 13.18 gram in 1 litre of water, a 1 x SSC solution is prepared with a concentration of 0.15 M NaCl and 0.015 M Trisodium citrate.
- after dissolving 263.56 gram in 1 litre of water, a 20 x SSC solution is prepared with a concentration of 3.0 M NaCl and 0.3 M Trisodium citrate.
- to avoid precipitation no higher concentrations of 20x SSC stock solutions are recommended.

20 ltr pack, to prepare 20 l (1X) solution or 1 l (20X) solution

S 1511.0020 \_\_\_\_\_ 263.56 g

200 ltr pack, to prepare 200 l (1X) solution or 10 l (20X) solution

S 1511.0200 \_\_\_\_\_ 2635.6 g

S 1512

## SSPE-BUFFER

A homogeneous mixture of molecular grade Sodium chloride, Sodium phosphate and EDTA disodium to prepare SSPE-buffer. Suitable for use in nucleic acid hybridisation.

NaCl	0.15 M	8.77 g/l
NaH <sub>2</sub> PO <sub>4</sub> ·H <sub>2</sub> O	0.01 M	1.38 g/l
EDTA-Na <sub>2</sub> ·2H <sub>2</sub> O	0.001 M	0.37 g/l
		10.52 g/l

pH (water, 20°C): 8.3 ± 0.2

Sambrook, J., et al., Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory (1989), p.B.13.

- after dissolving 10.52 gram in 1 litre of water, a 1 x SSC solution is prepared with a concentration of 0.15 M NaCl, 0.01 M Sodium phosphate and 0.01 M Na<sub>2</sub>EDTA.
- after dissolving 210.4 gram in 1 litre of water, a 20 x SSC solution is prepared with a concentration of 3.0 M NaCl and 0.2 M Sodium phosphate and 0.02 mol Na<sub>2</sub>EDTA.
- to avoid precipitation no higher concentrations of 20x SSC stock solutions are recommended.

20 ltr pack, to prepare 20 l (1X) solution or 1 l (20X) solution

S 1512.0020 \_\_\_\_\_ 210.4 g

200 ltr pack, to prepare 200 l (1X) solution or 10 l (20X) solution

S 1512.0200 \_\_\_\_\_ 2103.6 g

S 1357

## STARCH FROM POTATOES

Moisture : 20%  
pH : 5.0 - 8.0

- store dry at room temperature
- CAS 9005-84-9

S 1357.1000 1 kg

S 1324

## STARCH FROM RICE

- store dry at room temperature
- CAS 9005-84-9

S 1324.1000 1 kg

S 0162

## STERILLIUM

- store at room temperature
- miscible in water
- R: 10 S: 16
- UN 1987

S 0162.1000 1 l

S 0148

STREPTOMYCIN SULPHATE 

$(C_{21}H_{39}N_7O_{12})_2 \cdot 3H_2SO_4 = 1457.4$

Streptomycin is an aminoglycoside antibiotic and has a bactericidal action against many gram-negative bacteria. Aminoglycosides are transported into sensitive bacterial cells by an active transport process. Within the cell, it binds to the 30S subunit (S12 protein), inhibiting protein synthesis and generating errors in the transcription of the genetic code.

Assay : > 720 IU/mg

- store dry at 2-8°C
- soluble in water
- R: 22-61 S: 36/37/39-45-53
- CAS 3810-74-0

S 0148.0050 50 g

S 0148.0100 100 g

S 0809

## SUCROSE

$C_{12}H_{22}O_{11} = 342.3$

Assay : > 99.7%

White to off-white crystalline powder

- store dry at room temperature
- soluble in water
- CAS 57-50-1

S 0809.1000 1 kg

S 0809.5000 5 kg

S 0809.9025 25 kg

2 x 25 kg

4 x 25 kg

S 0149

SULPHAMETHOXAZOLE 

$C_{10}H_{11}N_3O_3S = 253.3$

Sulphamethoxazole is a bacteriostatic antibiotic. It has a similar structure as p-aminobenzoic acid and interferes with the synthesis of nucleic acids in sensitive micro-organisms by blocking the conversion of p-aminobenzoic acid to the coenzyme dihydrofolic acid, a reduced form of folic acid.

- store at room temperature
- soluble in ethanol
- R: 36/37/38-43
- S: 26-36
- CAS 723-46-6

S 0149.0025 25 g

S 0149.0100 100 g

T 1359

## TALC

Hydrated Magnesium Silicate, approximately  $3MgO \cdot 4SiO_2 \cdot H_2O$

Assay : 17.0 – 19.5% Mg

- store at room temperature
- CAS 14807-96-6

T 1359.1000 1 kg

T 1359.5000 5 kg

T 1360

## TAURINE

2-aminoethanesulfonic acid  
 $C_2H_7NO_3S = 125.1$

Assay \_\_\_\_\_ : > 98%  
 Crystalline

- store at room temperature
- soluble in water (12°C / 65 g/l)
- CAS 107-35-7

T 1360.0100 \_\_\_\_\_ 100 g

T 1507

## TBE-BUFFER

Dry homogeneous powdered TBE-buffer.

A homogeneous mixture of molecular grade Tris base, boric acid and  $Na_2EDTA \cdot 2H_2O$  for use in gel electrophoresis.

Tris base	0.089 M	10.78 g/l
Boric acid	0.089 M	5.50 g/l
$Na_2EDTA \cdot 2H_2O$	0.002 M	0.74 g/l
		17.02 g/l

pH (water, 20°C):  $8.3 \pm 0.1$

Sambrook, J., et al., Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory (1989), p.6.7, B.23.

- after dissolving 17.02 gram in 1 litre of water, a 1x TBE solution is prepared with a concentration of 0.089 M Tris Base, 0.089 M Borate and 0.002 M  $Na_2EDTA$ .
- after dissolving 170.2 gram in 1 litre of water, a 10x TBE solution is prepared with a concentration of 0.89 M Tris Base, 0.89 Borate and 0.02 M  $Na_2EDTA$ .
- to avoid precipitation no higher concentrations of TBE stock solutions are recommended.

10 l pack,  
 To prepare 10 l (1X) solution or 1 l (10X) solution  
 T 1507.0010 \_\_\_\_\_ 170.2 g

100 l pack,  
 To prepare 100 l (1X) solution or 10 l (10X) solution  
 T 1507.0100 \_\_\_\_\_ 1702.0 g

1000 l pack,  
 To prepare 1000 l (1X) solution or 100 l (10X) solution  
 T 1507.1000 \_\_\_\_\_ 17020.0 g

T 1508

## TE-BUFFER

Dry homogeneous powdered TE-Buffer.

A homogeneous mixture of molecular grade Tris base and  $Na_2EDTA \cdot 2H_2O$  to prepare TE buffer.

Tris base	10.0 mM	1.21 g/l
$Na_2EDTA \cdot 2H_2O$	1.0 mM	0.37 g/l
		1.58 g/l

pH (water, 20°C):  $8.0 \pm 0.1$

- after dissolving 1.58 gram in 1 litre of water, a 1x TE solution is prepared with a concentration of 10.0 mM Tris Base and 1.0 mM  $Na_2EDTA$ .
- after dissolving 15.84 gram in 1 litre of water, a 10x TE solution is prepared with a concentration of 100 mM Tris Base and 10 mM  $Na_2EDTA$ .
- to avoid precipitation no higher concentrations of TE stock solutions are recommended.

100 l pack,  
 To prepare 100 l (1X) solution or 10 l (10X) solution  
 T 1508.0100 \_\_\_\_\_ 158.35 g

1000 l pack,  
 To prepare 1000 l (1X) solution or 100 l (10X) solution  
 T 1508.1000 \_\_\_\_\_ 1583.5 g

T 0150

TETRACYCLINE  
HYDROCHLORIDE

$C_{22}H_{24}N_2O_8 \cdot HCl = 480.9$

Tetracycline is a bacteriostatic antibiotic with activity against gram-positive and gram-negative bacteria. Within the cell tetracycline binds reversibly to the 30S subunit of the ribosome, preventing the binding of aminoacyl transfer RNA and inhibiting protein synthesis and hence cell growth. Used as a selective marker for the transformation of plasmids encoding for tetracycline resistance (Tetr) such as pBR322, pBR325 and pMB9.

- store dry at room temperature
- slightly soluble in water, soluble in ethanol
- protect from light
- R: 36/37/38-63-64
- S: 22-36/37-39
- CAS 64-75-5

T 0150.0025 \_\_\_\_\_ 25 g  
 T 0150.0100 \_\_\_\_\_ 100 g

T 0614

## THIAMINE HYDROCHLORIDE

 $C_{12}H_{17}ClN_4OS$  = 337.3

Assay \_\_\_\_\_ : &gt; 98.5%

- store at room temperature
- soluble in water
- CAS 67-03-8

T 0614.0025	25 g
T 0614.0100	100 g
T 0614.0250	250 g
T 0614.1000	1 kg

T 0916

THIDIAZURON  $C_9H_8N_4OS$  = 220.2

Cytokinin like growth regulator

- powder storage at room temperature
- soluble in DMSO
- liquid storage at 2-8°C
- sterilization by filtration
- concentration: 0.001-0.05 mg/l
- R: 36/37/38 S: 22-26-36
- CAS 51707-55-2

T 0916.0250	250 mg
T 0916.0500	500 mg
T 0916.1000	1 g

T 0151

THIMEROSAL  $C_9H_9HgNaO_2S$  = 404.8

Thimerosal is a bacteriostatic and fungistatic mercurial agent.

- store at room temperature
- soluble in water
- R: 33-26/27/28-50/53 S: 2-13-36-45-28-60/61
- CAS 54-64-8
- UN 2025

T 0151.0010	10 g
T 0151.0025	25 g

Infiltration of *Agrobacterium* into tobacco leaves.  
Agroinfiltration is used for rapid functional gene analysis in plants.  
Dr. Jan Schaart, Wageningen UR Plant Breeding

T 0719

## L-THREONINE

 $C_4H_9NO_3$  = 119.1

Assay \_\_\_\_\_ : &gt; 99%

- store at room temperature
- soluble in water (20°C / 90 g/l)
- CAS 72-19-5

T 0719.0025	25 g
T 0719.0100	100 g
T 0719.0500	500 g

T 0180

TICARCILLIN DISODIUM  $C_{15}H_{14}N_2Na_2O_6S_2$  = 428.4

Ticarcillin is an inhibitor of bacterial cell wall synthesis. It inhibits the cross-linking of peptidoglycan by binding and inactivating of transpeptidases. High activity against gram-negative bacteria such as *Agrobacterium* strains.  $\beta$ -lactamase sensitive.

Assay \_\_\_\_\_ : &gt; 95%

- store dry at 2-8°C
- soluble in water
- R: 42/43 S:22-24/25-36
- CAS 4697-14-7

T 0180.0001	1 g
T 0180.0010	10 g



T 0190

**TICARCILLIN DISODIUM/  
CLAVULANATE POTASSIUM****Ticarcillin and clavulanic acid mixed in a ratio of 15:1**

Ticarcillin is an inhibitor of bacterial cell wall synthesis. It inhibits the cross-linking of peptidoglycan by binding and inactivating of transpeptidases. High activity against gram-negative bacteria such as *Agrobacterium* strains.  $\beta$ -lactamase sensitive. Clavulanic acid is a specific inhibitor of  $\beta$ -lactamase and protects ticarcillin against inactivation by  $\beta$ -lactamase. A very effective combination against resistant *Agrobacterium* species.

- store dry at 2-8°C
- soluble in water
- hygroscopic
- R: 42/43 S: 22-24/25-36
- CAS (Ticarcillin disodium): 4697-14-7
- CAS (Clavulanate potassium): 61177-45-5

T 0190.0002	2 g
T 0190.0010	10 g
T 0190.0025	25 g

T 0153

**TOBRAMYCIN SULPHATE**

Tobramycin is an aminoglycoside antibiotic and has a bactericidal action against many gram-negative bacteria.

Aminoglycosides are taken into sensitive bacterial cells by an active transport process. Within the cell, they bind to the 30S and to some extent to the 50S subunits of the bacterial ribosome, inhibiting protein synthesis and generating errors in the transcription of the genetic code.

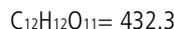
Assay \_\_\_\_\_ : > 634  $\mu\text{g}/\text{mg}$

- store dry at 2-8°C
- soluble in water (20°C / 50 g/l)
- R: 23/24/25-61 S: 22-36/37/39-45-53
- CAS 49842-07-1

T 0153.0001	1 g
T 0153.0005	5 g



T 1395

**TREHALOSE ANHYDROUS**

Assay \_\_\_\_\_ : > 99%

- store at room temperature
- soluble in water
- CAS: 99-20-7

T 1395.0002	25 g
T 1395.0010	100 g
T 1395.0025	500 g

T 0941

**Meta-TOPOLIN**

Cytokinin growth regulator

Meta-topolin [6-(3-hydroxybenzylamino)purine] is an aromatic cytokinin. It was first isolated from poplar leaves. Its name is derived from "topol", the Czech word for poplar. The metabolism of meta-topolin is similar to that of other cytokinins. Just as zeatin and BAP, meta-topolin may undergo ribosylation at position 9 without a significant effect on the activity. In *Spathiphyllum floribundum*, shoot production in media with BAP and meta-topolin is very similar. However, after transfer to the soil, the shoots produced with meta-topolin root much better during acclimatization.

S.P.O. Werbrouck, M. Strnad, H.A Van Onckelen and P.C. Debergh, Meta-topolin, an alternative to benzyladenine in tissue culture?. *Physiol. Plant.* 98: 291-297 (1996).

J. Holub, J. Hanus, D.E. Hanke and M. Strnad, Biological activity of cytokinins derived from ortho- and meta-hydroxybenzyladenine. *Plant Growth Reg.* 26: 109-115 (1998)

Assay (HPLC) \_\_\_\_\_ : > 99%

- Store dry at 2-8°C
- CAS 75737-38-1

T 0941.0100	100 mg
T 0941.0500	500 mg
T 0941.1000	1 g
T 0941.5000	5 g

T 0915

**2,4,5-TRICHLOROPHENOXYACETATE  
ACID POTASSIUM SALT** $C_8H_4Cl_3O_3K = 263.6$ 

- soluble in water
- powder storage at room temperature
- liquid storage at 2-8°C
- sterilization: autoclavable
- concentration: 0.01-5.0 mg/l
- R: 20/21/22 S: 20/21-36/37/39
- CAS 37785-57-2
- UN 3077

T 0915.0025 25 g

T 0929

**2,3,5-TRIIODOBENZOIC ACID**

TIBA,

 $C_7H_3I_3O_2 = 499.8$ 

Noncompetitive inhibitor of polar auxin transport

Assay : &gt; 95%

- protect from moisture and light
- store between -25°C and -15°C
- soluble in 1N NaOH
- R: 22 S: 24/25-36
- CAS 88-82-4

T 0929.0005 5 g

T 0929.0010 10 g

T 1361

**TRIETHANOLAMINE**

2,2',2''-Nitrilotriethanol, Free Base

 $C_6H_{15}NO_3 = 149.2$ 

1 liter = 1.12 kg (25°C)

Assay	: > 98%
pKa (at 25°C)	: 7.8
pH range	: 7.3-8.3
Water	: < 0.5%

- store dry at room temperature
- soluble in water
- R: 36/37/38 S: 26-36
- CAS 102-71-6

T 1361.0500 500 ml

T 1361.1000 1 l

T 0154

**TRIMETHOPRIM** $C_{14}H_{18}N_4O_3 = 290.3$ 

Trimethoprim is active against gram-negative and gram-positive aerobic bacteria. The antibiotic is a dihydrofolate reductase inhibitor. It inhibits the conversion of dihydrofolic acid to tetrahydrofolic acid, which is necessary for the synthesis of amino acids, purines, thymidines and ultimately DNA synthesis. Resistance may develop very fast.

Assay : &gt; 98.5%

- store dry at room temperature
- soluble in propyleneglycol
- R: 20/21/22
- S: 22-36/37/39
- CAS 738-70-5, UN 2811

T 0154.0005 5 g

T 0154.0025 25 g

T 0928

**TRIFLURALIN** $C_{13}H_{16}F_3N_3O_4 = 335.3$ 

Disrupts Mitosis by inhibiting the formation of microtubules

- store at room temperature
- soluble in acetone
- R: 36-43-50/53
- S: 24-37-60-61
- CAS 1582-09-8
- UN 3077

T 0928.0250 250 mg

T 0181

**TRIMETHOPRIM LACTATE** $C_{14}H_{18}N_4O_3 \cdot C_3H_6O_3 = 380.4$ 

- store dry at room temperature
- soluble in water
- R: 20/21/22
- S: 22-36/37/39
- CAS 23256-42-0, UN 2811

T 0181.0250 250mg

T 0181.1000 1 g

T 1501

## TRIS, ULTRAPURE



Tris(hydroxymethyl)aminomethane  
2-Amino-2-hydroxy-methyl-1,3,propanediol  
 $C_4H_{11}NO_3 = 121.1$

A highly purified quality of Tris with excellent properties for molecular biology and biological buffers

Purity, dried substance	: > 99.9%
pH (1M in water)	: 10.5-11.5

- store at room temperature
- soluble in water (25°C / >700 g/l)
- R: 36/37/38
- S: 26-36
- CAS 77-86-1

T 1501.1000	1 kg
T 1501.5000	5 kg
T 1501.9010	10 kg
T 1501.9025	25 kg
T 1501.9025	2x 25 kg

T 1513

## TRIS HYDROCHLORIDE



Tris HCl, Tris(hydroxymethyl)aminomethane-Hydrochloride  
 $C_4H_{11}NO_3.HCl = 157.6$

A highly purified quality of Tris HCl with excellent properties for molecular biology.

Purity, dried substance	: > 99%
pKa (20°C)	: 8.0 - 8.4
pH (0.5 M in water, 25°C)	: 3.5 - 5.0
Useful pH range	: 7 - 9

- store at room temperature
- soluble in water (20°C / >100 g/l)
- R: 36/37/38
- S: 26-36
- CAS 1185-53-1

T 1513.0100	100 g
T 1513.0250	250 g
T 1513.0500	500 g
T 1513.1000	1 kg

T 1332

## TRYPTONE

Pancreatic digest of casein

total nitrogen (TN)	: 12.5 – 13.5%
amino nitrogen (AN)	: 3.0 – 4.0%
pH (5% solution)	: 6.5 – 7.5

- store dry at room temperature
- hygroscopic
- soluble in water

T 1332.0100	100 g
T 1332.0500	500 g
T 1332.1000	1 kg

T 0720

## L-TRYPTOPHAN

$C_{11}H_{12}N_2O_2 = 204.4$

Assay	: > 98.5%
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- store at room temperature
- soluble in water (20°C / 10 g/l)
- CAS 73-22-3

T 0720.0025	25 g
T 0720.0100	100 g



T 0721

## L-TYROSINE

 $C_9H_{11}NO_3 = 181.2$ 

Assay \_\_\_\_\_ : &gt; 99%

- store at room temperature
- soluble in water (20°C / 0.4 g/l)
- CAS 60-18-4

T 0721.0100 \_\_\_\_\_ 100 g

T 0721.0500 \_\_\_\_\_ 500 g

T 0721.1000 \_\_\_\_\_ 1 kg

U 1363

## UREA

 $CH_4N_2O = 60.1$ 

Assay \_\_\_\_\_ : &gt; 99%

- store at room temperature
- soluble in water (1080 g/l / 20°C)
- S: 22-24/25
- CAS 57-13-6

U 1363.1000 \_\_\_\_\_ 1 kg

U 1363.5000 \_\_\_\_\_ 5 kg

V 0170

## VALIDAMYCIN A

 $C_{20}H_{35}NO_{13} = 497.5$ 

Inhibition of Trehalase activity. Enhances trehalose accumulation in transgenic plants.

Oscar J.M. Goddijn et al., Plant Physiol (1997) 113: 181-190.

- store at 2-8°C
- soluble in DMSO and ethanol
- S: 36/37
- CAS 37248-47-8

V 0170.0001 \_\_\_\_\_ 1 g

V 0722

## L-VALINE

 $C_5H_{11}NO_2 = 117.1$ 

Assay \_\_\_\_\_ : &gt; 98.5%

- store at room temperature
- soluble in water (20°C / 85 g/l)
- CAS 72-18-4

V 0722.0100 \_\_\_\_\_ 100 g

V 0722.0500 \_\_\_\_\_ 500 g

V 0155

VANCOMYCIN  
HYDROCHLORIDE

$C_{66}H_{75}Cl_2N_9O_{24}.HCl = 1485.7$   
plant cell culture tested

Vancomycin is a glycopeptide antibiotic. It inhibits the formation of the peptidoglycan polymers of the bacterial cell wall. Unlike penicillins that act primarily to prevent the crosslinking of peptidoglycans that give the cell its strength, vancomycin prevents the transfer and addition of the muramylpentapeptide building blocks that form the peptidoglycan molecule itself. Vancomycin is often used in combination with cefotaxime or carbenicillin to obtain a synergism in antimicrobial activity against bacteria. Especially used for Agrobacterium species with a high  $\beta$ -lactamase production.

Potency \_\_\_\_\_ : &gt; 1050 IU/mg

- store dry at 2-8°C in airtight containers protected from light
- soluble in water (20°C / 200 g/l)
- R: 20/21/22-36/37-43 S: 36/37/39-45-47
- CAS 1404-93-9

V 0155.0001 \_\_\_\_\_ 1 g

V 0155.0005 \_\_\_\_\_ 5 g

V 0155.0025 \_\_\_\_\_ 25 g

X 0808

## D-XYLOSE

 $C_5H_{10}O_5 = 150.1$ 

Assay \_\_\_\_\_ : &gt; 99%

- store dry at room temperature
- soluble in water
- CAS 58-86-6

X 0808.0100 \_\_\_\_\_ 100 g

X 0808.0500 \_\_\_\_\_ 500 g

Y 1333

## YEAST EXTRACT

A dried yeast autolysate with a high content of amino nitrogen and water soluble B-complex vitamins. Due to its low NaCl content this quality is well suited for plant tissue culture.

Typical analysis (% w/w)

total nitrogen (TN)	: 10.0 - 11.8
amino nitrogen (AN)	: 4.8 - 6.3
sodium chloride	: < 0.5%
pH (8.3% solution)	: 6.8 - 7.2

- store dry at room temperature
- soluble in water
- CAS 8013-01-2

Y 1333.0100	100 g
Y 1333.0500	500 g
Y 1333.1000	1 kg

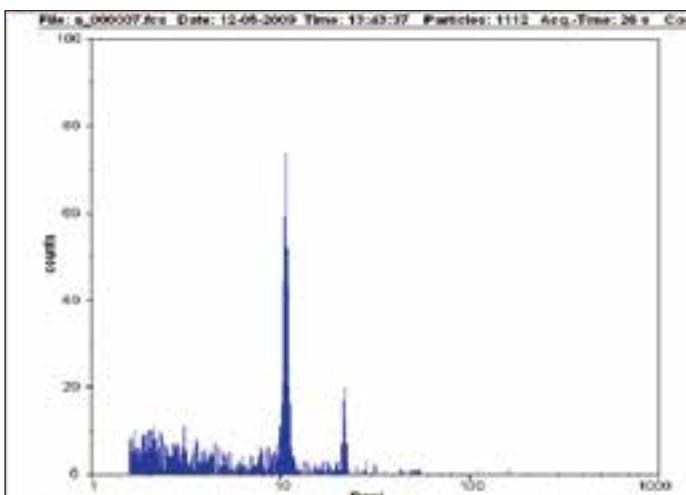
Y 1709

## YPD AGAR

Glucose.H <sub>2</sub> O	: 20 g/l
Peptone	: 20 g/l
Yeast extract	: 10 g/l
Microbiological tested Agar	: 10 g/l

- store dry at room temperature
- dissolve 60 g in 1 l distilled water and adjust the pH to 7.2.

Y 1709.0100	100 g
Y 1709.0500	500 g



Flow Cytometry: Ploidy analyses on isolated Brassica nuclei from leaf Analysis on logarithmic scale. The first peak is 2C level ( 2x nuclei in G0/G1 phase. The second peak is 4C level ( 2x nuclei in G2 phase, or generated by endoreduplication)

Y 1708

## YPD BROTH

Glucose.H <sub>2</sub> O	: 20 g/l
Peptone	: 20 g/l
Yeast extract	: 10 g/l

- store dry at room temperature
- dissolve 50 g in 1 l distilled water and adjust the pH to 7.2.

Y 1708.0100	100 g
Y 1708.0500	500 g

Z 0917

## ZEATIN, trans isomer

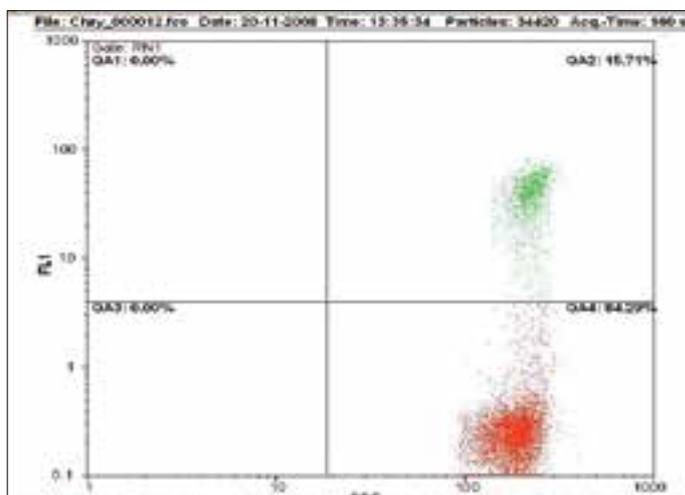
C<sub>10</sub>H<sub>13</sub>N<sub>5</sub>O = 219.2

Assay : > 98.0%

Off white to yellow crystals

- soluble in 1N NaOH
- store powder between -25°C and -15°C
- store liquid between -25°C and -15°C
- sterilisation : filtration
- concentration : 0.01-5.0 mg/l
- S: 22-24/25
- CAS 1637-39-4

Z 0917.0050	50 mg
Z 0917.0100	100 mg
Z 0917.0250	250 mg
Z 0917.0500	500 mg
Z 0917.1000	1 g



Flow Cytometry: Viability/Vitality analyses of pollen in Chrysanthemum Pollen is stained with FDA, green fluorescence (FL1) is quantified, and plotted against scatter signal. The green population are FDA stained pollen, the red population are non-viable/dead pollen.

Iribov B.V., The Netherlands

Z 0937

**ZEATIN RIBOSIDE, trans isomer**



Zeatin ribose was used for plant regeneration from tomato, Brassica nigra and Vigna sublobata protoplasts. Bhadra SK et al., PCR 14: 175-179 (1994). Hossain M et al. PCTOC 42: 141-146 (1995). Narasimhulu SM et al. PCTOC 32 (1): 35-39 (1993). Zeatin ribose has been efficiently used for direct and efficient regeneration from leaf explants of potato. From all cytokinins tested, Zeatin riboside produced the maximum number of shoots per explant. Yadav NR and Sticklen MB. PCR 14: 645-647 (1995). Somatic embryogenesis of tomato calli was induced on medium supplemented with Zeatin riboside. Chen LZ, Breeding Sci 44 (3): 257, (1994). Zeatin riboside was effectively used for direct initiation of shoot cultures from axils of bracts from Aloe, Gasteria, and Haworthia species. Richwine AM et al. HortScience 30 (7): 1443, (1995).

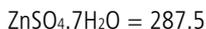
Assay (dried substance) : > 97%

- soluble in water
- powder storage between -25°C and -15°C
- liquid storage between -25°C and -15°C
- sterilization: filtration
- CAS 6025-53-2

Z 0937.0025	25 mg
Z 0937.0050	50 mg
Z 0937.0100	100 mg
Z 0937.0250	250 mg
Z 0937.0500	500 mg
Z 0937.1000	1 g

Z 0526

**ZINC SULPHATE  
HEPTAHYDRATE**



Assay : > 99%

- store at room temperature
- soluble in water (20°C / 960 g/l)
- R: 22-41-50/53 S: 22-26-39-46-60-61
- CAS 7446-20-0
- UN 3077

Z 0526.0500	500 g
Z 0526.1000	1 kg

Hardening of TC plants. Compartment with first fase after tissue culture. Humidity controlled with fog system.

Cosmo Plant, joint hardening facility of Iribov, Allplant and Maatschap Holtmaat.

Z 0186

**ZEOCIN™**



Zeocin™ is produced by Streptomyces verticillus and part of the structurally related group of bleomycin/phleomycin type antibiotics. The antibiotic is applied as a selective agent in transformation experiments with mammalian cells, plant cells and yeast. The cytotoxic action results from the ability to cause fragmentation of DNA. The antibiotic binds to DNA through its amino-terminal peptide, and the activated complex generates free radicals that are responsible for scission of the DNA chain. Zeocin™ is used as a selective agent for the incorporation of the Sh ble gene that encodes a 13,665 dalton protein. By binding to the antibiotic, the protein prevents binding of Zeocin™ to DNA. Zeocin™ is a trade mark of Cayla.

- store at 2-8°C
- soluble in water
- R: 22-40-42/43 S:24/25-36/37/39
- CAS 11006-33-0

Z 0186.0250	250 mg
Z 0186.1000	1 g



F 3001

### FORCEPS, 23 cm

This stainless steel forceps, with a length of 23 cm, has been especially developed for the handling of plantlets in Plant Tissue culture. By means of the long thin extended legs, the distance between the hand, the sterile plantlets and culture vessel has been lengthened significantly, hereby drastically reducing the risks of contamination. With its long thin legs it is easy maneuvering in long narrow culture tubes and due to the length, the bottom can be reached without contacting the sterile rim of the tubes. Its light weight and the required low pressure by hand to close the forceps give it a fine ergonomic performance without fatiguing the hand.

F 3001.0001 1 piece

F 3003

### FORCEPS EXTENDED, 30 cm

This extended forceps allows a maximum distance between the hand and the plantlets minimizing the risks of contamination to nil. The forceps is specially designed to work in combination with the Ergonomic scalpel handle, S3110. While handed the length of both tools is about equal providing a symmetric and ergonomically balanced work situation.

F 3003.0001 1 piece

S 3101

### SCALPEL HANDLE

The distance between the handle and the blade of this scalpel (18 cm) has been lengthened in this design to reduce the risk of contamination. Because of its low weight and ergonomic shape it is a handy tool for cutting plantlets.

S 3101.0001 1 piece

S 3110

### ERGONOMIC SCALPEL HANDLE

In cooperation with tissue culture laboratories Duchefa Biochemie B.V. has developed a new ergonomically shaped scalpel handle to facilitate a good and well balanced firm grip of the tool while cutting plantlets. The hexagonal shaped grip with a diameter of 10 mm positions the fingers in an ergonomically position allowing a firm hold without cramping fingers and wrist. To avoid weight the grip is made hollow and is in a good weight balance with the extended shaft. By extending the shaft the risk of contamination caused by manual contact is minimized and a safe distance to the plant material is guaranteed.

Dimensions: Overall length 24 cm, grip length 11 cm, shaft length 13 cm, weight 41 gram.

S 3110.0001 1 piece



S 3200

## SCALPEL BLADES NO. 10

S 3200.0001	1 Box contains 100 pieces (non-sterile).
1- 10 boxes	price per box
11- 25 boxes	price per box
26- 50 boxes	price per box
51-100 boxes	price per box

S 3201

## SCALPEL BLADES NO. 11

S 3201.0001	1 Box contains 100 pieces (non-sterile).
1- 10 boxes	price per box
11- 25 boxes	price per box
26- 50 boxes	price per box
51-100 boxes	price per box

R 3002

## REST

Stainless steel rest for holding sterile forceps and scalpel handles in a convenient position. Length : 20 cm Height : 3 cm

R 3002.0001	1 piece
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G 3302

## GLASS BEADS FOR STERILIZER

- diameter : 1.5-2 mm

G 3302.0500	500 gram
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P 3202

## PAPER CUTTING PAD, 12.5 x 19 cm

Paper cutting pads are used for sterile cutting of plantlets in laminar flows. A sterile sealed plastic package contains 30 gamma radiated paper cutting pads.

P 3202.0001	
10	packages of 30 cutting pads
100	packages of 30 cutting pads
1000	packages of 30 cutting pads



G 3301

## GLASS BEAD STERILIZER

Model "Lab Associates"

- weight: 3.5 kg
- outside dimensions: 15 x 12 x 15 cm
- tube dimensions (diameter. x height) : 5.5 x 12 cm
- operating temperature of 275° C
- thermostat controlled 200 W / 220 Volt or 110 Volt (upon request)
- including glass beads

G 3301.0001	Glass Bead Sterilizer
1	piece
2-4	pieces
5-9	pieces
10	pieces



W 1607

**CULTURE TUBES "DE WIT"**

Polycarbonate, Gamma Radiated  
Heigh 130 mm, diameter middle 27 mm, diameter bottom 10 mm.

Culture Tubes "De Wit" are specifically designed for in Vitro Tissue Culture. The conical shape of the tubes provides enough space to grow while using a limited quantity of medium.

*Culture Tubes "De Wit" are sterile packed per 75 pieces.*

W 1607.0750	750 pieces	(10 x 75 pieces)
W 1607.1500	1500 pieces	(20 x 75 pieces)
W 1607.2250	2250 pieces	(30 x 75 pieces)
W 1607.3000	3000 pieces	(40 x 75 pieces)
W 1607.3750	3750 pieces	(50 x 75 pieces)
	> 3750 pieces	

T 1608

**"DE WIT TRAY"**

White polystyreen foam tray (60 x 40 cm) with 240 holes (20 x 12) to plug in "De Wit tubes"

T 1608.0010	Box of 10 pieces
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S 3301

**LEUCOPORE TAPE, 2.5 cm x 9.2 m**

Leucopore Tape is a non woven ventilating tape impermeable for bacteria. Due to these properties it can be used for sealing petridishes and tissue containers allowing ventilation without the plates drying.

L 3301.0001	1 roll of leucopore tape 2.5 cm x 9.2 m
1	roll
5	rolls
10	rolls

L 3302

**LEUCOPORE TAPE, 1.25 cm x 9.2 m**

L 3302.0001	1 roll of leucopore tape 1.25 cm x 9.2 m
1	roll
5	rolls
10	rolls

The goods E1650, E1654, E1674, W1607 and T1608 are shipped Ex Works (EXW) to all destinations. Transportation charges will vary with the destination, weight, and content of each shipment and will be subcharged accordingly on the corresponding invoice.

## THE FULL-GAS ECO2 BOX AND OS 140 BOX

A new generation of tissue culture vessels with a revolutionary breathing system, your guarantee for carefree micropropagation!

### Description:

- All boxes are equipped with a "breathing" hermetic cover.
- The cover is constructed out of parallel strips of cristal-clear plastic with intermittent narrow strips of filter material welded between them. This results in two parallel batteries of filters.
- Each filter battery consists of a double row of filter wicks, i.e. micro-channels filled with hydrofobic filter material.
- To adjust gas exchange two different types of colored filters are available.

Type	Color	Filter length
L	White	3.5 mm
XXL	Green	7.0 mm

Gas exchange will increase as a result of filter length.

### Its advantages:

- Adjustable gas exchange: this occurs by means of depth filtration through the numerous filter wicks. The length of these filter wicks can be adapted to the needs of the plant species being raised, thus avoiding vitrification.
- No danger of infection: the hermetic cover and the resilient filter material, which forms a perfect barrier against pests and secondary contamination.
- Recyclable: 100%, filter, vessel and cover are made of polypropylene.
- Eco 2 box and OS 140 box are not autoclavable.

## E 1650 / E 1654

### ECO2 BOX OVAL MODEL WITH HERMETIC COVER AND BREATHING STRIP

- Properties : crystal-clear polypropylene.
- Dimensions : vessel height: 80 mm  
vessel base: 125 mm L x 65 mm W  
vessel top and cover : 150 mm L x 90 mm W
- Packaging : vessels: 25 p. / sealed bag (350 (14 x 25))  
covers: 25 p. / sealed bag (350 (14 x 25))  
vessels and covers together in 1 carton.

Price per box of 350 complete sets:

E 1650.0001	White filter (L)
E 1654.0001	Green filter (XXL)

## E 1674

### OS 140 BOX + ODS FILTER: ROUND MODEL WITH HERMETIC COVER AND BREATHING STRIP

- Properties : crystal-clear polypropylene.
- dimensions : vessel height: 140 mm
- vessel base : 90 mm diameter
- vessel top and cover : 115 mm diameter
- packaging : vessels: 15 p. / sealed bag, (180 (12 x 15))

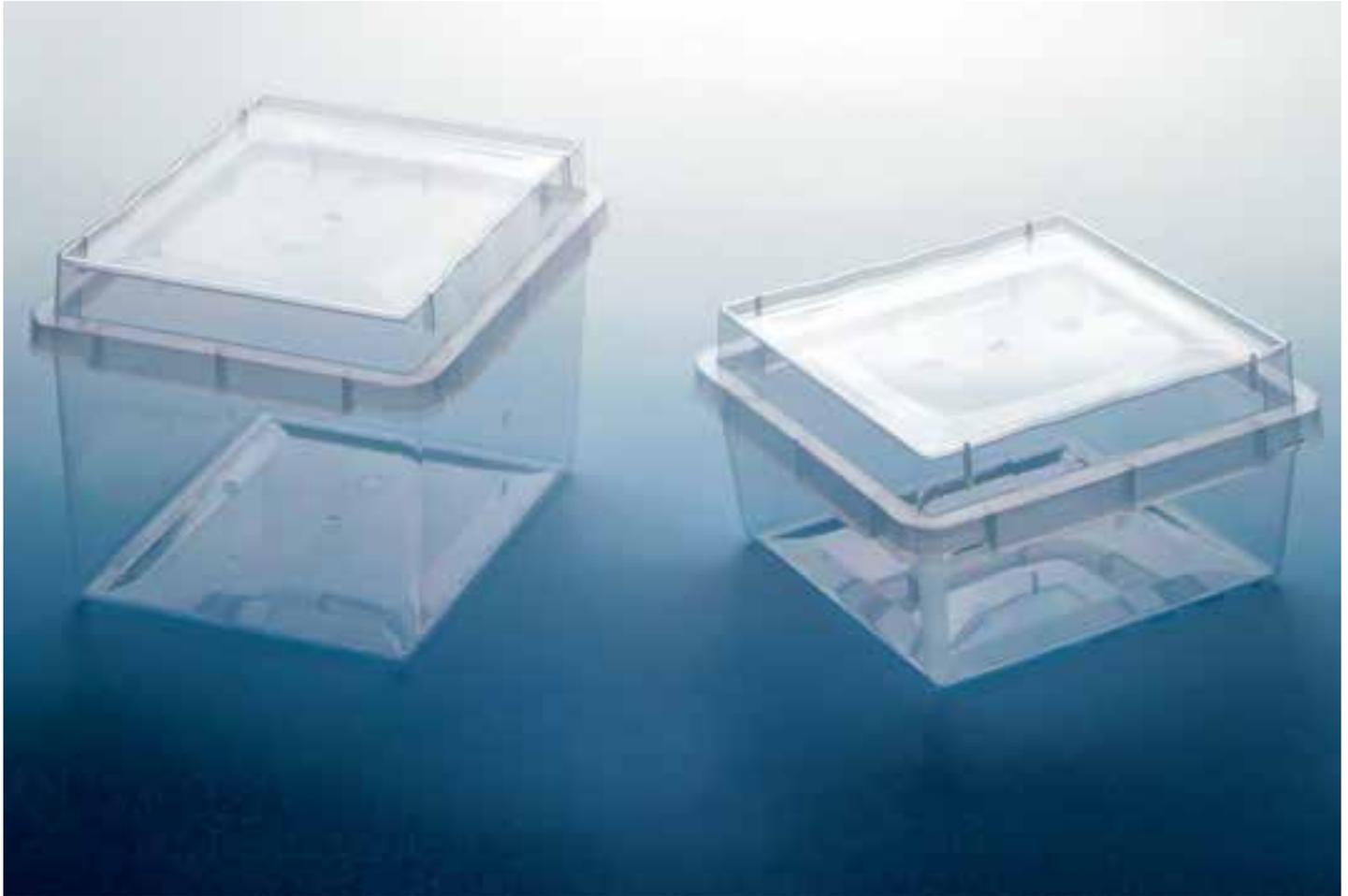
Price per box of 180 complete sets:

E 1674.0001	Green filter (XXL)
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S 1680/S1685

## STERI VENT CONTAINER



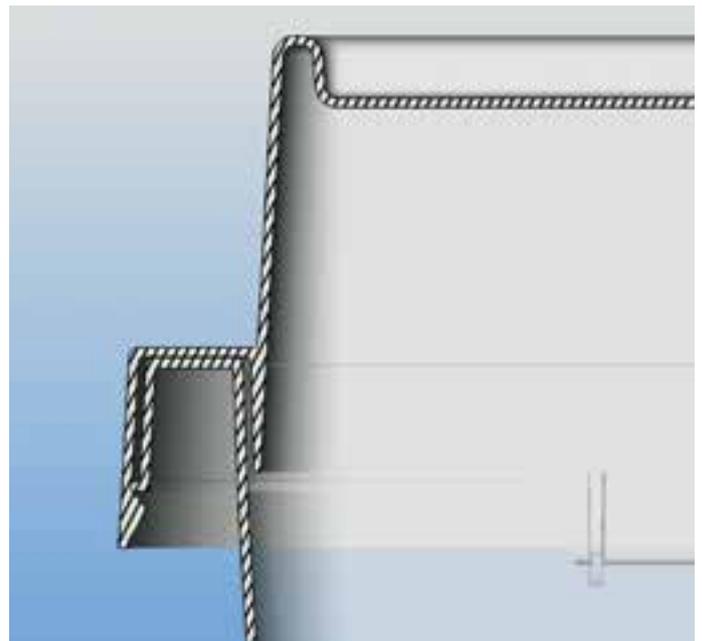
The newly developed Steri Vent Container is the successor of the successful Vitro Vent container.

Its completely new design contains many functional and ergonomical improvements. The Steri Vent is made of highly purified and totally transparent polypropylene, which results in a firm and crystal clear plant tissue culture container. Steri Vent containers are sterilized during the production process and do not need gamma irradiation, which causes discoloration of the polypropylene and detrimental chemical reactions.

### Closure

The newly developed labyrinth closure guarantees a hermetically closed container for Bacteria, Yeast, Fungi, Mites and Trips.

Although hermetically closed, the Steri Vent allows a continuous ventilation with the outer atmosphere. There is a continuous exchange of fresh air from the outside and volatile components from the inner side of the container. Another positive result of this air replacement is a severely reduced rate of condensation within the Steri Vent.



**Ergonomics**

The Steri Vent is a rectangular shaped container available in two sizes.

High Model with dimensions (lxbxh), 107 x 94 x 96 mm

Low model with dimensions (lxbxh), 107 x 94 x 65 mm

See picture below.

Both sizes allow a very efficient use of the available space in the climate room.

The lid is designed in such a way that the raised hood in the middle of the lid functions as a grip that avoids contamination when touching the container while closing. The manner to take hold of the hood is easily recognized by its curved shape, which allows fast and easy opening and closing.

Inside the containers are small circular rigs to permit a smooth de-stacking of the sterile containers packed in polypropylene bags.

Spacers on the outside of the bottom of the Steri Vent provide a fixed space between two piled containers with an improved aeration between separate piles of containers.

**Transport costs**

These goods are shipped Ex Works (EXW) to all destinations. Transportation charges will vary with the destination, weight, and content of each shipment and will be subcharged accordingly on the corresponding invoice.

**Pack sizes**

Steri Vent containers are packed and sold in sealed sterile polypropylene bags. Three different sleeves are available;

**S 1681.0032**

sleeve includes 32 Steri Vent lids.

15 sleeves are packed in one carton box.

**S 1682.0048**

sleeve includes 48 Steri Vent Low model containers.

15 sleeves are packed in one carton box.

**S 1686.0032**

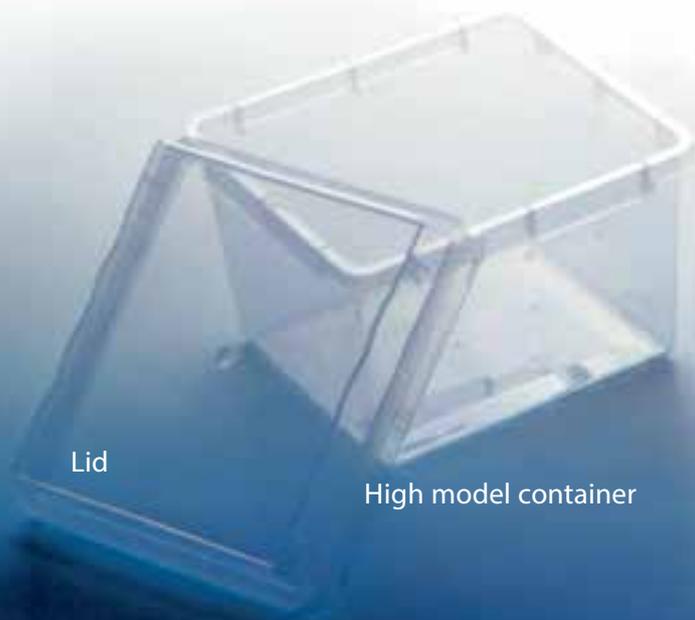
sleeve includes 32 Steri Vent High model containers.

15 sleeves are packed in one carton box.

Sleeves are packed in a solid carton box including polypropylene inside layer. Each carton box contains 15 cases of solely S1681.0032, S1682.0048 or S1686.0032

Steri vent High Model (Height 96 mm)			
		Lids	High container
		S 1681.0032	S 1686.0032
480	pcs	15 sleeves	15 sleeves
960	pcs	30 sleeves	30 sleeves
1920	pcs	60 sleeves	60 sleeves
2880	pcs	90 sleeves	90 sleeves
5760	pcs	180 sleeves	180 sleeves

Steri Vent Low Model (Height 65 mm)			
		Lids	Low container
		S 1681.0032	S 1682.0048
1440	pcs	45 sleeves	30 sleeves
2880	pcs	90 sleeves	60 sleeves



## STERILIZATION OF NUTRIENT MEDIUM IN FLOW

EnbioJet Sterilizer uses a modern technology called Direct Energy Transfer (DET), which involves an immediate transfer of microwave energy to medium flowing through a Teflon chamber. DET technology guarantees that all of the medium is heated to a constant high temperature within only a few seconds.

Main advantages:

- Possibility of a flexible efficiency increase up to 400 l/h
- Up to 75% energy savings
- Time saving
- Ease of operation
- Perfect Temperature Control
- Limited exposure of the nutrient medium to high temperature - sterilization effect within several seconds
- 30-percent lower agar consumption
- 100-percent microbiological efficiency

### Process parameters

Nutrient sterilization process parameters	Value
Capacities	90 l/h – 400 l/h
Input temperature	60 °C
Process temperature	132 °C
Output temperature	40 °C

### Technical parameters

EnbioJet technical parameters	Value
Power installed	16 kW
Average energy consumption	9 kW
Maximum process temperature	145 °C
Cooling water	5 l/min
Compressed air	5 bar

### Sterilization process in EnbioJet

Nutrient medium is pumped by the EnbioJet pump, and then it flows through a Teflon (PTFE) pipe section. There, energy coming from microwaves is supplied to the medium. The medium is heated to the temperature of 132 °C, and within several seconds the sterility effect is achieved. The validation performed using the *Bacillus Subtillis* and *Geobacillus stearothermophilus* strains confirmed the efficiency of sterilization in 132 °C within 10 seconds.

Sterile nutrient medium flows from the EnbioJet system to the dispensing system. As the process of sterilization in EnbioJet is effected in the flow, simultaneous pouring of the medium is possible. In order to ensure process continuity and stable sterilizer operation, the dispenser should be equipped with a buffer tank. It enables to hold the sterilized nutrient medium in a situation of a momentary dispensing delay.





Media sterilization in seconds with Enbiojet significantly reduce ingredient decomposition. Microwaves eliminate temperature gradients within the medium being processed and hence risks of under- or over-heating. Additionally, the sterilization and dispensing of the medium is conducted in one step, with savings of up to 50% in time and 50 to 75% in energy consumption compared to using either a media preparatory system or autoclaves. Input efficiencies arise from the mentioned very short exposure of the media to a high temperature and much lower thermal decomposition of fragile components. Medium pH also remains very stable and predictable. The sterilizing capacity of EnbioJet is 90 to 400 L/h.

The equipment has 3 automatic programs:

- FLUSHING, STERILIZATION - programs for flushing and sterilization with superheated steam of the equipment itself and connected dispenser process lines.
- PRODUCTION - the program used for sterilizing the nutrient medium.

The programs are controlled via the menu on the LCD panel, which is built into the device. Software makes it possible to record and archive all process data.





## Philips GreenPower LEDs save up to 60% energy

LED lighting is known to offer a number of benefits in horticulture, including increased yield, enabling earlier flowering and speeding up root growth, and, last but not least, substantial energy savings.

**PHILIPS**  
sense and simplicity

# Philips GreenPower LEDs save up to 60% energy

LEDs are used most effectively if the spectrum and light level are exactly tuned to the crop and growth conditions. In the past years, Philips conducted more than 30 field tests to determine the optimal spectrum and light level for multilayer production. This results in the GreenPower LED production module reducing energy consumption and creating a more uniform light distribution.

The **GreenPower LED Production module** for multilayer applications (typical 50-150  $\mu\text{mol/s/m}^2$ ) can replace conventional TL lighting (36W or 58W) reducing energy consumption up to 60%. For most applications, the modules with deep red and blue can be used. Next to energy efficiency, LEDs provide less heat and a more uniform light distribution.

## For most common installations a LED alternative is available:

The modules have the same length as the 36W TL (122 cm.) or 58W TL (152 cm.). An existing installation with 2x36W or 2x58W TL can be replaced by only one module producing a comparable light level.

## Features:

- Plug & play, integrated driver 230V
- Easy to install
- Long service life
- Light weight design
- IP66



Existing TL installation	Replace by LED module	Result At comparable light level	Payback time
1x36W	1x 122 cm 16W	Up to 60% energy saving	Less than 3 years
1x58W	1x 152 cm 23W		
2x36W	1x 122 cm 30W		
2x58W	1x 152 cm 45W		

## Technical data GreenPower LED production module:

Philips GreenPower LED production module	Photon flux typical $\mu\text{mol/s}$ per module	Power consumption W	Lifetime hours	Photon flux maintenance %	Length cm.	Order code
<b>Deep red/blue</b>						
GreenPower LED production DR/B 120 LO	24	16	25.000	90%	122	9290 004 87103
GreenPower LED production DR/B 120	47	30	25.000	90%	122	9290 004 86903
GreenPower LED production DR/B 150 LO	35	23	25.000	90%	152	9290 004 87603
GreenPower LED production DR/B 150	70	45	25.000	90%	152	9290 004 87403
<b>Deep red/white (if work light is needed)</b>						
GreenPower LED production DR/W 120	47	30	15.000	90%	122	9290 004 87003
GreenPower LED production DR/W 150	70	45	15.000	90%	152	9290 004 87503
<b>Deep red (if no blue is needed for growth)</b>						
GreenPower LED production DR 120	47	30	25.000	90%	122	9290 004 86803
GreenPower LED production DR 150	70	45	25.000	90%	152	9290 004 87303

## Next to the GreenPower LED production module Philips offers following solutions:

The **GreenPower LED Research module** is designed for research and field tests. With this module, the growth light level and spectrum (deep red, blue and far red) can be tuned exactly for different test plans.

**GreenPower LED string** is used in multilayer applications like tissue culture, storage and transport, where low uniform light levels are required (typical 10-25  $\mu\text{mol/s/m}^2$ ). The GreenPower LED string white is ideal as growth lighting (through efficient blue in the spectrum) and working light. The GreenPower LED string blue and deep red complete the range.

For more information please check [www.philips.com/horti](http://www.philips.com/horti) or contact Mr. Jan Dijkman ([jan.dijkman@philips.com](mailto:jan.dijkman@philips.com)).



# MEDIA FOR PHYTOPATHOLOGY

Duchefa Biochemie B.V. produces an extensive range of phytopathology media and media used in seed health testing. Since production takes place in our own laboratories, Duchefa Biochemie B.V. is also able to manufacture custom made media according to laboratory specifications. Obviously, strict secrecy is guaranteed.

## POWDERED MEDIA

Powdered media are extremely hygroscopic and must be protected from atmospheric moisture. Be sure the glass bottle containing the powdered medium is carefully closed after opening. Otherwise the remaining contents will deteriorate.

Store the dry medium at 2-8°C and keep well closed.

Preparing the media in a concentrated form is not recommended. Some salt complexes may precipitate in a concentrated solution.

## CUSTOM MADE MEDIUM

As a manufacturer of powdered media Duchefa Biochemie B.V. has the ability to produce almost any medium desired. Many of our relations are using custom made media fitting to their own specific purposes, that are produced by Duchefa Biochemie B.V. If you are interested to have your own medium, please contact us or send the Custom Made Medium form.

1. **Name:** Please mention your full name, address, fax and telephone number, so we can contact you if anything proves to be unclear.
2. **Name and/or Product number** of the custom-made medium
3. **Formulation:** The formulation of the medium will be stated in mg/l or molarity. To prevent possible mistakes we prefer to have the concentration in both ways. Please be accurate in your description, for instance: magnesium sulphate anhydrous or magnesium sulphate heptahydrate.
4. **Quantity:** To guarantee absolute homogeneity a minimal quantity per production of one kilogram custom made medium (or it's equivalent in litres) is required.
5. **Delivery Schedule:** Most custom made media will be supplied within two weeks. Larger quantities can be dispatched in portions if desired.
6. **Declaration of discretion:** Before sending us your formulation Duchefa Biochemie B.V. is prepared to send you a declaration in which absolute secrecy will be assured. After receipt of the undersigned declaration simply send your formulation. Please contact us if such a declaration is required.

## PRICES

The prices of most custom-made media are equal to the prices of our standard media. Favourable discounts will be granted on bulk quantities. However, additions of specific components to the media could have their influence on the price. Please indicate the details on the custom-made medium form and send it by mail, fax or e-mail to:

### DUCHEFA BIOCHEMIE B.V.

*We will contact you after receipt.*

## DISCLAIMER

Although described in literature as selective media for certain phytopathological micro-organisms Duchefa Biochemie B.V. strongly recommends that the enduser tests, each medium for its selective properties and nutritional requirements growth of mentioned micro-organisms. The use of positive controls and negative controls during the cultivation of pathogenic micro-organisms is strongly recommended. Duchefa B.V. does not accept any liability for the outcome of any test by using the phytopathology media as produced by Duchefa Biochemie B.V.

## BEAN

*Pseudomonas syringae* pv. *syringae*

KBBC

MSP

MT

*Pseudomonas savastanoi* pv. *phaseolicola*

mKB

MSP

MT

*Xanthomonas axonopodis* pv. *phaseoli*

MT

mXCP1

PTSA

## BRASSICA

*Xanthomonas campestris* pv. *campestris*

mCS20ABN

mFS

*Xanthomonas campestris* pv. *armoraciae*

mCS20ABN

mFS

## CARROTS

*Xanthomonas campestris* pv. *carotae*

mD5A

mKM

mTBM

## LEEK

*Pseudomonas syringae* pv. *porri*

PSM

KBBC

## PEA

*Pseudomonas syringae* pv. *pisi*

SNAC

KBBC

## PEPPER

*Xanthomonas campestris* pv. *vesicatoria*

mTMB

MXV

CKTM

## TOMATO

*Clavibacter michiganensis* subsp. *michiganensis*

mSCM

D2ANX

*Pseudomonas syringae* pv. *tomato*

KBBC

KBZ

*Xanthomonas campestris* pv. *vesicatoria*

mTMB

MXV

CKTM

## BACTERIAL MEDIUM

bacteria

KB

YDC

CDA

CDB

## FUNGAL MEDIUM

fungi

MA

CDA

CDB

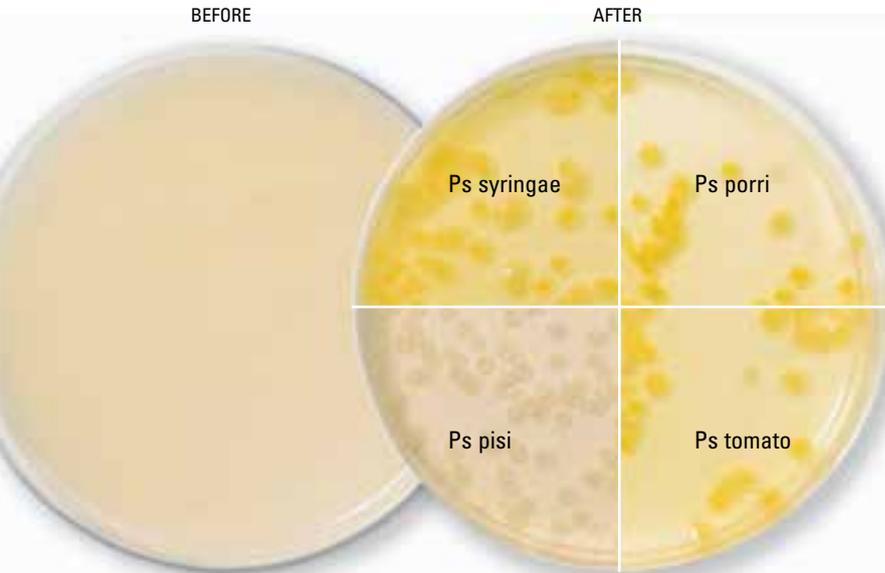
# Phytopathology

Bacteria Screening Medium 523	<b>B1713</b>	177
CKTM Medium	<b>C5140</b>	168
Czapek Dox Agar, CDA	<b>C1715</b>	174
Czapek Dox Broth, CDB	<b>C1714</b>	175
D2ANX Medium	<b>D5128</b>	170
KB medium (King's B Medium)	<b>K5165</b>	172
KBBC Medium	<b>K5120</b>	154
KBZ Medium	<b>K5129</b>	171
Leifert and Waites sterility test Medium	<b>L1716</b>	178
Luria Broth Agar, Miller	<b>L1718</b>	179
Luria Broth Base, Miller	<b>L1717</b>	180
Malt Agar (MA)	<b>L1719</b>	176
mCS20ABN Medium	<b>C5122</b>	159
mD5A Medium	<b>D5124</b>	161
mFS Medium	<b>F5123</b>	160
mKM Medium	<b>K5125</b>	162
MSP Medium	<b>M5167</b>	155
MT Medium	<b>M5133</b>	156
mTBM medium	<b>T5132</b>	163
mTMB Medium	<b>T5126</b>	166
mXCP1 Medium	<b>X5121</b>	157
MXV medium	<b>M5131</b>	167
PSM medium	<b>P5134</b>	164
PTSA Medium	<b>P5135</b>	158
SCM Medium	<b>S5127</b>	169
SNAC medium	<b>S5130</b>	165
YDC medium	<b>Y5136</b>	173

# PHYTOPATHOLOGY

# K5120 KBBC Medium

Crop: **Bean, Leek, Pea, Tomato**  
 Disease: **Bacterial brown spot (bean)**  
 Pathogen: ***Pseudomonas syringae* pv. *syringae***  
***Pseudomonas syringae* pv. *porri***  
***Pseudomonas syringae* pv. *pisii***  
***Pseudomonas syringae* pv. *tomato***



*Pseudomonas syringae* pv. *syringae* (*Pss*) is the causal organism of bacterial brown spot of beans. This bacterium is seed borne and therefore its detection on seeds is important. KBBC medium is a rather selective medium to detect *Pss* on seeds of beans. This medium is based on King's B Medium (K5165), however in KBBC Medium boric acid (1.5 g/liter), cephalaxin and nystatin are added. Nystatin is used to control fungi. As an alternative, cycloheximide, a more potent fungicide, can be used. KBBC is much more selective than MSP (M5167) and in general the recovery of *Pss* is smaller on KBBC than on MSP. *Pspha*, unlike *Pss*, will not grow on KBBC. Therefore, the chance of detection of *Pss* is higher when both complementary media are used. Detection of *Pss* is performed by the dilution plating of bacterial extract on KBBC and MSP. Then *Pss*-suspected isolates are transferred to KB medium. Finally, the identification of suspected colonies can be performed by a pathogenicity assay or PCR. Colonies of *Pss* on KBBC are 3-4 mm in diameter, flat, circular, translucent, creamy white and show blue fluorescence under UV light. This medium can also be used for the detection of seed borne *Ps porri*, *Ps pisi* and *Ps tomato* on seed of resp. leek, pea and tomato.

## COMPOSITION OF MEDIA K5120: KBBC MEDIUM

COMPOUND	GRAM/LITER
Agar	15.0
Di-potassium hydrogen phosphate (K <sub>2</sub> HPO <sub>4</sub> )	1.5
Boric acid (H <sub>3</sub> BO <sub>3</sub> )	1.5
Magnesium sulphate anhydrous (MgSO <sub>4</sub> anhydrous)	0.73
Proteose Peptone	20.0

## METHOD

- Dissolve 38.7 grams of ingredients in distilled water and adjust volume to 970 ml.
- Add 30 ml glycerol (50%) and mix.
- Adjust pH to 7.2.
- Autoclave the solutions (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:  
80 mg cephalaxin monohydrate (C0110)  
35 mg nystatin (N0138) or 100 mg cycloheximide (C0176)
- Allow medium to cool down to ca. 45 °C – 50 °C and add antibiotics to the solution.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

Mohan, S.K. and Schaad, N.W. 1987. An improved agar plating assay for detecting *Pseudomonas syringae* pv. *syringae* and *Pseudomonas syringae* pv. *phaseolicola* in contaminated bean seed. *Phytopathology* 77: 1390-1395.

## K5120 KBBC MEDIUM

K5120.1000 1 kg

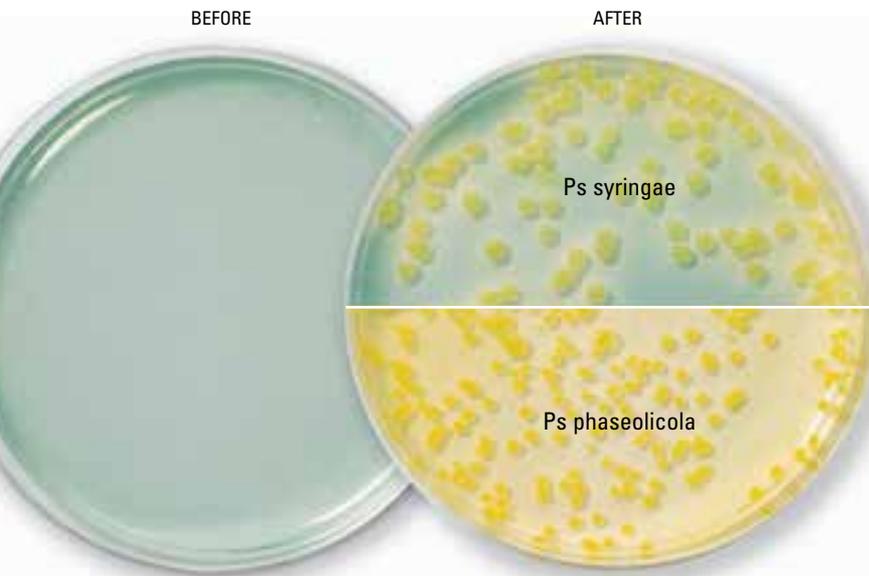
For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# M5167 MSP Medium

Crop: **Bean (*Phaseolus vulgaris*)**

Disease: **Bacterial brown spot and halo blight**

Pathogen: ***Pseudomonas syringae* pv. *syringae***  
***Pseudomonas savastanoi* pv. *phaseolicola***



MSP (Modified Sucrose Peptone) medium is a suitable medium for the detection of *Pseudomonas savastanoi* pv. *phaseolicola* (*Psp*) and *Pseudomonas syringae* pv. *syringae* (*Pss*). Addition of bromothymol blue gives this medium a blue appearance. The color of bacterial colonies is influenced by this compound. The assay starts with dilution plating of bacterial extract from seeds on MSP. Then suspected colonies from MSP can be transferred to King's B Medium (K5165). Finally, the identity of suspected isolates is confirmed by a pathogenicity test or PCR.

Colonies of *Psp* and *Pss* are ca. 3 mm in diameter, circular, raised, globose, glistening and light yellow with a denser center. The medium around *Psp* colonies turns light yellow after three days of incubation.

## COMPOSITION OF MEDIA M5167: MSP MEDIUM

COMPOUND	GRAM/LITER
Agar	20.0
Di-potassium hydrogen phosphate (K <sub>2</sub> HPO <sub>4</sub> )	0.5
Peptone special	5.0
Magnesium sulphate anhydrous (MgSO <sub>4</sub> anhydrous)	0.13
Sucrose	20.0

## METHOD

- Dissolve 45.6 grams of ingredients in distilled water and adjust volume to 1000 ml.
- Adjust pH to 7.4.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Prepare sterile solutions and add the following amounts per liter medium:
  - 80 mg cephalexin monohydrate (C0110)
  - 35 mg nystatin (N0138)
  - 10 mg vancomycin HCl (V0155)
  - 15 mg bromothymol blue
- Allow medium to cool down to ca. 45 °C – 50 °C and add antibiotics to the solution.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

Mohan, S.K. and Schaad, N.W. 1987. An improved agar plating assay for detecting *Pseudomonas syringae* pv. *syringae* and *Pseudomonas syringae* pv. *phaseolicola* in contaminated bean seed. *Phytopathology* 77: 1390-1395.

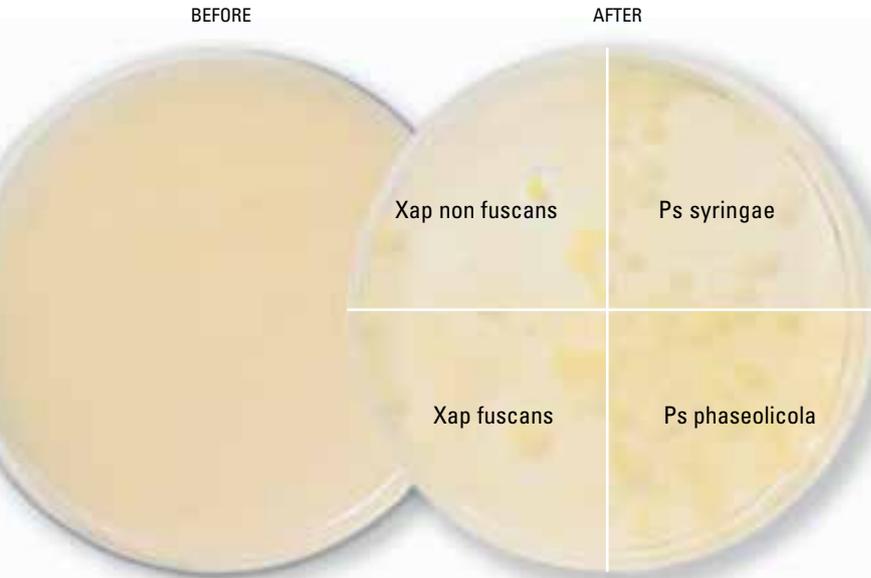
## M5167 MSP MEDIUM

M5167.1000                      1 kg

For prepared and ready to use plates of this medium contact:  
Tritium Microbiologie      Tel : 040-2051615  
Rooijakkersstraat 6        Fax : 040-2051395  
5652 BB Eindhoven        Email : info@tritium-microbiologie.nl  
The Netherlands

# M5133 MT Medium

Crop: **Bean (*Phaseolus vulgaris*)**  
 Disease: **Bacterial brown spot, common blight and halo blight**  
 Pathogen: ***Pseudomonas syringae* pv. *syringae*  
*Pseudomonas savastanoi* pv. *phaseolicola*  
*Xanthomonas axonopodis* pv. *phaseoli***



The MT (Milk-Tween) Medium is a semi-selective medium for the detection of *Pseudomonas syringae* pv. *syringae* (*Pss*), *Pseudomonas savastanoi* pv. *phaseolicola* (*Psp*) and *Xanthomonas axonopodis* pv. *phaseoli* (*Xap*) in bean seed. The medium relies on the ability of the micro-organisms to hydrolyze casein. Suspected isolates are transferred to YDC (*Xap*) or KB (*Pss* and *Psp*). Finally, the identity of suspected colonies is determined by PCR or a pathogenicity test. The colonies of *Psp* and *Pss* are cream white, flat circular, 4-5 mm in diameter and produce a blue fluorescent pigment under UV light. *Xap* colonies (3 – 3.5 mm in diameter) are yellow, non fluorescent and typical two zones surround colonies: a bigger, clear zone of casein hydrolysis and a smaller zone of Tween 80 lipolysis. *Xap* var. *fuscans* (1 – 2 mm in diameter) produces a brown pigment within 5 days.

**COMPOSITION OF MEDIA  
M5133: MT MEDIUM**

COMPOUND	GRAM/LITER
Proteose Peptone	10.0
Calcium chloride anhydrous (CaCl <sub>2</sub> anhydrous)	0.25
Tyrosine	0.5
Agar	15.0

**METHOD**

- Dissolve 25.7 grams of ingredients in distilled water and adjust volume to 800 ml.
- Dissolve 10 ml Tween 80 in distilled water and adjust volume to 100 ml.
- Dissolve 10 g of skim milk powder in 100 ml distilled water.
- Autoclave the solutions separately (121 °C, 15 psi for 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:  
 80 mg cephalixin monohydrate (C0110)  
 35 mg nystatin (N0138)  
 10 mg vancomycin HCl (V0155)
- Allow medium to cool down to ca. 45 °C – 50 °C and add the Tween, skim milk powder and antibiotics solutions.
- Mix gently to avoid air bubbles and pour plates (20 ml per 9.0 cm plate).

**Reference:**

Goszczyńska and Serfontein, 1998 "Milk-Tween agar, a semiselective medium for isolation and differentiation of *Pseudomonas syringae* pv. *syringae*, *Pseudomonas syringae* pv. *phaseolicola* and *Xanthomonas axonopodis* pv. *phaseoli* ", Journal of Microbiological Methods 32: 65-72.

**M5133 MT MEDIUM**

K5133.1000                      1 kg

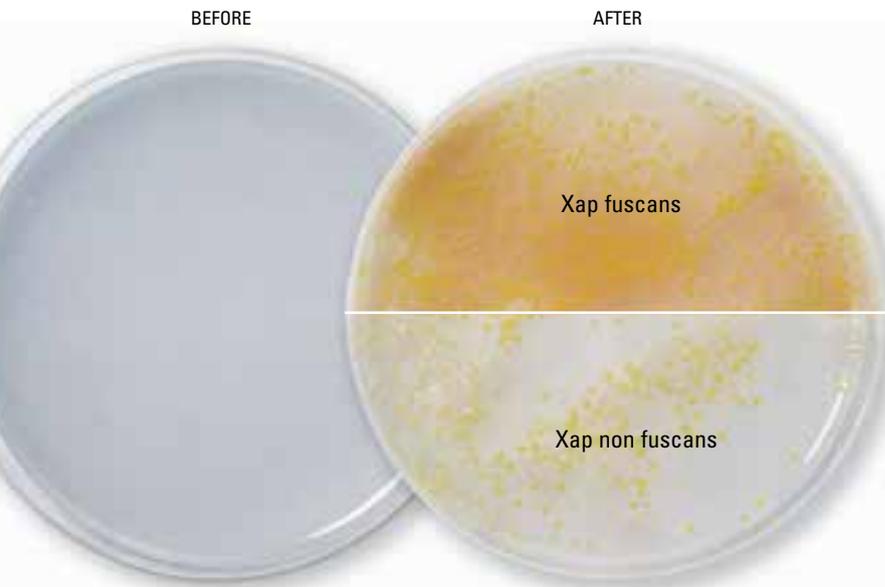
For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie      Tel : 040-2051615  
 Rooijakkersstraat 6      Fax : 040-2051395  
 5652 BB Eindhoven      Email : info@tritium-microbiologie.nl  
 The Netherlands

# X5121 mXCP1 Medium

Crop: **Bean (*Phaseolus vulgaris*)**

Disease: **Common blight**

Pathogen: ***Xanthomonas axonopodis* pv. *phaseoli***



The mXCP1 (modified *Xanthomonas Campestris* pv. *Phaseoli*) medium is a semi-selective medium for the detection of *Xanthomonas axonopodis* pv. *phaseoli* (*Xap*) in bean seed. Both the fuscans and non-fuscans type of *Xap* grow on mXCP1. However the production of the fuscous pigment only becomes visible after a relatively long incubation. Modification of the medium was necessary because of poor recovery of isolates of the *Xap* var. fuscans type. Recognition of putative *Xap* colonies relies on the ability of the *Xanthomonas axonopodis* pv. *phaseoli* to hydrolyze starch. The colonies of *Xanthomonas axonopodis* pv. *phaseoli* on the mXCP1 plate are surrounded by a clear zone of starch hydrolysis.

Detection of *Psp* and *Xap* is often performed in combi-assay. *Xap* is detected by dilution plating of bacterial extract from seeds on mXCP1. Then suspected colonies from mXCP1 should be transferred to YDC. Finally, the identity of suspected isolates is confirmed by a pathogenicity test or PCR.

*Xap* colonies are yellow mucoid, convex and surrounded by a clear zone of starch hydrolysis. Colonies of var. fuscans are distinguished by brown pigmentation.

## COMPOSITION OF MEDIA X5121: mXCP1 MEDIUM

COMPOUND	GRAM/LITER
Peptone special	10.0
Potassium bromide (KBr)	10.0
Calcium chloride anhydrous (CaCl <sub>2</sub> anhydrous)	0.25
Agar	20.0
Soluble Starch	20.0
Crystal Violet	0.0015

## METHOD

- Dissolve 60.2 grams of the ingredients in distilled water and adjust volume to 900 ml.
- Dissolve 10 ml Tween 80 in distilled water and adjust volume to 100 ml.
- Autoclave the solutions (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 10 mg cephalixin monohydrate (C0110)
  - 3 mg 5-fluorouracil (F0123)
  - 0.1 mg tobramycin sulphate (T0153)
  - 35 mg nystatin (N0138)
- Allow medium to cool down to ca. 45 °C – 50 °C, mix solutions and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).
- Store plates for 4 days at 4° C to improve visibility of starch hydrolysis.

## Reference:

McGuire, R.G., Jones, J.B. and Sasser, M. 1986. Tween media for semiselective isolation of *Xanthomonas campestris* pv. *vesicatoria* from soil and plant material. *Plant Dis.* 70: 887 - 891

## X5121 mXCP1 MEDIUM

X5121.1000

1 kg

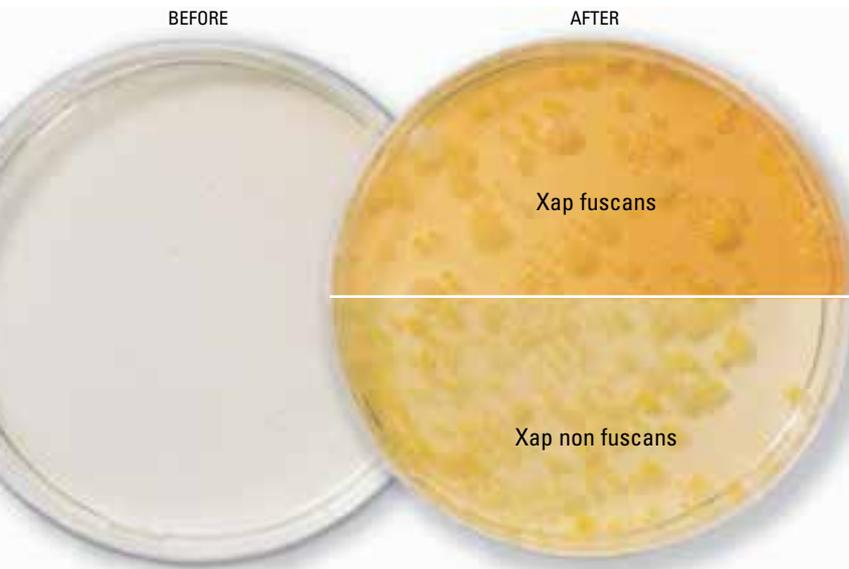
For prepared and ready to use plates of this medium contact:  
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 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# P5135 PTSA Medium

Crop: **Bean (*Phaseolus vulgaris*)**

Disease: **Common blight**

Pathogen: ***Xanthomonas axonopodis* pv. *phaseoli***



PTSA (Peptone Tyrosine Sodium chloride Agar) is a semi-selective medium for the detection of *Xanthomonas axonopodis* pv. *phaseoli* in bean seed. The medium is not very selective in comparison with mXCP1, but especially colonies from the var. *fuscans* are easily recognized on this medium because of their excessive production of visible brown pigment. The non-fuscans isolates of *Xap* grow well on PTSA medium but their recognition is much more difficult due to the lack of pigment production. For relatively clean seed lots, PTSA medium is useful, but for saprophyte-rich samples mXCP1 is much more suitable. *Xap* is detected by dilution plating of bacterial extract from seeds on PTSA. Then suspected colonies from PTSA should be transferred to YDC. Finally, the identity of suspected isolates is confirmed by a pathogenicity test or PCR. Colonies of *Xap* var. *fuscans* are distinguished by brown pigmentation.

## COMPOSITION OF MEDIA P5135: PTSA MEDIUM

COMPOUND	GRAM/LITER
Peptone special	10.0
L-tyrosine	1.0
Soluble starch	2.0
Sodium chloride (NaCl)	5.0
Agar	15.0

## METHOD

- Dissolve 33.0 grams of ingredients in distilled water and adjust volume to 1000 ml.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down to ca. 45 °C – 50 °C.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

Van Vuurde J.W.L., Van den Bovenkamp, G.W. and Birnbaum, Y. 1983. Immunofluorescence microscopy and enzyme-linked immunosorbent assay as potential routine tests for the detection of *Pseudomonas syringae* pv. *phaseolicola* and *Xanthomonas campestris* pv. *phaseoli* in bean seeds. Seed Sc. & Technol. 11: 547 -559

## P5135 PTSA MEDIUM

P5135.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# C5122

## mCS20ABN Medium

(extra phosphate and Agar)

Crop: **Brassica**

Disease: **Black rot and bacterial leaf spot**

Pathogen: ***Xanthomonas campestris* pv. *campestris* and *Xanthomonas campestris* pv. *armoraciae***



CS20ABN has been developed by Chang et al. to isolate *Xanthomonas campestris* pv. *campestris* (*Xcc*) from crucifer seeds. The original medium recipe allowed the quick isolation of most isolates of *Xcc*. However, the recovery of some isolates of *Xcc* was poor due to pH-dependent sensitivity to neomycin. In the modified version, the pH is lowered to 6.5 by the addition of extra potassium dihydrogen phosphate.

This modification improved the recovery of some neomycin-sensitive isolates considerably.

Contaminated seed lots can be detected by dilution plating of the bacterial extract on mCS20ABN and mFS. Suspected isolates are then transferred to YDC. Finally, the identity of the suspected isolates can be determined by a pathogenicity test using brassica seedlings.

The colonies of *Xcc* and *Xanthomonas campestris* pv. *armoraciae* are yellow, mucoid and surrounded by a zone of starch hydrolysis.

### COMPOSITION OF MEDIA

#### C5122: mCS20ABN MEDIUM

COMPOUND	GRAM/LITER
Agar	18.0
Soluble starch	25.0
Soya Peptone	2.0
Tryptone	2.0
Potassium dihydrogen phosphate (KH <sub>2</sub> PO <sub>4</sub> )	2.8
Di-ammonium hydrogen phosphate ((NH <sub>4</sub> ) <sub>2</sub> HPO <sub>4</sub> )	0.8
Magnesium sulphate anhydrous (MgSO <sub>4</sub> anhydrous)	0.1952
L-glutamine	6.0
L-histidine	1.0
Glucose monohydrate	1.0

### METHOD

- Dissolve 58.8 grams of ingredients in 900 ml distilled water.
- Adjust pH to 6.5 and adjust volume to 1000 ml.
- pH should be 6.5 and not higher!
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 35 mg nystatin (N0138)
  - 40 mg neomycin (M0135)
  - 100 mg bacitracin (B0106)
- Allow medium to cool down to ca. 45 °C – 50 °C and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).
- Store plates for 4 days at 4 °C to improve visibility of starch hydrolysis.

### Reference:

Chang, C.J., Donaldson, R., Crowley, M, and Pinnow, D. 1991. A new semiselective medium for the isolation of *Xanthomonas campestris* pv. *campestris*. *Phytopathology* 81:449-453.

### C5122 mCS20ABN MEDIUM

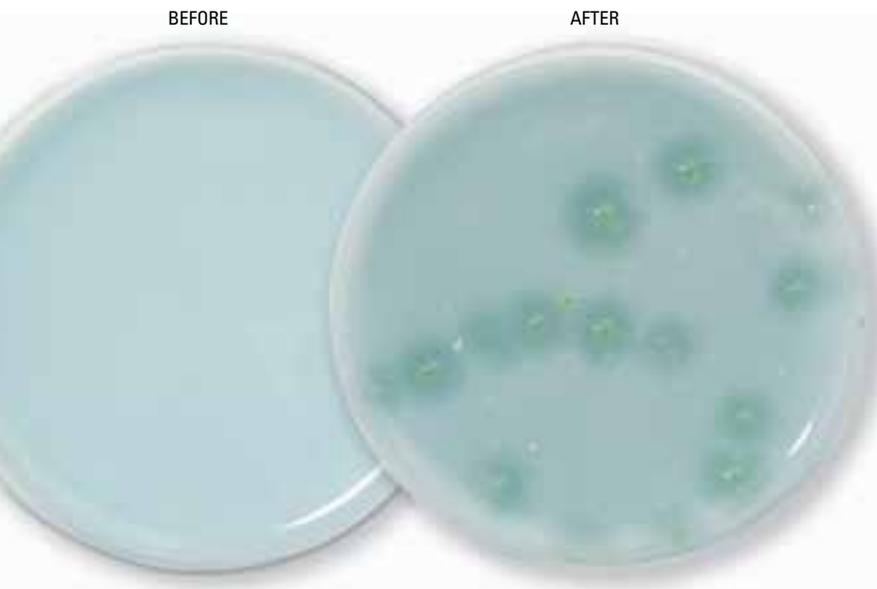
C5122.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# F5123

## mFS Medium

Crop: **Brassica**Disease: **Black rot and bacterial leaf spot**Pathogen: ***Xanthomonas campestris* pv. *campestris***  
***Xanthomonas campestris* pv. *armoraciae***

mFS (modified Fieldhouse Sasser medium) has been developed to detect black rot in brassica. This medium is complementary to mCS20ABN (C5122) due to some alternative antibiotics. Modifications concern the addition of extra starch and omission of gentamycin.

Contaminated seed lots can be detected by dilution plating of the bacterial extract on mCS20ABN and mFS. Suspected isolates are then transferred to YDC. Finally, the identity of the suspected isolates can be determined by a pathogenicity test using brassica seedlings.

The colonies of *Xanthomonas campestris* pv. *campestris* (*Xcc*) and *Xanthomonas campestris* pv. *armoraciae* (*Xca*) on mFS medium are pale green to transparent, mucoid and surrounded by a small zone of starch hydrolysis. Colonies are in general smaller than on mCS20ABN and may show remarkable variation in size and may be visible only after 5-6 days.

### COMPOSITION OF MEDIA

#### F5123: mFS MEDIUM

COMPOUND	GRAM/LITER
Soluble starch	25.0
Yeast Extract	0.1
Di-potassium hydrogen phosphate ( $K_2HPO_4$ )	0.8
Potassium dihydrogen phosphate ( $KH_2PO_4$ )	0.8
Potassium nitrate ( $KNO_3$ )	0.5
Magnesium sulphate anhydrous ( $MgSO_4$ anhydrous)	0.0488
Agar	15.0

### METHOD

- Dissolve 42.2 grams of ingredients in distilled water and adjust volume to 950 ml and adjust pH to 6.8.
- Add 1.5 ml methyl green (1 % aq.) and adjust volume to 1000 ml with distilled water.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Prepare the following sterile solutions of vitamins, amino acids and antibiotics per liter medium:
  - 35 mg nystatin (N0138)
  - 3 mg D-methionine (M0715)
  - 1 mg pyridoxine-HCl (P0612)
  - 50 mg cephalixin monohydrate (C0110)
  - 30 mg trimethoprim (T0154)
- Allow medium to cool down to ca. 45 °C – 50 °C and add solutions.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).
- Store plates for 4 days at 4 °C to improve visibility of starch hydrolysis.

### Reference:

Yuen, G.Y., Alvarez, A.M., Benedict, A.A., and Trotter, K.J. 1987. Use of monoclonal antibodies to monitor the dissemination of *Xanthomonas campestris* pv. *campestris*. *Phytopathology* 77:366-370.

### F5123 mFS MEDIUM

F5123.1000

1 kg

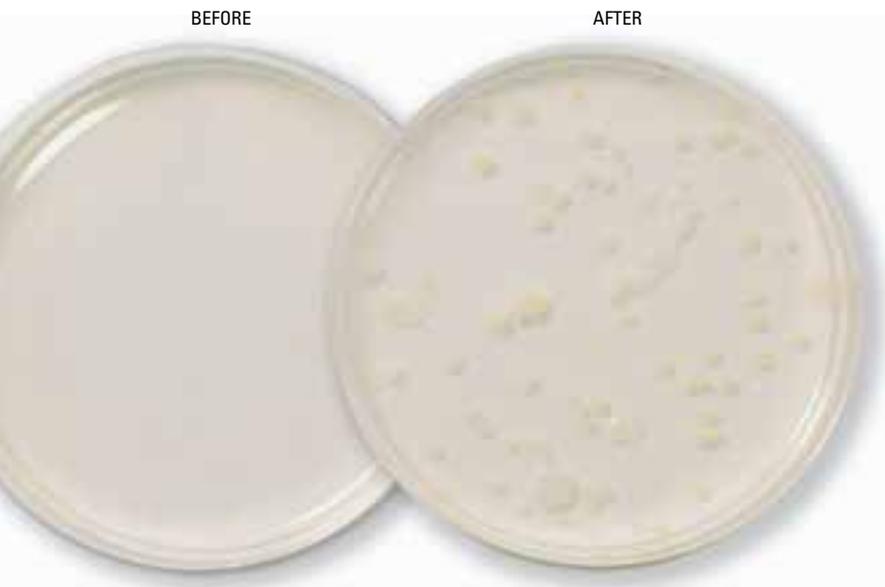
For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# D5124 mD5A Medium

Crop: **Carrot (*Daucus carota*)**

Disease: **Bacterial leaf blight**

Pathogen: ***Xanthomonas hortorum* pv. *carotae***



mD5A (modified D-5 Agar medium) is used to detect seed borne *Xanthomonas campestris* pv. *carota* (*Xccar*), the causal organism of bacterial blight of carrots. Contaminated seed lots can be detected by dilution plating of the bacterial extract on mD5A and another semi-selective medium. Suspected isolates are then transferred to YDC. Finally, the identity of the suspected isolates can be determined by PCR. Colonies of *Xccar* on mD5A medium look straw-yellow, glistening, round, smooth, convex and are 2–3 mm in diameter.

## COMPOSITION OF MEDIA D5124: mD5A MEDIUM

COMPOUND	GRAM/LITER
Agar	15.0
Sodium dihydrogen phosphate (NaH <sub>2</sub> PO <sub>4</sub> )	0.9
Di-potassium hydrogen phosphate (K <sub>2</sub> HPO <sub>4</sub> )	3.0
Magnesium sulphate anhydrous (MgSO <sub>4</sub> anhydrous)	0.15
Ammonium chloride (NH <sub>4</sub> Cl)	1.0

## METHOD

- Dissolve 20.1 grams of ingredients in distilled water and adjust volume to 900 ml and adjust pH to 6.4.
- Dissolve 10.0 grams of D-cellobiose in distilled water and adjust volume to 100 ml.
- Autoclave the solutions separately (121 °C, 15 psi, 15 minutes).
- Prepare the following sterile amino acids and antibiotics solutions and add the following amounts per liter medium:
  - 5 mg L-glutamic acid (G0707)
  - 1 mg L-methionine (M0715)
  - 35 mg nystatin (N0138)
  - 10 mg cephalixin monohydrate (C0110)
  - 10 mg bacitracin (B0106)
- Allow medium to cool down to ca. 45 °C – 50 °C and add solutions.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

Kuan, T.L., Minsavage, G.V. and Gabrielson, R.L. 1985. Detection of *Xanthomonas campestris* pv. *carotae* in carrot seed. Plant disease 61758-61760.  
Cubeta, M.S. and Kuan, T.L. 1986 Comparison of MD5 and XCS media and development of MD5A medium for detection of *Xanthomonas hortorum* p.v. *carotae* in carrot seed, Phytopathology 76: 1109 (Abstract)

## D5124 mD5A MEDIUM

D5124.1000

1 kg

For prepared and ready to use plates of this medium contact:  
Tritium Microbiologie Tel : 040-2051615  
Rooijakkersstraat 6 Fax : 040-2051395  
5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
The Netherlands

# K5125 mKM Medium

Crop: **Carrot (*Daucus carota*)**Disease: **Bacterial leaf blight**Pathogen: ***Xanthomonas hortorum* pv. *carotae***

mKM medium (modified KM-1 medium) is used to detect *Xanthomonas hortorum* pv. *carotae* (*Xccar*). Contaminated seed lots can be detected by dilution plating of the bacterial extract on mD5A and another semi-selective medium. Suspected isolates are then transferred to YDC. Finally, the identity of the suspected isolates can be determined by PCR. The colonies of *Xccar* on mKM plates are light-yellow cream, light brown to peach yellow, glistening, round and about 2 – 4 mm in diameter.

## COMPOSITION OF MEDIA K5125: mKM MEDIUM

COMPOUND	GRAM/LITER
Agar	18.0
Potassium dihydrogen phosphate (KH <sub>2</sub> PO <sub>4</sub> )	1.2
Di-potassium hydrogen phosphate (K <sub>2</sub> HPO <sub>4</sub> )	1.2
Ammonium chloride (NH <sub>4</sub> Cl)	1.0
Lactose monohydrate	10.0
Threhalose anhydrous.	4.0
2-Thiobarbituric acid	0.2
Yeast Extract	0.5

## METHOD

- Dissolve 36.1 grams of the ingredients in distilled water and adjust volume to 1000 ml and adjust pH to 6.6.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 35 mg nystatin (N0138)
  - 10 mg cephalixin monohydrate (C0110),
  - 50 mg bacitracin (B0106)
  - 2 mg tobramycin sulphate (T0153)
- Allow medium to cool down to ca. 45 °C – 50 °C and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

Kim, H.K., Sasser, M. and Sands, D.C. 1982. Selective medium for *xanthomonas hortorum* pv. *translucens* Phytopathology 72:936. (Abstrn)

## K5125 mKM MEDIUM

K5125.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# T5132

## mTBM Medium

Crop: **Carrot (*Daucus carota*)**

Disease: **Bacterial leaf blight**

Pathogen: ***Xanthomonas hortorum* pv. *carotae***

BEFORE

AFTER



mTBM Medium (modified TBM medium) is used to detect *Xanthomonas hortorum* pv. *carotae* (*Xcca*). Other semi-selective media for *Xanthomonas campestris* pv. *carotae* are mKM Medium (K5125) and mD5A Medium (D5124). The colonies of *Xanthomonas hortorum* pv. *carotae* on mTBM plates are white or yellow or white-yellow, glistening round, convex with entire margins and surrounded by a large clear zone of casein hydrolyses.

### COMPOSITION OF MEDIA

#### T5132: mTBM MEDIUM

COMPOUND	GRAM/LITER
Agar	15.0
Boric acid (H <sub>3</sub> BO <sub>3</sub> )	0.3
Potassium bromide (KBr)	10.0
Peptone	10.0

### METHOD

- Dissolve 35.3 grams of ingredients in distilled water and adjust volume to 800 ml and adjust pH to 7.4.
- Dissolve 10 ml of Tween 80 in distilled water and adjust to 100 ml.
- Dissolve 10 g of skim milk powder in distilled water and adjust volume to 100 ml.
- Autoclave the solutions separately (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 20 mg nystatin (N0138)
  - 65 mg cephalixin monohydrate (C0110)
  - 12 mg 5-fluorouracil (F0123)
- Allow solution to cool down to ca. 45 °C – 50 °C and mix the solutions.
- Mix gently to avoid air bubbles and pour plates (20 ml per 9.0 cm plate).

### Reference:

McGuire, R.G., Jones, J.B. and Sasser, M. 1986. Tween medium for semiselective isolation of *Xanthomonas hortorum* pv. *vesicatoria* from soil and plant material. *Plant Dis.* 70; 887 – 891.

### T5132 mTBM MEDIUM

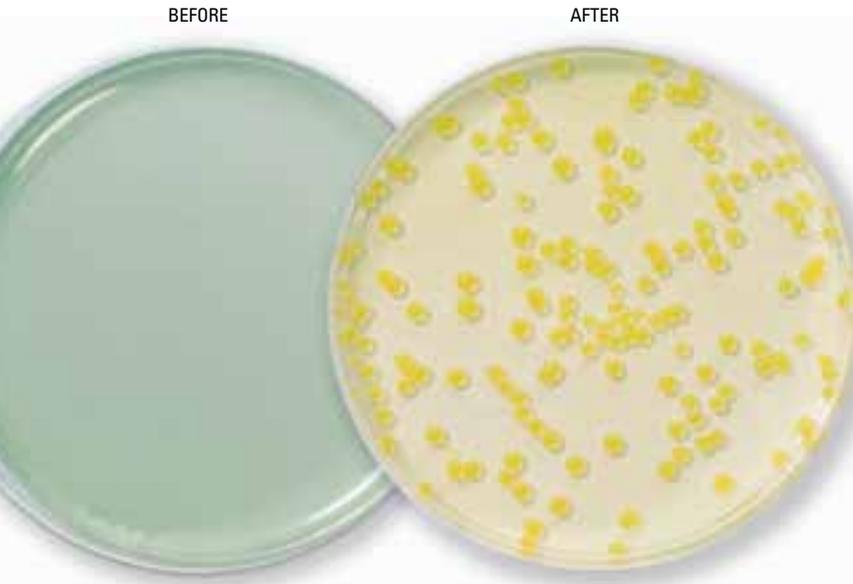
T5132.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# P5134

## PSM Medium

Crop: **Leek**Disease: **Bacterial blight of leek**Pathogen: ***Pseudomonas syringae* pv. *porri***

*Pseudomonas syringae* pv. *porri* (*Pspo*) is the causal organism of bacterial blight of leek. This pathogen can be seed-borne and therefore the testing of seeds of leek is common. Seeds of leek can be saprophyte-rich and this might disguise the presence of *Pspo*. Detection of this bacterium is performed by dilution plating on highly selective media such as KBBC and PSM (Pseudomonas Syringae Medium). Putative *Pspo* colonies are then transferred to KB. Thereafter the identity of the suspected colonies is determined by immunofluorescence microscopy. Finally, the identity is determined by a *Pspo*-specific PCR or a pathogenicity assay using seedlings of leek.

On PSM the colonies of *Pspo* are 2-4 mm in diameter, circular with smooth edge, translucent, creamy-yellow to transparent white. Note that the color of *Pspo* colonies is rather variable since the accumulation of bromothymol blue per colony is strongly dependent on the total number of colonies per plate.

### COMPOSITION OF MEDIA P5134: PSM MEDIUM

COMPOUND	GRAM/LITER
Sucrose	20.0
Peptone special	5.0
Di-potassium hydrogen phosphate ( $K_2HPO_4$ )	0.5
Magnesium sulphate anhydrous ( $MgSO_4$ )	0.13
Agar	20.0

### METHOD

- Dissolve 45.6 grams of ingredients in 970 ml distilled water, adjust pH to 7.5 and adjust volume to 990 ml.
- Add 1 gram of boric acid to 10 ml of distilled water.
- Autoclave the solutions separately (121 °C, 15 psi, 15 minutes).
- Prepare sterile solutions and add the following amounts per liter medium:
  - 80 mg cephalixin monohydrate (C0110)
  - 35 mg nystatin (N0138)
  - 10 mg vancomycin HCl (V0155)
  - 15 mg bromothymol blue
- Allow medium to cool down to ca. 45 °C – 50 °C and add boric acid and antibiotic solutions to mixture of the ingredients.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

### Reference:

Koike, S.T., Barak, J.D., Henderson, D.M., and Gilbertson, R.L. 1999. Bacterial blight of leek: A new disease in California caused by *Pseudomonas syringae*. Plant Dis. 83:165-170.

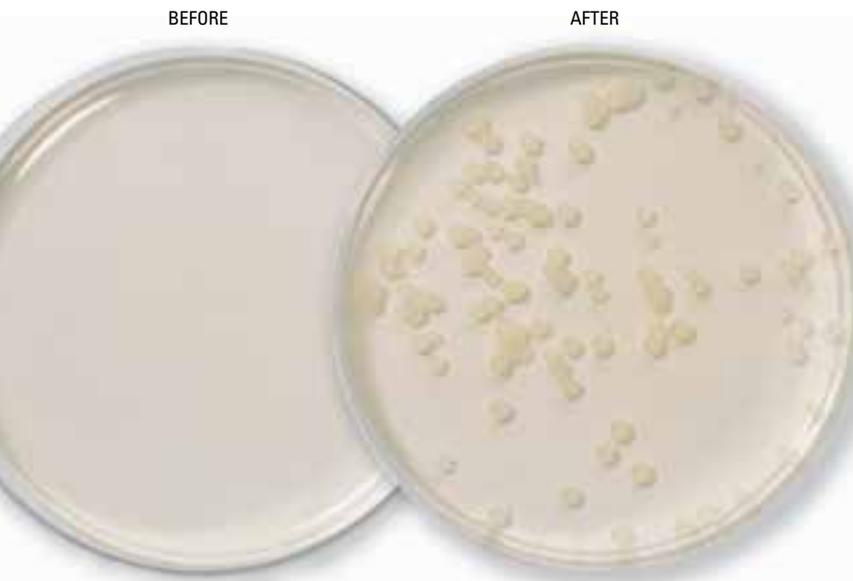
### P5134 PSM MEDIUM

P5134.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# S5130 SNAC Medium

Crop: **Pea**Disease: **Bacterial blight of pea**Pathogen: ***Pseudomonas syringae* pv. *pisii***

*Pseudomonas syringae* pv. *pisii* (*Psp*) is the causal organism of bacterial blight of pea. The use of clean seeds is an important measure for controlling this disease. SNAC is derived from the SNA medium. The selectivity of the medium was increased by the addition of boric acid and antibiotics. In general dilution plating on semi-selective medium such as SNAC and/or KBBC is used for the detection of *Psp*. Then suspected colonies are transferred to KB. Through immunofluorescence microscopy, PCR or a pathogenicity assay the identity of suspected isolates can be confirmed.

Colonies of *Psp* on SNAC are white to transparent mucoid and dome-shaped.

## COMPOSITION OF MEDIA S5130: SNAC MEDIUM

COMPOUND	GRAM/LITER
Tryptone	5.0
Peptone	3.0
Sodium chloride (NaCl)	5.0
Sucrose	50.0
Agar	15.0

## METHOD

- Dissolve 75.0 grams of ingredients in distilled water and adjust volume to 990 ml.
- Add 1 gram of boric acid to 10 ml of distilled water.
- Autoclave the solutions separately (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 80 mg cephalixin monohydrate (C0110)
  - 35 mg nystatin (N0138)
- Allow medium to cool down to ca. 45 °C – 50 °C and add boric acid and antibiotic solutions.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

Franken, A.A.J.M., and van den Bovenkamp, G.W. 1990. The application of the combined use of immunofluorescence microscopy and dilution plating to detect *Pseudomonas syringae* pv. *pisii* in pea seeds. In proceedings of the 7th ICPP pp. 871-875.

## S5130 SNAC MEDIUM

S5130.1000

1 kg

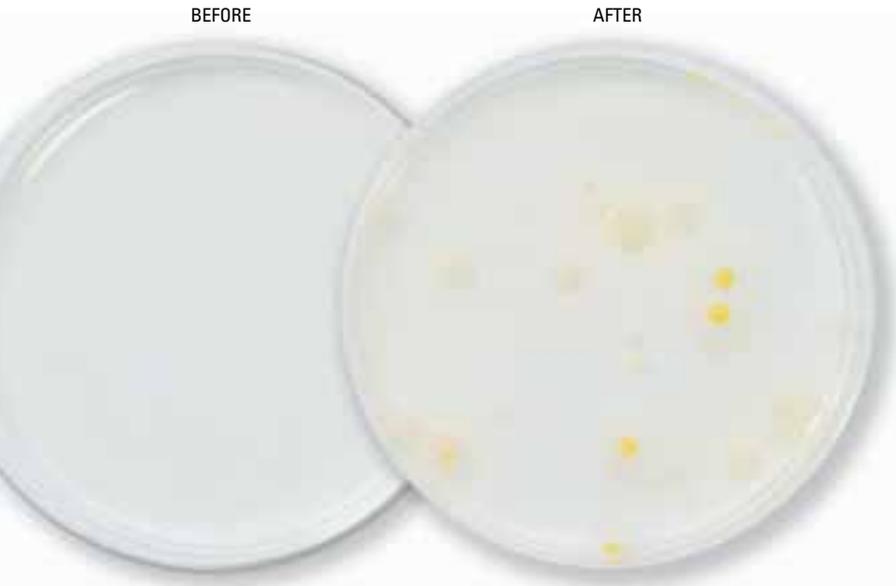
For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# T5126 mTMB Medium

Crop: **Pepper (*Capsicum annuum*)**  
**Tomato (*Lycopersicon lycopersicum*)**

Disease: **Bacterial spot**

Pathogen: ***Xanthomonas campestris* pv. *vesicatoria***  
***Xanthomonas vesicatoria***



Bacterial spot is an important bacterial disease of peppers. Two different bacteria, *Xanthomonas campestris* pv. *vesicatoria* (*Xcv*) and *Xanthomonas vesicatoria* (*Xv*) can incite this seed borne disease. mTMB (modified Tween Medium B) is a semi-selective medium for detection of *Xcv* and *Xv* on seeds of pepper and tomato. The colonies of *Xcv* and *Xv* on mTMB plates are yellow, slightly mucoid, mounded and round. *Xcv* utilizes Tween 80 and in 3-7 days a white crystalline halo usually forms around the yellow colony. Contaminated seed lots can be detected by dilution plating of the bacterial extract on CKTM, mKM or MXV. Suspected isolates are then transferred to YDC. Finally, the identity of the suspected isolates can be determined by a pathogenicity test or PCR.

## COMPOSITION OF MEDIA T5126: mTMB MEDIUM

COMPOUND	GRAM/LITER
Agar	15.0
Potassium bromide (KBr)	10.0
Boric acid (H <sub>3</sub> BO <sub>3</sub> )	0.1
Calcium chloride anhydrous (CaCl <sub>2</sub> anhydrous)	0.25
Peptone	10.0

## METHOD

- Dissolve 35.3 grams of ingredients in distilled water and adjust volume to 900 ml.
- Dissolve 10 ml of Tween 80 in distilled water and adjust volume to 100 ml.
- Autoclave the solutions separately (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 65 mg cephalixin monohydrate (C0110)
  - 12 mg 5-fluorouracil (F0123)
  - 0.2 mg tobramycin sulphate (T0153)
  - 100 mg cycloheximide (C0176)
- Allow medium to cool down to ca. 45 °C – 50 °C, mix the solutions and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

McGuire, R.G., Jones, J.B., and Sasser, M. 1986. Tween medium for semiselective isolation of *Xanthomonas campestris* pv. *veiscatoria* from soil and plant material. Plant Dis. 70:887-891.

## T5126 mTMB MEDIUM

T5126.1000                      1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie      Tel : 040-2051615  
 Rooijakkersstraat 6        Fax : 040-2051395  
 5652 BB Eindhoven        Email : info@tritium-microbiologie.nl  
 The Netherlands

# M5131 MXV Medium

Crop:	<b>Pepper (<i>Capsicum annuum</i>)</b> <b>Tomato (<i>Lycopersicon lycopersicum</i>)</b>
Disease:	<b>Bacterial spot</b>
Pathogen:	<b><i>Xanthomonas campestris</i> pv. <i>vesicatoria</i></b> <b><i>Xanthomonas vesicatoria</i></b>



Bacterial spot is an important bacterial disease of peppers. Two different bacteria, *Xanthomonas campestris* pv. *vesicatoria* (*Xcv*) and *Xanthomonas vesicatoria* (*Xv*) can incite this seed borne disease. MXV medium is a semi-selective medium for detection of *Xcv* and *Xv* on seeds of pepper and tomato. The colonies of *Xcv* on MXV plates utilize Tween 80 and are yellow and mucoid. Contaminated seed lots can be detected by dilution plating of the bacterial extract on mTMB, CKTM or mKM. Suspected isolates are then transferred to YDC. Finally, the identity of the suspected isolates can be determined by a pathogenicity test or PCR.

## COMPOSITION OF MEDIA M5131: MXV MEDIUM

COMPOUND	GRAM/LITER
Agar	15.0
Potassium dihydrogen phosphate (KH <sub>2</sub> PO <sub>4</sub> )	0.8
Di-potassium hydrogen phosphate (K <sub>2</sub> HPO <sub>4</sub> )	0.8
Ammonium chloride (NH <sub>4</sub> Cl)	1.0
Lactose	10.0
Threhalose	4.0
Thiobarbituric acid	0.1
Yeast Extract	0.5

## METHOD

- Dissolve 32.2 grams of the ingredients in distilled water, adjust volume to 900 ml and adjust pH to 6.6.
- Dissolve 10 ml of Tween 80 in distilled water and adjust volume to 100 ml.
- Autoclave the solutions separately (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 32.5 mg cephalixin monohydrate (C0110)
  - 100 mg bacitracin (B0106)
  - 6 mg 5-fluorouracil (F0123)
  - 10 mg neomycin sulphate (M0135)
  - 0.2 mg tobramycin sulphate (T0153)
  - 100 mg cycloheximide (C0176)
- Allow medium to cool down to ca. 45 °C – 50 °C, mix the solutions and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

McGuire, R.G., Jones, J.B., and Sasser, M. 1986. Tween medium for semiselective isolation of *Xanthomonas campestris* pv. *veiscatoria* from soil and plant material. *Plant Dis.* 70:887-891.

## M5131 MXV MEDIUM

M5131.1000                      1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie      Tel : 040-2051615  
 Rooijakkersstraat 6        Fax : 040-2051395  
 5652 BB Eindhoven        Email : info@tritium-microbiologie.nl  
 The Netherlands

# C5140 CKTM Medium

Crop: **Pepper (*Capsicum annuum*)**  
**Tomato (*Lycopersicon lycopersicum*)**

Disease: **Bacterial spot**

Pathogen: ***Xanthomonas campestris* pv. *vesicatoria***



CKTM medium is a semi-selective medium, which is used in combination with modified TMB medium (T5126) or MXV medium (M5131) to detect *Xanthomonas campestris* pv. *vesicatoria* (*Xcv*) in seeds of pepper and tomato. *Xcv* colonies on plates containing CKTM media are yellow, mucoid, mounded and round.

**COMPOSITION OF MEDIA**  
**C5140: CKTM MEDIUM**

COMPOUND	GRAM/LITER
Soya Peptone	2.0
Tryptone	2.0
Glucose anhydrous	1.0
L-glutamine	6.0
L-histidine	1.0
Di-ammonium hydrogen phosphate ((NH <sub>4</sub> ) <sub>2</sub> HPO <sub>4</sub> )	0.8
Potassium dihydrogen phosphate (KH <sub>2</sub> PO <sub>4</sub> )	1.0
Magnesium sulfate anhydrous (MgSO <sub>4</sub> anh)	0.2
Agar	15.0

**METHOD**

- Dissolve 29.0 grams of the ingredients in distilled water and adjust volume to 900 ml.
- Dissolve 10 ml of Tween 80 in distilled water and adjust volume to 100 ml.
- Autoclave the solutions separately (121 °C, 15 psi for 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 65 mg cephalixin monohydrate (C0110)
  - 12 mg 5-fluorouracil (F0123)
  - 0.4 mg tobramycin sulphate (T0153)
  - 100 mg cycloheximide (C0176)
  - 100 mg bacitricin (B0106)
  - 10 mg neomycin sulphate (M0135)
- Allow medium to cool down to ca. 45 °C – 50 °C, mix the solutions and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

**Reference:**

Sijam, K., Chang, C.J. and Gitaitis, R.D. 1992. A medium for differentiation tomato and pepper strains of *Xanthomonas campestris* pv. *vesicatoria*. *Canad. J. Plant Pathol.* 90: 208-213.

**C5140 CKTM MEDIUM**

C5140.1000                      1 kg

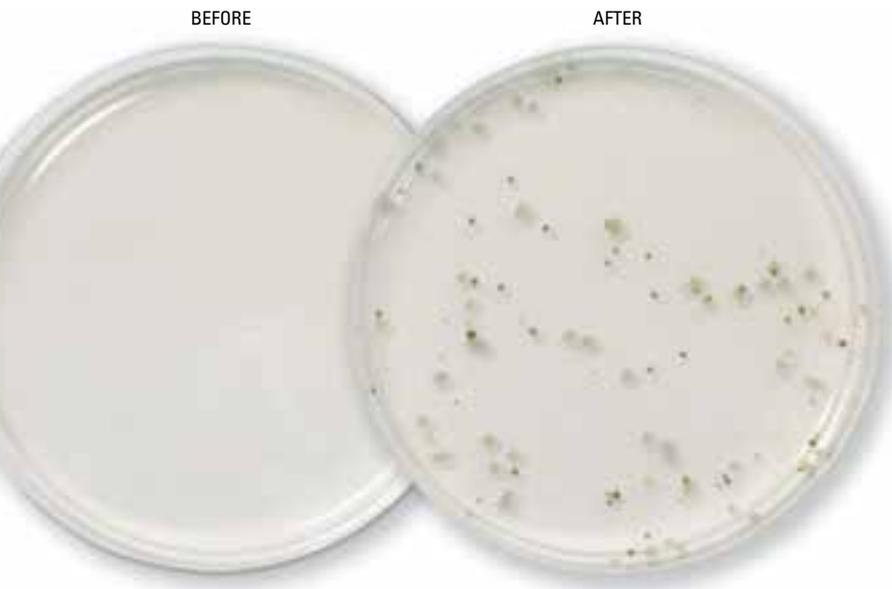
For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie      Tel : 040-2051615  
 Rooijakkersstraat 6        Fax : 040-2051395  
 5652 BB Eindhoven        Email : info@tritium-microbiologie.nl  
 The Netherlands

# S5127 SCM Medium

Crop: **Tomato (*Lycopersicon lycopersicum*)**

Disease: **Bacterial canker**

Pathogen: ***Clavibacter michiganensis* subsp. *michiganensis***



Bacterial canker is the most important bacterial disease of tomato. The causal organism is *Clavibacter michiganensis* subsp. *michiganensis* (*Cmm*) and this bacterium can be introduced by contaminated seeds. For the detection of *Cmm*, tomato seeds are first soaked in buffer. Then a stomacher is used for the release of bacteria from the seeds. After the concentration of the bacteria, dilution plating on two semi-selective media is performed. SCM medium is such a semi-selective media. Actually, there are several modifications in use concerning the used carbon source, LiCl and the addition of antibiotics. This medium is used in combination with D2ANX medium (D5128). After dilution plating suspected isolates are transferred to YDC. Finally the identity of suspected isolates is determined by a pathogenicity test or PCR. The colonies of *Clavibacter michiganensis* subsp. *michiganensis* on SCM are small, light to dark grey, glistening, fluidal and often irregularly shaped.

## COMPOSITION OF MEDIA S5127: mSCM MEDIUM

COMPOUND	GRAM/LITER
Agar	18.0
Potassium dihydrogen phosphate (KH <sub>2</sub> PO <sub>4</sub> )	0.5
Di-potassium hydrogen phosphate (K <sub>2</sub> HPO <sub>4</sub> )	2.0
Magnesium sulphate anhydrous (MgSO <sub>4</sub> anhydrous)	0.122
Boric acid (H <sub>3</sub> BO <sub>3</sub> )	1.5
Yeast Extract	0.1
Sucrose	10.0

## METHOD

- Dissolve 32.2 grams of ingredients in distilled water, adjust volume to 1000 ml and adjust pH to 7.3.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Prepare sterile solutions and add the following amounts per liter medium:
  - 100 mg nicotinic acid (N0611)
  - 30 mg nalidixic acid (N0134)
  - 100 mg cycloheximide (C0176)
  - 10 mg potassium tellurite (1 ml of 1% tellurite solution)
- Allow medium to cool down to ca. 45 °C – 50 °C and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

Fatmi, M. and Schaad, N.W. 1988. Semiselective agar medium for isolation of *Clavibacter michiganense* subsp. *michiganense* from tomato seeds. *Phytopathology* 78:121-126.

## S5127 SCM MEDIUM

S5127.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# D5128 D2ANX Medium

Crop: **Tomato (*Lycopersicon lycopersicum*)**

Disease: **Bacterial canker**

Pathogen: ***Clavibacter michiganensis* subsp. *michiganensis***

BEFORE

AFTER



D2ANX is a semi-selective medium, which is used to detect *Clavibacter michiganensis* subsp. *michiganensis* (*Cmm*). This medium, with a relatively low selectivity, is often used in combination with the more selective mSCM medium (S5127). Despite the slow growth of *Cmm* colonies the evaluation of plates can already be performed after 6-7 days of incubation. On mSCM, the growth is more slow and *Cmm* colonies can only be seen after about 9-10 days. On D2ANX, *Cmm* colonies are glistening, yellow and mucoid.

## COMPOSITION OF MEDIA D5128: D2ANX MEDIUM

COMPOUND	GRAM/LITER
MgSO <sub>4</sub> anhydrous	0.15
Glucose anhydrous	10.0
Yeast Extract	2.0
Agar	18.0
Tris HCl	1.2
Boric acid (H <sub>3</sub> BO <sub>3</sub> )	1.0
Ammonium chloride (NH <sub>4</sub> Cl)	1.0
Casein hydrolysate	4.0

## METHOD

- Dissolve 37.3 grams of ingredients in distilled water, adjust volume to 1000 ml and adjust pH to 7.4.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Prepare sterile antibiotic solutions and add the following amounts per liter medium:
  - 28 mg nalidixic acid (N0134)
  - 100 mg cycloheximide (C0176)
  - 10 mg polymixin B sulphate (P0145)
- Allow solutions to cool down to ca. 45 °C – 50 °C and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).
- R: 36/37/38

## Reference:

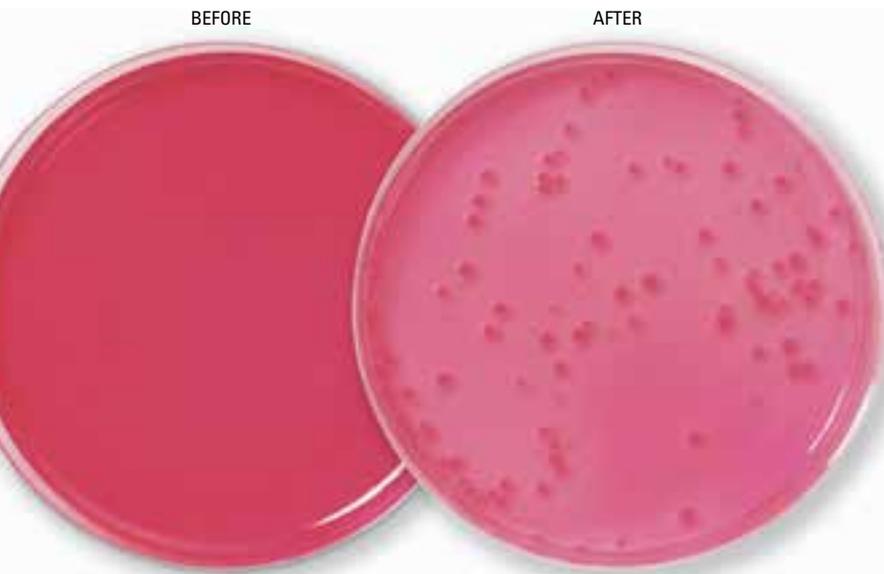
Kado, C.I., and Heskett, M.G. 1970. Selective media for the isolation of *Agrobacterium*, *Corynebacterium*, *Erwinia*, *Pseudomonas* and *Xanthomonas*. *Phytopathology* 60:969-976.

## D5128 D2ANX MEDIUM

D5128.1000                      1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie      Tel : 040-2051615  
 Rooijakkersstraat 6        Fax : 040-2051395  
 5652 BB Eindhoven        Email : info@tritium-microbiologie.nl  
 The Netherlands

# K5129 KBZ Medium

Crop: **Tomato**Disease: **Bacterial speck**Pathogen: ***Pseudomonas syringae* pv. *tomato***

Bacterial speck of tomatoes is caused by the bacterium *Pseudomonas syringae* pv. *tomato* (*Pst*). The bacterium can be introduced by the use of *Pst*-contaminated seeds. Therefore, detection of *Pst* in seeds of tomato is common. For the detection of *Pst*, seeds are first soaked in buffer. Then a stomacher is used for the release of bacteria from the seeds. The bacteria are concentrated by centrifugation. Then dilution plating on two semi-selective media KBZ and KBBC is performed. Suspected colonies are transferred to KB and finally identified by PCR or a pathogenicity assay. *Pst* forms small, flat and pink-colored colonies on KBZ after ca. 5 days.

## COMPOSITION OF MEDIA K5129: KBZ MEDIUM

COMPOUND	GRAM/LITER
Agar	15.0
Di-potassium hydrogen phosphate (K <sub>2</sub> HPO <sub>4</sub> )	1.5
Magnesium sulphate anhydrous (MgSO <sub>4</sub> anhydrous)	0.73
Proteose	20.0

## METHOD

- Dissolve 37.2 grams of ingredients in distilled water, adjust volume to 960 ml and adjust pH to 7.5.
- Prepare 30 ml of 50 % glycerol.
- Dissolve 1.5 g boric acid in 10 ml distilled water.
- Autoclave the solutions separately (121 °C, 15 psi, 15 minutes).
- Prepare sterile solutions and add the following amounts per liter medium:
  - 160 mg cephalaxin monohydrate (C0110)
  - 1,4 mg triphenyltetrazoliumchloride
  - 100 mg cycloheximide (C0176)
  - 18 mg paraosanilin
- Allow medium to cool down to ca. 45 °C – 50 °C, mix the solutions and add antibiotics.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

## Reference:

King, E.O. Ward, M.K. and Raney, D.E. 1954. Two simple media for the demonstration of pyocyanin and fluorescein. J. Lab. Clin. Med. 44:301-307.

## K5129 KBZ MEDIUM

K5129.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# K5165 KB Medium

Medium: **General bacterial medium**

Purpose: **Subculturing of numerous bacterial species**



KB (King's B) is a non-selective medium and used to subculture suspected isolates. Addition of antibiotics such as cephalaxine will make the medium (mKB) suitable for the detection of several *Pseudomonads* such as *Pseudomonas syringae* pv. *syringae* and *Pseudomonas savastanoi* pv. *phaseolicola* (see photo).

King's B medium is amongst others used for detection and subculturing of fluorescent *pseudomonads* from seeds and plants. Pathovars of *Pseudomonas syringae* produce a blue fluorescent pigment that becomes visible under UV light.

## COMPOSITION OF MEDIA K5165: KB MEDIUM

COMPOUND	GRAM/LITER
Agar	15.0
Di-potassium hydrogen phosphate ( $K_2HPO_4$ )	1.5
Magnesium sulphate anhydrous ( $MgSO_4$ anhydrous)	0.73
Proteose	20.0

## METHOD

- Dissolve 37.2 grams of ingredients in distilled water, adjust volume to 980 ml and adjust pH to 7.5.
  - Add 20 ml of 50% glycerol.
  - Autoclave the solution (121 °C, 15 psi, 15 minutes).
  - Allow medium to cool down to ca. 45 °C – 50 °C.
  - Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).
- **Optional: addition of 50 mg cephalaxin and 35 mg nystatin per liter to allow selectivity for pseudomonads (mKB).**

## Reference:

King, E.O. Ward, M.K. and Raney, D.E. 1954. Two simple media for the demonstration of pyocyanin and fluorescein. J. Lab. Clin. Med. 44:301-307.

## K5165 KB MEDIUM

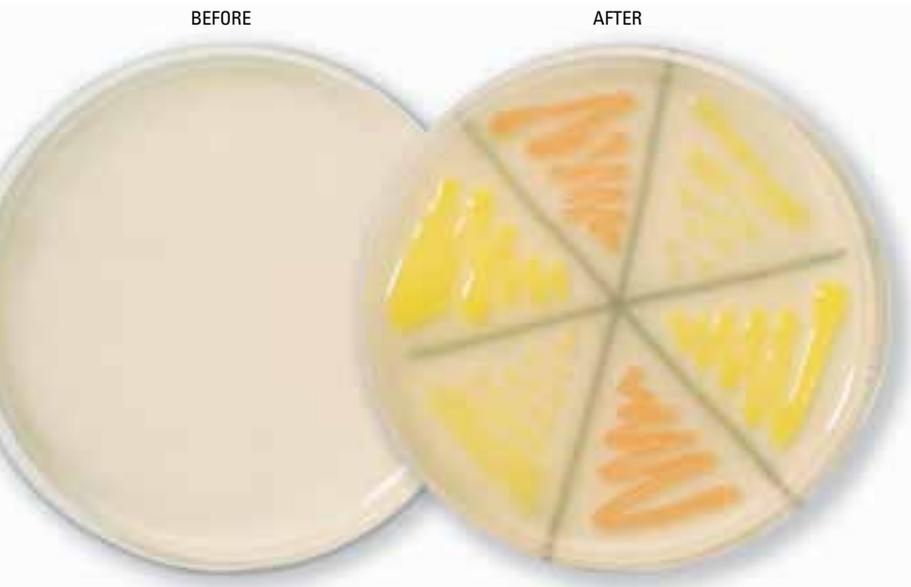
K5165.1000 1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# Y5136 YDC Medium

Medium: **General bacterial medium**

Purpose: **Subculturing bacteria such as xanthomonads and clavibacters**



YDC (Yeast extract-dextrose-CaCO<sub>3</sub>) medium is a non-selective media. YDC is used amongst others for subculturing suspected xanthomonads (yellow) and clavibacters (orange) after dilution on semi-selective media (see photo).

## COMPOSITION OF MEDIA Y5136: YDC MEDIUM

COMPOUND	GRAM/LITER
Yeast Extract	10.0
Calcium carbonate (CaCO <sub>3</sub> )	20.0
Agar	15.0
Glucose anhydrous	20.0

## METHOD

- Dissolve 65.0 grams of ingredients in distilled water, adjust volume to 1000 ml and adjust pH to 6.9.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down to ca. 45 °C – 50 °C.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).
- During pouring of medium mix the CaCO<sub>3</sub> thoroughly.

## Reference:

Wilson, E.E. Zeitoun, F.M. Fredrickson, D.L. 1967. Bacterial phloem canker, a new disease of Persian walnut trees. *Phytopathology* 57:618-621.

## Y5136 YDC MEDIUM

Y5136.1000

1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands



Medium: **General fungal and bacterial medium**

Purpose: **Cultivation of fungi and bacteria**



Czapex Dox Agar medium is used for the cultivation of those fungi and bacteria that are able to utilize sodium nitrate as the sole source of nitrogen.

**COMPOSITION OF MEDIA  
C1715: CZAPEK DOX AGAR, CDA**

COMPOUND	GRAM/LITER
Agar	12.0
Ferrous sulphate	0.01
Magnesium glycerophosphate	0.5
Potassium chloride	0.5
Potassium sulphate	0.35
Sodium nitrate	2.0
Sucrose	30.0

**METHOD**

- Dissolve 45.5 grams of ingredients in distilled water and adjust volume to 1000 ml.
- The final pH has to be  $6.8 \pm 0.2$ .
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down to ca. 45 °C – 50 °C.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

**Reference:**

Tuite, J. 1969. Plant pathological methods - fungi and bacteria. Burgess publishing co., Minneapolis, MN. 293 pp.

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

<b>C1715 CZAPEK DOX AGAR, CDA</b>	
C1715.0100	100 g
C1715.0500	500 g
C1715.1000	1000 g

# C1714

## Czapek Dox Broth, CDB

Medium: **General fungal and bacterial medium**

Purpose: **Cultivation of fungi and bacteria**

BEFORE

AFTER



Czapek Dox Broth medium is used for the cultivation of those fungi and bacteria that are able to utilize sodium nitrate as the sole source of nitrogen.

### COMPOSITION OF MEDIA

#### C1714: CZAPEK DOX BROTH, CDB

COMPOUND	GRAM/LITER
Ferrous sulphate	0.01
Magnesium glycerophosphate	0.5
Potassium chloride	0.5
Potassium sulphate	0.35
Sodium nitrate	2.0
Sucrose	30.0

- Dissolve 33.4 grams of ingredients in distilled water and adjust volume to 1000 ml.
- The final pH has to be  $6.8 \pm 0.2$ .
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down.

#### Reference:

Tuite, J. 1969. Plant pathological methods - fungi and bacteria. Burgess publishing co., Minneapolis, MN. 293 pp.

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie    Tel : 040-2051615  
 Rooijakkersstraat 6    Fax : 040-2051395  
 5652 BB Eindhoven    Email : info@tritium-microbiologie.nl  
 The Netherlands

#### C1714 CZAPEK DOX BROTH, CDB

C1714.0500	500 g
C1714.1000	1000 g

# L1719

## Malt Agar, MA

Medium: **General fungal medium**

Purpose: **Culturing of fungi**



Malt Agar medium is a non-selective multipurpose medium for cultivation of numerous fungi. Lowering the pH of the medium below 5.5 results in the inhibition of bacteria and permits good recovery of yeasts and moulds. Growth of bacteria can be reduced by the addition of antibiotics.

### COMPOSITION OF MEDIA L1719 MALT AGAR, MA

COMPOUND	GRAM/LITER
Agar	30.0
Malt extract	15.0

### METHOD

- Dissolve 45 grams of ingredients in distilled water and adjust volume to 1000 ml.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down to ca. 45 °C – 50 °C.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

### Reference:

Tuite, J. 1969. Plant pathological methods - fungi and bacteria. Burgess publishing co., Minneapolis, MN. 293 pp.

### L1719 MALT AGAR, MA

L1719.0100	100 g
L1719.0500	500 g
L1719.1000	1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

**B1713****Bacteria Screening Medium 523**Medium: **General bacterial medium**Purpose: **Cultivation of bacteria****COMPOSITION OF MEDIA  
B1713: BACTERIA SCREENING  
MEDIUM 523**

COMPOUND	GRAM/LITER
Casein hydrolysate	8.0
Magnesium sulphate heptahydrate	0.15
Potassium phosphate monobasic	2.0
Yeast Extract	4.0
Sucrose	10.0
Agar	8.0

**METHOD**

- Dissolve 32.15 grams of ingredients in distilled water and adjust volume to 1000 ml.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down to ca. 45 °C – 50 °C.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

**Reference:**Viss, et al., *In Vitro Cell. Dev. Biol.*, 27P, **42** (1991)**B1713 BACTERIA SCREENING MEDIUM 523**

B 1713.0100	100 g
B 1713.0500	500 g
B 1713.1000	1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

# L1716

## Leifert and Waites Sterility Test Medium

Medium: **General bacterial medium**

Purpose: **Sterility test medium for bacteria**

In the Duchefa Biochemie's Leifert and Waites Sterility Test, Medium Beef extract 3.0 g/l has been replaced by 7,0 g/l Meat extract to obtain a more clear and stable medium.

### COMPOSITION OF MEDIA L1716: LEIFERT AND WAITES STERILITY TEST MEDIUM

COMPOUND	GRAM/LITER
Meat Extract	7.0
Glucose	5.0
MS medium + vitamins	2.2
Peptone	4.0
Sodium chloride	2.0
Sucrose	15.0
Yeast Extract	10.0

### METHOD

- Dissolve 45.2 grams of ingredients in distilled water and adjust volume to 1000 ml.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down to ca. 45 °C – 50 °C.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

### Reference:

Leifert, et al., J. Applied Bacteriology, 67, 353-361 (1989)

### L1716 LEIFERT AND WAITES STERILITY TEST MEDIUM

L 1716.0100	100 g
L 1716.0500	500 g
L 1716.1000	1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

**L1718****Luria Broth Agar, Miller**Medium: **General bacterial medium**Purpose: **Cultivation of bacteria****COMPOSITION OF MEDIA  
L1718: LURIA BROTH AGAR, MILLER**

COMPOUND	GRAM/LITER
Sodium chloride	0.5
Tryptone	10.0
Yeast Extract	5.0
Agar	15.0

**METHOD**

- Dissolve 30.5 grams of ingredients in distilled water and adjust volume to 1000 ml.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down to ca. 45 °C – 50 °C.
- Mix gently to avoid air bubbles and pour plates (15-20 ml per 9.0 cm plate).

**L1718 LURIA BROTH AGAR, MILLER**

L 1718.0100	100 g
L 1718.0500	500 g
L 1718.1000	1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie Tel : 040-2051615  
 Rooijakkersstraat 6 Fax : 040-2051395  
 5652 BB Eindhoven Email : info@tritium-microbiologie.nl  
 The Netherlands

**L1717****Luria Broth Base, Miller**Medium: **General bacterial medium**Purpose: **Cultivation of bacteria****COMPOSITION OF MEDIA  
L1717: LURIA BROTH BASE, MILLER**

COMPOUND	GRAM/LITER
Sodium chloride	0.5
Tryptone	10.0
Yeast Extract	5.0

**METHOD**

- Dissolve 16.5 grams of ingredients in distilled water and adjust volume to 1000 ml.
- Autoclave the solution (121 °C, 15 psi, 15 minutes).
- Allow medium to cool down.

**L1717 LURIA BROTH BASE, MILLER**

L 1717.0100	100 g
L 1717.0500	500 g
L 1717.1000	1 kg

For prepared and ready to use plates of this medium contact:  
 Tritium Microbiologie    Tel : 040-2051615  
 Rooijakkersstraat 6    Fax : 040-2051395  
 5652 BB Eindhoven    Email : info@tritium-microbiologie.nl  
 The Netherlands

Cat. nr.	Description of medium	Pathogen	ANTIBIOTICS (mg per liter medium)										
			Against gram positive, like Clavibacter						Against gram negative like Pseudomonas en Xanthomonas.				
			Bacitracin	Cephalexin monohydrate	Vancomycin HCl	Trimethoprim	Nalidixic acid	Neomycin sulphate	Polymixin B sulphate	Tobramycin sulphate	5-Fluorouracil	Cycloheximide	Nystatin
			B0106	C0110	V0155	T0154	N0134	M0135	P0145	T0153	F0123	C0176	N0138
K5120	KBBC	<i>Pseudomonas syringae</i> pv. <i>syringae</i> , pv. <i>porri</i> , pv. <i>pisii</i> , pv. <i>tomato</i>		80									35
M5167	MSP	<i>Pseudomonas savastanoi</i> pv. <i>phaseolicola</i> , <i>Pseudomonas syringae</i> pv. <i>syringae</i>		80	10								35
M5133	MT	<i>Pseudomonas syringae</i> pv. <i>syringae</i> , <i>Pseudomonas savastanoi</i> pv. <i>phaseolicola</i> , <i>Xanthomonas axonopodis</i> pv. <i>phaseoli</i>		80	10								35
X5121	mXCP1	<i>Xanthomonas axonopodis</i> pv. <i>phaseoli</i>		10						0.1	3		35
No antibiotics added													
P5135	PTSA	<i>Xanthomonas axonopodis</i> pv. <i>phaseoli</i>											
C5122	mCS20ABN	<i>Xanthomonas campestris</i> pv. <i>campestris</i> , <i>Xanthomonas campestris</i> pv. <i>armoraciae</i>	100					40					35
F5123	mFS	<i>Xanthomonas campestris</i> pv. <i>campestris</i> , <i>Xanthomonas campestris</i> pv. <i>armoraciae</i>		50		30							35
D5124	mD5A	<i>Xanthomonas campestris</i> pv. <i>carotae</i>	10	10									35
K5125	mKM	<i>Xanthomonas campestris</i> pv. <i>carotae</i>	50	10						2			35
T5132	mTBM	<i>Xanthomonas campestris</i> pv. <i>carotae</i>		65							12		20
P5134	PSM	<i>Pseudomonas syringae</i> pv. <i>porri</i>		80	10								35
S5130	SNAC	<i>Pseudomonas syringae</i> pv. <i>pisii</i>		80									35
T5126	mTMB	<i>Xanthomonas campestris</i> pv. <i>vesicatoria</i> , <i>Xanthomonas vesicatoria</i>		65						0.2	12	100	
M5131	MXV	<i>Xanthomonas campestris</i> pv. <i>vesicatoria</i> , <i>Xanthomonas vesicatoria</i>	100	32,5				10		0.2	6	100	
C5140	CKTM	<i>Xanthomonas campestris</i> pv. <i>vesicatoria</i>	100	65				10		4	12	100	
S5127	mSCM	<i>Clavibacter michiganensis</i> subsp. <i>michiganensis</i>					30					100	
D5128	DZANX	<i>Clavibacter michiganensis</i> subsp. <i>michiganensis</i>					28		10			100	
K5129	KBZ	<i>Pseudomonas syringae</i> pv. <i>tomato</i>		160								100	
K5165	mKB	Used for culturing <i>Pseudomonas</i>		50								100	35
K5165	KB	Used for culturing bacteria	No antibiotics added										
Y5136	YDC	Used for culturing bacteria like <i>Xanthomonas</i> anti <i>Clavibacteris</i>	No antibiotics added										
P1721	Potato Dextrose Agar, PDA	General fungal medium	No antibiotics added										
L1719	Malt Agar	General fungal medium	No antibiotics added										
P1722	Potato Dextrose Broth, PDB	General fungal medium	No antibiotics added										
C1715	CDA	General fungal and bacterial medium	No antibiotics added										
C1714	CDB	General fungal and bacterial medium	No antibiotics added										

## SUMMARY OF DUCHEFA GENERAL TERMS AND CONDITIONS OF SALE

### 1. Definitions and Scope

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Different arrangements with Duchefa agents or personnel and/or stated in purchase orders or letters, as well as any general terms and conditions of Customers are valid only if and to the extent that they have been accepted or confirmed by Duchefa in writing.

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### 8. Liability

Except in pursuance of the guarantee obligation as described in article 10, and in pursuance of peremptory law provisions, Duchefa is not liable for direct, indirect or consequential damage on the part of a Customer or third party resulting from the products supplied by Duchefa.

Duchefa is not liable for any damage a Customer might suffer as a result of the fact that the products the Customer bought from Duchefa prove not to be suitable for the use to which the Customer wishes to apply the products, unless the Customer has been expressly advised by Duchefa in writing in this regard

Duchefa is not liable for damage caused by the actions or omissions of Customers themselves or by persons appointed by Customers or for whom Customers are otherwise responsible.

Duchefa is not liable for damage that might occur to Customers themselves, or to persons appointed by Customers or for whom Customers are otherwise responsible, as a result of the fact that Customers, or persons appointed by Customers or for whom Customers are otherwise responsible, when applying and/or processing the products supplied by Duchefa fail to observe the legal regulations and/or the directions for use and/or the packaging directions in force, as found in product specifications, Material Safety Data Sheets (MSDS's), catalogues, lists, measurements, weights and the like.

Duchefa is not liable for damage that is the result of Customers furnishing incorrect or incomplete information or materials. Extra work Duchefa has to perform and extra expenses Duchefa has to incur as a result of such actions or omissions on the part of Customers can be charged to them at the Duchefa hourly rates then in force.

Any liability on the part of Duchefa for damage resulting from work performed by third parties on the products supplied by Duchefa, or as a result of which the proper operation of the products supplied by Duchefa is affected is expressly excluded.

Any liability of Duchefa resulting from an imputable shortcoming on the part of Duchefa shall be limited at all times to at most the net invoice value of the products supplied by Duchefa, save in the event of willful intent or gross negligence on the part of Duchefa.

Claims for damages have to be reported to Duchefa by registered mail within eight days of the damage occurring, or of the date on which a Customer became aware of the damage, as the case may be, failing which Duchefa can no longer assume liability for this damage.

### 9. Complaints

Customers are required to inspect the products supplied by Duchefa immediately after they receive them. Any complaints have to be reported to Duchefa in writing by registered mail, giving a detailed description of the nature and the grounds for the complaint, within eight days of the products being received or the work or services being rendered, as the case may be. Once this term has expired, Customers are deemed to have approved the goods, work or services, and will have forfeited any right (including that of defence) in this respect. If after the term has expired, Duchefa wishes on the basis of leniency to investigate the correctness of the complaint, this investigation and/or the work flowing from it can never result in any liability on the part of Duchefa.

In the event of a complaint, Duchefa will do all in its power to review the complaint within a reasonable time and to remedy the complaint where necessary. Customers are required at all times to give Duchefa the opportunity to examine the correctness of the complaint.

In the event that the objections of Customers are found by Duchefa to be justified, Duchefa has the right, at its discretion, to substitute products of the same kind, to apply the necessary improvements, or to apply a reasonable reduction in the price.

Customers do not have the right to claim dissolution, annulment of the contract or damages. Customers are not entitled on the grounds of the complaint relating to a specific product or a specific service to delay payment or refuse payment of other products or services on which the complaint does not have any bearing.

No matter what the reason, goods sold to customers can be returned to Duchefa only after prior written authorization and shipment and other instructions from Duchefa. Customers are required to observe strictly the directions concerning the storage and handling of the products supplied. Storage, freight and all related expenses are for the account and risk of Customers. The products supplied by Duchefa may only be returned for the account and risk of Duchefa after its express written permission.

### 10. Guarantee

Communications by or on behalf of Duchefa on the quality, the composition, the handling (in the broadest sense of the word as well as presented in the Material Safety Data Sheets (MSDS's)), application possibilities, properties and the like of the products supplied by Duchefa do not bind Duchefa unless these communications are made expressly, in the form of a written guarantee.

Any claim under a guarantee lapses if the products of Duchefa are not kept and/or stored in accordance with the stipulations that apply to the safekeeping of such products.

Any guarantee obligation lapses if Customers themselves make modifications or repairs to the products supplied by Duchefa or have these modifications and repairs made by third parties, or if the products supplied are not used or applied in accordance with the (legal) regulations and/or intended purpose, or if the products supplied are and/or have been improperly handled (in violation with amongst others Material Safety Data Sheet (MSDS's) requirements) or maintained in any other manner.

## 11. Retention of title

All products sold and supplied, even if the transaction was C.O.D., remain the property of Duchefa until the amounts a Customer owes Duchefa in this respect have been settled in full, including the collection costs and interest forming part of these amounts owed.

Customers are not entitled to transfer title to the products to third parties, whether or not for purposes of collateral security, unless they acquired title to the products by accession in pursuance of section 14, Book 5 of the Netherlands Civil Code, by confusion in pursuance of section 16, Book 5 of the Netherlands Civil Code, or by specification in pursuance of section 16, Book 5 of the Netherlands Civil Code. Customers nevertheless have the power of disposition over the products in order to process or treat them, or to resell them in the context of their normal business activities.

For as long as title to the products supplied by Duchefa has not been passed to a Customer, the Customer is obliged to insure these products for an adequate amount and at the customary conditions, and to agree in this respect that Duchefa is named as the insured. Any damage compensation claimable from the insurer concerning goods that belong to Duchefa, the Customer hereby passes on to Duchefa.

## 13. Prices

Prices are in EUR (€) and exclude VAT. Packaging expenses, packing, transport and insurance if any are not included. Work in excess of the work contractually agreed and increases in volume are quoted separately.

**13.2** Duchefa is entitled to charge the Customer in full for any price increases occurring between the time the proposal is issued or until the contract is concluded and the time of supply. Cost increases include: increase in freight rates, taxes, import and export duties or other levies, increase in wages and social security charges, currency fluctuations, and increase in raw material and energy prices.

In order to meet all restrictions and regulations which govern national and international transport of chemical products, Duchefa tries to ship all orders without delay while minimizing costs of delivery within these regulatory guidelines.

All orders with a destination within the European Union (E.U.) and a value of 275,- EUR (€) or more, are supplied Delivered Duty Paid (DDP). All orders with a destination within the E.U. and a value of less than 275,- EUR (€) are surcharged with an extra 17,50 EUR (€) for delivery. Transportation charges will vary with the destination, weight, and content of each shipment.

All orders with a destination outside the E.U. are shipped Ex Works (EXW). Transportation charges will vary with the destination, weight, and content of each shipment will be subcharged accordingly on the corresponding invoice.

All orders for hazardous chemicals will incur separate hazardous air freight charges. Special packaging may be necessary for safe delivery of certain hazardous chemicals. Separate special packaging charges will vary with hazardous product properties, weight, volume and destination. These extra hazardous good transport charges will be added to your invoice.

All freight charges, administrative costs and special packaging charges are available upon request at order entry and are indicated on our invoices.

## 14. Payment

Payments by Customers shall be made within 30 days of the invoice date, unless agreed otherwise. Payment shall be in EUR (€) to Duchefa at a Dutch bank in the Netherlands.

Any reliance of Customers upon set-off or suspension shall be excluded.

Customers who fail to pay promptly shall be deemed to be in default without any notice or judicial intervention to this effect. In that event, Customers shall be charged the higher of 1% and the statutory rate of interest per month on the

amount owing. Moreover, without prejudice to the further rights accruing to Duchefa under the law or the contract, in the event of Customers failing to pay promptly, Duchefa shall, at its discretion, be entitled either to suspend further supplies or dissolve the contract without any judicial intervention and to repossess either directly or indirectly, at the expense of Customers, all the products Duchefa supplied to them or all the products for which they failed to pay.

Where a Customer has exceeded the payment term, the Customer shall pay Duchefa any collection charges, whether incurred in or out of court, including the expense claims submitted by the adviser or advisers appointed by Duchefa for the collection. The out-of-court collection charges shall amount to at least 15% of the total amount the Customer owes Duchefa, subject to a minimum of EUR (€) 150,- excluding VAT.

Every payment by the Customer shall first be applied to the interest owing, then to the expenses incurred on the collection of the amount owing, and finally to the principal.

Complaints concerning invoices have to be reported to Duchefa in writing within eight days of the date of the invoice, failing which Customers shall be deemed to have accepted the invoice as being correct.

In the event of delivery in the interim, Duchefa is entitled to send an invoice for the work in question, which invoice has to be settled in accordance with the provisions laid down in these General Terms and Conditions. Failure on the part of the Customer to pay promptly shall entitle Duchefa to suspend any further work for the Customer.

## 23. Applicable law

Only Dutch law shall apply to these General Terms and Conditions, to all contracts and to all agreements stemming from them, to which these General Terms and Conditions apply in full or in part. Part 3, Title 4, Book 6 of the Netherlands Civil Code is declared explicitly applicable.

## 24. Adjudication of disputes

All disputes between the parties, arising from the contract(s) of sale entered into between them, which cannot be resolved through consultation between the parties, shall be submitted exclusively to the court of jurisdiction in Haarlem, the Netherlands, being the court in the district in which Duchefa is established, unless Duchefa opts to bring the dispute before another court.

## 25. Translations

In the event of any differences in meaning or interpretation, as the case may be, between the Dutch-language text of these General Terms and Conditions and translations thereof, the Dutch-language text prevails.

Filed at the Office of the District Court at Haarlem, The Netherlands, on June 2006 under number: 15/2006

**UPON REQUEST WE WILL SEND THE COMPLETE GENERAL TERMS AND CONDITIONS OF SALE.**

## INDICATION OF PARTICULAR RISKS

<b>R:1</b>	Explosive when dry
<b>R:2</b>	Risk of explosion by shock, friction, fire or other sources of ignition
<b>R:3</b>	Extreme risk of explosion by shock, friction, fire or other sources of ignition
<b>R:4</b>	Forms very sensitive explosive metallic compounds
<b>R:5</b>	Heating may cause an explosion
<b>R:6</b>	Explosive with or without cont. with air
<b>R:7</b>	May cause fire
<b>R:8</b>	Cont. with combust. mat. may cause fire
<b>R:9</b>	Explos. when mixed with combustible mat.
<b>R:10</b>	Flammable
<b>R:11</b>	Highly flammable
<b>R:12</b>	Extremely flammable
<b>R:14</b>	Reacts violently with water
<b>R:15</b>	Contact with water liberates extremely flammable gases
<b>R:16</b>	Explosive when mixed with oxidizing substances
<b>R:17</b>	Spontaneously flammable in air
<b>R:18</b>	In use, may form flammable/explosive vapour-air mixture
<b>R:19</b>	May form explosive peroxides
<b>R:20</b>	Harmful by inhalation
<b>R:21</b>	Harmful in contact with skin
<b>R:22</b>	Harmful if swallowed
<b>R:23</b>	Toxic by inhalation
<b>R:24</b>	Toxic in contact with skin
<b>R:25</b>	Toxic if swallowed
<b>R:26</b>	Very toxic by inhalation
<b>R:27</b>	Very toxic in contact with skin
<b>R:28</b>	Very toxic if swallowed
<b>R:29</b>	Contact with water liberates toxic gas
<b>R:30</b>	Can become highly flammable in use
<b>R:31</b>	Contact with acids liberates toxic gas
<b>R:32</b>	Contact with acids liberates very toxic gas
<b>R:33</b>	Danger of cumulative effects
<b>R:34</b>	Causes burns
<b>R:35</b>	Causes severe burns
<b>R:36</b>	Irritating to eyes
<b>R:37</b>	Irritating to respiratory system
<b>R:38</b>	Irritating to skin
<b>R:39</b>	Danger of very serious irreversible effects
<b>R:40</b>	Possible risk of irreversible effects
<b>R:41</b>	Risk of serious damage to eyes
<b>R:42</b>	May cause sensitization by inhalation
<b>R:43</b>	May cause sensitization by skin contact
<b>R:44</b>	Risk of explosion if heated under confinement
<b>R:45</b>	May cause cancer
<b>R:46</b>	May cause heritable genetic damage
<b>R:48</b>	Danger of serious damage to health by prolonged exposure
<b>R:49</b>	May cause cancer by inhalation
<b>R:50</b>	Very toxic to aquatic organisms
<b>R:51</b>	Toxic to aquatic organisms
<b>R:52</b>	Harmful to aquatic organisms
<b>R:53</b>	May cause long-term adverse effects in the aquatic environment
<b>R:54</b>	Toxic to flora
<b>R:55</b>	Toxic to fauna
<b>R:56</b>	Toxic to soil organisms
<b>R:57</b>	Toxic to bees
<b>R:58</b>	May cause long-term adverse effects in the environment
<b>R:59</b>	Dangerous for the ozone layer
<b>R:60</b>	May impair fertility
<b>R:61</b>	May cause harm to the unborn child
<b>R:62</b>	Possible risk of impaired fertility

<b>R:63</b>	Possible risk of harm to the unborn child
<b>R:64</b>	May cause harm to breast-fed babies
<b>R:65</b>	harmful: may cause lung-damage if swallowed

## COMBINATION OF PARTICULAR RISKS

<b>R:14/15</b>	Reacts violently with water, liberating extremely flammable gases
<b>R:15/29</b>	Contact with water liberates toxic, extremely flammable gas
<b>R:20/21</b>	Harmful by inhalation and in contact with skin
<b>R:20/21/22</b>	Harmful by inhalation, in contact with skin and if swallowed
<b>R:20/22</b>	Harmful by inhalation and if swallowed
<b>R:21/22</b>	Harmful in contact with skin and if swallowed.
<b>R:23/24</b>	Toxic by inhalation and in cont. with skin
<b>R:23/24/25</b>	Toxic by inhalation, in contact with skin and if swallowed
<b>R:23/25</b>	Toxic by inhalation and if swallowed
<b>R:24/25</b>	Toxic in contact with skin and if swallowed.
<b>R:26/27</b>	Very toxic by inhalation and in cont. with skin
<b>R:26/27/28</b>	Very toxic by inhalation, in contact with skin and if swallowed
<b>R:26/28</b>	Very toxic by inhalation and if swallowed.
<b>R:27/28</b>	Very toxic in cont. with skin and if swallowed.
<b>R:36/37</b>	Irritating to eyes and respiratory system
<b>R:36/37/38</b>	Irritating to eyes, respiratory system and skin
<b>R:36/38</b>	Irritating to eyes and skin
<b>R:37/38</b>	Irritating to respiratory system and skin
<b>R:39/23</b>	Toxic: danger of very serious irreversible effects through inhalation
<b>R:39/23/24</b>	Toxic: danger of very serious irreversible effects through inhalation and in contact with skin
<b>R:39/23/24/25</b>	Toxic: danger of very serious irreversible effects through inhalation, in contact with skin and if swallowed
<b>R:39/23/25</b>	Toxic: danger of very serious irreversible effects through inhalation and if swallowed
<b>R:39/24</b>	Toxic: danger of very serious irreversible effects in contact with skin
<b>R:39/24/25</b>	Toxic: danger of very serious irreversible effects in contact with skin and if swal.
<b>R:39/25</b>	Toxic: danger of very serious irreversible effects if swallowed
<b>R:39/26</b>	Very toxic: danger of very serious irrevers. effects through inhalation
<b>R:39/26/27</b>	Very toxic: danger of very serious irreversible effects through inhalation and in contact with skin
<b>R:39/26/27/28</b>	Very toxic: danger of very serious irreversible effects through inhalation, in contact with skin and if swallowed
<b>R:39/26/28</b>	Very toxic: danger of very serious irreversible effects through inhalation and if swallowed
<b>R:39/27</b>	Very toxic: danger of very serious irreversible effects in contact with skin
<b>R:39/27/28</b>	Very toxic: danger of very serious irreversible effects in contact with skin and if swallowed
<b>R:39/28</b>	Very toxic: danger of very serious irreversible effects if swallowed
<b>R:40/20</b>	Harmful: possible risk of irreversible effects through inhalation
<b>R:40/20/21</b>	Harmful: possible risk of irreversible effects through inhalation and in contact with skin
<b>R:40/20/21/22</b>	Harmful: possible risk of irreversible effects through inhalation, in contact with skin and if swallowed
<b>R:40/20/22</b>	Harmful: possible risk of irrevers. effects through inhalation and if swallowed.
<b>R:40/21</b>	Harmful: possible risk of irreversible effects in contact with skin
<b>R:40/21/22</b>	Harmful: possible risk of irreversible effects in contact with skin and if swallowed
<b>R:40/22</b>	Harmful: possible risk of irreversible effects if swallowed
<b>R:42/43</b>	May cause sensitization by inhalation and skin contact
<b>R:48/20</b>	Harmful: danger of serious damage to health by prolonged exposure through inhalation

- R:48/20/21** Harmful: danger of serious damage to health by prolonged exposure through inhalation and in contact with skin
- R:48/20/21/22** Harmful: danger of serious damage to health by prolonged exposure through inhalation, in contact with skin and if swallowed
- R:48/20/22** Harmful: danger of serious damage to health by prolonged exposure through inhalation and if swallowed
- R:48/21** Harmful: danger of serious damage to health by prolonged exposure in contact with skin
- R:48/21/22** Harmful: danger of serious damage to health by prolonged exposure in contact with skin and if swallowed
- R:48/22** Harmful: danger of serious damage to health by prolonged exposure if swal.
- R:48/23** Toxic: danger of serious damage to health by prolonged exposure through inhalation
- R:48/23/24** Toxic: danger of serious damage to health by prolonged exposure through inhalation and in contact with skin
- R:48/23/24/25** Toxic: danger of serious damage to health by prolonged exposure through inhalation, in contact with skin and if swallowed
- R:48/23/25** Toxic: danger of serious damage to health by prolonged exposure through inhalation and if swallowed
- R:48/24** Toxic: danger of serious damage to health by prolonged exposure in contact with skin
- R:48/24/25** Toxic: danger of serious damage to health by prolonged exposure in contact with skin and if swallowed
- R:48/25** Toxic: danger of serious damage to health by prolonged exposure if swal.
- R:50/53** Very toxic to aquatic organisms, may cause long-term adverse effects in the aquatic environment
- R:51/53** Toxic to aquatic organisms, may cause long-term adverse effects in the aquatic environment
- R:52/53** Harmful to aquatic organisms, may cause long-term adverse effects in the aquatic environment

## INDICATION OF SAFETY PRECAUTIONS REQUIRED

- S:1** Keep locked up
- S:2** Keep out of the reach of children
- S:3** Keep in a cool place
- S:4** Keep away from living quarters
- S:5** Keep contents under...(appror. liquid to be specified by the manuf.)
- S:6** Keep under... (inert gas to be specified by the manufacturer)
- S:7** Keep container tightly closed
- S:8** Keep container dry
- S:9** Keep container in a well ventilated place
- S:12** Do not keep the container sealed
- S:13** Keep away from food, drink and animal feeding stuffs
- S:14** Keep away from...(incomp. mater. to be indicated by the manufacturer)
- S:15** Keep away from heat
- S:16** Keep away from sources of ignition- No Smoking
- S:17** Keep away from combustible material
- S:18** Handle and open container with care
- S:20** When using do not eat or drink
- S:21** When using do not smoke
- S:22** Do not breathe dust
- S:23** Do not breathe gas/fumes/vapour/spray (appropriate wording to be specified by the manufacturer)
- S:24** Avoid contact with skin
- S:25** Avoid contact with eyes
- S:26** In case of contact with eyes, rinse immediately with plenty of water and seek medical advise
- S:27** Take off immediately all contaminated clothing

- S:28** After contact with skin, wash immediately with plenty of (to be specified by the manufacturer)
- S:29** Do not empty into drains
- S:30** Never add water to this product
- S:33** Take precautionary measures against static discharges
- S:35** This material and its container must be disposed of in a safe way
- S:36** Wear suitable protective clothing
- S:37** Wear suitable gloves
- S:38** In case of insufficient ventilation, wear suitable respiratory equipment
- S:39** Wear eye/face protection
- S:40** To clean the floor and all objects contaminated by this material use ...(to be specified by the manufacturer)
- S:41** In case of fire and/or explosion do not breathe fumes
- S:42** During fumigation/spraying wear suitable respiratory equipment (appropriate wording to be specified)
- S:43** In case of fire, use ... (indicate in the space the precise type of fire-fighting equipment. If water increases the risk, add -"Never use water")
- S:45** In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible)
- S:46** If swallow. seek medic. advice immed. and show this container or label
- S:47** Keep at temperature not exceeding ...C (to be specified by the manuf.)
- S:48** Keep wet with ... (appropriate material to be specified by the manuf.)
- S:49** Keep only in the original container
- S:50** Do not mix with ... (to be specified by the manufacturer)
- S:51** Use only in well-ventilated areas
- S:52** Not recommended for interior use on large surface areas
- S:53** Avoid exposure - obtain special instructions before use
- S:56** Disp. of this mat. and its container at hazard or special waste collect. point
- S:57** Use appropriate container to avoid environmental contamination
- S:59** Refer to manufacturer/supplier for information on recovery/recycling
- S:60** This material and its container must be disposed of as hazardous.waste
- S:61** Avoid release to the envir. Refer to special instruct./safety data sheet
- S:62** If swallowed, do not induce vomiting: seek medical advice immediately and show this container or label

## COMBINATION OF SAFETY PRECAUTIONS REQUIRED

- S:1/2** Keep locked up and out of the reach of children
- S:3/7** Keep container tightly closed in a cool place
- S:3/9/14** Keep in a cool, well-ventilated place away from ... (incompatible materials to be indicated by the manufacturer)
- S:3/9/14/49** Keep only in the original container in a cool, well ventilated place away from ...(incompat. materials to be indicated by the manufacturer)
- S:3/9/49** Keep only in the original container in a cool, well ventilated place
- S:3/14** Keep in a cool place away from ... (incompatible materials to be indicated by the manufacturer)
- S:7/8** Keep container tightly closed and dry
- S:7/9** Keep container tightly closed and in a well-ventilated place
- S:7/47** Keep container tightly closed and at a temperature not exceeding ...C (to be specified by the manufacturer)
- S:20/21** When using do not eat, drink or smoke
- S:24/25** Avoid contact with skin and eyes
- S:29/56** Do not empty into drains, dispose of this material and its container at hazardous or special waste collection point
- S:36/37** Wear suitable protective clothing and gloves
- S:36/37/39** Wear suitable protective clothing, gloves and eye/face protection
- S:36/39** Wear suitable protective clothing and eye/face protection
- S:37/39** Wear suitable gloves and eye/face protection
- S:47/49** Keep only in the original container at a temperature not exceeding ...C (to be specified by the manufacturer)

# Index (alphabetically)

Product name	Cat. no.	CAS no.	Page	Product name	Cat. no.	CAS no.	Page
1-Naphtylphosphate sodium monohydrate	N1350	81012-89-7	117	Cupric sulphate pentahydrate	C0508	7758-99-8	95
2,3,5-Triiodobenzoic acid	T0929	88-82-4	135	Cyanocobalamin	C0726	68-19-9	96
2,4,5-Trichlorophenoxyacetic acid	T0915	37785-57-2	135	Cycloheximide	C0176	66-81-9	96
2,4-Dichlorophenoxyacetic acid (2,4 D)	D0911	94-75-7	97	Czapek Dox Agar, CDA	C1715		174
24-Epibrassinolide	E0940	78821-43-9	99	Czapek Dox Broth, CDB	C1714		175
2-IP	D0906	2365-40-4	97	D(+)-Pantothenate calcium	C0604	137-08-6	120
2-iP riboside	D0934	7724-76-7	98	D(+)-Biotine	B0603	58-85-5	85
2-Nitrophenyl-β-D-galactopyranoside	O1409	369-07-3	118	D2ANX medium	D5128		170
4-CPPU	C0943	68157-60-8	95	Daishin agar	D1004	9002-18-0	78
5-Fluoro orotic acid (5-FOA)	F0176	703-95-7	101	D-Cycloserine	C0119	68-41-7	96
5-Fluoro uracil (5-FU)	F0123	51-21-8	101	De Greef and Jacobs medium	D0205		38
6-Benzylaminopurine (6-BAP)	B0904	1214-39-7	84	De Greef and Jacobs medium including vitamins	D0206		38
6-Benzylaminopurine riboside	B0930	4294-16-0	84	De Wit <sup>®</sup> Tray 10 pcs.	T1608		142
6-Mercaptopurine monohydrate	M0129	6112-76-1	113	Dextran sulphate sodium	D1342	9011-18-1	96
8-Hydroxyquinoline	H0168	148-24-3	106	D-Fructose	F0801	57-48-7	102
Abisic acid (S-ABA)	A0941	21293-29-8	77	D-Galactose	G0810	59-23-4	103
Acetylsalicylic acid	A1366	50-78-2	77	D-Glucose monohydrate	G0802	10/15996	104
Acyclovir	A0183	59277-89-3	77	Dicamba	D0920	1918-00-9	97
Adenine hemisulphate dihydrate	A0908	321-30-2	77	Dihydrozeatin (DHZ)	D0933	14894-18-9	97
Adenosine	A1334	58-61-7	77	Dimethylsulfoxide (DMSO)	D1370	67-68-5	98
Adenosine-5-triphosphate	A1335	987-65-5	78	di-Potassium hydrogen phosphate	P0573	11/4/7758	124
Agarose SPI	A1203	9012-36-6	79	Di-Sodium hydrogen phosphate dihydrate	S0537	10028-24-7	128
Aluminium chloride hexahydrate	A0532	7784-13-6	80	Dithioerythritol (DTE)	D1308	6892-68-8	98
Amiprofos methyl	A0185	36001-88-4	80	Dithiothreitol (DTT)	D1309	12/3/3483	98
Ammonium chloride	A0528	12125-02-9	80	DKW/Juglans medium	D0246		39
Ammonium dihydrogen phosphate	A1338	7722-76-1	80	DKW/Juglans medium including vitamins	D0247		39
Ammonium nitrate	A0501	6484-52-2	81	DKW/Juglans vitamin mixture	D0414		39
Ammonium sulphate	A0502	7783-20-2	81	D-Luciferin	L1349	2591-17-5	110
Amoxicillin sodium / Clavulanate potassium	A0189		81	D-Mannitol	M0803	69-65-8	112
Amoxicillin trihydrate	A0101	61336-70-7	81	D-Mannose	M1392	3458-28-4	113
Amphotericin B	A0103	1397-89-3	82	Doxorubicin HCl 0.2% in 0.9% NaCl solution (5ml)	D0120	25316-40-9	99
Amphotericin B suspension	A0192	1397-89-3	82	Doxycycline HCl	D0121	10592-13-9	99
Ampicillin sodium	A0104	69-52-3	82	D-Ribose	R0806	50-69-1	126
a-naphtalene acetic acid	N0903	86-87-3	116	D-Sorbitol	S0807	50-70-4	129
Anderson's Rhododendron	A0201		34	D-Xylose	X0808	58-86-6	137
Anderson's Rhododendron including vitamins	A0202		34	ECO2 box green filter (oval model 80mm H)	E1654		143
Apramycin sulphate	A0164	65710-07-8	82	ECO2 box white filter (oval model 80mm H)	E1650		143
Atrazine	A0156	1912-24-9	83	EDTA disodium dihydrate	E0511	6381-92-6	100
Bacitracin	B0106	1405-87-4	84	Ergonomic Scalp Handle	S3110		140
Bacteria Screening medium 523	B1713		177	Eriksson (ER) medium	E0207		40
Banana powder	B1304		84	Eriksson (ER) medium including vitamins	E0208		40
BES	B1514	10191-18-1	85	Eriksson (ER) vitamin mixture	E0402		40
Bis-Tris buffer grade	B1516	6976-37-0	85	Erythromycin	E0122	114-07-8	100
Bleomycin sulphate	B0107	9041-93-4	86	Esculin	E1343	531-75-9	100
Blue-Gal	B1414	97753-82-7	88	Fe-EDDHA	F0527		100
b-Naphtoxyacetic acid	N0912	120-23-0	116	FeNaEDTA	E0509	15708-41-5	100
Boric acid	B0503	10043-35-3	86	Ferrous sulphate heptahydrate	F0512	7782-63-0	100
Bromoxynil	B0157	1689-84-5	88	Fluridon	F0919	59756-60-4	101
Calcium carbonate	C0529	471-34-1	88	Flurprimidol	F0935	56425-91-3	102
Calcium chloride dihydrate	C0504	10035-04-8	88	Folic acid	F0608	59-30-3	102
Calcium citrate tetrahydrate	C0530	5785-44-4	89	Folinic acid calcium pentahydrate (old C0607)	F0619	41927-89-3	102
Calcium gluconate monohydrate	C0531	299-28-5	89	Forceps extended, 30 cm	F3003		140
Calcium nitrate tetrahydrate	C0505	13477-34-4	89	Forceps, 23 cm	F3001		140
Calcium phosphate tribasic	C0506	7758-87-4	89	G-418 disulphate	G0175	108321-42-2	102
Carbenicillin disodium	C0109	4800-94-6	90	Gamborg B5 medium	G0209		41
Carboxin	C0160	5234-68-4	90	Gamborg B5 medium including vitamins	G0210		41
Carrageenan Iota Type	C1006	7/1/9062	89	Gamborg B5 vitamin mixture	G0415		41
Casein hydrolysate	C1301	9000-71-9	90	Gelcarin GP- 812	G1007	7/1/9000	89
Cefotaxime sodium	C0111	64485-93-4	90	Gelrite™	G1101	71010-52-1	103
Cellulase R-10	C8001	9012-54-8	91	Gentamycin sulphate	G0124	1405-41-0	103
Cellulase RS	C8003	9012-54-8	91	Gibberellic acid 4+7	G0938	468-44-0/510-75-8	104
Cephalexin monohydrate	C0110	15686-71-2	90	Gibberellic acid A3	G0907	77-06-5	104
Cetrimide	C1397	8044-71-1	92	Glass Bead Sterilizer (incl. Glass beads)	G3301		141
Cetrimonium	C1393	57-09-0	92	Glass Beads for Sterilizer	G3302		141
CHAPS	C1374	75621-03-3	92	Glutathione reduced	G1346	70-18-8	105
Charcoal activated	C1302	64365-11-3	92	Glycerol	G1345	56-81-5	105
CHÉE & POOL basal salt medium	C0248		35	Glycine	G0709	56-40-6	105
CHÉE & POOL medium including vitamins	C0249		35	Glyphosate	G0158	1071-83-6	105
Chloramphenicol	C0113	56-75-7	92	Gresshof & Doy (DBM2) vitamin mixture	G0404		42
Chlorhexidine digluconate	C0114	18472-51-0	93	Gresshoff & Doy medium	G0211		42
Chlorhexidine HCl	C0115	3697-42-5	93	Gresshoff & Doy medium including vitamins	G0212		42
Chloroxylenol ("Dettol")	D0161	88-04-0	94	Griseofulvin	G0167	126-07-8	105
Chlorsulfuron	C0177	64902-72-3	94	Guanidine HCl	G1375	50-01-1	106
Chlortetracycline HCl	C0116	64-72-2	94	Heller medium	H0213		43
Choline chloride	C0605	67-48-1	94	HEPES	H1504	7365-45-9	106
CHU (N6) Medium	C0203		36	Hygromycine B	H0192	31282-04-9	107
CHU (N6) Medium including vitamins	C0204		36	Indole-3-acetic acid (IAA)	I0901	87-51-4	107
CHU vitamine mixture	C0401		36	Indole-3-butyric acid (IBA)	I0902	133-32-4	107
Citric acid monohydrate	C1303	5949-29-1	94	IPTG	I1401	367-93-1	108
CKTM medium	C5140		168	Jasmonic acid	J0936	6894-38-8	108
CLC / Ipomoea CP medium	C0228		37	Kanamycin sulphate monohydrate	K0126	25389-94-0	108
CLC / Ipomoea EP medium	C0229		37	KAO & Michayluk medium	K0214		44
Clindamycin HCl	C0117	21462-39-5	95	KB medium (King's B medium)	K5165		172
Cobalt chloride hexahydrate	C0507	7791-13-1	95	KBBC medium	K5120		154
Colchicine	C1305	64-86-8	95	KBZ medium	K5129		171
Colistin sulphate	C0118	1264-72-8	95	Kinetin	K0905	525-79-1	109
Culture tubes "De Wit" (W1614 bottom / W1615 top)	W1607		142	Knudson C Orchid medium	K0215		45

Product name	Cat. no.	CAS no.	Page	Product name	Cat. no.	CAS no.	Page
Lactose monohydrate	L1372	10039-26-6	109	Murashige & Skoog medium mod. No. 1A	M0232		55
L-Alanine	A0703	56-41-7	80	Murashige & Skoog medium mod. No. 1B	M0233		55
L-Arginine	A0704	74-79-3	83	Murashige & Skoog medium mod. No. 2A	M0234		56
L-Ascorbic acid	A0602	50-81-7	83	Murashige & Skoog medium mod. No. 2B	M0235		56
L-Asparagine monohydrate	A0725	5794-13-8	83	Murashige & Skoog medium mod. No. 3A	M0236		57
L-Aspartic acid	A0705	56-84-8	83	Murashige & Skoog medium mod. No. 3B	M0237		57
LB Agar High salt	L1706		109	Murashige & Skoog medium mod. No. 4	M0238		58
LB Agar Low salt	L1705		109	Murashige & Skoog medium mod. No. 5	M0239		58
LB Broth High salt	L1704		109	Murashige & Skoog medium shoot multiplication	BM0244		62
LB Broth Low salt	L1703		109	Murashige & Skoog medium vitamin mixture	M0409		50
L-Cysteine HCl monohydrate	C0706	4/6/7048	96	mXCP1 medium	X5121		157
LED Lighting			148	MXV medium	M5131		167
Leifert and Waites sterility test medium	L1716		178	Myo-Inositol	I0609	87-89-8	107
Leucopore tape, 1.25 cm x 9.2 m	L3302		142	Nalidixic acid	N0134	389-08-2	116
Leucopore tape, 2.5 cm x 9.2 m	L3301		142	Naphthylphthalamic acid	N0926	132-66-1	117
L-Glutamic acid	G0707	56-86-0	104	N-Benzyl-9-(tetrahydropyranyl)-adenine (BPA)	B0932	2312-73-4	85
L-Glutamine	G0708	56-85-9	104	Neomycin sulphate	M0135	1405-10-3	117
L-Histidine	H0710	71-00-1	106	Nicotinamide	N0610	98-92-0	117
Lincomycin HCl monohydrate	L0127	7179-49-9	110	Nicotinic acid	N0611	59-67-6	118
Lindemann Orchid medium	L0216		46	Nitro Blue Tetrazolium (NBT)	N1411	38184-50-8	118
Linsmaier & Skoog medium	L0230		47	Nitsch medium	N0223		63
Linsmaier & Skoog vitamin mixture	L0406		47	Nitsch medium including vitamins	N0224		63
L-Isoleucine	I0711	73-32-5	108	Nitsch vitamin mixture	N0410		63
Litvay medium	L0217		48	NLN medium	N0252		64
Litvay medium including vitamins	L0218		48	NLN medium vitamin mixture	N0253		64
Litvay vitamin mixture	L0407		48	Nystatine	N0138	1400-61-9	119
L-Leucine	L0712	61-90-5	110	Orchimax including activated charcoal	O0262		65
L-Lysine HCl	L0714	657-27-2	110	Orchimax without activated charcoal	O0257		65
L-Methionine	M0715	63-68-3	113	Oryzoline	O1318	19044-88-3	119
L-Ornithine HCl	O1351	3184-13-2	119	OS140 box green filter (round model 140mm H)	E1674		143
Low Melting Agarose PPC	L1204	9012-36-6	79	Oxytetracycline HCl	O0140	2058-46-0	119
L-Phenylalanine	P0716	63-91-2	121	Paclotubrazol	P0922	76738-62-0	120
L-Proline	P0717	147-85-3	125	p-Aminobenzoic acid	A0601	150-13-0	80
L-Serine	S0718	56-45-1	127	Paper Cutting Pad	P3202		141
L-Threonine	T0719	72-19-5	133	Paromomycin sulphate	P0141	1263-89-4	120
L-Tryptophan	T0720	73-22-3	136	p-Chlorophenoxyacetic acid (4-CPA)	C0909	122-88-3	94
L-Tyrosine	T0721	60-18-4	137	Pectolyase Y-23	P8004	9033-35-6	120
Luria Broth Agar, Miller	L1718		179	Penicillin G sodium	P0142	69-57-8	121
Luria Broth Base, Miller	L1717		180	Peptone	P1328	73049-73-7	121
L-Valine	V0722	72-18-4	137	Peptone water	P1707	73049-73-7	121
Macerozyme R-10	M8002	9032-75-1	111	Peptone water, buffered	B1702	73049-73-7	121
Macro salt mixture B5	M0304		73	Phleomycin	P0187	11006-33-0	122
Macro salt mixture MS	M0305		74	Phloroglucinol	P1353	108-73-6	122
Macro salt mixture Nitsch	M0306		74	Phosphinotricin	P0159	77182-82-2	122
Magenta-GlcA cyclohexylammonium salt	M1412	144110-43-0	87	Phyto agar	P1003	9002-18-0	78
Magnesium chloride hexahydrate	M0533	7791-18-6	111	Picloram	P0914	2/1/1918	122
Magnesium sulphate heptahydrate	M0513	10034-99-8	111	PIPES	P1505	5625-37-6	123
Maleic hydrazide	M0921	123-33-1	111	Plant agar	P1001	9002-18-0	78
Malic acid	M1315	617-48-1	111	Plant Propagation by Tissue Culture	P5001		75
Malt Agar (MA)	L1719		176	p-Nitrophenyl-b-D-glucuronide	N1408	10344-94-2	118
Malt extract	M1327	8002-48-0	112	Polyethylene Glycol 400	P0813	25322-68-3	123
Maltose monohydrate	M0811	6363-53-7	112	Polyethylene Glycol 4000	P0804	25322-68-3	123
Manganese sulphate monohydrate	M0514	10034-96-5	112	Polyethylene Glycol 6000	P0805	25322-68-3	123
McCown Woody Plant medium	M0219		49	Polymixine B sulphate	P0145	1405-20-5	123
McCown Woody Plant medium including vitamins	M0220		49	Polyoxyethylenesorbitan monolaurate	P1362	9005-64-5	123
McCown Woody Plant vitamin mixture	M0408		49	Polyoxyethylenesorbitan monooleate	P1365	9005-65-6	124
mCS20ABN medium	C5122		159	Polyvinyl pyrrolidone (PVP 10)	P1368	9003-39-8	124
mD5A medium	D5124		161	Potassium chloride	P0515	7447-40-7	124
MES monohydrate	M1503	4432-31-9	113	Potassium dihydrogen phosphate	P0574	7778-77-0	124
meta-Topoline	T0941	75737-38-1	134	Potassium hydroxide	P0517	1310-58-3	124
Methotrexate	M0130	59-05-2	114	Potassium iodide	P0518	7681-11-0	125
Methyl jasmonate	M0918	39924-52-2	114	Potassium nitrate	P0519	7757-79-1	125
Metronidazole	M0131	443-48-1	114	Potassium sulphate	P0535	7778-80-5	125
mFS medium	F5123		160	Propyleneglycol	P1391	57-55-6	125
Miconazole nitrate	M0132	22832-87-7	115	PSM medium	P5134		164
Micro agar	M1002	9002-18-0	78	PTSA medium	P5135		158
Micro salt mixture B5	M0302		73	Putrescine HCl	P0927	333-93-7	125
Micro salt mixture MS	M0301		74	Pyridoxine HCl	P0612	58-56-0	126
Micro salt mixture Nitsch	M0303		74	Quoirin & Lepoivre medium	Q0250		66
Minocycline HCl	M0172	13614-98-7	115	Quoirin & Lepoivre medium including vitamins	Q0251		66
Mitomycin C	M0133	50-07-7	115	Raffinose pentahydrate	R0812	17629-30-0	126
mKM medium	K5125		162	Rest	R3002		141
MOPS	M1502	1132-61-2	115	Ribavirin	R0182	36791-04-5	126
MSP medium	M5167		155	Riboflavine	R0613	83-88-5	126
MT medium	M5133		156	Rifampicin	R0146	13292-46-1	127
mTBM medium	T5132		163	Rugini Olive medium	R0258		67
mTMB medium	T5126		166	Salicylic acid	S1367	69-72-7	127
MTT	M1415	298-93-1	116	Salmon Gal	S1403	138182-21-5	93
MUG trihydrate	M1404	6160-80-1	114	Salmon-XGlcA cyclohexylammonium salt	S1407	138182-20-4	93
Murashige & Miller medium stage I & II	M0243		61	Scalpel Blades No. 10	S3200		141
Murashige & Skoog med. Finer&Nagasawa	M0240		59	Scalpel Blades No. 11	S3201		141
Murashige & Skoog med. incl. mod. vitamins	M0245		51	Scalpel Handle	S3101		140
Murashige & Skoog med. van der Salm	M0241		60	Scalpel Handle Ergonomic	S3110		140
Murashige & Skoog med. van der Salm / vit.	M0242		60	Schenk & Hildebrandt medium	S0225		68
Murashige & Skoog medium	M0221		50	Schenk & Hildebrandt vitamin mixture	S0411		68
Murashige & Skoog medium incl. MES buffer	M0254		54	SCM medium	S5127		169
Murashige & Skoog medium incl. Nitsch vitamins	M0256		53	Seaplaque™ agarose	S1202	9012-36-6	79
Murashige & Skoog medium incl. vitamins/MES	M0255		54	Silver nitrate	S0536	7761-88-8	127
Murashige & Skoog medium including B5 vitamins	M0231		52	S-medium	S0261		69
Murashige & Skoog medium including vitamins	M0222		50	SNAC medium	S5130		165

Product name	Cat. no.	CAS no.	Page	Product name	Cat. no.	CAS no.	Page
Sodium alginate	S1320	9005-38-3	128	Ticarcillin disodium / clavulanate potassium	T0190		134
Sodium chloride	S0520	7647-14-5	128	Tobramycine sulphate	T0153	49842-07-1	134
Sodium dihydrogen phosphate dihydrate	S0522	13472-35-0	128	Trehalose Anhydrous	T1395	99-20-7	134
Sodium dodecyl sulphate (SDS)	S1377	151-21-3	128	Triethanolamine	T1361	102-71-6	135
Sodium hydroxide	S0523	1310-73-2	129	Trifluralin	T0928	1582-09-8	135
Sodium molybdate dihydrate	S0525	10102-40-6	129	Trimethoprim	T0154	738-70-5	135
Sodium nitrate	S0524	7613-99-4	129	Trimethoprim lactate	T0181	23256-42-0	135
Sodium thiosulphate	S0538	7772-98-7	129	TRIS HCl	T1513	1185-53-1	136
Soya peptone	S1330	73049-73-7	129	TRIS ultrapure	T1501	77-86-1	136
Spectinomycin HCl pentahydrate	S0188	22189-32-8	130	tri-Sodium citrate dihydrate	S0521	4/3/6132	128
Spermidine	S1369	124-20-9	130	Tryptone	T1332		136
SSC-buffer	S1511		130	Urea	U1363	57-13-6	137
SSPE-buffer	S1512		130	Vacin & Went medium	V0226		71
Starch from potatoes	S1357	9005-84-9	131	Validamycin A	V0170	37248-47-8	137
Starch from rice	S1324	9005-84-9	131	Vancomycin HCl	V0155	1404-93-9	137
Sterillium	S0162		131	Westvaco WV5 medium incl. vitamins	W0260		70
Sterivent Container			144	White medium	W0227		72
Sterivent high	S1685		144	X-Gal	X1402	7240-90-6	86
Sterivent low	S1680		144	X-GlcA cyclohexylammonium salt	X1405	114162-64-0	87
Streptomycin sulphate	S0148	3810-74-0	131	X-GlcA sodium salt trihydrate	X1406	129541-41-9	87
Sucrose	S0809	57-50-1	131	X-phos disodium salt (BCIP disodium salt)	X1410	102185-33-1	87
Sulphamethoxazole	S0149	723-46-6	131	X-Phos p-Toluidine salt (BCIP p-Toluidine salt)	X1413	6/9/6578	88
Talc	T1359		131	YDC medium	Y5136		173
Taurine	T1360	107-35-7	132	Yeast extract	Y1333	1/2/8013	138
TBE buffer	T1507		132	YPD Agar	Y1709		138
TE-buffer	T1508		132	YPD Broth	Y1708		138
Tetracycline HCl	T0150	64-75-5	132	Zeatin	Z0917	1637-39-4	138
Thiamine HCl	T0614	67-03-8	133	Zeatin riboside	Z0937	6025-53-2	139
Thidiazuron	T0916	51707-55-2	133	Zeocin™	Z0186	11006-33-0	139
Thimerosal	T0151	54-64-8	133	Zinc sulphate heptahydrate	Z0526	7446-20-0	139
Ticarcillin disodium	T0180	4697-14-7	133				

## Index (catalogue number)

Cat. no.	Product name	CAS no.	Page	Cat. no.	Product name	CAS no.	Page
A0101	Amoxicillin trihydrate	61336-70-7	81	C0176	Cycloheximide	66-81-9	96
A0103	Amphotericin B	1397-89-3	82	C0177	Chlorsulfuron	64902-72-3	94
A0104	Ampicillin sodium	69-52-3	82	C0203	CHU (N6) Medium		36
A0156	Atrazine	1912-24-9	83	C0204	CHU (N6) Medium including vitamins		36
A0164	Apramycin sulphate	65710-07-8	82	C0228	CLC / Ipomoea CP medium		37
A0183	Acyclovir	59277-89-3	77	C0229	CLC / Ipomoea EP medium		37
A0185	Amiprophos methyl	36001-88-4	80	C0248	CHÉE & POOL basal salt medium		35
A0189	Amoxicillin sodium / Clavulanate potassium		81	C0249	CHÉE & POOL medium including vitamins		35
A0192	Amphotericin B suspension	1397-89-3	82	C0401	CHU vitamine mixture		36
A0201	Anderson's Rhododendron		34	C0504	Calcium chloride dihydrate	10035-04-8	88
A0202	Anderson's Rhododendron including vitamins		34	C0505	Calcium nitrate tetrahydrate	13477-34-4	89
A0501	Ammonium nitrate	6484-52-2	81	C0506	Calcium phosphate tribasic	7758-87-4	89
A0502	Ammonium sulphate	7783-20-2	81	C0507	Cobalt chloride hexahydrate	7791-13-1	95
A0528	Ammonium chloride	12125-02-9	80	C0508	Cupric sulphate pentahydrate	7758-99-8	95
A0532	Aluminium chloride hexahydrate	7784-13-6	80	C0529	Calcium carbonate	471-34-1	88
A0601	p-Aminobenzoic acid	150-13-0	80	C0530	Calcium citrate tetrahydrate	5785-44-4	89
A0602	L-Ascorbic acid	50-81-7	83	C0531	Calcium gluconate monohydrate	299-28-5	89
A0703	L-Alanine	56-41-7	80	C0604	D(+) Pantothenate calcium	137-08-6	120
A0704	L-Arginine	74-79-3	83	C0605	Choline chloride	67-48-1	94
A0705	L-Aspartic acid	56-84-8	83	C0706	L-Cysteine HCl monohydrate	4/6/7048	96
A0725	L-Asparagine monohydrate	5794-13-8	83	C0726	Cyanocobalamin	68-19-9	96
A0908	Adenine hemisulphate dihydrate	321-30-2	77	C0909	p-Chlorophenoxyacetic acid (4-CPA)	122-88-3	94
A0941	Absisic acid (S-ABA)	21293-29-8	77	C0943	4-CPUP	68157-60-8	95
A1203	Agarose SPI	9012-36-6	79	C1006	Carrageenan Iota Type	7/1/9062	89
A1334	Adenosine	58-61-7	77	C1301	Casein hydrolysate	9000-71-9	90
A1335	Adenosine-5-triphosphate	987-65-5	78	C1302	Charcoal activated	64365-11-3	92
A1338	Ammonium dihydrogen phosphate	7722-76-1	80	C1303	Citric acid monohydrate	5949-29-1	94
A1366	Acetylsalicylic acid	50-78-2	77	C1305	Colchicine	64-86-8	95
B0106	Bacitracin	1405-87-4	84	C1374	CHAPS	75621-03-3	92
B0107	Bleomycin sulphate	9041-93-4	86	C1393	Cetrinonium	57-09-0	92
B0157	Bromoxynil	1689-84-5	88	C1397	Cetrimide	8044-71-1	92
B0503	Boric acid	10043-35-3	86	C1714	Czapek Dox Broth, CDB		175
B0603	D(+)-Biotine	58-85-5	85	C1715	Czapek Dox Agar, CDA		174
B0904	6-Benzylaminopurine (6-BAP)	1214-39-7	84	C5122	mCS20ABN medium		159
B0930	6-Benzylaminopurine riboside	4294-16-0	84	C5140	CKTM medium		168
B0932	N-Benzyl-9-(tetrahydropyranil)-adenine (BPA)	2312-73-4	85	C8001	Cellulase R-10	9012-54-8	91
B1304	Banana powder		84	C8003	Cellulase RS	9012-54-8	91
B1414	Blue-Gal	97753-82-7	88	D0120	Doxorubicin HCl 0.2% in 0.9% NaCl solution (5ml)	25316-40-9	99
B1514	BES	10191-18-1	85	D0121	Doxycycline HCl	10592-13-9	99
B1516	Bis-Tris buffer grade	6976-37-0	85	D0161	Chloroxyleneol ("Dettol")	88-04-0	94
B1702	Peptone water, buffered	73049-73-7	121	D0205	De Greef and Jacobs medium		38
B1713	Bacteria Screening medium 523		177	D0206	De Greef and Jacobs medium including vitamins		38
C0109	Carbenicillin disodium	4800-94-6	90	D0246	DKW/Juglans medium		39
C0110	Cephalexin monohydrate	15686-71-2	90	D0247	DKW/Juglans medium including vitamins		39
C0111	Cefotaxime sodium	64485-93-4	90	D0414	DKW/Juglans vitamin mixture		39
C0113	Chloramphenicol	56-75-7	92	D0906	2-iP	2365-40-4	97
C0114	Chlorhexidine digluconate	18472-51-0	93	D0911	2,4-Dichlorophenoxyacetic acid (2,4 D)	94-75-7	97
C0115	Chlorhexidine HCl	3697-42-5	93	D0920	Dicamba	1918-00-9	97
C0116	Chlortetracycline HCl	64-72-2	94	D0933	Dihydrozeatin (DHZ)	14894-18-9	97
C0117	Clindamycin HCl	21462-39-5	95	D0934	2-iP riboside	7724-76-7	98
C0118	Colistin sulphate	1264-72-8	95	D1004	Daishin agar	9002-18-0	78
C0119	D-Cycloserine	68-41-7	96	D1308	Dithioerythritol (DTE)	6892-68-8	98
C0160	Carboxin	5234-68-4	90	D1309	Dithiothreitol (DTT)	12/3/3483	98

Cat. no.	Product name	CAS no.	Page	Cat. no.	Product name	CAS no.	Page
D1342	Dextran sulphate sodium	9011-18-1	96	L3301	Leucopore tape, 2.5 cm x 9.2 m		142
D1370	Dimethylsulfoxide (DMSO)	67-68-5	98	L3302	Leucopore tape, 1.25 cm x 9.2 m		142
D5124	mD5A medium		161	M0129	6-Mercaptopurine monohydrate	6112-76-1	113
D5128	D2ANX medium		170	M0130	Methotrexate	59-05-2	114
E0122	Erythromycin	114-07-8	100	M0131	Metronidazole	443-48-1	114
E0207	Eriksson (ER) medium		40	M0132	Miconazole nitrate	22832-87-7	115
E0208	Eriksson (ER) medium including vitamins		40	M0133	Mitomycin C	50-07-7	115
E0402	Eriksson (ER) vitamin mixture		40	M0135	Neomycin sulphate	1405-10-3	117
E0509	FeNaEDTA	15708-41-5	100	M0172	Minocycline HCl	13614-98-7	115
E0511	EDTA disodium dihydrate	6381-92-6	100	M0219	McCown Woody Plant medium		49
E0940	24-Epibrassinolide	78821-43-9	99	M0220	McCown Woody Plant medium including vitamins		49
E1343	Esculin	531-75-9	100	M0221	Murashige & Skoog medium		50
E1650	ECO2 box white filter (oval model 80mm H)		143	M0222	Murashige & Skoog medium including vitamins		50
E1654	ECO2 box green filter (oval model 80mm H)		143	M0231	Murashige & Skoog medium including B5 vitamins		52
E1674	OS140 box green filter (round model 140mm H)		143	M0232	Murashige & Skoog medium mod. No. 1A		55
F0123	5-Fluoro uracil (5-FU)	51-21-8	101	M0233	Murashige & Skoog medium mod. No. 1B		55
F0176	5-Fluoro orotic acid (5-FOA)	703-95-7	101	M0234	Murashige & Skoog medium mod. No. 2A		56
F0512	Ferrous sulphate heptahydrate	7782-63-0	100	M0235	Murashige & Skoog medium mod. No. 2B		56
F0527	Fe-EDDHA		100	M0236	Murashige & Skoog medium mod. No. 3A		57
F0608	Folic acid	59-30-3	102	M0237	Murashige & Skoog medium mod. No. 3B		57
F0619	Folinate calcium pentahydrate (old C0607)	41927-89-3	102	M0238	Murashige & Skoog medium mod. No. 4		58
F0801	D-Fructose	57-48-7	102	M0239	Murashige & Skoog medium mod. No. 5		58
F0919	Fluridon	59756-60-4	101	M0240	Murashige & Skoog med. Finer&Nagasawa		59
F0935	Flurprimidol	56425-91-3	102	M0241	Murashige & Skoog med. van der Salm		60
F3001	Forceps, 23 cm		140	M0242	Murashige & Skoog med. van der Salm / vit.		60
F3003	Forceps extended, 30 cm		140	M0243	Murashige & Miller medium stage I & II		61
F5123	mFS medium		160	M0244	Murashige & Skoog medium shoot multiplication B		62
G0124	Gentamycin sulphate	1405-41-0	103	M0245	Murashige & Skoog med. incl. mod. vitamins		51
G0158	Glyphosate	1071-83-6	105	M0254	Murashige & Skoog medium incl. MES buffer		54
G0167	Griseofulvin	126-07-8	105	M0255	Murashige & Skoog medium incl. vitamins/MES		54
G0175	G-418 disulphate	108321-42-2	102	M0256	Murashige & Skoog medium incl. Nitsch vitamins		53
G0209	Gamborg B5 medium		41	M0301	Micro salt mixture MS		74
G0210	Gamborg B5 medium including vitamins		41	M0302	Micro salt mixture B5		73
G0211	Gresshoff & Doy medium		42	M0303	Micro salt mixture Nitsch		74
G0212	Gresshoff & Doy medium including vitamins		42	M0304	Macro salt mixture B5		73
G0404	Gresshof & Doy (DBM2) vitamin mixture		42	M0305	Macro salt mixture MS		74
G0415	Gamborg B5 vitamin mixture		41	M0306	Macro salt mixture Nitsch		74
G0707	L-Glutamic acid	56-86-0	104	M0408	McCown Woody Plant vitamin mixture		49
G0708	L-Glutamine	56-85-9	104	M0409	Murashige & Skoog medium vitamin mixture		50
G0709	Glycine	56-40-6	105	M0513	Magnesium sulphate heptahydrate	10034-99-8	111
G0802	D-Glucose monohydrate	10/15996	104	M0514	Manganese sulphate monohydrate	10034-96-5	112
G0810	D-Galactose	59-23-4	103	M0533	Magnesium chloride hexahydrate	7791-18-6	111
G0907	Gibberellic acid A3	77-06-5	104	M0715	L-Methionine	63-68-3	113
G0938	Gibberellic acid 4+7	468-44-0/510-75-8	104	M0803	D-Mannitol	69-65-8	112
G1007	Gelcarin GP- 812	7/1/9000	89	M0811	Maltose monohydrate	6363-53-7	112
G1101	Gelrite™	71010-52-1	103	M0918	Methyl jasmonate	39924-52-2	114
G1345	Glycerol	56-81-5	105	M0921	Maleic hydrazide	123-33-1	111
G1346	Gluthathione reduced	70-18-8	105	M1002	Micro agar	9002-18-0	78
G1375	Guanidine HCl	50-01-1	106	M1315	Malic acid	617-48-1	111
G3301	Glass Bead Sterilizer (incl. Glass beads)		141	M1327	Malt extract	8002-48-0	112
G3302	Glass Beads for Sterilizer		141	M1392	D-Mannose	3458-28-4	113
H0168	8-Hydroxyquinoline	148-24-3	106	M1404	MUG trihydrate	6160-80-1	114
H0192	Hygromycine B	31282-04-9	107	M1412	Magenta-GlcA cyclohexylammonium salt	144110-43-0	87
H0213	Heller medium		43	M1415	MTT	298-93-1	116
H0710	L-Histidine	71-00-1	106	M1502	MOPS	1132-61-2	115
H1504	HEPES	7365-45-9	106	M1503	MES monohydrate	4432-31-9	113
I0609	Myo-Inositol	87-89-8	107	M5131	MXV medium		167
I0711	L-Isoleucine	73-32-5	108	M5133	MT medium		156
I0901	Indole-3-acetic acid (IAA)	87-51-4	107	M5167	MSP medium		155
I0902	Indole-3-butyric acid (IBA)	133-32-4	107	M8002	Macerozyme R-10	9032-75-1	111
I1401	IPTG	367-93-1	108	N0134	Nalidixic acid	389-08-2	116
J0936	Jasmonic acid	6894-38-8	108	N0138	Nystatine	1400-61-9	119
K0126	Kanamycin sulphate monohydrate	25389-94-0	108	N0223	Nitsch medium		63
K0214	KAO & Michayluk medium		44	N0224	Nitsch medium including vitamins		63
K0215	Knudson C Orchid medium		45	N0252	NLN medium		64
K0905	Kinetin	525-79-1	109	N0253	NLN medium vitamin mixture		64
K5120	KBBC medium		154	N0410	Nitsch vitamin mixture		63
K5125	mKM medium		162	N0610	Nicotinamide	98-92-0	117
K5129	KBZ medium		171	N0611	Nicotinic acid	59-67-6	118
K5165	KB medium (King's B medium)		172	N0903	a-naphtalene acetic acid	86-87-3	116
L0127	Lincomycin HCl monohydrate	7179-49-9	110	N0912	b-Naphtoxyacetic acid	120-23-0	116
L0216	Lindemann Orchid medium		46	N0926	Naphthylphthalamic acid	132-66-1	117
L0217	Litvay medium		48	N1350	1-Naphtylphosphate sodium monohydrate	81012-89-7	117
L0218	Litvay medium including vitamins		48	N1408	p-Nitrophenyl-b-D-glucuronide	10344-94-2	118
L0230	Linsmaier & Skoog medium		47	N1411	Nitro Blue Tetrazolium (NBT)	38184-50-8	118
L0406	Linsmaier & Skoog vitamin mixture		47	O0140	Oxytetracycline HCl	2058-46-0	119
L0407	Litvay vitamin mixture		48	O0257	Orchimax without activated charcoal		65
L0712	L-Leucine	61-90-5	110	O0262	Orchimax including activated charcoal		65
L0714	L-Lysine HCl	657-27-2	110	O1318	Oryzaline	19044-88-3	119
L1204	Low Melting Agarose PPC	9012-36-6	79	O1351	L-Ornithine HCl	3184-13-2	119
L1349	D-Luciferin	2591-17-5	110	O1409	2-Nitrophenyl-b-D-galactopyranoside	369-07-3	118
L1372	Lactose monohydrate	10039-26-6	109	P0141	Paromomycin sulphate	1263-89-4	120
L1703	LB Broth Low salt		109	P0142	Penicillin G sodium	69-57-8	121
L1704	LB Broth High salt		109	P0145	Polymixine B sulphate	1405-20-5	123
L1705	LB Agar Low salt		109	P0159	Phosphinotricin	77182-82-2	122
L1706	LB Agar High salt		109	P0187	Phleomycin	11006-33-0	122
L1716	Leifert and Waites sterility test medium		178	P0515	Potassium chloride	7447-40-7	124
L1717	Luria Broth Base, Miller		180	P0517	Potassium hydroxide	1310-58-3	124
L1718	Luria Broth Agar, Miller		179	CP0518	Potassium iodide	7681-11-0	125
L1719	Malt Agar (MA)		176	P0519	Potassium nitrate	7757-79-1	125

at. no.	Product name	CAS no.	Page	Cat. no.	Product name	CAS no.	Page
P0535	Potassium sulphate	7778-80-5	125	S1511	SSC-buffer		130
P0573	di-Potassium hydrogen phosphate	114/7758	124	S1512	SSPE-buffer		130
P0574	Potassium dihydrogen phosphate	7778-77-0	124	S1680	Sterivent low		144
P0612	Pyridoxine HCl	58-56-0	126	S1685	Sterivent high		144
P0716	L-Phenylalanine	63-91-2	121	S3101	Scalpel Handle		140
P0717	L-Proline	147-85-3	125	S3110	Ergonomic Scalple Handle		140
P0804	Polyethylene Glycol 4000	25322-68-3	123	S3110	Scalpel Handle Ergonomic		140
P0805	Polyethylene Glycol 6000	25322-68-3	123	S3200	Scalpel Blades No. 10		141
P0813	Polyethylene Glycol 400	25322-68-3	123	S3201	Scalpel Blades No. 11		141
P0914	Picloram	2/1/1918	122	S5127	SCM medium		169
P0922	Paclobutrazol	76738-62-0	120	S5130	SNAC medium		165
P0927	Putrescine HCl	333-93-7	125	T0150	Tetracycline HCl	64-75-5	132
P1001	Plant agar	9002-18-0	78	T0151	Thimerosal	54-64-8	133
P1003	Phyto agar	9002-18-0	78	T0153	Tobramycine sulphate	49842-07-1	134
P1328	Peptone	73049-73-7	121	T0154	Trimethoprim	738-70-5	135
P1353	Phloroglucinol	108-73-6	122	T0180	Ticarcillin disodium	4697-14-7	133
P1362	Polyoxyethylenesorbitan monolaurate	9005-64-5	123	T0181	Trimethoprim lactate	23256-42-0	135
P1365	Polyoxyethylenesorbitan monooleate	9005-65-6	124	T0190	Ticarcillin disodium / clavulanate potassium		134
P1368	Polyvinyl pyrrolidone (PVP 10)	9003-39-8	124	T0614	Thiamine HCl	67-03-8	133
P1391	Propyleneglycol	57-55-6	125	T0719	L-Threonine	72-19-5	133
P1505	PIPES	5625-37-6	123	T0720	L-Tryptophan	73-22-3	136
P1707	Peptone water	73049-73-7	121	T0721	L-Tyrosine	60-18-4	137
P3202	Paper Cutting Pad		141	T0915	2,4,5-Trichlorophenoxyacetic acid	37785-57-2	135
P5134	PSM medium		164	T0916	Thidiazuron	51707-55-2	133
P5135	PTSA medium		158	T0928	Trifluralin	1582-09-8	135
P8004	Pectolyase Y-23	9033-35-6	120	T0929	2,3,5-Triiodobenzoic acid	88-82-4	135
Q0250	Quoirin & Lepoivre medium		66	T0941	meta-Topoline	75737-38-1	134
Q0251	Quoirin & Lepoivre medium including vitamins		66	T1332	Tryptone		136
R0146	Rifampicin	13292-46-1	127	T1359	Talc		131
R0182	Ribavirin	36791-04-5	126	T1360	Taurine	107-35-7	132
R0258	Rugini Olive medium		67	T1361	Triethanolamine	102-71-6	135
R0613	Riboflavine	83-88-5	126	T1395	Trehalose Anhydrous	99-20-7	134
R0806	D-Ribose	50-69-1	126	T1501	TRIS ultrapure	77-86-1	136
R0812	Raffinose pentahydrate	17629-30-0	126	T1507	TBE buffer		132
R3002	Rest		141	T1508	TE-buffer		132
S0148	Streptomycin sulphate	3810-74-0	131	T1513	TRIS HCl	1185-53-1	136
S0149	Sulphamethoxazole	723-46-6	131	T1608	De Wit' Tray 10 pcs.		142
S0162	Sterillium		131	T5126	mTMB medium		166
S0188	Spectinomycin HCl pentahydrate	22189-32-8	130	T5132	mTBM medium		163
S0225	Schenk & Hildebrandt medium		68	U1363	Urea	57-13-6	137
S0261	S-medium		69	V0155	Vancomycin HCl	1404-93-9	137
S0411	Schenk & Hildebrandt vitamin mixture		68	V0170	Validamycin A	37248-47-8	137
S0520	Sodium chloride	7647-14-5	128	V0226	Vacin & Went medium		71
S0521	tri-Sodium citrate dihydrate	4/3/6132	128	V0722	L-Valine	72-18-4	137
S0522	Sodium dihydrogen phosphate dihydrate	13472-35-0	128	W0227	White medium		72
S0523	Sodium hydroxide	1310-73-2	129	W0260	Westvaco WV5 medium incl. vitamins		70
S0524	Sodium nitrate	7613-99-4	129	W1607	Culture tubes "De Wit" (W1614 bottom / W1615 top)		142
S0525	Sodium molybdate dihydrate	10102-40-6	129	X0808	D-Xylose	58-86-6	137
S0536	Silver nitrate	7761-88-8	127	X1402	X-Gal	7240-90-6	86
S0537	Di-Sodium hydrogen phosphate dihydrate	10028-24-7	128	X1405	X-GlcA cyclohexylammonium salt	114162-64-0	87
S0538	Sodium thiosulphate	7772-98-7	129	X1406	X-GlcA sodium salt trihydrate	129541-41-9	87
S0718	L-Serine	56-45-1	127	X1410	X-phos disodium salt (BCIP disodium salt)	102185-33-1	87
S0807	D-Sorbitol	50-70-4	129	X1413	X-Phos p-Toluidine salt (BCIP p-Toluidine salt)	6/9/6578	88
S0809	Sucrose	57-50-1	131	X5121	mXCP1 medium		157
S1202	Seaplaque™ agarose	9012-36-6	79	Y1333	Yeast extract	1/2/8013	138
S1320	Sodium alginate	9005-38-3	128	Y1708	YPD Broth		138
S1324	Starch from rice	9005-84-9	131	Y1709	YPD Agar		138
S1330	Soya peptone	73049-73-7	129	Y5136	YDC medium		173
S1357	Starch from potatoes	9005-84-9	131	Z0186	Zeocin™	11006-33-0	139
S1367	Salicylic acid	69-72-7	127	Z0526	Zinc sulphate heptahydrate	7446-20-0	139
S1369	Spermidine	124-20-9	130	Z0917	Zeatin	1637-39-4	138
S1377	Sodium dodecyl sulphate (SDS)	151-21-3	128	Z0937	Zeatin riboside	6025-53-2	139
S1403	Salmon Gal	138182-21-5	93		Sterivent Container		144
S1407	Salmon-XGlcA cyclohexylammonium salt	138182-20-4	93				

## Index (CAS no.)

CAS no.	Product name	Cat. no.	Page	CAS no.	Product name	Cat. no.	Page
2/1/1918	Picloram	P0914	122	25322-68-3	Polyethylene Glycol 6000	P0805	123
12/3/3483	Dithiothreitol (DTT)	D1309	98	25322-68-3	Polyethylene Glycol 400	P0813	123
10/1/5996	D-Glucose monohydrate	G0802	104	25389-94-0	Kanamycine sulphate monohydrate	K0126	108
4/3/6132	tri-Sodium citrate dihydrate	S0521	128	2591-17-5	D-Luciferin	L1349	110
6/9/6578	X-Phos p-Toluidine salt (BCIP p-Toluidine salt)	X1413	88	298-93-1	MTT	M1415	116
4/6/7048	L-Cysteine HCl monohydrate	C0706	96	299-28-5	Calcium gluconate monohydrate	C0531	89
11/4/7758	di-Potassium hydrogen phosphate	P0573	124	31282-04-9	Hygromycine B	H0192	107
1/2/8013	Yeast extract	Y1333	138	3184-13-2	L-Ornithine HCl	O1351	119
7/1/9000	Gelcarin GP- 812	G1007	89	321-30-2	Adenine hemisulphate dihydrate	A0908	77
7/1/9062	Carrageenan Iota Type	C1006	89	333-93-7	Putrescine HCl	P0927	125
10028-24-7	Di-Sodium hydrogen phosphate dihydrate	S0537	128	3458-28-4	D-Mannose	M1392	113
10034-96-5	Manganese sulphate monohydrate	M0514	112	36001-88-4	Amiprophos methyl	A0185	80
10034-99-8	Magnesium sulphate heptahydrate	M0513	111	36791-04-5	Ribavirin	R0182	126
10035-04-8	Calcium chloride dihydrate	C0504	88	367-93-1	IPGT	I1401	108
10039-26-6	Lactose monohydrate	L1372	109	369-07-3	2-Nitrophenyl-b-D-galactopyranoside	O1409	118
10043-35-3	Boric acid	B0503	86	3697-42-5	Chlorhexidine HCl	C0115	93
10102-40-6	Sodium molybdate dihydrate	S0525	129	37248-47-8	Validamycin A	V0170	137
10191-18-1	BES	B1514	85	37785-57-2	2,4,5-Trichlorophenoxyacetic acid	T0915	135
102185-33-1	X-phos disodium salt (BCIP disodium salt)	X1410	87	3810-74-0	Streptomycin sulphate	S0148	131
102-71-6	Triethanolamine	T1361	135	38184-50-8	Nitro Blue Tetrazolium (NBT)	N1411	118
10344-94-2	p-Nitrophenyl-b-D-glucuronide	N1408	118	389-08-2	Nalidixic acid	N0134	116
10592-13-9	Doxycycline HCl	D0121	99	39924-52-2	Methyl jasmonate	M0918	114
1071-83-6	Glyphosate	G0158	105	41927-89-3	Folinate calcium pentahydrate (old C0607)	F0619	102
107-35-7	Taurine	T1360	132	4294-16-0	6-Benzylaminopurine riboside	B0930	84
108321-42-2	G-418 disulphate	G0175	102	4432-31-9	MES monohydrate	M1503	113
108-73-6	Phloroglucinol	P1353	122	443-48-1	Metronidazole	M0131	114
11006-33-0	Phleomycin	P0187	122	468-44-0/510-75-8	Gibberellic acid 4+7	G0938	104
11006-33-0	Zeocin™	Z0186	139	4697-14-7	Ticarillin disodium	T0180	133
1132-61-2	MOPS	M1502	115	471-34-1	Calcium carbonate	C0529	88
114-07-8	Erythromycin	E0122	100	4800-94-6	Carbenicillin disodium	C0109	90
114162-64-0	X-GlcA cyclohexylammonium salt	X1405	87	49842-07-1	Tobramycine sulphate	T0153	134
1185-53-1	TRIS HCl	T1513	136	50-01-1	Guanidine HCl	G1375	106
120-23-0	b-Naphtoxyacetic acid	N0912	116	50-07-7	Mitomycin C	M0133	115
12125-02-9	Ammonium chloride	A0528	80	50-69-1	D-Ribose	R0806	126
1214-39-7	6-Benzylaminopurine (6-BAP)	B0904	84	50-70-4	D-Sorbitol	S0807	129
122-88-3	p-Chlorophenoxyacetic acid (4-CPA)	C0909	94	50-78-2	Acetylsalicylic acid	A1366	77
123-33-1	Maleic hydrazide	M0921	111	50-81-7	L-Ascorbic acid	A0602	83
124-20-9	Spermidine	S1369	130	51-21-8	5-Fluoro uracil (5-FU)	F0123	101
126-07-8	Griseofulvin	G0167	105	51707-55-2	Thidiazuron	T0916	133
1263-89-4	Paromomycin sulphate	P0141	120	5234-68-4	Carboxin	C0160	90
1264-72-8	Colistin sulphate	C0118	95	525-79-1	Kinetin	K0905	109
129541-41-9	X-GlcA sodium salt trihydrate	X1406	87	531-75-9	Esculin	E1343	100
1310-58-3	Potassium hydroxide	P0517	124	54-64-8	Thimerosal	T0151	133
1310-73-2	Sodium hydroxide	S0523	129	5625-37-6	PIPES	P1505	123
132-66-1	Naphthylphthalamic acid	N0926	117	56-40-6	Glycine	G0709	105
13292-46-1	Rifampicin	R0146	127	56-41-7	L-Alanine	A0703	80
133-32-4	Indole-3-butyric acid (IBA)	I0902	107	56425-91-3	Flurprimidol	F0935	102
13472-35-0	Sodium dihydrogen phosphate dihydrate	S0522	128	56-45-1	L-Serine	S0718	127
13477-34-4	Calcium nitrate tetrahydrate	C0505	89	56-75-7	Chloramphenicol	C0113	92
13614-98-7	Minocycline HCl	M0172	115	56-81-5	Glycerol	G1345	105
137-08-6	D(+) Pantothenate calcium	C0604	120	56-84-8	L-Aspartic acid	A0705	83
138182-20-4	Salmon-XGlcA cyclohexylammonium salt	S1407	93	56-85-9	L-Glutamine	G0708	104
138182-21-5	Salmon Gal	S1403	93	56-86-0	L-Glutamic acid	G0707	104
1397-89-3	Amphotericin B	A0103	82	57-09-0	Cetrimonium	C1393	92
1397-89-3	Amphotericin B suspension	A0192	82	57-13-6	Urea	U1363	137
1400-61-9	Nystatine	N0138	119	57-48-7	D-Fructose	F0801	102
1404-93-9	Vancomycin HCl	V0155	137	57-50-1	Sucrose	S0809	131
1405-10-3	Neomycin sulphate	M0135	117	57-55-6	Propyleneglycol	P1391	125
1405-20-5	Polymixine B sulphate	P0145	123	5785-44-4	Calcium citrate tetrahydrate	C0530	89
1405-41-0	Gentamycin sulphate	G0124	103	5794-13-8	L-Asparagine monohydrate	A0725	83
1405-87-4	Bacitracin	B0106	84	58-56-0	Pyridoxine HCl	P0612	126
144110-43-0	Magenta-GlcA cyclohexylammonium salt	M1412	87	58-61-7	Adenosine	A1334	77
147-85-3	L-Proline	P0717	125	58-85-5	D(+)-Biotine	B0603	85
148-24-3	8-Hydroxyquinoline	H0168	106	58-86-6	D-Xylose	X0808	137
14894-18-9	Dihydrozeatin (DHZ)	D0933	97	59-05-2	Methotrexate	M0130	114
150-13-0	p-Aminobenzoic acid	A0601	80	59-23-4	D-Galactose	G0810	103
151-21-3	Sodium dodecyl sulphate (SDS)	S1377	128	59277-89-3	Acyclovir	A0183	77
15686-71-2	Cephalexin monohydrate	C0110	90	59-30-3	Folic acid	F0608	102
15708-41-5	FeNaEDTA	E0509	100	5949-29-1	Citric acid monohydrate	C1303	94
1582-09-8	Trifluralin	T0928	135	59-67-6	Nicotinic acid	N0611	118
1637-39-4	Zeatin	Z0917	138	59756-60-4	Fluridon	F0919	101
1689-84-5	Bromoxynil	B0157	88	60-18-4	L-Tyrosine	T0721	137
17629-30-0	Raffinose pentahydrate	R0812	126	6025-53-2	Zeatin riboside	Z0937	139
18472-51-0	Chlorhexidine digluconate	C0114	93	6112-76-1	6-Mercaptopurine monohydrate	M0129	113
19044-88-3	Oryzaline	O1318	119	61336-70-7	Amoxicillin trihydrate	A0101	81
1912-24-9	Atrazine	A0156	83	6160-80-1	MUG trihydrate	M1404	114
1918-00-9	Dicamba	D0920	97	617-48-1	Malic acid	M1315	111
2058-46-0	Oxytetracycline HCl	O0140	119	61-90-5	L-Leucine	L0712	110
21293-29-8	Absisic acid (S-ABA)	A0941	77	6363-53-7	Maltose monohydrate	M0811	112
21462-39-5	Clindamycin HCl	C0117	95	63-68-3	L-Methionine	M0715	113
22189-32-8	Spectinomycin HCl pentahydrate	S0188	130	6381-92-6	EDTA disodium dihydrate	E0511	100
22832-87-7	Miconazole nitrate	M0132	115	63-91-2	L-Phenylalanine	P0716	121
2312-73-4	N-Benzyl-9-(tetrahydropyranyl)-adenine (BPA)	B0932	85	64365-11-3	Charcoal activated	C1302	92
23256-42-0	Trimethoprim lactate	T0181	135	64485-93-4	Cefotaxime sodium	C0111	90
2365-40-4	2-iP	D0906	97	64-72-2	Chlortetracycline HCl	C0116	94
25316-40-9	Doxorubicin HCl 0.2% in 0.9% NaCl solution (5ml)	D0120	99	64-75-5	Tetracycline HCl	T0150	132
25322-68-3	Polyethylene Glycol 4000	P0804	123	6484-52-2	Ammonium nitrate	A0501	81

CAS no.	Product name	Cat. no.	Page	CAS no.	Product name	Cat. no.	Page
64-86-8	Colchicine	C1305	95	7757-79-1	Potassium nitrate	P0519	125
64902-72-3	Chlorsulfuron	C0177	94	7758-87-4	Calcium phosphate tribasic	C0506	89
65710-07-8	Apramycin sulphate	A0164	82	7758-99-8	Cupric sulphate pentahydrate	C0508	95
657-27-2	L-Lysine HCl	L0714	110	7761-88-8	Silver nitrate	S0536	127
66-81-9	Cycloheximide	C0176	96	7772-98-7	Sodium thiosulphate	S0538	129
67-03-8	Thiamine HCl	T0614	133	7778-77-0	Potassium dihydrogen phosphate	P0574	124
67-48-1	Choline chloride	C0605	94	7778-80-5	Potassium sulphate	P0535	125
67-68-5	Dimethylsulfoxide (DMSO)	D1370	98	7782-63-0	Ferrous sulphate heptahydrate	F0512	100
68157-60-8	4-CPPU	C0943	95	7783-20-2	Ammonium sulphate	A0502	81
68-19-9	Cyanocobalamin	C0726	96	7784-13-6	Aluminium chloride hexahydrate	A0532	80
68-41-7	D-Cycloserine	C0119	96	77-86-1	TRIS ultrapure	T1501	136
6892-68-8	Dithioerythritol (DTE)	D1308	98	7791-13-1	Cobalt chloride hexahydrate	C0507	95
6894-38-8	Jasmonic acid	J0936	108	7791-18-6	Magnesium chloride hexahydrate	M0533	111
69-52-3	Ampicillin sodium	A0104	82	78821-43-9	24-Épibrassinolide	E0940	99
69-57-8	Penicillin G sodium	P0142	121	8002-48-0	Malt extract	M1327	112
69-65-8	D-Mannitol	M0803	112	8044-71-1	Cetrimide	C1397	92
69-72-7	Salicylic acid	S1367	127	81012-89-7	1-Naphthylphosphate sodium monohydrate	N1350	117
6976-37-0	Bis-Tris buffer grade	B1516	85	83-88-5	Riboflavin	R0613	126
70-18-8	Glutathione reduced	G1346	105	86-87-3	a-naphtalene acetic acid	N0903	116
703-95-7	5-Fluoro orotic acid (5-FOA)	F0176	101	87-51-4	Indole-3-acetic acid (IAA)	I0901	107
71-00-1	L-Histidine	H0710	106	87-89-8	Myo-Inositol	I0609	107
71010-52-1	Gelrite™	G1101	103	88-04-0	Chloroxyleneol ("Dettol")	D0161	94
7179-49-9	Lincomycin HCl monohydrate	L0127	110	88-82-4	2,3,5-Triiodobenzoic acid	T0929	135
72-18-4	L-Valine	V0722	137	9000-71-9	Casein hydrolysate	C1301	90
72-19-5	L-Threonine	T0719	133	9002-18-0	Daishin agar	D1004	78
723-46-6	Sulphamethoxazole	S0149	131	9002-18-0	Micro agar	M1002	78
7240-90-6	X-Gal	X1402	86	9002-18-0	Plant agar	P1001	78
73049-73-7	Peptone water, buffered	B1702	121	9002-18-0	Phyto agar	P1003	78
73049-73-7	Peptone	P1328	121	9003-39-8	Polyvinyl pyrrolidone (PVP 10)	P1368	124
73049-73-7	Peptone water	P1707	121	9005-38-3	Sodium alginate	S1320	128
73049-73-7	Soya peptone	S1330	129	9005-64-5	Polyoxyethylenesorbitan monolaurate	P1362	123
73-22-3	L-Tryptophan	T0720	136	9005-65-6	Polyoxyethylenesorbitan monooleate	P1365	124
73-32-5	L-Isoleucine	I0711	108	9005-84-9	Starch from rice	S1324	131
7365-45-9	HEPES	H1504	106	9005-84-9	Starch from potatoes	S1357	131
738-70-5	Trimethoprim	T0154	135	9011-18-1	Dextran sulphate sodium	D1342	96
7446-20-0	Zinc sulphate heptahydrate	Z0526	139	9012-36-6	Agarose SPI	A1203	79
7447-40-7	Potassium chloride	P0515	124	9012-36-6	Low Melting Agarose PPC	L1204	79
74-79-3	L-Arginine	A0704	83	9012-36-6	Seaplaque™ agarose	S1202	79
75621-03-3	CHAPS	C1374	92	9012-54-8	Cellulase R-10	C8001	91
75737-38-1	meta-Topoline	T0941	134	9012-54-8	Cellulase RS	C8003	91
7613-99-4	Sodium nitrate	S0524	129	9032-75-1	Macerozyme R-10	M8002	111
7647-14-5	Sodium chloride	S0520	128	9033-35-6	Pectolyase Y-23	P8004	120
76738-62-0	Paclitaxel	P0922	120	9041-93-4	Bleomycin sulphate	B0107	86
7681-11-0	Potassium iodide	P0518	125	94-75-7	2,4-Dichlorophenoxyacetic acid (2,4 D)	D0911	97
77-06-5	Gibberellic acid A3	G0907	104	97753-82-7	Blue-Gal	B1414	88
77182-82-2	Phosphinotricin	P0159	122	987-65-5	Adenosine-5-triphosphate	A1335	78
7722-76-1	Ammonium dihydrogen phosphate	A1338	80	98-92-0	Nicotinamide	N0610	117
7724-76-7	2-iP riboside	D0934	98	99-20-7	Trehalose Anhydrous	T1395	134

