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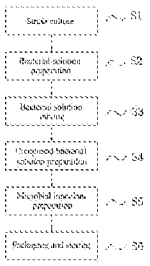
(54) **MICROBIAL INOCULANT FOR COMPOSTING LIVESTOCK AND POULTRY MANURE AND PREPARATION METHOD THEREOF**

(57) The present invention discloses a microbial inoculant for composting livestock and poultry manure and a preparation method thereof. The present invention relates to the technical field of microbial inoculants, including the following components: bacillus subtilis, azotobacter vinelandii, paecilomyces lilacinus, bacillus coagulans, trichoderma harzianum, streptomyces microflavus, trichoderma, organophosphorus bacteria, potassium bacteria and rhizobium astragali, by the use of this inoculant, it may not only have the ability of improving the survivability in a soil environment, converting nitrogen and potassium in soil, improving the soil fertility, and stimulating the crop growth, but also reduce the amount of chemical fertilizers, inhibit the reproduction of germs, and achieve the three-in-one effect of increasing the production and income, inhibiting diseases, and improving the quality, so the input cost is reduced.



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MICROBIAL INOCULANT FOR COMPOSTING LIVESTOCK AND POULTRY MANURE AND PREPARATION METHOD THEREOF

TECHNICAL FIELD

The present invention relates to the technical field of microbial inoculants, in particular to a microbial inoculant for composting livestock and poultry manure and a preparation method thereof.

BACKGROUND ART

Existing microbial inoculants for composting livestock and poultry manure have relatively tedious production steps and relatively complicated operations, which not only increase the input cost, but also produce finished products with relatively weak bactericidal and bacteriostatic effects, and the survivability in a soil environment is relatively poor, so it may not help the growth of crops better. For this reason, persons skilled in the art provide a microbial inoculant for composting livestock and poultry manure and a preparation method thereof.

SUMMARY

In view of the deficiencies of the prior art, the present invention provides a microbial inoculant for composting livestock and poultry manure and a preparation method thereof, as to solve problems that existing microbial inoculants for composting livestock and poultry manure are relatively high in input cost, finished products produced are relatively weak in bactericidal and bacteriostatic effects, and the survivability in a soil environment is relatively poor, so it may not help the growth of crops better.

In order to achieve the above purpose, the present invention is achieved by the following technical schemes: a microbial inoculant for composting livestock and poultry manure, including the following components: bacillus subtilis, azotobacter vinelandii, paecilomyces lilacinus, bacillus coagulans, trichoderma harzianum,

streptomyces microflavus, trichoderma, organophosphorus bacteria, potassium bacteria and rhizobium astragali; and

the microbial inoculant for composting the livestock and poultry manure is prepared from the following components in parts by mass: 2.0-3.0 parts of the bacillus subtilis, 2.6-3.7 parts of the azotobacter vinelandii, 1.8-2.4 parts of the paecilomyces lilacinus, 3.3-4.0 parts of the bacillus coagulans, 2.1-3.1 parts of the trichoderma harzianum, 4.0-4.2 parts of the streptomyces microflavus, 0.6-0.8 parts of the trichoderma, 0.3-0.4 parts of the organophosphorus bacteria, 0.3-0.4 parts of the potassium bacteria and 0.5-1.0 part of the rhizobium astragali.

As a further technical scheme of the present invention, it is prepared from the following components in parts by mass: 2.0 parts of the bacillus subtilis, 2.6 parts of the azotobacter vinelandii, 1.8 parts of the paecilomyces lilacinus, 3.3 parts of the bacillus coagulans, 2.1 parts of the trichoderma harzianum, 4.0 parts of the streptomyces microflavus, 0.6 parts of the trichoderma, 0.3 parts of the organophosphorus bacteria, 0.3 parts of the potassium bacteria and 0.5 parts of the rhizobium astragali.

As a further technical scheme of the present invention, it is prepared from the following components in parts by mass: 2.5 parts of the bacillus subtilis, 3.0 parts of the azotobacter vinelandii, 2.1 parts of the paecilomyces lilacinus, 3.6 parts of the bacillus coagulans, 2.6 parts of the trichoderma harzianum, 4.1 parts of the streptomyces microflavus, 0.7 parts of the trichoderma, 0.35 parts of the organophosphorus bacteria, 0.35 parts of the potassium bacteria and 0.7 parts of the rhizobium astragali.

As a further technical scheme of the present invention, it is prepared from the following components in parts by mass: 3.0 parts of the bacillus subtilis, 3.7 parts of the azotobacter vinelandii, 2.4 parts of the paecilomyces lilacinus, 4.0 parts of the bacillus coagulans, 3.1 parts of the trichoderma harzianum, 4.2 parts of the streptomyces microflavus, 0.8 parts of the trichoderma, 0.4 parts of the organophosphorus bacteria, 0.4 parts of the potassium bacteria and 1.0 part of the rhizobium astragali.

As a further technical scheme of the present invention, the streptomyces microflavus is mainly fermented from a liquid culture

medium of soybean cake powder, glucose, a calcium hydrochloride and a sodium chloride.

As a further technical scheme of the present invention, the content of the soybean cake powder is 3 g, the content of the glucose is 3 g, the content of the calcium hydrochloride is 0.5 g, and the content of the sodium chloride is 0.4 g.

As a further technical scheme of the present invention, the azotobacter vinelandii and the potassium bacteria are both cultured in an Ashby medium, the trichoderma is cultured in a potato sucrose medium, the organophosphorus bacteria are cultured in a lecithin medium, and the rhizobium astragali is cultured in a yeast juice sucrose medium.

The preparation method for the above microbial inoculant for composting the livestock and poultry manure includes the following steps:

S1. strain culture: inoculating the bacillus subtilis, azotobacter vinelandii, paecilomyces lilacinus, bacillus coagulans, trichoderma harzianum, streptomyces microflavus, trichoderma, organophosphorus bacteria, potassium bacteria and rhizobium astragali respectively in each culture medium for culturing;

S2. bacterial solution preparation: putting the various bacterial strains cultured in the Step S1 into a drug bottle of a seed culture medium in a mode of slant surface inoculation, and using a liquid culture medium for culturing separately, to obtain bacterial solution of the bacterial strain;

S3. bacterial solution mixing: putting all types of the bacterial solution prepared in the Step S2 into a fermentation tank loaded with seeds, stirring and mixing, to obtain composite seed solution;

S4. compound bacterial solution preparation: inoculating all the seed solution in the Step S3 into a seed tank loaded with a fermentation culture medium for fermenting and culturing, to obtain compound bacterial solution;

S5. microbial inoculant preparation: drying and powder-spraying the compound bacterial solution in the Step S4, to obtain a finished product microbial inoculant; and

S6. packaging and storing: adding the finished product micro-

bial inoculant in the Step S5 to a filling machine, packaging it into a container by using the filling machine, and putting it in a storage.

BENEFICIAL EFFECT

The present invention provides a microbial inoculant for composting livestock and poultry manure and a preparation method thereof. Compared with the prior art, it has the following beneficial effects:

1. A microbial inoculant for composting livestock and poultry manure and a preparation method thereof, by using this inoculant, it may not only have the ability of improving the survivability in the soil environment, converting nitrogen and potassium in soil, improving the soil fertility, and simulating the crop growth, but also reduce the amount of chemical fertilizers, inhibit the reproduction of germs, and achieve the three-in-one effect of increasing the production and income, inhibiting diseases, and improving the quality, so the input cost is reduced.

2. A microbial inoculant for composting livestock and poultry manure and a preparation method thereof, by adding the trichoderma harzianum, a cell wall degrading enzyme of pathogenic bacteria may be secreted, and the cell wall of the pathogenic bacteria may be destroyed, thereby the pathogenic bacteria are killed; by adding the paecilomyces lilacinus, it has the efficacy of preventing and controlling a plurality of nematodes; and by adding the bacillus coagulans, it has an anti-insect function to plants, thereby the immunity of crops is improved, and the disease resistance of the crops is strengthened.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 is a flow diagram of a preparation method of the present invention.

DETAILED DESCRIPTION OF THE EMBODIMENTS

Embodiment 1

The present invention provides a technical scheme: a microbial inoculant for composting livestock and poultry manure, which is

prepared from the following components in parts by mass: 2.0 parts of bacillus subtilis, 2.6 parts of azotobacter vinelandii, 1.8 parts of paecilomyces lilacinus, 3.3 parts of bacillus coagulans, 2.1 parts of trichoderma harzianum, 4.0 parts of streptomyces microflavus, 0.6 parts of trichoderma, 0.3 parts of organophosphorus bacteria, 0.3 parts of potassium bacteria and 0.5 parts of rhizobium astragali.

The streptomyces microflavus is mainly fermented from a liquid culture medium of soybean cake powder, glucose, a calcium hydrochloride and a sodium chloride.

The content of the soybean cake powder is 3 g, the content of the glucose is 3 g, the content of the calcium hydrochloride is 0.5 g, and the content of the sodium chloride is 0.4 g.

The azotobacter vinelandii and the potassium bacteria are both cultured in an Ashby medium, the trichoderma is cultured in a potato sucrose medium, the organophosphorus bacteria are cultured in a lecithin medium, and the rhizobium astragali is cultured in a yeast juice sucrose medium.

A preparation method for the above microbial inoculant for composting the livestock and poultry manure includes the following steps:

S1. strain culture: inoculating the bacillus subtilis, azotobacter vinelandii, paecilomyces lilacinus, bacillus coagulans, trichoderma harzianum, streptomyces microflavus, trichoderma, organophosphorus bacteria, potassium bacteria and rhizobium astragali respectively in each culture medium for culturing;

S2. bacterial solution preparation: putting the various bacterial strains cultured in the Step S1 into a drug bottle of a seed culture medium in a mode of slant surface inoculation, and using a liquid culture medium for culturing separately, to obtain bacterial solution of the bacterial strain;

S3. bacterial solution mixing: putting all types of the bacterial solution prepared in the Step S2 into a fermentation tank loaded with seeds, stirring and mixing, to obtain composite seed solution;

S4. compound bacterial solution preparation: inoculating all the seed solution in the Step S3 into a seed tank loaded with a

fermentation culture medium for fermenting and culturing, to obtain compound bacterial solution;

S5. microbial inoculant preparation: drying and powder-spraying the compound bacterial solution in the Step S4, to obtain a finished product microbial inoculant; and

S6. packaging and storing: adding the finished product microbial inoculant in the Step S5 to a filling machine, packaging it into a container by using the filling machine, and putting it in a storage.

Embodiment 2

The present invention provides a technical scheme: a microbial inoculant for composting livestock and poultry manure, which is prepared from the following components in parts by mass: 2.5 parts of bacillus subtilis, 3.0 parts of azotobacter vinelandii, 2.1 parts of paecilomyces lilacinus, 3.6 parts of bacillus coagulans, 2.6 parts of trichoderma harzianum, 4.1 parts of streptomyces microflavus, 0.7 parts of trichoderma, 0.35 parts of organophosphorus bacteria, 0.35 parts of potassium bacteria and 0.7 parts of rhizobium astragali.

The streptomyces microflavus is mainly fermented from a liquid culture medium of soybean cake powder, glucose, a calcium hydrochloride and a sodium chloride.

The content of the soybean cake powder is 3 g, the content of the glucose is 3 g, the content of the calcium hydrochloride is 0.5 g, and the content of the sodium chloride is 0.4 g.

The azotobacter vinelandii and the potassium bacteria are both cultured in an Ashby medium, the trichoderma is cultured in a potato sucrose medium, the organophosphorus bacteria are cultured in a lecithin medium, and the rhizobium astragali is cultured in a yeast juice sucrose medium.

A preparation method for the above microbial inoculant for composting the livestock and poultry manure is the same as Embodiment 1.

Embodiment 3

The present invention provides a technical scheme: a microbial inoculant for composting livestock and poultry manure, which is prepared from the following components in parts by mass: 3.0 parts

of bacillus subtilis, 3.7 parts of azotobacter vinelandii, 2.4 parts of paecilomyces lilacinus, 4.0 parts of bacillus coagulans, 3.1 parts of trichoderma harzianum, 4.2 parts of streptomyces microflavus, 0.8 parts of trichoderma, 0.4 parts of organophosphorus bacteria, 0.4 parts of potassium bacteria and 1.0 part of rhizobium astragali.

The streptomyces microflavus is mainly fermented from a liquid culture medium of soybean cake powder, glucose, a calcium hydrochloride and a sodium chloride.

The content of the soybean cake powder is 3 g, the content of the glucose is 3 g, the content of the calcium hydrochloride is 0.5 g, and the content of the sodium chloride is 0.4 g.

The azotobacter vinelandii and the potassium bacteria are both cultured in an Ashby medium, the trichoderma is cultured in a potato sucrose medium, the organophosphorus bacteria are cultured in a lecithin medium, and the rhizobium astragali is cultured in a yeast juice sucrose medium.

A preparation method for the above microbial inoculant for composting the livestock and poultry manure includes the following steps:

S1. strain culture: inoculating the bacillus subtilis, azotobacter vinelandii, paecilomyces lilacinus, bacillus coagulans, trichoderma harzianum, streptomyces microflavus, trichoderma, organophosphorus bacteria, potassium bacteria and rhizobium astragali respectively in each culture medium for culturing;

S2. bacterial solution preparation: putting the various bacterial strains cultured in the Step S1 into a drug bottle of a seed culture medium in a mode of slant surface inoculation, and using a liquid culture medium for culturing separately, to obtain bacterial solution of the bacterial strain;

S3. bacterial solution mixing: putting all types of the bacterial solution prepared in the Step S2 into a fermentation tank loaded with seeds, stirring and mixing, to obtain composite seed solution;

S4. compound bacterial solution preparation: inoculating all the seed solution in the Step S3 into a seed tank loaded with a fermentation culture medium for fermenting and culturing, to ob-

tain compound bacterial solution;

S5. microbial inoculant preparation: drying and powder-spraying the compound bacterial solution in the Step S4, to obtain a finished product microbial inoculant; and

S6. packaging and storing: adding the finished product microbial inoculant in the Step S5 to a filling machine, packaging it into a container by using the filling machine, and putting it in a storage.

I. Test method for antagonism of actinomyces microflavus:

① a tube of the azotobacter vinelandii, the organophosphorus bacteria, the potassium bacteria, the rhizobium astragali and the trichoderma is taken each;

② bacterial cells are washed out respectively with sterile water, 1 mm of bacterial suspension is absorbed each, mixed uniformly with an uncured agar medium and poured into a culture dish; and

③ after being cured, fermentation broth is dipped with a circular filter paper sheet with a diameter of 1 cm and it sticks on the medium, after being placed at 26-28°C for 5-7 days, the size of an inhibition zone is observed.

Results are shown in the following table:

Bacterial strain	Diameter of inhibition zone (mm)
Azotobacter vinelandii	0(dense bacterial lawn with a width of 6 mm)
Organophosphorus bacteria	0
Potassium bacteria	0
Rhizobium astragali	0
Trichoderma	17-18

From the test results in the above table, it may be seen that the actinomyces microflavus has the apparent inhibitory effect on the trichoderma, but has no inhibitory effect on the potassium bacteria, the organophosphorus bacteria and the rhizobium astragali, and not only has no inhibitory effect on the azotobacter vinelandii, but also causes the azotobacter vinelandii to generate a dense bacterial lawn with a thickness of 6 mm around the filter paper sheet, so it has the effect of stimulating growth.

It should be noted that, in this article, relationship terms

such as first and second are only used to distinguish one entity or operation from another entity or operation, and do not necessarily require or imply any such actual relationship or sequence existing between these entities or operations. In addition, terms "including", "containing" or any other variations thereof are intended to encompass non-exclusive inclusion, such that a process, a method, an object or a device including a series of elements includes not only those elements, but also includes other elements that are not explicitly listed, or elements inherent to such a process, method, object or device.

Although the embodiments of the present invention are already shown and described, it may be understood by those skilled in the art that various changes, modifications, replacements and variations may be made to these embodiments without departing from the principle and spirit of the present invention, and the scope of the present invention is defined by the appended claims and equivalents thereof.

C O N C L U S I E S

1. Microbieel inoculant voor het composteren van mest van vee en pluimvee, met het kenmerk, dat het wordt bereid uit de volgende bestanddelen in massadelen: 2,0 tot 3,0 delen bacillus subtilis, 2,6 tot 3,7 delen azotobacter vinelandii, 1,8 tot 2,4 delen paecilomyces lilacinus, 3,3 tot 4,0 delen bacillus coagulans, 2,1 tot 3,1 delen trichoderma harzianum, 4,0 tot 4,2 delen streptomyces microflavus, 0,6 tot 0,8 delen trichoderma, 0,3 tot 0,4 delen organofosforbacteriën, 0,3 tot 0,4 delen kaliumbacteriën en 0,5 tot 1,0 onderdeel van rhizobium astragali.

2. Microbieel inoculant voor het composteren van mest van vee en pluimvee volgens conclusie 1, met het kenmerk, dat de streptomyces microflavus hoofdzakelijk wordt gefermenteerd uit een vloeibaar kweekmedium van sojabonenkoekpoeder, glucose, een calciumhydrochloride en een natriumchloride.

3. Microbieel inoculant voor het composteren van mest van vee en pluimvee volgens conclusie 1 of 2, met het kenmerk, dat een bereidingswijze de volgende stappen omvat:

S1. stamcultuur: het enten van de bacillus subtilis, azotobacter vinelandii, paecilomyces lilacinus, bacillus coagulans, trichoderma harzianum, streptomyces microflavus, trichoderma, organofosfor bacteriën, kaliumbacteriën en rhizobium astragali respectievelijk in elk kweekmedium voor het kweken;

S2. bereiding van bacteriële oplossing: het plaatsen van de verschillende bacteriestammen die in stap S1 zijn gekweekt in een medicijnfles van een zaadkweekmedium in een modus van inoculatie met schuin oppervlak, en het gebruiken van een vloeibaar kweekmedium voor afzonderlijk kweken, om een bacteriële oplossing van de bacteriestam te verkrijgen;

S3. menging van bacteriële oplossing: alle typen van de in stap S2 bereide bacteriële oplossing in een met zaden beladen fermentatietank doen, roeren en mengen, om een samengestelde zaadoplossing te verkrijgen;

S4. bereiding van samengestelde bacteriële oplossing: het enten van alle zaadoplossing in stap S3 in een zaadtank die is beladen met een fermentatie kweekmedium voor fermentatie en kweken, om een samengestelde bacteriële oplossing te verkrijgen;

S5. bereiding van microbieel inoculant: het drogen en poedersproeien van de samengestelde bacteriële oplossing in stap S4, om een microbieel inoculant als eindproduct te verkrijgen; en

S6. verpakking en opslag: het microbiële inoculant van het eindproduct in de stap S5 toevoegen aan een vulmachine, het in een container verpakken met behulp van de vulmachine en het in een opslag plaatsen.

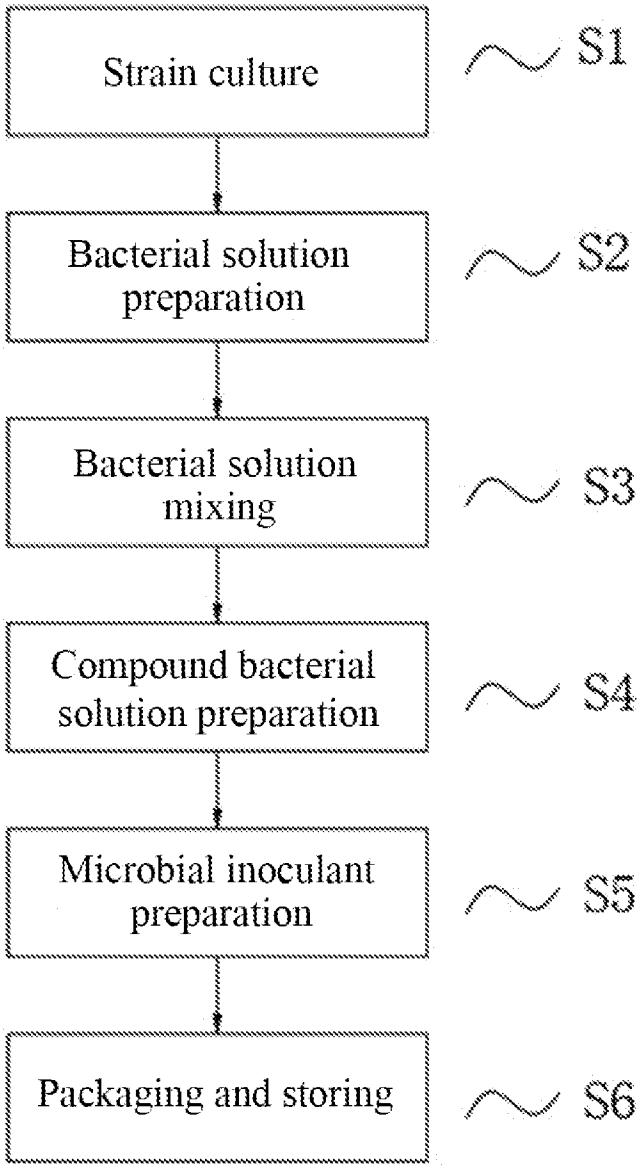
Strain culture
S1
Bacterial solution preparation
S2
Bacterial solution mixing
S3
Compound bacterial solution preparation
S4
Microbial inoculant preparation
S5
Packaging and storing
S6

FIG. 1

Rijksdienst voor Ondernemend Nederland

OCTROOIAANVRAAG NR.:
NO 142214
NL 2032828

ONDERZOEKSRAPPORT

BETREFFENDE HET RESULTAAT VAN HET ONDERZOEK NAAR DE STAND VAN DE TECHNIEK

RELEVANTE LITERATUUR

Categorie[1]	Literatuur met, voor zover nodig, aanduiding van speciaal van belang zijnde tekstgedeelten of figuren.	Van belang voor conclusie(s) nr:	Classificatie(IPC)
Y	CN 108 484 267 A (UNIV NORTHEAST AGRICULTURAL) 4 september 2018 (2018-09-04) * conclusies; voorbeelden * -----	1-3	INV. C05F17/20 C12N1/14 C12N1/20
Y	WO 2017/184810 A1 (KOCH AGRONOMIC SERVICES LLC) 26 oktober 2017 (2017-10-26) * bladzijde 3, regel 29 - bladzijde 11, regel 16; conclusies * -----	1-3	
Y	GREFF BABETT ET AL: "Influence of microbial inoculants on co-composting of lignocellulosic crop residues with farm animal manure: A review", JOURNAL OF ENVIRONMENTAL MANAGEMENT, deel 302, 16 november 2021 (2021-11-16), bladzijde 114088, XP093032716, AMSTERDAM, NL ISSN: 0301-4797, DOI: 10.1016/j.jenvman.2021.114088 * bladzijde 4, kolom 1, alinea 1 - bladzijde 6, kolom 1, alinea 3; tabel 1 * ----- -/--	1-3	

Onderzochte gebieden van de techniek

C05F
C12R
C05G
C12N

Indien gewijzigde conclusies zijn ingediend, heeft dit rapport betrekking op de conclusies ingediend op:

Plaats van onderzoek:	Datum waarop het onderzoek werd voltooid:	Bevoegd ambtenaar:
München	20 maart 2023	Burkhardt, Peter

[1] NDERLINCATEGORIE VAN DE VERMELDE LITERATUUR

X: de conclusie wordt als niet nieuw of niet inventief beschouwd ten opzichte van deze literatuur
Y: de conclusie wordt als niet inventief beschouwd ten opzichte van de combinatie van deze literatuur met andere geciteerde literatuur van dezelfde categorie, waarbij de combinatie voor de vakman voor de hand liggend wordt geacht
A: niet tot de categorie X of Y behorende literatuur die de stand van de techniek beschrijft
O: niet-schriftelijke stand van de techniek
P: tussen de voorrangsdatum en de indieningsdatum gepubliceerde literatuur
T: na de indieningsdatum of de voorrangsdatum gepubliceerde literatuur die niet bezwarend is voor de octrooiaanvrage, maar wordt vermeld ter verheldering van de theorie of het principe dat ten grondslag ligt aan de uitvinding
E: eerdere octrooi(aanvrage), gepubliceerd op of na de indieningsdatum, waarin dezelfde uitvinding wordt beschreven
D: in de octrooiaanvrage vermeld
L: om andere redenen vermelde literatuur
&: lid van dezelfde octrooifamilie of overeenkomstige octrooipublicatie

RELEVANTE LITERATUUR

Categorie[1]	Literatuur met, voor zover nodig, aanduiding van speciaal van belang zijnde tekstgedeelten of figuren.	Van belang voor conclusie(s) nr:
Y	ZHOU CHENG ET AL: "A new strategy for co-composting dairy manure with rice straw: Addition of different inocula at three stages of composting", WASTE MANAGEMENT., deel 40, 30 maart 2015 (2015-03-30), bladzijden 38-43, XP093032768, US ISSN: 0956-053X, DOI: 10.1016/j.wasman.2015.03.016 * bladzijde 39, kolom 2, alinea 2 - bladzijde 40, kolom 1, alinea 1 *	1-3
A	CN 111 646 837 A (HUBEI LYUDAO AGRICULTURE DEV CO LTD) 11 september 2020 (2020-09-11) * het gehele document *	1-3
A	CN 1 137 505 A (SUN KEYONG [CN]) 11 december 1996 (1996-12-11) * het gehele document *	1-3

[1] CATEGORIE VAN DE VERMELDE LITERATUUR

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Y: de conclusie wordt als niet inventief beschouwd ten opzichte van de combinatie van deze literatuur met andere geciteerde literatuur van dezelfde categorie, waarbij de combinatie voor de vakman voor de hand liggend wordt geacht
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O: niet-schriftelijke stand van de techniek
P: tussen de voorrangsdatum en de indieningsdatum gepubliceerde literatuur
T: na de indieningsdatum of de voorrangsdatum gepubliceerde literatuur die niet bezwarend is voor de octrooiaanvrage, maar wordt vermeld ter verheldering van de theorie of het principe dat ten grondslag ligt aan de uitvinding
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D: in de octrooiaanvrage vermeld
L: om andere redenen vermelde literatuur
&: lid van dezelfde octrooifamilie of overeenkomstige octrooipublicatie

AANHANGSEL BEHORENDE BIJ HET RAPPORT BETREFFENDE HET ONDERZOEK NAAR DE STAND VAN DE TECHNIEK, UITGEVOERD IN DE OCTROOIAANVRAGE NR.

NO 142214
NL 2032828

Het aanhangsel bevat een opgave van elders gepubliceerde octrooiaanvragen of octrooien (zogenaamde leden van dezelfde octrooifamilie), die overeenkomen met octrooischriften genoemd in het rapport.
De opgave is samengesteld aan de hand van gegevens uit het computerbestand van het Europees Octrooibureau per
De juistheid en volledigheid van deze opgave wordt noch door het Europees Octrooibureau, noch door het Bureau voor de Industriële eigendom gegarandeerd;; de gegevens worden verstrekt voor informatiedoeleinden.

20-03-2023

In het rapport genoemd octrooigeschrift	Datum van publicatie	Overeenkomend(e) geschrift(en)	Datum van publicatie
CN 108484267 A	04-09-2018	GEEN	
WO 2017184810 A1	26-10-2017	US 2019124917 A1	02-05-2019
		WO 2017184810 A1	26-10-2017
CN 111646837 A	11-09-2020	GEEN	
CN 1137505 A	11-12-1996	GEEN	

EPO FORM P0466

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SCHRIFTELIJKE OPINIE

DOSSIER NUMMER	INDIENINGSDATUM	VOORRANGSDATUM	AANVRAAGNUMMER
NO142214	23.08.2022		NL2032828

CLASSIFICATIE
INV. C05F17/20 C12N1/14 C12N1/20

AANVRAGER
Shandong Agricultural University, et al

Deze schriftelijke opinie bevat een toelichting op de volgende onderdelen:

- ☒ Onderdeel I Basis van de schriftelijke opinie
- ☐ Onderdeel II Voorrang
- ☐ Onderdeel III Vaststelling nieuwheid, inventiviteit en industriële toepasbaarheid niet mogelijk
- ☐ Onderdeel IV De aanvraag heeft betrekking op meer dan één uitvinding
- ☒ Onderdeel V Gemotiveerde verklaring ten aanzien van nieuwheid, inventiviteit en industriële toepasbaarheid
- ☐ Onderdeel VI Andere geciteerde documenten
- ☐ Onderdeel VII Overige gebreken
- ☒ Onderdeel VIII Overige opmerkingen

DE BEVOEGDE AMBTENAAR
Burkhardt, Peter

Aanvraag nr.:

SCHRIFTELIJKE OPINIE

NL2032828

Onderdeel I Basis van de Schriftelijke Opinie

1. Deze schriftelijke opinie is opgesteld op basis van de meest recente conclusies ingediend voor aanvang van het onderzoek.

2. Deze motivering is opgesteld, met betrekking tot **nucleotide- en/of aminozuursequenties** die genoemd worden in de aanvraag, op basis van een sequentielijst die:

   a. ☐ is opgenomen in de aanvraag zoals deze oorspronkelijk is ingediend

   b. ☐ aangeleverd is na de indieningsdatum ten behoeve van het onderzoek

      ☐ en vergezeld ging van een verklaring dat de sequentielijst niet meer informatie bevat dan de aanvraag zoals deze oorspronkelijk is ingediend.

3. ☐ Deze motivering is opgesteld, met betrekking tot nucleotide- en/of aminozuursequenties die genoemd worden in de aanvraag, voor zover een zinvolle motivering gevormd kon worden zonder een sequentielijst die voldeed aan WIPO standaard ST.26.

4. Overige opmerkingen:

Onderdeel V Gemotiveerde verklaring ten aanzien van nieuwheid, inventiviteit en industriële toepasbaarheid

1. Verklaring

Nieuwheid	Ja:	Conclusies 1-3
	Nee:	Conclusies
Inventiviteit	Ja:	Conclusies
	Nee:	Conclusies 1-3
Industriële toepasbaarheid	Ja:	Conclusies 1-3
	Nee:	Conclusies

2. Citaties en toelichting:

   Zie aparte bladzijde

Onderdeel VIII Overige opmerkingen

De volgende opmerkingen met betrekking tot de duidelijkheid van de conclusies, beschrijving, en figuren, of met betrekking tot de vraag of de conclusies nawerkbaar zijn, worden gemaakt:

Zie aparte bladzijde

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1 The following documents (D) are listed in this written opinion. The numbering follows the order of the Search Report.

D1 CN 108484267 (University Northeast Agriculture)
D2 WO 2017/184810 (Koch Agronomic Services)
D3 Greff *et al.*, 2022. J. Env. Mgmt. 30:114088
D4 Zhou *et al*, 40:38-43. Waste Mgmt. 40:38-43
D5 CN 111646837 (Hubei Livdao Agricultural Development Co Ltd.)
D6 CN 111646837 (Sun *et al.*)

2 Claim 1 is directed to a microbial inoculant comprising *Bacillus subtilis*, *Azotobacter vinelandii, Paecilomyces lilacinus*, *Bacillus coagulans*, *Trichoderma harzianum*, *Streptomyces microflavus*, *Trichoderma*, so called organophosphorous bacteria, so called potassium bacteria and *Rhizobium astragali*.

3 The closest prior art to the subject-matter of claim 1 is any of D1 (Examples; claims) or D2 (page 3, line 29 - page 11, line 16; claims). Both documents disclose microbial inoculants comprising different microorganisms. The objective technical problem may thus be formulated as the provision of an alternative microbial composition.

4 The composition of claim 1 appears to solve that technical problem. The solution, however, lacks an inventive step because the provision of an alternative *per se* is not inventive.

5 A wide variety of alternative compositions were disclosed in D3 (page 4, first column, first paragraph - page 6, first column, third paragraph; Table 1) and D4 (page 39, second column, second paragraph - page 40, first column, first paragraph). The provision of just a further alternative lacks an inventive step

6 In order to be patentable, the selection of an alternative must not be arbitrary but must be justified by the technical purpose, i.e. a hitherto unknown or unexpected technical effect which is caused by those features distinguishing the method of the invention from the prior art. This is not the case at present.

7 Claim 1 does not involve an inventive step. The same holds true for dependent claims 2 and 3.

Re Item VIII

Certain observations on the application

1 The subject-matter of claim 1 is completely unclear insofar it relates to "organophosphorus bacteria" or "potassium bacteria". Both designations are unknown to the person skilled in the art. Moreover, an undue burden is placed on others trying to carry out the claimed invention.