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(71) Applicant: BIOBEST BELGIUM N.V. [BE/BE]; Ilse Velden 18, 2260 Westerlo (BE).

(72) Inventors: WÄCKERS, Felix; St Paullusstraat 43, 2000 Antwerpen (BE). PUT, Kurt; Vliegeneindestraat 73, 3582 Koersel-Beringen (BE). ARIJS, Yves; Gooreinde 63, 2260 Westerlo (BE). VAN DEN BORRE, Yannick; Paddegatstraat 15, 1880 Nieuwenrode (BE). VANDUFFEL, Mathias; Biezenstraat 10, 3040 Loonbeek (BE). DE VRIEZE, Sander; Magnoliadreef 12, 9700 Oudenaarde (BE). PEKAS, Tolis; Kouden Eerd 28, 2260 Tongerlo (BE).

(74) Agents: PAEMEN, Liesbet et al.; De Clercq & Partners, E. Gevaertdreef 10a, 9830 Sint-Martens-Latem (BE).

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(54) Title: IMPROVED PLANT BIOLOGICAL CONTROL BY PREDATORY MITES.

(57) Abstract: The present invention relates to methods for the biological control of crop pests by promoting the growth and development of predatory mites by increasing the attractiveness of the plant to the predatory mite, particularly in the case of plants lacking trichomes. The methods comprise applying a liquid fiber composition, preferably comprising a suitable adhesive agent, to a plant or part thereof and contacting a population of predatory mites with said plant.



IMPROVED PLANT BIOLOGICAL CONTROL BY PREDATORY MITES

FIELD OF THE INVENTION

The present invention in general relates to the field of biological plant protection, particularly biological plant protection using predatory mites. More particularly, the present invention relates to methods and compositions to promote the growth and development of the predatory mite population on a plant or plant part, as well as their application in controlling crop pests.

10 BACKGROUND OF THE INVENTION

Plant pests such as thrips, spider mites and whitefly cause considerable damage to various plants and crops, resulting in significant economic loss to growers and higher prices for consumers. Although plant pests may be controlled using chemical pesticides, this is not always possible or desirable. Indeed, the widespread use of chemical pesticides can result in health and environmental problems, and in the appearance of resistant insect varieties.

The use of natural predators of plant pests for biological plant pest control already finds widespread use. In particular, beneficial arthropods such as predatory mites are often used in agriculture for biological control of plant pests such as thrips or spider mites. Typically, the predatory mites are applied to the plants manually, via controlled release systems, or via other methods.

The presence of leaf trichomes, particularly non-glandular trichomes like leaf hairs, or (tuft) domatia on plant or plant parts is of great importance for maintaining a sufficient mite population, by providing oviposition sites, protection, shelter and/or a more suitable microclimate for the predatory mites. However, many plants or plant parts lack (non-glandular) leaf trichomes, leaf hairs or (tuft) domatia, such as is the case for certain perennial fruit crops, like cultivated pear, citrus and some grape cultivars, and for certain ornamental plants. Accordingly, the use of predatory mites for protecting plants and plant parts lacking these structures can be problematic, because predatory mite populations are often not able to establish.

Loughner *et al.* (Entomologia Experimentalis et Applicata 134: 78–88, 2010) discloses that increasing habitat complexity via the use of fiber patches as artificial trichomes strongly reduced the predatory mite dispersal behavior: the mites rapidly dispersed away from leaves that lacked these structures. However, in this study, the fiber patches were individually placed on the plant leaves, which is too cumbersome and impractical when upscaling from an individual plant to orchard or greenhouse level.

There thus remains a need in the art for methods and means for establishing, maintaining and/or promoting a predatory mite population on a plant or plant part/structures, particularly on a plant or plant part/structure lacking natural, non-glandular leaf trichomes, like hairs, in order for a better biological plant pest control. In particular, said methods and means should provide a solution to said need which is easily applicable, has a low-cost and is suitable for a large-scale utilization on a large number of plants, such as e.g. in orchards, fields or greenhouses.

SUMMARY OF THE INVENTION

The present invention relates to novel methods and means to support the biological control of plant pests on a crop plant or ornamental plant or part thereof, particularly a plant or plant part or plant structure lacking leaf trichomes, leaf hairs or domatia, by promoting and/or maintaining the predatory mite population on said plant or plant part, comprising applying a liquid composition comprising a fiber to said plant or plant part.

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Indeed, the inventors have found that fibers can be applied onto leaves of a plant via a liquid dispersion of said fibers and when applied as such will promote the maintenance of a predatory mite population on a plant crop.

In particular embodiments, the invention provides methods for increasing the attractiveness of a plant or plant part/structure for a predatory mite population, said methods comprising applying a liquid composition comprising a fiber to said plant or plant part/structure. In particular embodiments, said liquid composition comprises between 0.01 to 5.0 wt% fibers. In particular embodiments, the liquid composition is an aqueous composition.

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In particular embodiments, the length of the fiber ranges from 0.5 to 100 mm. In particular embodiments, the fiber is crimped or curled. In further particular embodiments, the fiber has a textured (i.e. uneven) surface.

In particular embodiments, the liquid composition comprising a fiber further comprises an adhesive compound. In particular embodiments, the adhesive compound is a sugar, a sugar alcohol, a soluble starch or a derivative thereof, arabic gum, xanthan gum, pectin or colloidal silica.

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In particular embodiments, the liquid composition comprising a fiber further comprises a fungus reducing agent, a pest repellent, and/or a plant metabolism modifying agent.

In particular embodiments, the liquid composition comprising a fiber is applied by spraying or nebulization. In particular embodiments, the liquid composition is applied using a motor driven liquid sprayer apparatus as provided herein.

- 5 In particular embodiments of the methods provided herein, the plant is a herbaceous plant, a vine, an ornamental plant or a tree, fruits or fruiting structures, or a plant structure, such as resting structures such as bulbs, tubers roots. In further particular embodiments, the plant is a pear tree or a Citrus spp. tree.
- 10 The methods described herein are of particular interest in the biological control of pests on plants or plant structures by predatory mites. The mites can be either naturally occurring on the plant or is a mite population applied to the plant. Accordingly, in particular embodiments, the methods further comprise contacting said plant with said predatory mite population before, after or simultaneously with the application of the liquid
- 15 fiber composition. In particular embodiments, the methods provided herein are used with predatory mites of the family *Phytoseiidae*.

Also provided herein are liquid compositions comprising a fiber for providing a fiber on a plant or plant structure, preferably wherein the length of the fiber ranges from 0.5 to 100

20 mm. In particular embodiments, said liquid composition further comprises one or more of the following compounds: (i) an adhesive compound, (ii) a nutritional source for predatory mites; (iii) a fungal reducing compound; (iv) a pest repellent; (v) a plant metabolism modifying agent. Also provided herein is a container comprising said liquid composition.

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Further provided are kits for the biological control of pests on a plant or plant structure comprising a fiber composition, preferably a liquid or aqueous fiber composition, and, provided separately, a predatory mite population, or a source thereof. In particular

30 embodiments, the length of the fiber ranges from 0.5 to 100 mm and/or the fibers is crimped or curled.

In particular embodiments, the kits further comprise one or more of the following compounds: (i) an adhesive compound, (ii) a nutritional source for said predatory mites; (iii) a fungal reducing compound; (iv) a pest repellent; (v) a plant metabolism modifying agent. In particular embodiments, the predatory mite provided in the kit is a member of

35 the family *Phytoseiidae*.

Another aspect of the present invention relates to a motor driven liquid sprayer apparatus configured for spraying a liquid fiber composition in an uniform way, comprising: (i) a reservoir configured for containing the liquid to be sprayed, wherein said container comprises an outlet passage for the liquid to be sprayed; a motor powered
5 pump assembly for dispensing the liquid through a spraying means equipped with a spray nozzle, wherein the inlet port of said pump assembly is connected with the outlet passage of said reservoir by tubings and wherein the outlet port of said pump assembly is connected with the spraying means; characterized in that said motor driven liquid sprayer apparatus further comprises a means for regulating or interrupting the liquid flow
10 adapted to avoid fiber induced liquid blockages; and/or a stirring means or a means for suspending the fiber in the liquid solution located in said liquid reservoir. In particular embodiments, the motor driven liquid sprayer apparatus comprises (a) an axial flow impeller as a stirring means; and/or (b) a spray nozzle having one or more openings with a diameter of 100-150 μm ; and/or (c) a pinch valve or ball valve as a means for
15 regulating or interrupting the liquid flow.

The above and other characteristics, features and advantages of the concepts described herein will become apparent from the following detailed description, which illustrates, by way of example, the principles of the invention.

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BRIEF DESCRIPTION OF THE DRAWINGS

The following description of the figures of specific embodiments of the invention is merely exemplary in nature and is not intended to limit the present teachings, their application or uses.

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Figure 1 is a schematic representation of part of a motor driven liquid sprayer apparatus for use in a particular embodiment of the present invention.

Figure 2 is a schematic representation of the reservoir of a motor driven liquid sprayer apparatus, adapted for fiber application according to a particular embodiment of the
30 present invention, with a side view shown on the left and a front view on the right.

DETAILED DESCRIPTION OF THE INVENTION

While potentially serving as a guide for understanding, any reference signs in the claims shall not be construed as limiting the scope thereof.

35 As used herein, the singular forms “a”, “an”, and “the” include both singular and plural referents unless the context clearly dictates otherwise.

The terms “comprising”, “comprises” and “comprised of” as used herein are synonymous with “including”, “includes” or “containing”, “contains”, and are inclusive or open-ended and do not exclude additional, non-recited elements or method steps. The terms “comprising”, “comprises” and “comprised of” when referring to recited components,
5 elements or method steps also include embodiments which “consist of” said recited components, elements or method steps.

Furthermore, the terms first, second, third and the like in the description and in the claims, are used for distinguishing between similar elements and not necessarily for describing a sequential or chronological order, unless specified. It is to be understood
10 that the terms so used are interchangeable under appropriate circumstances and that the embodiments described herein are capable of operation in other sequences than described or illustrated herein.

The values as used herein, when referring to a measurable value such as a parameter, an amount, a temporal duration, and the like, particularly when used in combination with
15 the term “about”, is meant to encompass variations of $\pm 10\%$ or less, preferably $\pm 5\%$ or less, more preferably $\pm 1\%$ or less, and still more preferably $\pm 0.1\%$ or less of and from the specified value, insofar such variations are appropriate to ensure one or more of the technical effects provided herein. It is to be understood that each value as used herein is itself also specifically, and preferably, disclosed.

Unless otherwise defined, all terms used in disclosing the concepts described herein, including technical and scientific terms, have the meaning as commonly understood by one of ordinary skill in the art. By means of further guidance, definitions for the terms used in the description are included to better appreciate the teaching of the present disclosure. The terms or definitions used herein are provided solely to aid in the
25 understanding of the teachings provided herein.

Reference throughout this specification to “one embodiment” or “an embodiment” means that a particular feature, structure or characteristic described in connection with the embodiment is included in at least one embodiment provided herein. Thus, appearances of the phrases “in one embodiment” or “in an embodiment” in various places throughout
30 this specification are not necessarily all referring to the same embodiment, but may. Furthermore, the particular features, structures or characteristics may be combined in any suitable manner, as would be apparent to a person skilled in the art from this disclosure, in one or more embodiments. Furthermore, while some embodiments described herein include some but not other features included in other embodiments,
35 combinations of features of different embodiments are also provided herein, and form different embodiments, as would be understood by those in the art. For example, in the

appended claims, any of the features of the claimed embodiments can be used in any combination.

The applicants have identified methods and means which address one or more of the
5 disadvantages of the prior art detailed above. Indeed, it has been found that spraying an aqueous composition comprising fibers, even at low fiber contents, is a surprisingly effective way to apply fibers to plants and plant parts, such as leaves, fruits or fruiting structures, or resting structures such as bulbs, tubers roots, thus creating an environment suitable for establishing, maintaining and/or promoting a predatory mite
10 population on said plant or plant part. Indeed, the inventors have found that dispersing the fibers in dry conditions in an air stream was ill suited for the large-scale application of the fibers to the plants: the dry fibers entangle easily or form lumps in applicator devices and the fibers are only loosely attached to the leaves resulting in a very uneven fiber adherence to the plant, or even no adherence. Moreover, the inhalation of the airborne
15 fibers by the operator is an undesired side-effect.

In contrast, particularly upon stirring or shaking of the liquid fiber composition, the fibers are well dispersed in the liquid: the fibers can be evenly applied with the liquid spray and their application rate can be precisely quantified. In addition, the wet fibers remain better attached to the plant parts.

20 As dedicated spraying systems are widely used in the cultivation of plants in fields, orchards and greenhouses, utilization of a liquid composition comprising fibers as set out herein is easily applicable.

Without being bound by theory, the fibers may provide artificial oviposition sites and shelter for the predatory mites, resulting in a more secure environment for the mites and
25 their eggs.

Advantageously, an adhesive compound may be present in the liquid composition comprising a fiber. This allows increasing the adherence between fiber and plant part, thus avoiding the removal of the fibers from said plant by wind or other environmental phenomena. Moreover, in particular embodiments, such as where the adhesive is a
30 sugar, said adhesive can function as a food source for the mites.

As used herein, the term “predatory mite” refers to mites who feed on other organisms, in particular arthropods, including thrips, white fly, other mite species and the like. In particular embodiments, predatory mites comprise phytoseiid predatory mites, i.e.
35 members of the family *Phytoseiidae*. Phytoseiid predatory mites are widely used for biological control of pests such as spider mites, thrips, and whitefly, particularly in

greenhouse crops. In particular embodiments, the predatory mites are from a genus selected from the group consisting of *Euseius*, *Amblyseius*, *Neoseiulus*, *Iphiseius*, *Indoseiulus*, *Kampimodromus*, *Typhlodromalus*, *Phytoseius* and *Typhlodromus*. Non-limiting examples of mite species suitable for use in the presently disclosed biological control methods and means include *Euseius stipulatus*, *Eusius galicus*, *Amblyseius cucumeris*, *Typhlodromus pyri*, *Amblyseius swirskii*, and *Iphiseius degenerans*. In particular embodiments, said mite population is applied to said plant before or after applying the liquid fiber composition as described herein. In particular embodiments, the predatory mite population is a natural mite population.

10

In a first aspect, the present invention provides methods for increasing the attractiveness and suitability of a plant for a predatory mite, the methods comprising applying a liquid composition comprising a fiber to a plant or part thereof. Indeed, it has been found that applying a liquid composition comprising a fiber to a plant, will increase the attractiveness of the plant for the predatory mite. More particularly, it has been found that the number of eggs laid by the predatory mites on the leaves increased after having applied fibers to the plant in this manner. Accordingly, the invention provides methods for increasing the reproduction rate of a predatory mite on a plant. As the number of predatory mites on a plant will affect the biological control ensured by said predatory mites, the application also provides methods for the biological control of pests on a plant by predatory mites, which methods comprise applying a liquid or aqueous composition comprising a fiber to a plant or part thereof. In particular embodiments, said methods also comprise introducing a mite population on said plant or part thereof. More in particular, the present invention relates to methods for promoting or maintaining a predatory mite population on a crop plant, comprising applying a liquid fiber composition to a plant or part thereof and introducing a mite population on said crop plant or part thereof. The mite population may be introduced on or allowed to contact with said plant or part thereof before, after or simultaneously with the step of applying the liquid fiber composition to said plant or part thereof. In particular embodiments, the method for the biological control of pests on a plant by predatory mites comprises applying, for instance by spraying, a liquid composition comprising a fiber to said plant or part thereof, wherein mite eggs are attached to said fiber prior to fiber application/spraying. Also, in particular embodiments, the present invention provides a method for promoting or maintaining a predatory mite population on a plant comprising applying a liquid fiber composition to a plant or part thereof wherein said plant contains a predatory mite population.

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In the context of the present invention, fibers are applied onto a plant using a liquid carrier. The term "liquid" is used to describe a physical state that is fluid without being gaseous. A liquid can flow freely and has a definite volume. Preferred liquid carriers include water, oil, including but not limited to vegetable oil, glycerol and the like. In particular embodiments, the liquid composition comprising fibers is an aqueous composition comprising fibers, with the aqueous phase acting as carrier. The term "aqueous" as used herein means that more than 50 percent by volume is water, preferably more than 60, 70, 80 or 90 percent by volume is water, such as 100 percent by volume is water. Aqueous compositions may further comprise organic liquids which are miscible with water.

In the context of the different aspects and embodiments of the present invention, methods and products involving the use both natural and synthetic insoluble fibers are provided. Natural fibers include cellulose based fibers, like cotton fibers, hemp, kapok, or jute fibers, flax fibers, and the like, animal hair like wool or alpaca, arthropod produced fibers, such as silk, but also feathers. Synthetic fibers include polyamide fibers, such as nylon, polyester fibers, acrylic fibers, polylactide fibers, fibers made from regenerated cellulose, and the like. The fibers may have a coarse or smooth surface. Preferably, said fiber is particularly suitable as an oviposition substrate. Preferably, said fiber is easily dispersible in an aqueous solution. In particular embodiments, said fiber is an acrylic or polylactide fiber.

The fibers may be present in the liquid composition as individual, isolated fibers or structured in threads, yarns, fiber clusters or attached to a joint base. In particular embodiments, the fibers are crimped or curled. The crimping or curling may be a natural characteristic of the fiber or may be the result of mechanical crimping, spiral crimping, or another type. A fiber may have a combination of two or more types of crimping. In certain embodiments, the crimping or curling of the fiber may occur when the fiber is in the dry state, such as when the liquid carrier has evaporated or drained away. Advantageously, because of their crimped or curled nature, parts of the crimped fibers protrude from the leaf or plant part surface, thereby providing better suited oviposition and shelter sites for the predatory mites population. In particular embodiments, the fibers have a textured (uneven) surface, which may provide better suited oviposition sites for the predatory mites population.

The fibers may be hollow or solid. In particular embodiments, the fibers are porous or hollow having an internal cavity, wherein the internal cavity optionally comprises one or

more components, including but not limited to an adhesive agent, a food source, a pheromone, fungal reducing compound; a pest repellent; a plant metabolism modifying agent. etc, advantageously allowing the controlled release of said component within the cavity.

- 5 The fibers can be characterized in size and/or shape. This further emphasizes that the fibers envisaged are non-soluble fibers in that it is required that their shape is substantially maintained in the liquid composition. In particular embodiments, the fiber length ranges from about 0.1 to 100 mm or 0.5 to 25 mm, more preferably ranges from about 1 or 2 mm to about 10 or 15 mm.
- 10 The fibers may have a circular or other cross-sectional shape, such as elliptical, multi-lobed and the like. In particular embodiments, the average fiber diameter is in the range of about 0.5 to about 100 microns.

In particular embodiments, the fiber has a linear density between 0.5 and 4000 dtex, more preferably has a linear density between 0.5 and 200 dtex, such as between 0.5 and

- 15 5 dtex or between 50 and 150 dtex.

Tex is a unit of measure for the linear mass density of fibers and is defined as the mass in grams per 1000 meters. The most commonly used unit is actually the decitex, abbreviated dtex, which is the mass in grams per 10,000 meters. One can calculate the diameter of a filament given its weight in dtex with the following formula:

$$20 \quad \varnothing = \sqrt{\frac{4 \times 10^{-6} \cdot \text{dtex}}{\pi \rho}},$$

wherein ρ is the density of the material.

- In particular embodiments, the fiber content of the aqueous composition provided herein ranges from 0.01 to 5.0 wt%, more preferably ranges from 0.01 to 1.0 or 2.0 wt%, even more preferably ranges from 0.02 to 0.5 wt%, most preferably ranges from 0.025 to 0.2
- 25 wt%. In a particularly preferred embodiment, the fiber content in the aqueous fiber composition is about 0.05 wt%.

- Advantageously, the liquid or aqueous composition comprising a fiber as provided in particular embodiments of the methods and means of the present invention further
- 30 comprises an adhesive compound for improving the adherence of the fiber to the plant or part thereof.

In particular embodiments, said adhesive compound is a natural, water soluble adhesive agent. Preferred adhesive compounds include water-soluble starch and derivatives thereof, sugars or sugar alcohols, such as sucrose, fructose, glucose, maltose,

trehalose, galactose, raffinose, mannitol or sorbitol, Arabic gum, xanthan gum, pectin, colloidal silica...

In particular embodiments, the adhesive compound is present in the solution at a concentration of 0.1 to 50 wt%, such as 0.1 to 10 wt% or 15 wt%, more preferably is
5 present in a concentration of 0.5 to 3 wt%, even more preferably is present in a concentration of about 1.0 to 2.0 wt%.

Advantageously, said adhesive may also act as a nutritional source for the predatory mites. Alternatively, the aqueous composition comprising a fiber as provided herein may
10 further comprise an additional nutritional source that is not an adhesive. In particular embodiments, said liquid composition does not comprise pollen, as pollen tend to form lumps in an aqueous environment, thus negatively affecting the quality, shelf life and application of the pollen and/or the fibers.

In particular embodiments, a fungus reducing agent may be added to the liquid fiber
15 composition, particularly aqueous fiber composition provided herein. A fungus reducing agent is any agent reducing fungal growth for example by slowing or preventing fungal growth such as by interfering with fungal metabolism or reducing fungal growth by destruction of fungal biomass. The fungus reducing agent may comprise chemical fungus reducing agents such as a natural or synthetic fungicide, for example a natural
20 fungicide selected from citral, neral, 2,3-epoxyneral, geranial, farnesal, α -acaradial, β -acaradial, or natamycin (pimaricin). Alternatively, a biological fungus reducing agent such as a microbial antagonist or a population of fungivorous mites may be introduced in said plant.

In particular embodiments, a pest repellent agent may be added to the liquid fiber
25 composition. The pest repellent may comprise natural or synthetic chemical repellentia such as essential oils or other aromatic compounds.

In particular embodiments, the liquid composition comprising fibers further includes one or more components modifying plant metabolism or promoting plant growth, such as a fertilizing compound, a plant hormone or an elicitor of plant resistance to disease and/or
30 pests.

In the methods provided herein, the liquid composition comprising a fiber is applied to a plant or part thereof. In particular embodiments, the liquid composition comprising a fiber is applied to a plant part or plant structure, i.e. one or more plant parts/structures
35 selected from leafs, stem, trunk, shoots, buds, fruits or fruiting structures, flowers or flowering structures, or resting structures such as bulbs, tubers roots. In particular

embodiments of the methods of the present invention, the liquid composition comprising a fiber is applied to the upper side and the underside of the leaves or other plant part in about equal proportions, or stated differently, after application about 40-60% of the fibers is present on each side of the leaf. In other particular embodiments of the present
5 inventions, the liquid composition comprising a fiber is applied preferentially to the upper side or underside of the leaves or other plant part, or stated differently, after application, one side of the leaves or other plant part have about 60 to 100% of the fibers present. Indeed, it has been found that at least some predatory mites prefer the underside of the leaves for laying eggs, such that application of the fibers to the underside of the leaves
10 further promotes the number of eggs laid on the plant. Accordingly, in particular embodiments, the fibers are applied more extensively to the underside of the leaves or other part of the plant. The methods provided herein wherein the composition further comprises an adhesive compound will promote adherence of the fibers to the underside of the plants. In particular embodiments, the methods for application of the liquid
15 composition ensure uniform distribution of the composition over the targeted plant or plant part.

The invention relates to applying a liquid composition comprising fibers to a plant part. More particularly this encompasses ensuring that the liquid composition is deposited
20 directly onto the plant or plant part. In particular embodiments of the methods of the present invention, the liquid composition comprising a fiber is applied to the plant by spraying. Accordingly, in preferred embodiments, the liquid composition is suitable for being applied by spraying or nebulization. Suitable spraying systems include but are not limited to moveable spraying systems comprising a reservoir for carrying the liquid
25 composition, such as portable by hand or on the back, or moveable via a trolley, trailer or tractor, or sprinkle systems. Advantageously, in the case of the biological control of pests on tree crops, like pear and Citrus spp, the liquid fiber composition provided herein can be applied by a moveable motor driven liquid sprayer, including but not limited to a portable motor driven backpack liquid sprayer, a motor driven trolley liquid sprayer or a
30 motor driven trailer/tractor liquid sprayer. The use of a motor driven liquid sprayer apparatus ensures a deep penetration of the liquid composition in the tree canopy and generates a turbulent air stream comprising the liquid. This way, the aqueous fiber composition of the present invention forms multiple small aerosol droplets, which due to the turbulent stream (which moves the tree leaves) and the small droplet size allow fiber
35 deposit on both the upper side and underside of the leaves throughout the plant.

Alternative methods for depositing a liquid composition on a plant or part thereof can also be provided such as dripping, pouring, or applying with specific means such as a brush, pipette, sponge etc;

5 As detailed above, in particular embodiments, the methods provided herein also involve the introduction of a predatory mite population on said plant. The term "introducing" as used herein in the context of introducing predatory mites, refers to the introduction of predatory mites on or near a plant, organism or item to be protected from pests. In particular embodiments, the methods involve contacting a mite population or one or
10 more individuals of a mite population with a plant or a plant part thereof. In particular embodiments, introducing a predatory mite population may comprise contacting or inoculating a plant or part thereof with sexually mature adults from both sexes. Additionally or alternatively, individuals of both sexes of other life stages, e.g. eggs, larvae and/or nymphs, which can mature to sexually mature adults can also be used. In
15 particular embodiments, the predatory mite population can be introduced via the liquid fiber composition as provided herein, wherein mite eggs are attached to said fiber. In particular embodiments, the liquid fiber composition as provided herein does not comprise living mites, in particular does not comprise larvae and/or nymphs or adult mites.

20

Another aspect of the present invention provides a liquid composition comprising a fiber, and optionally an adhesive as described for use in the methods herein above. In particular embodiments, said liquid fiber composition may comprise one or more of the following agents, as described herein:

- 25 - an adhesive compound, preferably a natural, water soluble adhesive agents. Preferred adhesive compounds include water-soluble starch and derivatives thereof, sugars or sugar alcohols, such as sucrose, fructose, glucose, maltose, trehalose, galactose, raffinose, mannitol, sorbitol, and the like;
- a nutritional source;
- 30 - a fungal reducing agent;
- a pest repellent
- plant metabolism modifying compounds

In particular embodiments, the present invention further provides a container comprising a liquid or aqueous composition comprising a fiber as provided herein and as described
35 above. In particularly preferred embodiments said container comprising a liquid or aqueous composition as provided herein is a part of a spraying apparatus. In particular

said container comprising a liquid or aqueous fiber composition corresponds to the reservoir or liquid holding means of a spraying apparatus. Preferably, said spraying apparatus is a motor driven liquid spraying apparatus, in particular is the motor driven liquid spraying apparatus according to another aspect of the present invention, as
5 detailed below.

However, it is also provided herein that in particular embodiments, the composition is distributed and/or stored as a dry composition, for mixing with a liquid before use. In particular embodiments, the dry composition comprises a fiber and an adhesive. Thus, in
10 particular embodiments, the composition comprising an adhesive and a fiber is provided for mixing with a liquid to obtain a liquid composition. In particular embodiments, where the liquid comprising the fibers is dried, the concentration of the adhesive on the fibers is between 80% and 100%. Also provided herein are methods for producing the liquid fiber composition provided herein which comprise combining fiber and an adhesive with a
15 liquid.

Further the application provides a kit comprising (i) a fiber composition, which upon addition of water or another liquid yields the liquid fiber composition as provided herein, and (ii) a predatory mite population or a source thereof. It will be understood that in
20 particular embodiments, the predatory mite population is provided separately from said fiber composition. In particular embodiments, said kit comprises a liquid or aqueous fiber composition as provided herein.

In particular embodiments, the fiber composition in the kit comprises a natural or
25 synthetic insoluble fiber, which may have a smooth or coarse surface, which may be solid or hollow and/or which may be crimped or curled. Examples of natural fibers include cellulose based fibers, like cotton fibers, hemp or jute fibers, flax fibers, and the like. Examples of synthetic fibers include polyamide fibers, such as nylon, polyester fibers, acrylic fibers, fibers made from regenerated cellulose, and the like. In particular
30 embodiments the fiber length ranges from about 0.5 to 100 mm, more preferably ranges from about 1 or 2 mm to about 10 or 15 mm. In particular embodiments, the average fiber diameter is in the range of from about 0.5 to about 100 microns.

In particular embodiments, said fiber composition may comprise one or more of the following agents, as described herein:

- 35 - an adhesive compound, preferably a natural, water soluble adhesive agents.
Preferred adhesive compounds include water-soluble starch and derivatives

thereof, sugars or sugar alcohols, such as sucrose, fructose, glucose, maltose, trehalose, galactose, raffinose, mannitol, sorbitol, and the like;

- a nutritional source;
- a fungal reducing agent;
- 5 - a pest repellent
- plant metabolism modifying compounds

In particular embodiments, the predatory mite population or source thereof is a container comprising a predatory mite population or source thereof. The container may be of any
10 type which is suitable for restraining mite individuals. It may be open, or closed. The shape of the container is not critical, and may for example be cuboid, cylindrical, etc. In particular embodiments, the container does not have a fixed shape, and may for example comprise a bag. If closed, the container may comprise means which facilitate exchange of metabolic gases and heat between its interior and its exterior, such as
15 ventilation holes, which may be covered e.g. with a mesh to prevent the escape of individuals of the mite population from the container. In particular embodiments, the container is adapted to the (controlled) release of mobile stages of the predatory mite in a plant. This is particularly suitable for the introduction of predatory mites. For example, the container may be provided with one or more apertures which can be opened and
20 closed. In particular said source of a predatory mite population comprises sexually mature adults from both sexes, and/or individuals of both sexes of other life stages, e.g. eggs, larvae and/or nymphs, which can mature to sexually mature adults; and/or fertilized female predatory mites.

25 As detailed above, the inventors have found that particular methods of spraying the composition comprising fibers described herein on the plants are particularly effective for ensuring adequate fiber application to the plants of interest. Accordingly, another aspect of the present invention provides a device for applying the fiber compositions provided herein. More particularly, the present invention also provides a moveable motor driven
30 liquid sprayer apparatus, preferably a trailer or trolley motor driven liquid sprayer apparatus or a portable motor driven liquid sprayer apparatus, such as a motor driven backpack liquid sprayer apparatus, adapted to spray the aqueous composition comprising fibers as provided herein in a uniform way.

Said motor driven liquid spraying apparatus comprises (i) a reservoir or container (1)
35 configured for containing the liquid to be sprayed, in particular the liquid fiber composition as provided herein, wherein said container (1) comprises an outlet passage

for the liquid to be sprayed; (ii) a motor powered pump assembly (not shown in figure 1) for dispensing the liquid through a spraying means, typically through a manually controlled wand (not shown in figure) equipped with a spray nozzle, wherein the inlet port of said pump assembly is connected with the outlet passage of said reservoir by tubings
5 and wherein the outlet port of said pump assembly is connected with the dispensing wand; further comprising (iii) means for regulating or interrupting the liquid flow adapted to avoid fiber induced liquid blockages, preferably a pinch valve or a ball valve (2); and/or (iv) a stirring means or a means for suspending the fiber in the liquid solution (3), preferably an axial-flow or radial-flow impeller, more preferably an axial-flow impeller,
10 located in said liquid reservoir (Figure 1).

In particular embodiments, the spray nozzle has one or more openings that correspond to the size of the fiber. For instance, in particular embodiments, the one or more openings of the spray nozzle have a diameter of 100-150 μm .

In preferred embodiments, the ratio of the diameter of the impeller (d), in particular an
15 axial flow impeller, to the diameter or width of the reservoir (1) (D) ranges from about 0.3 to about 0.6, preferably ranges from about 0.4 to about 0.5 (Figure 2). In particular embodiments, the impeller is positioned at a distance (C) from the bottom of the reservoir (1) ranging from about 1/3 of the impeller diameter to about 1/2 of the reservoir diameter or reservoir width.

20 In particular embodiments, the impeller is configured to limit or to avoid the fibers sticking or adhering to the impeller.

In particular embodiments, the reservoir (1) comprises baffles attached to the wall of the reservoir to interrupt the circular flow induced by the impeller, particularly in cylindrical reservoirs, and to promote the even dispersion of the fibers in the reservoir.

25 Said motor powered pump assembly may comprise an internal combustion engine or may be electric powered, such as a battery operated motor assembly.

In particular embodiments, said motorized liquid sprayer apparatus comprises fixing means to securely attach the sprayer apparatus to the back of a person, such as straps which may be attached to the upper and optionally also the lower portions of the sprayer
30 apparatus.

The methods and means for the biological control of plant pests by predatory mites described herein, may thus be used for protecting any plant, organism or item for which biological control of pests, more particularly mites is desired. Preferably said plant is a
35 herbaceous crop, a vine, an ornamental plant or a tree crop plant. In particular embodiments, the methods and means of the present invention is particularly suited for

the biological protection of plants or plant structures having little or no domatia or trichomes, in part having little or no non-glandular trichomes, such as hairs. Exemplary plants include herbaceous crops, such as some brassicas, grape, ornamental plants, such as roses, or tree crops (e.g. pear trees, *Citrus* spp.).

5

The following examples are provided for the purpose of illustrating the present invention and by no means are meant and in no way should be interpreted to limit the scope of the present invention.

10 EXAMPLES

Example 1. Behaviour of predatory mites on plants without trichomes

The numbers of eggs attached by *E. galicus* predatory mites on small citrus trees with or without fiber application was evaluated in three setups.

15

In a first set up, two groups of six plants were used. In the first group, 5 leaves of each citrus plant were inoculated with 4 fibers of about 1 – 1.5 cm and 5 predatory mites were introduced on each leaf with fibers. In the second (control group), 5 leaves were marked on each citrus plant, no fibers were applied and 5 predatory mites were introduced on each marked leaf. Plant pollen were provided as a source of nutrients for the mites.

20

After seven days, about one egg was found on each leaf having fibers, while no eggs were found on the control leaves (without fibers).

In a second setup, an aqueous 0.05% fiber composition was sprayed by a motorized sprayer on a number of small citrus trees (n=3). On each tree, 200 *E. galicus* predatory mites were introduced via a bio-box.

25

After thirteen days, about two eggs were found on leaves having fibers, while no eggs were found on the leaves of the citrus plants which had not been treated with the fiber composition.

30

In a third setup, the mite egg laying on the top or underside of a leaf of a citrus plant was evaluated. Three groups of six plants were used: a first group was used as the control group and no fibers were applied to the leaves, but 5 leaves were marked & 5 predatory mites were introduced on the marked leaves. In the second group, the top surface of 5 leaves of each plant were inoculated with 4 fibers of about 1 – 1.5 cm and 5 predatory mites were introduced on each leaf with fibers. The third group was treated as the

35

second group, but with the fibers applied on the underside of the leaves. For each group of plants, plant pollen were provided as a nutritional source for the mites.

No eggs were found on the leaves of the control group. On average about 0.4 eggs were found on the leaves with the fibers attached to the upper side, while on average about 1.6 mite eggs were found on leaves with the fibers attached to the underside of the leaf. This indicates that the *E. galicus* mite has a preference to lay its eggs on the underside of the leaves.

Example 2. Fiber application to plants

Fiber application to plants (in pots) was evaluated with different sprayer systems. Typically, spraying systems in agriculture (such as spray booms and spray wands) have color coded nozzles (SO 10625:2005).

Initially, as fibers may block the nozzles, the effect of the presence of fibers in water on the spraying quality and efficiency was evaluated for a series of 0.05 wt% aqueous fiber compositions (fiber length 5 – 8 mm) for different nozzle types. Two fibers were used: Dralon X, a white, smooth synthetic (acrylic) fiber; and “fiber 2-1”, a natural brown, rough fiber. The results are presented in Table 1.

Table 1.

Nozzle type	Dralon X	Fiber 2-1
White	OK	OK
Gray	OK	OK (upto fiber concentration to 0.02 wt%)
Brown	OK	Blocked nozzle
Red	OK	Blocked nozzle
Blue	OK	Blocked nozzle
Yellow	Blocked nozzle	Blocked nozzle
Green	Blocked nozzle	Blocked nozzle

Secondly, plants were sprayed with a 0.05wt% fiber-in-water dispersion, using a Birchmeier Profi Star 5 hand spraying system with Blue nozzle wherein all filters were removed from the hand spraying system, including the filter from the spraying wand/spraying nozzle. Spraying was at a pressure of 4 bar. Care was taken to fully disperse the fiber in the water by shaking the sprayer before use.

The results show that, for each fiber, about 50 to 55 fibers were on average found on the top side of each leaf of the citrus pot plant, while 0 to 10 fibers were found on the bottom side of the citrus pot plant.

This experiment showed that in order to apply fibers on the underside of the leaves the spraying wand should be pointed upwards, especially if a spraying system without air assistance is used. In addition, spraying systems with air assistance yield smaller water droplets which have the advantage of a better distribution of the water and fiber in the
5 plant canopy.

Example 3. Fiber adherence

In another experimental setup, the addition of different compounds to the fiber-in-water suspension was studied to improve the adherence of the fiber to the leaves.

10 4 different mixtures were tested: (1) DralonX fibre (0.05 wt%) in water; (2) DralonX fibre (0.05 wt%) in water + 1% starch; (3) DralonX fibre (0.05 wt%) in water + 0.5% sugar; (4) DralonX fibre (0.05 wt%) in water + 1% sugar.

Each composition was sprayed over a group of six citrus plants (height 40 cm) using a Birchmaier Profi Star 5 sprayer with gray nozzle. The effect of the wind was simulated
15 using a Makita BUB182 blower, positioned at 2m of the plants. 5 leaves were marked and the number of fibers were counted before and after the blowing treatment.

At lower blowing speed (and hence lower wind speed), all fibers remained on the leaves. At the maximum blower setting, about 1 fiber was lost per leaf when fibers were applied to the plants using the compositions comprising 0.5 and 1 % sugar, while fiber
20 application using the compositions comprising starch or no additives (only water) resulted in the loss of about 2 fibers per leaf. This experiment shows that adding a compound with adhesive properties to the fiber composition promotes the adherence of the fibers to the leaves.

In addition, when using starch as adhesive agent, white droplets/spots were visible on
25 the leaves.

Example 4. Fiber application via a motorized backpack spraying apparatus

Motorized sprayers that are portable on the back of a person are widely used in orchards for spraying crop trees. Indeed, trees are characterized by thick stems, lots of leaves and
30 a small number of trees per area. A motorized sprayer apparatus, particularly with air assistance, is ideally suited for the spraying of a liquid on both sides of the leaf, including those leaves in the center of the tree. Indeed, a turbulent air stream comprising the liquid is generated which leads to the formation of multiple small aerosol droplets, which are attracted by the leaves. The turbulent air stream also moves the leaves and branches in
35 different directions, ensuring an excellent dispersion of the sprayed liquid throughout the tree.

In the present example, the inventors have looked at the motorized spraying apparatus for applying the fibers to the crop plants.

First, some areas sensitive to blockage were adapted to reduce the risk of obstruction of the liquid flow by the fibers by replacing the existing valves with a pinch valve or a ball valve.

Secondly, some adaptations to a rectangular sprayer tank were made in order to keep the fibers well suspended. In certain instance, because the engine is located adjacent to the liquid tank, the engine vibrations are sufficient to keep the fibers in suspension. In an alternative setup, a motorized sprayer apparatus was developed wherein the sprayer tank was fitted with a DC engine with an axial-flow impeller (pitch blade impeller, 45° blades). This setup resulted in well dispersed fiber suspensions upon activation of the axial-flow impeller. An anti-ragging impeller may be used to avoid attachment of the fibers to the impeller.

Example 5. Fiber preference

Different fibers have been tested as oviposition substrates for predatory mites to assess if the mites show preference for a fiber type or size. The size/mass is expressed as dtex.

Series 1

Test setup:

Different kinds of synthetic fibers, varying in density/size were used. Dralon X is an acrylic fiber. The type II fibers were synthetic polylactide fibers.

The different fibers were attached on a plastic plate and placed in a circle or equidistant from a central point. Female mites (*Amblyseius swirskii*) and *Typha* pollen as a food source were placed in the central area. The fiber preference for egg laying was assessed by counting the number of eggs on the different fibers.

Table 2. Series 1: overview of fibers used.

Type I.1	Dralon X	251 bright 0.6 dtex 32 mm
Type I.2	Dralon X	351 bright 0.9 dtex 40 mm
Type I.3	Dralon X	250 bright 1.3 dtex 40 mm
Type I.4	Dralon X	250 bright 2.2 dtex 50 mm
Type I.5	Dralon X	100 bright 3.3 dtex 60 mm
Type II.1	Synthetic fiber, 50 dtex	
Type II.2	Synthetic fiber, 100 dtex (textured fiber)	

Type II.3	Synthetic fiber, 160 dtex
Type II.4	Synthetic fiber, 260 dtex
Type II.5	Synthetic fiber, 370 dtex
Type II.6	Synthetic fiber, 610 dtex
Type II.7	Synthetic fiber, 980 dtex
Type II.8	Synthetic fiber, 2170 dtex
Type II.9	Synthetic fiber, 3930 dtex

Results

Table 3. Number of eggs per fiber

Fiber type	Number of eggs (after 2 days)	Number of eggs (after 10 days)
Type I.1	12	58
Type I.2	0	75
Type I.3	18	42
Type I.4	3	84
Type I.5	10	45
Type II.1	3	37
Type II.2	0	21
Type II.3	0	18
Type II.4	0	24
Type II.5	2	33
Type II.6	4	26
Type II.7	1	33
Type II.8	2	25
Type II.9	5	21

5

A selection of the Type I & Type II fibers were further assessed, wherein the different fibers were placed in a hexagon formation, alternating between type I & Type II, with 15 female mites and pollen placed in the center. Unhatched eggs were counted after 2 and 13 days.

10

Table 4. Number of eggs per fiber type

Fiber type	Number of eggs (after 2 days)	Number of eggs (after 13 days)
Type II.3	3	16
Type I.5	0	9
Type II.2	9	42
Type I.4	0	2
Type II.1	7	44
Type I.1	3	2

Under the test conditions used, the best fibers for oviposition were the type II.1 50 dtex & and II.2 100 dtex synthetic fibers. In addition, Type II.2 fiber had a different fiber
 5 outline/shape, indicating that fiber architecture may play a role as well.

Series 2

Test setup:

Type II.2 fibers, either straight or curled, were attached on a plastic plate and placed
 10 equidistant from a central point. Female mites (*Amblyseius swirskii*) and *Typha* pollen as a food source were placed in the central area. The preference for egg laying was assessed by counting the number of eggs on the different fibers after 1 day and after 2 days.

15 Table 5. Number of eggs per fiber type

Fiber type	Number of eggs (after 1 day, mean of two plates)	Number of eggs (after 2 days, mean of two plates)
Type II.2, straight	16	27
Type II.1, curled	30	58

These results show that the curled fibers were visited circa twice as often for egg laying than the straight fibers. In addition, visual inspection showed that, in the case of the straight fibers, generally only one egg was founded at the ends of the fiber, while for the
 20 curled fibers, the eggs were positioned more uniformly over the fiber length. This was further confirmed in a separate series of experiments, which showed that thicker fibers, which were less curled/crimped, were substantially less used as oviposition substrate, compared to thin, curled fibers.

CLAIMS

1. A method for increasing the attractiveness of a plant or plant structure for a predatory mite population, said method comprising applying a liquid composition comprising a fiber to said plant or plant structure.
5
2. The method according to claim 1 wherein said liquid composition comprises between 0.01 to 5.0 wt% fibers.
- 10 3. The method according to claim 1 or 2 wherein the fiber has at least one of the following properties:
 - the fiber has a fiber length ranging from 0.5 to 100 mm;
 - the fiber is crimped or curled.
- 15 4. The method according to any of the preceding claims, wherein said liquid composition comprising a fiber further comprises an adhesive compound.
5. The method according to claim 4 wherein said adhesive compound is a sugar, a sugar alcohol, a soluble starch or a derivative thereof, arabic gum, xanthan gum, pectin or colloidal silica.
20
6. The method according to any one of the preceding claims, wherein the liquid composition comprising a fiber is applied by spraying or nebulization.
- 25 7. The method according to any of the preceding claims wherein the plant is a herbaceous plant, a vine, an ornamental plant, a tree or a plant structure such as a bulb, a tuber, or a fruit.
8. The method according to claim 7, wherein the plant is a pear tree or a Citrus spp. tree.
30
9. The method according to any one of the preceding claims, which is a method for the biological control of pests on plants by predatory mites.

10. The method according to claim 10, which further comprises contacting said plant or plant structure with said predatory mite population before, after or simultaneously with the application of the liquid fiber composition.
- 5 11. A kit for the biological control of pests on a plant comprising an aqueous fiber composition and, separated therefrom, a predatory mite population, or a source thereof, wherein the length of the fiber ranges from 0.5 to 100 mm and/or wherein the fiber is crimped or curled.
- 10 12. The kit according to claim 11 further comprising one or more of the following compounds: (i) an adhesive compound, (ii) a nutritional source for said predatory mites; (iii) a fungal reducing compound; (iv) a pest repellent; (v) a plant metabolism modifying agent.
- 15 13. A liquid composition comprising a fiber for providing a fiber on a plant or plant structure wherein the length of the fiber ranges from 0.5 to 100 mm and/or wherein the fiber is crimped or curled.
14. The liquid composition according to claim 13 further comprising one or more of
20 the following compounds: (i) an adhesive compound, (ii) a nutritional source for predatory mites; (iii) a fungal reducing compound; (iv) a pest repellent; (v) a plant metabolism modifying agent.
15. A container comprising the liquid composition according to claim 13 or 14.
25
16. The container according to claim 15, which is comprised within a liquid sprayer device.
17. A motor driven liquid sprayer apparatus configured for spraying a liquid fiber
30 composition in an uniform way, comprising:
(i) a reservoir (1) configured for containing the liquid to be sprayed, wherein said container (1) comprises an outlet passage for the liquid to be sprayed;
(ii) a motor powered pump assembly for dispensing the liquid through a spraying means equipped with a spray nozzle, wherein the inlet port of said pump assembly is connected with the outlet passage of said reservoir (1) by
35

tubings and wherein the outlet port of said pump assembly is connected with the spraying means;

characterized in that said motor driven liquid sprayer apparatus further comprises

- 5 (iii) a means for regulating or interrupting the liquid flow (2) adapted to avoid fiber induced liquid blockages; and/or
(iv) a stirring means or a means for suspending the fiber in the liquid solution (3), located in said liquid reservoir.

- 10 18. The motor driven liquid sprayer apparatus according to claim 17 wherein:
(a) said stirring means (3) is an axial flow impeller; and/or
(b) said spray nozzle has one or more openings with a diameter of 100-150 μm ; and/or
15 (c) said means for regulating or interrupting the liquid flow is a pinch valve or ball valve.

19. The motor driven liquid sprayer apparatus according to claim 17 or 18, which further comprises a liquid composition comprising a fiber for providing on a plant or plant structure.

20

20. The method according to claim 6 wherein the liquid composition comprising a fiber is applied using the motor driven liquid sprayer apparatus according to claim 17 or 18.

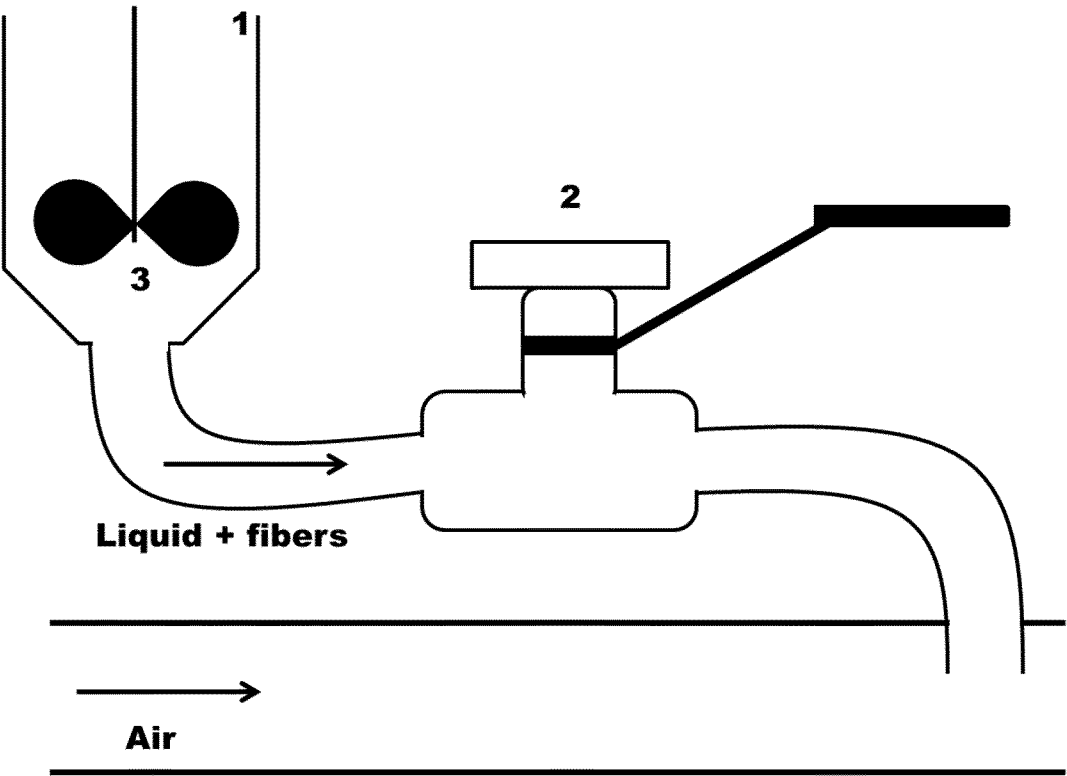


FIGURE 1

2/2

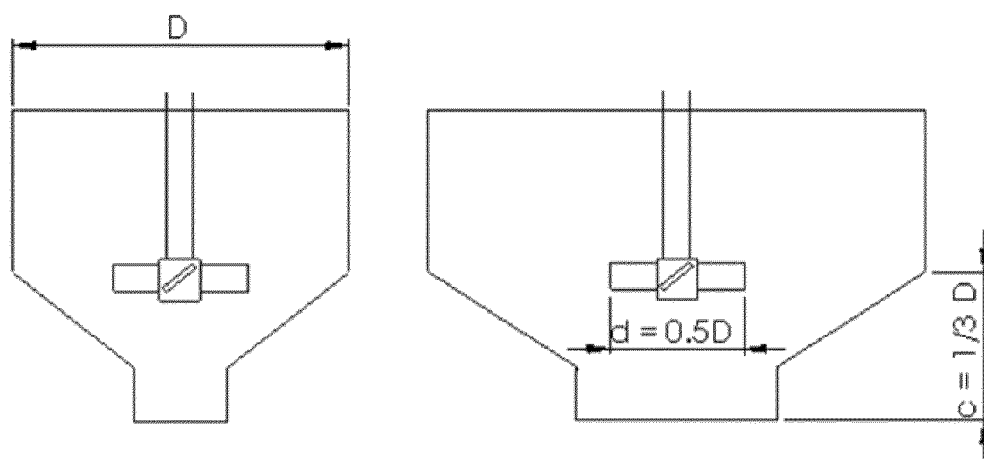


FIGURE 2