

**Improved Mixture of Useful Materials for Stock Culture in Soils that
are Infected with Nematodes**

The significance of in-soil plant-parasitic nematode strains in crop farming and the crop failures and growth depressions which they cause have been known to experts for decades. In order to combat these unwanted restrictions in arable farming, practice and scientific research have worked out a number of countermeasures. Thus, crop rotation – for example sugar beet, winter wheat and winter barley in a three-year cycle – can contribute towards easing the problems under discussion. Today, the use of synthetic nematocides is at least seriously restricted or prohibited over wide areas of agricultural land on account of the secondary damage which they cause. The incorporation of selected organic substances for improving soil or for controlling nematodes has been established practice for decades. The success of such measures is understood to involve a complex interaction between all soil organisms, see for example R.C. COOKE, 1963, **"Succession of Nematophagous Fungi During the Decomposition of Organic Matter in Soil"**, *Nature* 4863; 205 and S. HOFFMANN-HERGARTEN et al., 1993, **"Untersuchungen zur Steigerung der Wirkung nematodenfangender Pilze gegen den Heterodera-schachtii Frühbefall durch organische Dünger"**, *Zeitschrift für Pflanzenkrankheiten und Pflanzenschutz* 100 (2); 170-175.

The relevant literature is also concerned in particular with attempts to stimulate soil microorganism flora and, more particularly, strains which are capable of developing antagonistic and/or nematicidal effects against plant-pathogenic nematodes, cf. in particular **US-PS 5,057,141** and the literature cited therein, above all R. Rodriguez-Kabana et al. in **"Plant and Soil"**, 100:237 to 247 (1987). According to this particular literature reference, chitin-containing materials in admixture with other organic

nitrogen compounds, such as ammonium phosphate and urea, develop nematostatic and nematicidal activity in soils against plant-pathogenic nematode populations, but are not phytotoxic to the plants and actually serve as a food source. The teaching of **US-PS 4,536,207** goes in the same direction. This document describes the nematicidal activity of a chitin-protein complex compound which is obtained from a demineralized water-insoluble chitin material and a water-insoluble protein component. For practical application, these water-insoluble fine-particle complex compounds have to be mechanically incorporated in the soil in known manner.

The teaching of the present invention as described in the following is based on the prior art as represented by the US patents cited above. The key components of the mixtures described in these US patents are insoluble in water. This applies in particular to their chitin or chitin complex content. Accordingly, the incorporation of the insoluble material in upper soil layers can only be usefully undertaken in those parts of the soil which do not yet bear any plants. According to **US-PS 5,057,141**, a pronounced nematicidal effect is only observed in particular in the second successive plant cycle.

By contrast, the problem addressed by the technical teaching of hitherto unpublished earlier patent application **DE 198 24 358.8** was to provide a soil additive which could be introduced into the affected parts of the soil in liquid form and, more particularly, in the form of an aqueous preparation and which would lead there to rapid development of the antagonistic effects, above all of the microorganism flora active there, towards plant-pathogenic soil nematodes. At the same time, the mixture according to the invention would lend itself to introduction, more especially into the region around the plant roots, both before and also during the sowing or planting of the affected areas of soil and during subsequent plant growth. Accordingly, the teaching of the earlier application in question

sought to make it possible to strengthen the strains of microorganism flora, more especially bacteria and/or fungi, which are present in particular in the rhizosphere or mycorrhiza and hence in the immediate surface region of the plant roots and which are distinguished by antagonistic and/or
5 nematicidal activity towards unwanted nematode infestation of the plant roots.

By contrast, the technical teaching illustrated below is based on the object of being able once again to bring about a marked increase in efficacy with multicomponent mixtures of the type previously described in
10 the earlier patent application. The technical teaching extended in terms of the invention is based on the fact that chitosan or chitosan compounds, which are known to be obtainable by the deacetylation of chitin, also have a nematostatic or nematicidal action; cf., for example, **US-PS 5,139,949**. Although the primary product obtained by chitin deacetylation, namely high
15 molecular chitosan - especially with molecular weights in the region of 1 million or more - is as such insoluble or only poorly soluble in water and aqueous alkali solutions, it is known substantially to increase the solubility of this (these) chitosan component(s) by simple means. Thus, the chitosan-based components dissolve in aqueous solutions of organic and
20 inorganic acids. It is also known that chitosan with a degree of deacetylation of 50% is water-soluble. The water solubility can be increased by degrading the molecular weights to the range of chitosan oligomers; finally, the solubility can be adjusted over a wide pH range in water by means of simple chemical reactions - especially by
25 carboxymethylation to O-CM-chitosan, N-CM-chitosan and/or N/O-CM-chitosan. For details of this basic knowledge about chitosans, cf., for example, the publication entitled "**Applications of Chitin and Chitosan**", Goosen, Mattheus F.A. (Ed.), Lancaster: Technomic Publishing Company, 1996, chapter 1 entitled "**Applications and Properties of Chitosan**" Q. Li
30 et al., pages 3 to 29, particularly pages 8 and 9.

Against the background of this state of knowledge relating to possible flowable and particularly aqueous forms of application of chitosan, the further improved technical teaching illustrated below - based on the operating instructions in the above-cited earlier patent application DE 198 24 358.8 - becomes clear. The development in terms of the invention now provides the possibility of introducing, in addition to the nutrient or active substance mixtures with a nematostatic/nematicidal action described in the earlier patent application, chitosan-based mixing components with corresponding actions, in dissolved form, into the soil and especially into the region of the plants' root. This affords a further substantial increase in value in terms of the objective according to the invention, namely to protect plants from infestation by nematodes.

Subject of the Invention

Accordingly, the present invention relates to the use of a nutrient mixture of components (a), (b) and (c) defined hereinafter as a soil additive in crop cultivation in cultivated soils which are infected by plant-parasitic nematodes and/or in which the desired objective of optimizing the result of crop cultivation is endangered by corresponding nematode infestation. Components of class (a) are compounds of phosphorus containing at least partly lipophilic organic radicals which are used together with (b) urea and/or urea derivatives. The components of class (a) and class (b) are used in such quantity ratios of (a) to (b) that the ratio by weight of carbon (C) to nitrogen (N) does not exceed about 6:1. In a preferred embodiment, the ratio by weight of C to N is at most about 5:1. The further development according to the invention - in combination with the introduction of components (a) and (b) - provides for the simultaneous and/or staggered introduction of oligomeric and/or polymeric chitosans, which are referred to as components of class (c) in the following description of the invention. The staggered introduction of components (c) can take place either before

or after the introduction of components (a) and (b).

In another embodiment, the present invention relates to the use of phospholipids of vegetable origin in combination with urea and/or urea derivatives and in combination with oligomeric and/or polymeric chitosans as component (c) for the biological control of plant-parasitic soil nematodes by strengthening the correspondingly antagonistic soil potential - more especially by strengthening the growth of antagonistic and/or nematicidal rhizosphere bacteria and/or corresponding mycorrhiza strains - and at the same time promoting the growth of the cultivated plants/crops. In this embodiment, too, the mixtures according to the invention are preferably introduced into the endangered soil in the form of aqueous preparations before and/or more particularly during growth.

Details of the teaching according to the invention

Biological life processes and the discernible effects accompanying them are known to be the summary results of highly complex micro-organism interactions which can in turn can be determined by a number of external conditions or living conditions. Equal significance attaches in this regard to the growth and development both of microorganism flora and of soil fauna. This applies in particular to corresponding development processes in the soil and to the secondary results which they determine in the cultivation of plants in soil. Despite this general understanding, the possibility of permanently influencing these complex and interrelated life processes is still very limited.

DE 44 37 313 A1 describes a process for improving plant growth in agriculture, forestry and horticulture by stimulating the microorganisms that live in the soil in which the plants grow. The document in question proposes the use of certain mixtures based on phospholipids, for example lecithin, and phospholipid derivatives whose use for various purposes in agriculture had already been mentioned. Thus, European patent

application **EP 95 071** and International patent application **WO 89/8628** describe the use of phospholipids together with macronutrients as leaf fertilizers. International patent application **WO 93/1150** mentions the use of phospholipids as emulsifiers in fertilizer preparations. According to **DE 42**
5 **18 243**, mixtures of glycerophospholipids and urea can be used to activate certain hydrocarbon-consuming microorganisms so that they degrade mineral oil contamination in soil more quickly. Building on this knowledge, the teaching of **DE 44 37 313** proposes using phospholipids and phospholipid derivatives, above all in admixture with urea, to stimulate even
10 those microorganisms which live naturally in uncontaminated soil in order to promote the growth of the plants growing in the soils thus treated by a kind of general fertilizing effect.

Now, the invention is based on an observation which goes even further: the use of the above-mentioned mixtures to be described in detail
15 hereinafter provides for intervention in and hence biological control of the highly complex life system of the soil. The objective in this regard is the control and/or reduction of plant-parasitic nematode growth and, hence, the growth of selected strains of soil fauna by stimulation and hence growth promotion of selected strains of soil microorganism flora in the form of
20 bacteria and/or fungi.

It is known in principle that the so-called rhizosphere, i.e. the boundary layer of soil a few millimeters thick in the region of the plant roots and, in particular, the root hairs, harbours a high population of bacterial strains (microorganism flora) which, on the one hand, promote the
25 exchange of matter between the plant roots and the soil, but which on the other hand are also capable of protecting this life-sustaining in-soil part of the plants. The same applies to the mycorrhiza region of the interaction between plant roots and fungus populations in the soil. The teaching according to the invention is based inter alia on the observation that
30 addition of the soil additives according to the invention also strengthens

those microorganism populations and, hence, bacterial and/or fungal strains in the soil which develop nematostatic and/or nematicidal activity towards plant-parasitic nematodes and which therefore not only prevent or at least restrict the further build-up of nematode populations in the soil, but
5 also and above all limit the penetration of already existing nematodes into the plant roots. Irrespective of this and in addition to this hitherto unknown effect, the already known general fertilizing effect for promoting growth irrespective of nematodes remains intact for the use of the described mixtures.

10 This affords important advantages over the teaching according to US 5,057,141 and 4,536,207 cited above. The mixture of components (a), (b) and (c) introduced into the endangered areas of soil, preferably in the form of a liquid aqueous formulation, can be added to the soil both before and at least partly during the germination and/or growth phase(s) of the
15 plants to be protected. A corresponding treatment may be carried out just once and/or repeated as often as required, depending on demand. In another preferred embodiment of the invention, plant-physiologically compatible wetting agents, more especially of the o/w type, are added to the aqueous preparations of the soil additive. These wetting agents
20 support the spreading of the aqueous phase introduced in the upper layers of soil and, hence, especially in the growth region of the plant roots. It is possible in this way directly to stimulate the growth of the microorganism flora strains in the rhizosphere and/or corresponding mycorrhiza strains and hence to strengthen their nematostatic and/or nematicidal activity.
25 Accordingly, it will readily be appreciated that distinctly improved results can be achieved, even in the cultivation of crops particularly vulnerable to nematodes, such as sugar beet, by applying the teaching according to the invention.

The nutrient mixtures to be used in accordance with the invention
30 are characterized by a combination of selected representatives of class (a)

– compounds of phosphorus containing at least partly lipophilic organic radicals – and class (b) – urea and/or urea derivatives – as defined in the foregoing with the simultaneous and/or staggered concomitant use of the chitosans (c).

5 Preferred components of class (a) are esters of phosphoric acid with monohydric and/or polyhydric alcohols which have lipophilic radicals in their molecular structure. Partial esters of phosphoric acid are also particularly suitable, generally being used in the form of their (partial) salts.

Accordingly, suitable phosphoric acid esters are partial esters of
10 fatty alcohols which introduce the required lipophilic component into the phosphoric acid ester molecule through the hydrocarbon radical of the fatty alcohol. Partial esters of phosphoric acid with straight-chain fatty alcohols, of which at least a substantial proportion has been produced using C₈₋₁₀ fatty alcohols and/or lower ethoxylates thereof, are particularly suitable.
15 However, the phosphoric acid esters of higher fatty alcohols, for example containing 12 to 24 carbon atoms, are also suitable in principle, particular significance also attaching here to correspondingly olefinically unsaturated fatty alcohols.

However, particularly preferred phosphoric acid esters of class (a)
20 are phospholipids and phospholipid derivatives. It is known that phospholipids and their derivatives are amphiphilic substances which are obtained from vegetable or animal cells. Preferred phospholipids for the purposes of the teaching according to the invention are corresponding compounds of vegetable origin or phospholipid derivatives obtained
25 therefrom. Particularly preferred representatives of this class of components (a) are the glycerophospholipids which, normally, are also referred to as lecithin. The sphingophospholipids are less preferred. Known representatives which may be used in accordance with the invention are the diacyl phospholipids, phosphatidyl cholines, phosphatidyl
30 ethanolamines, phosphatidyl inositols, phosphatidyl serines, phosphatidyl

glycerols, phosphatidyl glycerophosphates, diphosphatidyl glycerol, N-acyl phosphatidyl ethanolamine and phosphatidic acid. Monoacyl phospholipids, lysophosphatidyl cholines, lysophosphatidyl ethanolamines, lysophosphatidyl inositols, lysophosphatidyl serines, lysophosphatidyl glycerols, lysophosphatidyl glycerophosphates, lysodiphosphatidyl glycerols, lyso-n-acyl phosphatidyl ethanolamines and lysophosphatidic acid are preferred. The phosphatidyl glycerides are obtainable on an industrial scale and available in large quantities. They are marketed as vegetable or animal lecithins and cephalins. These preparations are obtained, for example, from oils, such as corn oil, cottonseed oil or soybean oil. According to the invention, preferred components of class (a) are enzymatically hydrolyzed glycerophospholipids (enzymatically hydrolyzed lecithin) which have a more hydrophilic character through the elimination of a fatty acid ester. Only those products which have lost their phosphoric acid residue through the enzymatic hydrolysis are excluded.

According to the invention, urea and/or urea derivatives are introduced as an essential nitrogen source into the areas of soil to be protected as components of class (b) together with the phospholipids mentioned above. In its important embodiments, the teaching according to the invention is further characterized by an additional determining parameter which governs the quantity ratios in which components (a) and (b) are used. As a general rule, the mixtures of the particular components selected from these two classes are used in such quantity ratios that the calculable ratio by weight of carbon (C) to nitrogen (N) in the mixture of components (a) and (b) does not exceed about 6:1. Mixtures of component (a) to component (b) in which the ratio by weight of C to N is no more than 5:1 are preferred, mixtures in which that ratio by weight does not exceed 3:1 to 4:1 being particularly preferred. A suitable lower limit to the ratio by weight of C to N is 1:1 to 1.2:1, a maximum of about 2.5:1 and, more particularly, in the range from about 1.5:1 to 2:1 being particularly

preferred. Any nitrogen atoms present in component (a) are included in the calculation of the C:N ratio.

If phospholipids are used as representatives of the class (a) components – together with urea or urea derivatives as essential nitrogen
5 source – in the mixtures according to the invention, quantity ratios (expressed as parts by weight, based on the non-aqueous starting materials) of 3:1 to 1:3 parts of phospholipid to urea or urea derivative are normally suitable. In one preferred embodiment, substantially equal quantities of these basic components are used in admixture with one
10 another.

According to the invention, suitable urea derivatives are in particular the oligoureas or polyureas formed by condensation of the primary urea molecule. It is known that they are distinguished in practice by slow decomposition and hence by a long-term fertilizing effect. A long-term
15 effect such as this may be entirely desirable for the objective according to the invention of biogenic nematode control by strengthening the antagonistic and/or nematicidal potential of bacterial and/or fungal populations present in the soil. In general, however, urea as such is at least partly used as the nitrogen source. Reference is made in this
20 connection to another possible modification: other nitrogen sources known from fertilizer technology are also suitable as a constituent of the multicomponent mixtures to be used in accordance with the invention. Corresponding ammonium salts in particular are suitable in this regard, combinations of ammonium salts/urea being important nitrogen sources in
25 the context of the teaching according to the invention. Further references to suitable urea derivatives can be found, for example, in the above-cited US-PS 5,057,141, column 9, lines 40 to 50.

The chitosans or chitosan compounds (c) to be used for the purposes of the invention comprise the numerous components of this
30 (these) class(es) of substances known from the relevant published prior art

and mixtures thereof. In addition to the published prior art already mentioned, reference may be made to the following literature:

"Applications of Chitin and Chitosan", Mattheus F.A. Goosen (ed.) (Lancaster: Technomic Publishing Company, 1996) chapter 2 entitled

5 **"Applications of Chitin and Chitosan in the Ecological and Environmental Fields"**, edited by SHIGEHIRO HIRANO loc. cit., pages 31 to 54 and chapter 11 entitled **"New Applications of Chitin and Its Derivatives in Plant Protection"**, edited by HENRYK STRUSZCZYK et al., loc. cit, pages 171 to 184; and the publication in **"Biotechnology**

10 **Annual Review Volume 2"** Elsevier Science B.V. 1996, entitled **"Chitin Biotechnology Applications"**, edited by SHIGEHIRO HIRANO loc. cit., pages 237 to 258.

As already made clear on several occasions, the chitosans (c) to be used in accordance with the invention may be oligomers and/or polymers of

15 practically any desired molecular weight. In the manner already indicated, both low molecular weight oligomers and high molecular weight polymers can be converted into water-soluble forms which allow application to the soil and the spreading of the chitosans in the soil and especially into the region of the plants' roots. Particularly suitable chitosan compounds in this

20 connection can be those which also have sufficient water solubility especially in the neutral to weakly basic pH range and which have been modified in known manner, for example by salt formation with inorganic and/or organic acids, by lowering and regulation of the degree of polymerization or oligomerization and/or by derivatization of the chitosans

25 with solubilizing groups, for example carboxymethyl groups in the O- and/or N-position.

Although the cited published prior art also expressly describes the use of such chitosan compounds in the agricultural sector, including the use of corresponding aqueous chitosan formulations, the combination

30 essential to the invention, in terms of the multicomponent mixtures of

components (a), (b) and (c) defined according to the invention, coupled with the objective according to the invention, namely the effective biogenic control and limitation of the multiplication of parasitic soil nematodes in plant cultivation, is not disclosed in said published prior art relating to
5 chitosan or chitosan compounds which are also suitable according to the invention.

The introduction of chitosan components (c) into the soil regions to be treated can be carried out at the same time as the introduction of components (a) and (b) and/or at a different time. In one important
10 embodiment, however, provision is made for water-soluble chitosans or chitosan compounds to be introduced into the soil structure at least in part together with components (a) and (b). Another important feature pertaining precisely to the components (c) concomitantly used here according to the invention is that their spreading in the upper soil region, and especially in
15 the region of the plants' roots, is optimized. This normally implies the concomitant use of surfactant components, the preferred nature of which will be discussed below.

If chitosans are used in admixture with components (a) and (b), the quantities used here are at least 0.01% by weight, preferably at least
20 0.05% by weight and especially at least 0.1 to 1% by weight, based in each case on the water-free multicomponent mixture. The crucial point is that even such small quantities of chitosans can substantially influence the course of the vital processes in the soil; in particular, the growth of the healthy microbial flora is promoted and the growth of pests is suppressed.
25 Plant growth is promoted at the same time by stimulation/strengthening of the plants' own resistance.

The quantity in which the soil additive of components (a), (b) and (c) according to the invention is applied in each individual case is determined by a number of factors. These include inter alia the time at which the
30 multicomponent mixture is applied in dependence upon the planned or

existing cultivation program, the state of the soil, more especially in regard to the soil fauna already present and, in particular, the discernible nematode infestation, the sensitivity of the plants to be cultivated to the effect of plant-parasitic nematodes and the like. Suitable quantity ranges for the application and introduction of the soil additive according to the invention for protecting plants against nematode infestation are normally at least 0.2 to 0.5 g/m² soil surface area, based on the water-free combinations of phospholipids and urea or urea derivatives. Particularly preferred ranges for suitable quantities of the soil additives according to the invention, again based on the water-free mixtures, are above 1 g/m² to about 50-60 g/m² and preferably from 10 to 40 g/m² soil surface area.

The aqueous preparations of the soil additives preferably used in accordance with the invention are generally formulated as corresponding aqueous emulsions and/or dispersions. To produce these aqueous dispersions, to facilitate their penetration into the soil and, in particular, to spread the aqueous preparation in the soil, surface-active auxiliaries are also used in the aqueous preparations in the preferred embodiment of the invention. Suitable surface-active auxiliaries are, in particular, biologically compatible emulsifiers of the o/w type which, in turn, are degradable in particular through the usual metabolism processes of the soil microorganisms. Although corresponding anionic surfactants are suitable, nonionic surfactants that are rapidly and completely biodegradable are particularly preferred for the purposes of the invention.

Suitable anionic surfactants are, for example, soaps and biodegradable alkyl sulfates, more especially fatty alcohol sulfates. Suitable representatives are the partial esters of phosphoric acid with fatty alcohols and, in particular, corresponding partial esters with linear fatty alcohols of preferably natural origin and hence with an even number of carbon atoms. For example, corresponding esters of relatively short-chain fatty alcohols, for example containing 6 to 10 carbon atoms in the fatty alcohol molecule,

are suitable. However, alkyl phosphates with relatively long-chain fatty alcohol radicals, for example containing 12 to 24 carbon atoms, are also suitable. In the case under discussion, therefore, polyfunctional significance attaches to selected representatives of the class (a) components according to the invention. In addition to the interactions with the components of class (b) discussed above, use can be made here of the surface-active function of the phosphoric acid partial esters mentioned.

According to the invention, particularly preferred biodegradable surfactants are corresponding, at least predominantly nonionic compounds which are preferably based at least predominantly on natural materials and have preferred HLB values in the range from 10 to 18. According to the invention, a particularly preferred class of surfactants are the alkyl (oligo)glycoside compounds of which the alkyl chain derives at least predominantly from straight-chain fatty alcohols. Compounds of this type, which are now also known as APG components, are surface-active auxiliaries with a broad range of applications. It is known that APG-based wetting agents can be based entirely on natural materials. Reference is made to the extensive scientific knowledge and literature on the production and properties of APG compounds of the type in question, see for example the book by Hill et al. entitled **"Alkylpolyglycoside"**, VCH-Verlagsgesellschaft mbH, Weinheim 1997.

However, other biodegradable and compatible surfactant components with comparable properties are also suitable. These include in particular corresponding glucamines, glucamides or even sugar partial esters of monocarboxylic acids containing in particular 8 to 24 carbon atoms, sorbitan esters, for example of the sorbitan monostearate or sorbitan monooleate type, and also surfactants of biological origin. Examples of biological surfactants include sophorose lipid, trehalose lipid or the lipopeptides known as metabolism products or membrane constituents of a large number of microorganism strains. Further

particulars of biologically acceptable surface-active components can be found in DE 44 37 313 which has already repeatedly cited.

In another preferred embodiment of the invention, carbon sources for the growth of the microorganism flora are also added to the above-defined additives based on phospholipid/urea or urea derivative/chitosan and introduced into the affected area of soil, preferably together with the essential components mentioned above. The combined use of carriers containing P and/or N together with selected additional carbon sources for the growth of the microorganism flora is inter alia the subject of hitherto unpublished German patent application **DE 197 48 884.6** (H 3143), of which the disclosure is hereby included as part of the teaching of the present invention. Key statements on this additional class of carbon sources in the context of the multicomponent mixtures or soil additives according to the invention are summarized in the following. However, reference is made beforehand to an important preferred additional parameter for multicomponent mixtures of the type under discussion:

In the context of the teaching of the present invention, preferred multicomponent mixtures for introduction into the soil are those in which the quantity ratios of the C- and/or N-containing components used are adapted to one another in such a way that ratios by weight of C to N of about 6:1 and preferably of about 3 to 4:1 are not exceeded. It may even be of advantage to limit the C:N ratio present in the particular multicomponent mixture used to values of at most about 2 to 2.5:1.

Where the additional carbon sources mentioned above are used in the soil additives according to the invention, it can be important so far as the choice of components insoluble in the aqueous phase is concerned to select compounds which at least partly have pour points of or below 25 to 30°C and, more particularly, of or below 10 to 15°C. Suitable components such as these are, for example, olefinically unsaturated C₁₂₋₂₄ fatty alcohols, more particularly of natural origin. Particular significance attaches

in this regard to at least predominantly C_{16/18} fatty alcohols with a high degree of olefinic double bonds and solidification ranges of or below 20°C and preferably of or below 10 to 15°C.

Preferred representatives of such additional carbon sources are oil-
5 soluble, but biologically compatible organic compounds containing aliphatic and/or olefinically unsaturated and, in particular, at least predominantly linear hydrocarbon radicals containing at least 6 carbon atoms and, more particularly, at least 8 carbon atoms. Corresponding compounds functionalized with oxygen as hetero atom are particularly preferred. Typical
10 examples of such compounds are fatty alcohols and/or fatty acids or derivatives thereof, such as esters, more especially partial esters, ethers and/or amides. In the modification under discussion here where carbon sources are additionally used, additives which, in addition to carbon, also contain nitrogen in their molecular structure can be particularly suitable.
15 Typical examples are betaines, proteins and aminocarboxylic acids and derivatives and salts thereof.

As already mentioned, the use of additional carbon sources is a possible measure, but by no means a compulsory measure in the context of the teaching of the invention. The essence of the teaching of the
20 invention lies in strengthening the nematostatic/nematicidal effect of the multicomponent mixture – introduced as far as the rhizosphere through the liquid formulation – of components (a), (b) and (c) and, more particularly, of phospholipids and urea or urea derivatives. This effect is also achieved without using the additional carbon sources just mentioned.

25 The principle according to the invention of strengthening the growth of antagonistic and/or nematicidal microorganisms in the form of soil bacteria and/or fungi leads to rapidly pronounced effects, especially in cases where corresponding microorganism populations acting as starter cultures already exist in the areas of soil to be treated. Bearing in mind the
30 widespread occurrence of plant-parasitic nematode strains, a

correspondingly broad presence of microorganism populations suitable for the purposes of the invention can be assumed to exist in agricultural soil. However, use can additionally be made in known manner of contemporary scientific knowledge for introducing cultures of suitable bacterial and/or fungal strains with the required antagonistic or nematicidal effects as starter cultures into the area of soil to be treated. Reference may be made in this connection to general scientific knowledge, cf. for example the following publications: J. Coosemans (1991), **"Methods for Introducing *Verticillium chlamydosporium* into soil"** in: KERRY, B.R & CRUMP, D.H.: **Methods for Studying Nematophagus Fungi. IOBC/WPRS Bulletin XIV/2; 39-45** and A. Ciancio (1995), **"Observations on the Nematicidal Properties of Some Mycotoxins"**, *Fundam. Appl. Nematol.* 18(5); 451-454.

Another possible variant of the teaching of the invention lies in the combination of measures known from the prior art for controlling nematodes and the measures now being proposed in accordance with the present invention. In particular, the water-insoluble components or mixtures thereof, more particularly based on chitin or chitin/protein complexes, may first be incorporated in the area of soil to be treated, as proposed in the above-cited US-PSS 5,057,141 and 4,536,207 and the literature cited therein, the advantages of the multicomponent mixtures to be introduced in accordance with the invention in the form of an aqueous preparation then being additionally utilized, more especially in the course of plant growth. It will readily be appreciated that the biogenic strengthening and protective function of the microorganism strains to be strengthened in accordance with the invention are achieved in a particularly pronounced form by this combination of working steps.

In particular, however, the use of the components or multicomponent mixtures according to the invention may also be combined with the addition of conventional nematicides. These chemical agents, which are now

widely used, are the subject of numerous printed publications, cf. for example the book by H. Börner entitled **"Pflanzenkrankheiten und Pflanzenschutz (Plant Diseases and Plant Protection)"**, 5th Edition, Verlag Eugen Ulmer, Stuttgart, 1983, pages 128 to 132.

5 If the teaching according to the invention is applied in combination with conventional nematicides, the quantity in which these conventional chemicals are used can be substantially reduced without any danger of serious losses of effect. In this way, savings of known nematicides, which are normally produced by chemical syntheses, of at least 30% and
10 preferably of at least 50 to 75% can be made without any danger of losses of growth and/or yield.

 The multicomponent mixtures to be used in accordance with the invention may be prepared and stored in the form of aqueous concentrates which are diluted with water before application to the soil. According to the
15 invention, suitable concentrates of components (a) and (b) have the following composition for example: 10 to 30% by weight of lecithin and/or lecithin hydrolyzate; 10 to 30% by weight of urea or urea derivative; 1 to 10% by weight of surfactant, optionally other auxiliaries and, for the rest, water to 100% by weight. Concentrates of this type are preferably diluted
20 with water in a ratio of 1:20 to 1:100 before application to the soil to be protected. The aqueous preparation may be applied any known method. Both uniform distribution of the liquid phase over the soil surface and controlled injection, for example by the so-called Hydro-Ject process, are suitable. Even where the aqueous multicomponent mixtures are injected
25 into the soil, the surface-active components used in accordance with the invention promote horizontal spreading thereof. The same applies to the application of chitosan components (c) when they are introduced in admixture with components (a) and (b) and also when aqueous surfactant formulations containing only chitosans (c) are introduced.

30 When the multicomponent mixtures according to the invention are

applied or introduced with the simultaneous and/or staggered concomitant use of chitosans, different principles can come into play which indirectly influence the quantities of said multicomponent mixtures used in the operation concerned. Thus, in a first embodiment, the simultaneous and/or staggered introduction of the components or component mixtures defined according to the invention may be carried out only once over the particular plant growth period in question. However, the possibility, provided according to the invention, of working with water-soluble and/or water-emulsifiable multicomponent mixtures also opens the way to applying or introducing the nutrients or nutrient mixtures onto or into the soil several times at shorter or longer intervals. Thus it may be expedient repeatedly to apply comparatively small quantities of the nutrients according to the invention at intervals of at least 2 to 3 weeks, for example at intervals of 1 to 2 months. This procedure makes it possible to optimise efficacy even when only very low concentrations of nutrients or nutrient mixtures are introduced into the soil. Accordingly, the preferred lower limits to the quantities of nutrient mixtures according to the invention which are applied or introduced are at least 0.1 to 0.2 g/m². The earlier statements regarding the quantities which are preferably applied according to the invention are otherwise valid.

Examples

The following formulation I describes an aqueous, flowable and water-dilutable concentrate of chitosan-containing nutrient components in terms of the invention. The concentrate can be diluted with more water and can easily be introduced into the soil in this form for plant protection in terms of the teaching according to the invention.

The components denoted by trade names in this Table for formulation I are as follows:

Glucopon 600 UP	APG-based o/w wetting agent
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Lipotin NE

enzymatically hydrolyzed soya lecithin

- The aqueous chitosan solution used was a water-soluble chitosan polymer prepared by stirring 1 g of chitosan polymer with 5 ml of 1 molar HCl and 100 ml of water at room temperature until the chitosan had dissolved.

Formulation II

Raw material	Initial weight
Glucopon 600 UP	2 g
1% chitosan solution, acidic	58 ml
Lipotin NE	20 g
Urea	20 g
	100 g

- The aqueous active substance mixture indicated here contains 5.8 g of chitosan (solid) per kg of product.

- Suitable quantities of this product per application range, for example, from 2 g/m² to 40 g/m², a quantity of 20 g/m² preferably being chosen. This corresponds to a quantity of 0.116 g chitosan/m², based on chitosan added.

The above-described product mixture can be diluted, for example, with 6 to 50 times the quantity of water and can easily be introduced in this form onto or into the soil. The C:N:P ratio in this case is 17:10:0.4.