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(56) Related Art
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(54) **Title:** PROCESS FOR MANUFACTURING HYDROPHOBIC POLYMERS

(57) **Abstract:** Method for producing hydrophobic polymers, wherein a hydrophobic polymer is selected; a set of organisms is selected from among cells and/or cells products; aggregates are formed by working said cells and/or cell products in said polymers resulting in the formation of a so-called polymer-bio aggregate, wherein there is performed a new function of the thus polymer product. This invention further relates to the use of a so-called P.B.A. obtained therewith in specific applications.

Process for manufacturing hydrophobic polymers

5 **Field of the invention**

The present invention relates to a method for producing hydrophobic polymers incorporating living organisms and/or cell products.

10 **Prior art**

This type of method is known for non-permanent, biodegradable hydrophilic polymers with a melting point well below 100°C, in which temperature-sensitive tissue cells and organic molecules are incorporated. The polymer degrades after a
15 short time.

American patent US-5,985,354 in the name of MATHIOWITZ describes a method of the above type. The problem encountered in the known prior art is that the melting point of permanent, non-biodegradable polymers is well above 100°C at
20 normal pressure conditions for the incorporation of living material. The implantation of living, active organisms or microorganisms at temperatures of this level is impossible without fatal consequences for these organisms. Accordingly, it has to be assumed that the incorporation of living material during the production of a usable object from the base material, in this case the polymer, is not achievable,
25 even if the organisms which are introduced can subsequently perform useful activity at normal ambient temperatures. Possible activities in this respect are oxygen consumption or absorption, absorption of radiant energy, including what is known as "UV blocking", and the like. The above therefore demonstrates that currently there are considerable restrictions in the possible range of applications
30 for living cells in this type of polymer.

A further problem encountered in the known prior art is that the slow diffusion of cellular components and biomolecules in a moist environment is based on a
35 technology which is predicated on multi-wall microcapsules of hydrophilic, soluble or biodegradable polymers. However, there is currently no available technology which permits the slow diffusion of a gamma to biomolecules from a permanent

polymeric carrier without it being degraded in an aqueous and/or dry environment. This applies in particular to biomolecules of the hydrophobic fatty acid type, such as lipids and hydrocarbons.

5 Therefore, the known prior art mainly has the following shortcoming, namely that bio-encapsulation of cells in polymers is not possible above 100°C under standard pressure conditions, on the one hand, and that slow diffusion of cellular metabolites and related organic molecules from polymers is not possible without degradation of the polymer in a moist environment, on the
10 other hand.

The discussion of the background to the invention herein is included to explain the context of the invention. This is not to be taken as an admission that any of the material referred to was published, known or part of the common general
15 knowledge as at the priority date of any of the claims.

Where the terms "comprise", "comprises", "comprised" or "comprising" are used in this specification (including the claims) they are to be interpreted as specifying the presence of the stated features, integers, steps or components,
20 but not precluding the presence of one or more other features, integers, steps or components, or group thereof.

Summary of the invention

25 Viewed from one aspect, the present invention provides a method for producing polymers, wherein

- a polymer is selected;
- a set of organisms is selected from among cells, living organisms and/or cell products;
- 30 - aggregates are formed by working said cells and/or cell products in said polymers resulting in the formation of a so-called polymer-bio aggregate, wherein work is carried out at the working temperature range taken from the temperature interval of which the lower limit is set at substantially 100°C under substantially standard pressure conditions.

According to the present invention, there is proposed a method for producing hydrophobic polymers in which a hydrophobic polymer is selected, and furthermore a set of organisms is selected from cells and/or cell products. Remarkably, aggregates are then formed by working the said cells into the said polymer, resulting in the formation of what is known as a polymer-bio aggregate, referred to below as PBA, producing a novel function of the polymer product formed in this way.

According to a preferred embodiment of the invention, further to the main measure defined above, there is defined that the work is carried out in the operating temperature range taken from the temperature range for which the lower limit is set at virtually 100 °C under virtually standard pressure conditions, in particular at virtually one atmosphere.

According to a further preferred embodiment of the invention, there is defined that the abovementioned cells are selected from the category of what are known as the cysts and/or in a phase of inactive or dormant stages. A quite significant number of types of organisms or microorganisms can change from an active life form to what is known as a quiescent stage or spore, known as cysts. Said spores are able to withstand extreme environmental fluctuations in a latent form. In this state of anabiosis, they are able to withstand extremely dry conditions and temperatures well above 100 °C

Under suitable biotechnology conditions, these types can not only be cultivated but also converted, in controlled culture conditions, known as encysting, into usable spores for bio-encapsulation in a polymeric matrix.

During the production process of an industrial product, such as packaging material, textile fibers, granules or the like, said spores and the polymer are agglomerated within a short period of time during which the polymer is liquid, namely at a temperature above its melting point. This produces what is known as a polymer-bio aggregate, referred to below as "PBA".

As long as the product is not in use, the organisms of the bio-component in said PBA remain inactive. However, as soon as the living conditions become favorable, coinciding with the product starting to be used in association with an environment which is suitable for life in terms of temperature and relative humidity, the spores change into active, metabolizing cells under these favorable ambient conditions. For this period, the biologically active form will perform its intended function. As soon as the optimum conditions revert to conditions which are less than optimum, the active form returns to the spore.

The process remains reversible in accordance with a feedback mechanism which is controlled by the living environment of the organism in said PBA.

Thus, according to a particularly preferred embodiment of the invention, said cell products are selected from the category of so-called metabolites, i.e. the molecules which are biochemically synthesized by organisms under the abovementioned temperature working conditions.

According to yet another preferred embodiment of the present invention, the polymers are selected from non-biodegradable polymers. Reliable, slow and prolonged diffusion of organic molecules out of polymers into a moist or fluctuating environment can be realized without degradation of the polymer.

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An advantage obtained by virtue of the method defined by the present invention is mainly that the biological activity of the organisms incorporated in the so-called PBA produced in accordance with the invention imparts novel, previously unknown properties to the polymer. Said PBA hereby ensures the desired environment for which said PBA was made.

A further advantage consists in the standardized release of very specific biomolecules, such as so-called repellents, from a polymeric matrix, such as granules, textile fibers and the like, without said polymeric matrix being lost in a variable environment as a result of climatological instabilities, for example.

Further features and properties of the present invention are defined in further sub-claims.

Further details and particularities will emerge from the following description of a number of exemplary embodiments of the method according to the invention and its uses.

Description

In general terms, the present invention relates to a method for producing hydrophobic polymers which incorporate living organisms and/or cell products. A number of specific use examples are described below.

A particularly significant application area is in the food packaging sector which employs what is known as an oxygen barrier, with a PBA layer arranged as an intermediate layer in multi-layer packaging material for foodstuffs, such as PET bottles for beverages, such as beers or fruit juices for example. The polymer component of the PBA is in this case PET, while the PBA bio component is a type of yeast with a dry spore, such as for example *Saccharomyces*, which is able to withstand the high temperatures of the production process. The PBA remains inactive until the PET bottle has been filled. When the package is being filled with fruit juices or beer, for example, the internal environment of the PBA becomes water-saturated, with the result that the spores are activated to form respiring cells which consume all the oxygen present inside the bottle. As a result, all the oxygen is withdrawn from the contents under the influence of what is known as the O₂

scavenger. Also, all the external oxygen which can diffuse through the wall is captured by the yeast cells for respiration, which results in an efficient oxygen barrier.

5 A further example of a use consists in the action as a UV blocker, which works in a similar way to the above example. Instead of the yeast cells, there is incorporated in the PBA a type of alga, such as for example *Haematococcus*, the spores of which very intensively block UV light. A continuous layer of *Haematococcus* cells, haematocysts with a high concentration of astaxanthin, makes the PBA opaque to
10 UV light. This fact is utilized in moisture-resistant UV-proof films and polymer coverings.

A still further use consists in the combined application of both examples mentioned above in connection with food packaging with an oxygen barrier and a
15 UV blocker which is suitable for PET bottles as packaging for beers and fruit juices and the like. The PBA biocomponent is a calibrated mixture of *Saccharomyces* and *Haematococcus*. Said oxygen scavengers, such as yeast cells for example, represent a permanent oxygen barrier, while the UV blocker, such as a type of alga, for example, prevents photochemical degradation of the filling.

20 Yet another application consists in the absorption of energy from sunlight with a cooling effect which is similar to the example above relating to the so-called UV blocker. Instead of *Haematococcus*, the PBA incorporates a type of alga such as for example *Chlorococcus*, the active form of which, in the presence of a high
25 degree of moisture, participates very intensively in photosynthesis, consuming high-energy rays of the sunlight. A continuous layer of cells will provide the PBA with an energy-absorbing function, resulting in a non-heating, in other words cooling, effect at the bottom of the polymer. The above effect is utilized in moisture-resistant films and polymer coverings for sun-shielding purposes.

30 Finally there is the application example ranging from energy-absorbing cloth to perspiration-sensitive sports clothing which derives from the previous example. A PBA with a polymer component of polypropylene and a biocomponent of photosynthesizing organisms, such as a cyanobacterium or a unicellular alga type,
35 is extruded to form a textile fiber. The temperature-resistant spores of the algae, after they have been extruded to form a fiber, are processed to produce a textile

product. Use of fibers of this type in textile products ranges from covering fabrics, such as canvas, to sports clothing. For the absorption of moisture, for example sweat, the incorporated cells will convert the incident energy of sunlight into photosynthetic metabolites. As a result, the incident solar radiation is not converted into heat, but rather is extracted from the textile fiber, resulting in the desired cooling action. When drying out when no further sweat is being produced, the cells revert to their latent, inactive state. This is because the process is reversible.

10 Application examples relating to slow diffusion of cellular components and at least partially hydrophobic biomolecules in a moist environment are described below.

In a variant on the UV blocker from the above example, the active metabolite, astaxanthin, which very intensively blocks UV light, is incorporated in the PBA instead of the *Haematococcus* cells. As an alternative to the expensive component astaxanthin, it may be possible to use less expensive UV blockers. The diffusion rate of the UV blocker from the PBA in the middle layer of the polylamellar film to the periphery is regulated at a low to very low diffusion rate, depending on the quality and requirements. This fact is exploited in moisture-resistant UV-repellent films and polymer coverings, as well as for packaging material for food products. The polymer must in this case be durable and must not deteriorate in moist conditions.

In this context, an additional example of application consists in insect-repellent films, fibers – textile – and microgranules. This represents a variant on the above example. In this case, the bio component of the PBA is a bio-active organic molecule or a mixture of molecules, preferably hydrophobic substances, such as lipids, fatty acids, isoprene derivatives and hydrocarbons. In addition to a film or laminate, the processed product may also be a PBA which is processed to form a textile fiber or granule or microgranule, in which the biocomponent is released to the environment at a predetermined rate. This component has a specific repellent action to insects. Examples which have been tested include:

PBA with isoprene derivatives and/or branched hydrocarbons with a repellent activity to house dust mites. The PBA is extruded to form a textile fiber for weaving a fixed carpet and other products which have to be resistant to house dust mites; and

PBA with fatty acid components which are repellent to diptera, namely flies and mosquitoes, and biting and blood-sucking lice, Mallophaga, Anoplura, respectively, as well as the human head louse and poultry lice, cockroaches, ants and wasps. The PBA is granulated or extruded to form a textile fiber. The laden
5 granules are mixed into the animal's coat, to protect against myiasis, horsefly and the like, or are scattered on the nesting site of the host of the parasite in question. Laden fibers are processed to form a protective textile as a nightcap to kill head lice, or what is known as a tissue with which an object can be rubbed to protect against ants, cockroaches, flies and the like.

10

Further to the above example, another important application is the use of the PBAs as a crop protection agent, in particular as a herbicide or even as a fungicide.

15 The biological activity of the organisms incorporated in the PBA gives the polymer new properties which were not previously known. The PBA ensures the desired environment for which the PBA was made, such as for example an anaerobic environment, complete oxygen barrier, energy absorption of solar radiation, controlled release of metabolites and the like.

20

The interaction and exchange of various types of organisms or microorganisms and/or molecules in the bio component of the PBA can also yield a large number of possible applications.

THE CLAIMS DEFINING THE INVENTION ARE AS FOLLOWS:

1. Method for producing polymers incorporating living material, comprising
 - selecting a polymer;
 - 5 - selecting a set of organisms from among cells and living organisms; and/or selecting cell products of said organisms;
 - forming aggregates by implanting said organisms and/or cell products in said polymers resulting in the formation of a so-called polymer-bio aggregate (PBA), wherein implantation is carried out in the operating temperature range for which the lower limit is set at substantially 100°C under substantially standard pressure conditions, wherein
10 said cells are selected from among the category of the so-called cysts and/or in the phase of inactive or sleeping stages and said living organisms can change from active life form to a quiescent stage or spore.
- 15 2. Method according to claim 1, wherein said pressure is at substantially one atmosphere.
3. Method according to claim 1 or 2, wherein the organisms are selected from prokaryotes, bacteria or eukaryotes.
20
4. Method according to claim 1 or 2, wherein the organisms are selected from eukaryotes of the type protists, fungi, plants, and/or animals.
5. Method according to any one of claims 1 to 4, wherein said cells products are selected
25 from the category of so-called metabolites, being the molecules which are biochemically synthesized by organisms.
6. Method according to claim 1 or 2, wherein said organisms are unicellular.
- 30 7. Method according to claim 1 or 2, wherein said organisms are multicellular.
8. Method according to any one claims 1 to 7, wherein the polymers are selected from non-biodegradable polymers.
- 35 9. Method according to any one of claims 1 to 7, wherein the polymers are selected from the family of the polyolefins, or polyesters.

10. Method according to claim 9, wherein the polymers are selected from the family of the polyethylenes, or polypropylenes.
11. Method according to claim 9, wherein the polymer is PET.
12. Method according to any one of claims 1 to 11, wherein said organisms and/or cell products are imbedded in said polymer.
13. Method according to any one of claims 1 to 11, wherein said organisms and/or cell products are implanted into said polymer whilst the polymer is being produced.
14. Method according to any one of the claims 1 to 11, wherein said organisms and/or cell products are implanted into an existing polymer.
15. Method according to any one of the claims 1 to 11, wherein said organisms and/or cell products are blended in an existing polymer, forming an intermediate layer in a multilayer packaging material.
16. Method according to any one of claims 1 to 15 wherein the cysts and the polymer are aggregated within a short period of time during which the polymer is liquid, i.e. with its temperature above its melting point.
17. Method according to claim 1 or 2, wherein a PBA layer is arranged as an intermediate layer in a multilayer packaging material for foodstuffs.
18. Method according to claim 17, wherein the multilayer packaging material for foodstuffs is a PET bottle for beverages.
19. Method according to claim 17, wherein the polymer component of the PBA layer is composed of PET, and the organism is a type of yeast with a dry cyst, which is able to withstand high temperatures of the production process.
20. Method according to claim 19, wherein the yeast is *Saccharomyces*.
21. Method according to claim 19 or claim 20, wherein instead of and/or optionally in combination with the yeast cells, there is incorporated a type of alga in the PBA, the cysts of which block UV-light very intensively.

22. Method according to claim 21, wherein the selected alga is Haematococcus or Chlorococcus.

5 23. Method according to claim 22, wherein astaxanthin, a cell-product of Haematococcus, is incorporated in the PBA instead of said Haematococcus.

10 24. Use of the polymer-bio-aggregate obtained according to a method as defined in any one of claims 1 to 23, as a barrier, an insect-repellent agent, or a crop protection agent.

25. Use of the polymer-bio-aggregate obtained according to a method as defined in one of the claims 1 to 23, as an herbicide, or a fungicide.

15 26. Polymer-bio-aggregate produced by the method of any one of claims 1 to 23.