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(54) **Stable extracts from neem seeds**

Haltbare Extrakte aus Neemkernen

Extraits stables de graines de neem

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Description

This invention relates to stabilized extracts of the seeds of the neem tree (*Azadirachta indica* Juss.) which provide materials useful as insecticides, compositions containing such extracts and methods of their use.

The search for compositions which have a combination of excellent insecticidal activity and desirable low toxicity to plants and mammals is a continuing one because of factors such as the desire for compounds exhibiting greater insecticidal activity, better selectivity, low environmental impact, low production cost and effectiveness against insects resistant to many known insecticides.

Various parts of the neem (or nim) tree have long been used in India for their reputed medicinal or insecticidal properties. This subtropical tree is native to the arid regions of India, Pakistan, Sri Lanka and parts of Southeast Asia and Africa.

Although all parts of the neem tree appear to have natural resistance to pests and diseases, the seeds appear to have the greatest resistance. Formulations and extracts of the seeds have been shown to be effective against many species of crop pests including gypsy moths, Japanese beetles, aphids, tobacco budworms and boll weevils. For example, see *Chem. and Engineering News*, May 27, 1985, pp. 46-51 and *U.S. Dep. Agric., Agric. Rev. Man.*, ARM-NE-4. Neem seed extract is considered to be a broad-spectrum insecticide.

While neem seed extract has been shown to be active as an insecticide, it has not come into common use because of the known instability of the formulated extract. Moreover, undesirable aflatoxins, arising from fungal infection of the seed, may be present in the extracts.

Although laboratory scale procedures resulting in extract preparations containing over 25% azadirachtin have been described in the literature, a practical, economical and scalable method has been lacking.

Prior processes for obtaining crude neem extracts typically comprised the steps of:

a) extracting the ground seed with a polar organic solvent such as methanol or ethanol followed by filtration,
b) removing the solvent from the filtrate by evaporation under reduced pressure to yield a dry extract,
c) dissolving the dry extract in a mixture of a water-immiscible polar organic solvent, such as ethyl acetate, and aqueous saturated sodium chloride solution and
d) separating the organic layer, drying and evaporating the solvent to yield the semi-pure extract.

Such prior processes result in extracts containing a significant portion of hydrophobic organic impurities and no more than 10% azadirachtin. Higher purity preparations have been described but they require uneconomical, non-scalable chromatographic steps subsequent to those steps described above.

Several attempts have been made by various workers with the objective of removing the hydrophobic materials without resorting to chromatography. For example, *Journal of Liquid Chromatography*, 10 (6), 1151 (1987) shows an attempt to purify a crude methanolic extract by dissolving the extract in 50% aqueous methanol and then extracting the resulting solution with hexane. Other authors have described the removal of oil from the ground seed with a non-polar solvent such as hexane prior to the polar solvent extraction. However, these methods are not efficient since the crude methanolic or ethanolic extracts, apart from hexane soluble oils, also contain large quantities of hydrophobic impurities which are only sparingly soluble in non-polar solvents such as hexane.

In U.S. 4,556,562, an aqueous storage-stable neem seed extract composition is disclosed. In U.S. 4,946,681, a method is disclosed for stabilizing an alcoholic neem seed extract by removing water with molecular sieves.

Notwithstanding the above advances, there remains a need for an economical, stable composition effective to control pests, preferably a composition which is also free of aflatoxins.

It has now been discovered that an extract containing azadirachtin with improved stability can be prepared.

Accordingly in a first aspect the present invention provides a process for preparing storage stable neem seed extract which comprises:

a) deoiling ground neem seeds, neem kernels or partially dehulled seeds using hexane to obtain a deoiled cake,
b) extracting the deoiled cake with methanol followed by filtration,
c) either:

(i) removing the methanol to obtain a dry extract, dissolving 3 parts of the dry extract in 7 parts of methanol (w/w) and adding water to a final ratio of 35/65 methanol/water (v/v), or
(ii) evaporating the methanol to obtain a concentrate containing over 65% volatiles, and adding water to obtain a final methanol/water ratio of from 5/95 to 45/55, preferably 35/65 (v/v),

d) separating the aqueous methanolic phase from any solid impurities,
e) diluting the aqueous methanolic mixture with an aqueous salt solution,
f) extracting the dilute solution with a water immiscible solvent, preferably ethyl acetate, and

g) drying the resulting organic solution and, if desired, removing the solvent.

Simple alcoholic neem seed extracts as known in the art (U.S. 4,556,562) can be purified by the above process to yield extracts generally containing over 25% of azadirachtin. These extracts have improved stability when compared with less pure extracts containing 15% or less of azadirachtin.

Additionally, the stability of the extracts may be improved by treatment of the extract with a mild oxidizing agent such as alkaline solution of hydrogen peroxide, an alkyl peroxide, for example di-*n*-butyl peroxide, an acid peroxide, for example perbenzoic acid, sodium percarbonate or the like. Preferred solvents for this reaction are polar organic solvents such as ethyl acetate or butyl acetate. A preferred oxidizing agent is hydrogen peroxide. A preferred base used with the hydrogen peroxide is saturated sodium bicarbonate solution. Moreover, the extracts which have been treated with hydrogen peroxide have been found to be essentially free of aflatoxins which might be present in extracts from seeds contaminated with *Aspergillus niger*. In addition, when extracts as described in U.S. 4,943,434 are treated with a mild oxidizing agent as described above, they can be more easily hydrogenated to yield an extract containing dihydroazadirachtin with improved stability.

The extracts of this invention have improved susceptibility to azadirachtin decomposition. When treated extracts of this invention and non-treated extracts were placed in an oven at 54°C for several weeks and then tested to determine the azadirachtin content, it was found that the rate of decomposition of azadirachtin in the extract was significantly reduced, as was the sulfur content of the extract. These effects are believed to be due to precipitation of the hydrophobic impurities by water from the methanolic solution of the crude extract and/or treatment with the alkaline solution of hydrogen peroxide. A further aspect of the invention comprises the use of the above process to enhance the stability of neem seed extract.

The resulting extracts generally contain from about 1% to about 85% of azadirachtin and preferably from about 25% to about 45% of azadirachtin.

The stable extracts of this invention are also advantageously used in the preparation of storage stable formulations and compositions, and in another aspect the invention provides an insecticidal composition which comprises an agronomically acceptable carrier and an insecticidally effective amount of an extract produced by the above process. Examples of the preparation of compositions and formulations can be found in the American Chemical Society publication "Pesticidal Formulation Research," (1969), Advances in Chemistry Series, 86 and the Marcel Dekker, Inc. publication "Pesticide Formulations," (1973), Wade Van Valkenburg editor. In these compositions and formulations, the active substance is mixed with conventional inert agronomically acceptable (i.e., plant compatible and/or pesticidally inert) diluents or extenders such as solid types usable in conventional compositions or formulations as is well known in the art. If desired, adjuvants such as surfactants, stabilizers, antifoam agents and antidrift agents may also be added.

Examples of compositions and formulations according to this invention are those known in the art and include aqueous solutions and dispersions, oily solutions and oil dispersions, pastes, dusting powders, wettable powders, emulsifiable concentrates, flowables, granules, baits, invert emulsions, aerosol compositions and fumigating candles.

The compositions and formulations are prepared in a known manner. For example, the active compounds are extended with conventional dispersible liquid diluent carriers and/or dispersible solid carriers. If desired, carrier vehicle assistants can be used such as conventional surface-active agents including emulsifying agents and/or dispersing agents whereby, for example, organic solvents may be added as auxillary solvents in the case where water is used as a diluent.

Adhesives such as carboxymethyl cellulose and natural and synthetic polymers, for example gum arabic, polyvinyl alcohol, polyvinyl cellulose and polyvinyl acetate, in the form of powders, granules or latices can be used in the formulations to improve the adherence of the pesticide. Furthermore, a lubricant such as calcium stearate or magnesium stearate may be added to a wettable powder or to a mixture to be granulated.

The stabilized extract of the present invention may be employed alone and/or with such solid and/or liquid dispersible carrier vehicles and/or with other known compatible active agents, especially plant protection agents such as other insecticides, arthropodicides, nematicides, fungicides, bactericides, rodenticides, herbicides, fertilizers, growth-regulating agents, synergists and the like, if desired, or in the form of particular dosage preparations for specific applications made therefrom, such as solutions, emulsions, suspensions, powders, pastes and granules which are thus ready for use.

As concerns commercially marketed preparations, these generally contemplate carrier composition mixtures in which the stabilized extract is present in an amount substantially between 0.1% and 99% by weight and preferably between about 1% and 75% by weight of the mixture.

Some formulations are capable of confering an additional stabilizing effect on azadirachtin in the formulated form, possibly due to dispersion, protection from humidity, sunlight and the like.

The stabilized extract formulations can be applied as sprays by methods commonly employed such as conventional high-gallonage hydraulic sprays, low gallonage sprays, ultra low volume sprays, airblast sprays, aerial sprays and dusts.

Furthermore, the present invention contemplated methods of selectively killing, combatting or controlling pests which comprise contacting insects with a correspondingly combative or toxic amount (i.e., an insect controlling or an

insecticidally effective amount) of the stabilized extract of the invention alone or together with a carrier vehicle (composition or formulation) as noted above. The term "contacting" as employed in the specification and claims of this application is to be construed as applying to at least one of (a) such insects and (b) the corresponding habitat thereof (i.e., the locus to be protected, for example a growing crop or an area where a crop is to be grown) the active compound of this invention alone, as a constituent of a composition or formulation or as a constituent of a composition or formulation containing other insecticides or fungicides.

It will be realized, of course, that the concentration of the particular stabilized extract utilized in admixture with the carrier vehicle will depend upon such factors as the type of equipment employed, method of application, area to be treated, types of pests to be controlled and degree of infestation. In addition to the aforementioned ingredients, the preparations according to the invention may also contain other substances commonly used in preparations of this kind.

The following examples are presented to illustrate the invention and are not to be construed as limiting in scope. All parts and percentages are by weight unless otherwise indicated.

EXAMPLE 1: Preparation of Extract 1 Containing Unstabilized Azadirachtin

Destoned neem seeds, 1027 kilograms (kg), were deoiled using first a standard mechanical oil expeller and then by extraction with hexane in a standard counter-current Crown-Iron solvent extractor. Using the same solvent extractor, the 701 kg of deoiled seed was then extracted with methanol at a 1/3 ratio of seed/methanol (w/w).

The dilute methanolic extract was stripped to give 70 kg of semi-solids which were then transferred to a kettle containing 160 kg of brine and 160 kg of ethyl acetate. After 0.5 hour of mixing, the phases were allowed to separate. The upper organic phase then was washed with 40 kg of brine, dried over 2 kg of magnesium sulfate, filtered and stripped to give 13 kg of Extract 1 containing 1.3 kg (10%) azadirachtin. The azadirachtin in this extract rapidly decomposed at room temperature.

EXAMPLE 2: Preparation of Extract 2 Containing 5.9% of Azadirachtin

Extract 1, 25 grams (g), containing 5.1% azadirachtin was dissolved in 140 g of ethyl acetate. To this solution, 30 g of saturated sodium bicarbonate solution and then 6 g of 30% hydrogen peroxide were added and the mixture was stirred for 45 minutes at 45 to 50°C and then at 55°C for 15 minutes. After phase separation, the upper organic phase was washed with brine containing a small amount of sodium bisulfite. Thereafter, the upper organic phase was separated, dried over magnesium sulfate, filtered, evaporated under reduced pressure and dried to yield 20 g of Extract 2 containing 5.9% azadirachtin.

EXAMPLE 3: Preparation of Extract 3 Containing 7.6% of Azadirachtin

Extract 2, 2 g, was stirred with 20 g of hexane for one hour and then filtered. The filter cake was dried overnight under reduced pressure to yield Extract 3 containing 7.6% azadirachtin.

EXAMPLE 4: Preparation of Extract 4 Containing 31% of Azadirachtin

Neem seeds, 3200 kg, were partially dehulled and then deoiled by standard methodology using mechanical oil extrusion followed by hexane extraction. The fully deoiled neem cake, 1400 kg, was ground in 7000 kg of methanol. After removal of all solids, the clear methanolic extract was concentrated to 366 kg. To this concentrate, 596 kg of water was added with stirring and, after 30 minutes, the resulting precipitate filtered off. The clear liquid phase was diluted with 1080 kg of brine and extracted with 571.5 kg of ethyl acetate. The extraction was then repeated with 202 kg of ethyl acetate and the combined ethyl acetate phases were evaporated to give 31 kg of a concentrate. To this concentrate, 27 kg of saturated sodium bicarbonate solution containing 5.4 kg of 35% hydrogen peroxide was added and the mixture was stirred and brought up to 55°C. After 30 minutes at 55°C, the mixture was allowed to reach room temperature, the phases were separated and the upper organic phase was dried over sodium sulfate, filtered and evaporated to yield 12.8 kg of Extract 4 containing 31% azadirachtin.

EXAMPLE 5: Preparation of Wettable Powders

Wettable powders (WP) containing stabilized extracts of this invention were prepared as follows:

a. 20WP

Ingredients	Composition % by Weight
Stabilized extract (30% azadirachtin)	66.6
Sellogen® DFL[1]	2.0
Tamol® 731[2]	5.0
HiSil® 233[3]	26.4
	100.0

[1] Modified alkylnaphthalene sulfonate from Rhone-Poulenc
[2] Sodium salt of carboxylated polyelectrolyte from Rohm and Haas
[3] Silica from PPG

The formulation was pulverized with a hammer-mill through a 0.020 inch screen followed by air milling to an average particle size of 3 millimicrons to give a wettable powder. The wetting time for 2 g in water of 342 ppm hardness as calcium carbonate was 50 seconds. The suspensibility in 342 ppm water at 1% concentration was 94% after 30 minutes.

b. 75WP

Ingredients	Composition % by Weight
Stabilized extract (80% azadirachtin)	93.8
Aerosol® OTB[1]	1.0
Morwet® D425[2]	5.2
	100.0

[1] Dioctyl sodium sulfosuccinate from American Cyanamid
[2] Sodium naphthalene formaldehyde condensate from DeSoto, Inc.

The formulation was processed essentially as described in Example 5a to give a wettable powder with similar physical properties and efficacy as an insecticide for foliar pests.

c. 5WP

Ingredients	Composition % by Weight
Stabilized extract (20% azadirachtin)	25.0
Triton® X-120[1]	0.5
Toranil B[2]	1.0
Barden® Clay[3]	73.5
	100.0

[1] Octyl polyethoxyethanol on carrier from Union Carbide
[2] Calcium lignosulfate from Rhinelander Paper
[3] Kaolin clay from Huber

The formulation was processed essentially as described in Example 5a to give a wettable powder with similar physical properties and efficacy as an insecticide for foliar pests.

EXAMPLE 6: Stability Data and Sulfur Content

The extracts from Examples 1-4 and the 20WP from Example 5a were analyzed for azadirachtin content and sulfur and then placed in an oven at 54°C. They were analyzed for azadirachtin again after 14 and 28 days.

Azadirachtin content in the examples was determined by reverse phase HPLC using a sample of pure azadirachtin as a standard.

The results obtained are as shown:

Test Substance from Example	% Azadirachtin Content			% Azadirachtin Decomposition at Day 28	Ppm of Sulfur Content[1]
	Day 0	Day 14	Day 28		
1	5.1	1.3	0.0	100	6000
2	5.9	4.4	3.0	49	4200
3	7.6	6.6	5.7	25	4100
4	31.0	27.0	26.0	16	1500
5a	20.0	18.0	18.0	10	NA[2]

[1] Initial measurement (Day 0)
[2] Not analyzed

EXAMPLE 7: Biological Activity

After storage for 28 days at 0°C and 54°C, the extracts from examples 1, 2 and 4 and the 20WP from Example 5a were tested for biological activity.

For the test, lima bean (*Phaseolus limensis* var. Woods' Prolific) plants, 2-3 weeks old, were sprayed to run-off with the test solution. After drying, the treated leaves were detached and infested with second instar larvae of Southern Armyworms (*Spodoptera eridania*) at doses of 150, 38, 10 and 2.5 ppm using 10 insects/dose. The percent mortality was determined at 3 days and at 6 days.

The results obtained are as shown:

Test Substance from Ex.	28-Day Storage Temp. °C	% MORTALITY RESPONSE							
		3-DAY				6-DAY			
		150	38	10	2.5	150	38	10	2.5
1	0	90	80	10	10	100	100	100	10
	54	20	0	0	0	30	0	0	0
2	0	90	60	20	0	100	100	100	0
	54	100	40	0	0	100	100	20	0
4	0	90	70	30	0	100	100	10	10
	54	90	80	20	0	100	100	0	0
5a	0	100	80	20	10	100	100	100	0
	54	100	100	20	10	100	100	100	20

Claims

1. A process for preparing storage stable neem seed extract which comprises:

   a) deoiling ground neem seeds, neem kernels or partially dehulled seeds using hexane to obtain a deoiled cake,
   b) extracting the deoiled cake with methanol followed by filtration,
   c) either:

      (i) removing the methanol to obtain a dry extract, dissolving the dry extract in methanol, preferably at a weight ratio of extract to methanol of 3/7, and adding water to a final methanol/water ratio of from 5/95 to 45/55, preferably 35/65 (v/v), or
      (ii) evaporating the methanol to obtain a concentrate containing over 65% volatiles, and adding water to obtain a final methanol/water ratio of from 5/95 to 45/55, preferably 35/65 (v/v),

   d) separating the aqueous methanolic phase from any solid impurities,
   e) diluting the aqueous methanolic mixture with an aqueous salt solution,
   f) extracting the dilute solution with a water immiscible solvent, preferably ethyl acetate, and
   g) drying the resulting organic solution and, if desired, removing the solvent.

2. Process according to claim 1 comprising the further step of treating said extract with an oxidizing agent, preferably in a polar organic solvent, more preferably in ethyl acetate or butyl acetate.

3. Process according to claim 2 wherein the oxidising agent is an alkaline solution of hydrogen peroxide, or an alkyl peroxide, preferably di-*n*-butyl peroxide, or an acid peroxide, preferably perbenzoic acid or sodium percarbonate.

4. Process according to claim 2 or 3 wherein the alkaline solution is a sodium bicarbonate solution.

5. Process according to any preceding claim wherein the deoiling step (a) is carried out either by stirring with hexane followed by filtration, or by mechanically extruding the oil and optionally extracting the oil residue.

6. Process according to any preceding claim wherein no more than 25% of the azadirachtin contained in the stabilised extract decomposes after storage thereof at 54°C for 28 days.

7. Process according to any preceding claim wherein the resultant extract contains less than 4200 ppm of sulfur.

8. Process according to any preceding claim comprising the further step of converting the stabilized extract into a wettable powder.

9. An insecticidal composition which comprises an agronomically acceptable carrier and an insecticidally effective amount of an extract produced by the process of any preceding claim.

10. The use of a process as defined in any of claims 1 to 8 to stabilise a neem seeed extract.

Patentansprüche

1. Verfahren zur Herstellung von haltbaren Extrakten aus Neem-Samen, dadurch gekennzeichnet, daß man

   a) gemahlene Neem-Samen, Neem-Kerne oder teilweise enthülste Samen unter Verwendung von Hexan entölt um einen entölten Kuchen zu erhalten,
   b) den entölten Kuchen mit Methanol extrahiert und dann filtriert,
   c) entweder:

      (i) das Methanol entfernt, um einen trockenen Extrakt zu erhalten, den trockenen Extrakt in Methanol löst, vorzugsweise bei einem Gewichtsverhältnis vom Extrakt zu Methanol von 3:7 und Wasser zu einem Methanol-/Wasser-Endverhältnis von 5:95 bis 45:55, vorzugsweise 35:65 (Vol./Vol.) zusetzt,
      oder
      (ii) das Methanol verdampft um ein Konzentrat zu erhalten, das über 65% flüchtige Anteile enthält und Wasser zusetzt, um ein Methanol-/Wasser-Endverhältnis von 5:95 bis 45:55, vorzugsweise 35:65 (Vol./Vol.) zu erhalten,

   d) die wäßrig-methanolische Phase von irgendwelchen festen Verunreinigungen trennt,
   e) das wäßrig-methanolische Gemisch mit einer wäßrigen Salzlösung verdünnt,
   f) die verdünnte Lösung mit einem mit Wasser nicht mischbaren Lösungsmittel, vorzugsweise Ethylacetat, extrahiert, und
   g) die erhaltene organische Lösung trocknet und gewünschtenfalls das Lösungsmittel entfernt.

2. Verfahren nach Anspruch 1, dadurch gekennzeichnet, daß es die weitere Stufe der Behandlung dieses Extrakts mit einem Oxidationsmittel, vorzugsweise in einem polaren organischen Lösungsmittel, noch bevorzugter in Ethylacetat oder Butylacetat, umfaßt.

3. Verfahren nach Anspruch 2, dadurch gekennzeichnet, daß das Oxidationsmittel eine alkalische Lösung von Wasserstoffperoxid oder einem Alkylperoxid, vorzugsweise Di-n-Butylperoxid, oder ein Säureperoxid, vorzugsweise Perbenzoesäure oder Natriumpercarbonat ist.

4. Verfahren nach Anspruch 2, dadurch gekennzeichnet, daß die alkalische Lösung eine Natriumbicarbonatlösung ist.

5. Verfahren nach irgendeinem der vorhergehenden Ansprüche, dadurch gekennzeichnet, daß die Entölungsstufe (a) entweder durch Rühren mit Hexan gefolgt von Filtration oder durch mechanisches Auspressen des Öls und gegebenenfalls Extraktion des Ölrückstandes durchgeführt wird.

6. Verfahren nach irgendeinem der vorhergehenden Ansprüche, dadurch gekennzeichnet, daß sich nicht mehr als 25% des Azadirachtins, das im stabilisierten Extrakt enthalten ist, nach Lagerung desselben bei 54°C für 28 Tage zersetzen.

7. Verfahren nach irgendeinem der vorhergehenden Ansprüche, dadurch gekennzeichnet, daß der erhaltene Extrakt weniger als 4200 ppm Schwefel enthält.

8. Verfahren nach irgendeinem der vorhergehenden Ansprüche, dadurch gekennzeichnet, daß es die weitere Stufe der Überführung des stabilisierten Extrakts in ein benetzbares Pulver umfaßt.

9. Insektizide Zusammensetzung, dadurch gekennzeichnet, daß sie einen landwirtschaftlich annehmbaren Träger und eine insektizid wirksame Menge eines Extrakts, der nach dem Verfahren irgendeines der vorhergehenden Ansprüche hergestellt ist, enthält.

10. Anwendung eines Verfahrens gemäß irgendeinem der Ansprüche 1 bis 8 zur Stabilisierung eines Neem-Samenextrakts.

Revendications

1. Procédé pour préparer un extrait de graine de margousier stable au stockage, qui comprend les étapes consistant à:

   a) déshuiler des graines de margousier broyées, des amandes de margousier broyées ou des graines partiellement mondées de margousier broyées, en utilisant de l'hexane pour obtenir un tourteau déshuilé,
   b) extraire le tourteau déshuilé avec du méthanol, puis le filtrer;
   c) soit:

      (i) éliminer le méthanol pour obtenir un extrait sec, dissoudre l'extrait sec dans du méthanol, de préférence à un rapport pondéral de l'extrait au méthanol de 3/7, et ajouter de l'eau pour obtenir un rapport final méthanol/eau de 5/95 à 45/55, de préférence de 35/65 (en volume/volume), soit
      (ii) évaporer le méthanol pour obtenir un concentré contenant plus de 65% de matières volatiles et ajouter de l'eau pour obtenir un rapport méthanol/eau final de 5/95 à 45/55, de préférence de 35/65 (en volume/volume),

   d) séparer la phase méthanolique aqueuse de toute impureté solide,
   e) diluer le mélange méthanolique aqueux avec une solution aqueuse de sel,
   f) extraire la solution diluée avec un solvant non miscible avec l'eau, de préférence l'acétate d'éthyle, et
   g) sécher la solution organique résultante et, le cas échéant, éliminer le solvant.

2. Procédé selon la revendication 1, comprenant l'étape supplémentaire qui consiste à traiter ledit extrait avec un agent oxydant, de préférence dans un solvant organique polaire, mieux encore dans l'acétate d'éthyle ou l'acétate de butyle.

3. Procédé selon la revendication 2, dans lequel l'agent oxydant est une solution alcaline de peroxyde d'hydrogène ou un peroxyde d'alkyle, de préférence un peroxyde de di-n-butyle, ou un peroxyde d'acide, de préférence l'acide perbenzoïque ou le percarbonate de sodium.

4. Procédé selon la revendication 2 ou 3, dans lequel la solution alcaline est une solution de bicarbonate de sodium.

5. Procédé selon l'une quelconque des revendications précédentes, dans lequel l'étape de déshuilage (a) est réalisée soit par agitation avec de l'hexane, suivie d'une filtration, soit par extrusion mécanique de l'huile et éventuellement extraction du résidu huileux.

6. Procédé selon l'une quelconque des revendications précédentes, dans lequel pas plus de 25% de l'azadirachtine contenue dans l'extrait stabilisé ne se décompose après son stockage à 54°C pendant 28 jours.

7. Procédé selon l'une quelconque des revendications précédentes, dans lequel l'extrait résultant contient moins de 4 200 ppm de soufre.

8. Procédé selon l'une quelconque des revendications précédentes, comprenant l'étape supplémentaire de transformation de l'extrait stabilisé en poudre mouillable.

9. Composition insecticide qui comprend un véhicule acceptable en agronomie et une quantité efficace en tant qu'insecticide d'un extrait produit par le procédé selon l'une quelconque des revendications précédentes.

10. Utilisation d'un procédé tel que défini dans l'une quelconque des revendications 1 à 8 pour stabiliser un extrait de graines de margousier.

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