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(54) METHOD FOR PRODUCING TOBACCO-FLAVOR LIQUID, TOBACCO-FLAVOR LIQUID, TOBACCO ADDITIVE, AND FLAVOR INHALER

(57) This method for producing a tobacco-flavor liquid includes: extracting, from a tobacco material, a water-soluble component included in the tobacco material by using an aqueous solvent to obtain a tobacco extract liquid; culturing yeast in the tobacco extract liquid to obtain a yeast-containing culture liquid; and eluting, from the yeast included in the yeast-containing culture liquid, an active component included within fungal cells of the yeast by using an organic-solvent-containing elution solvent to obtain a tobacco-flavor liquid that includes the active component and the post-culturing tobacco extract liquid.

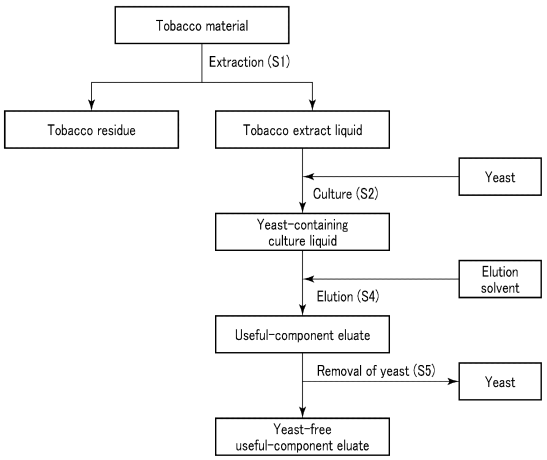

FIG. 1

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Description

FIELD

[0001] The present invention relates to a method for producing a tobacco flavor liquid, a tobacco flavor liquid, a tobacco additive, and a flavor inhaler.

BACKGROUND

[0002] Leaves of a tobacco plant are harvested and then subjected to various processes including drying in a farm house, subsequently one to several years of long-term aging in a leaf processing facility, and, subsequently to that, blending and cutting in a manufacturing facility. The processed leaves are then used to produce a flavor inhaler such as a cigarette. Through these various processes, leaves of a tobacco plant become a flavorful tobacco material. Such a tobacco material is referred to as "leaf tobacco" to enable distinction from the leaves of a tobacco plant. The leaf tobacco itself may be used as a flavor source of a flavor inhaler; alternatively, a tobacco extract liquid obtained by extracting a tobacco flavor component from the leaf tobacco may be used as a flavor source of a flavor inhaler.

[0003] Leaf tobacco contains various flavor components, and it is known that leaf tobacco or a tobacco extract liquid is subjected to chemical treatment or biological treatment to increase the amounts of these flavor components. For example, Patent Literature 1 discloses a method for preparing aromatized reconstituted tobacco from tobacco particles, and the method includes extracting tobacco particles with water to obtain an extract and a residue, thereafter fermenting the obtained extract with yeast to prepare a fermented liquid, preparing reconstituted tobacco from the obtained residue, and adding the fermented liquid to the reconstituted tobacco, thereby preparing aromatized reconstituted tobacco.

CITATION LIST

PATENT LITERATURE

[0004] Patent Literature 1: U.S. Patent No. 4895175

SUMMARY

TECHNICAL PROBLEM

[0005] An object of the present invention is to provide a technique relating to a tobacco extract liquid having increased amounts of useful components such as flavor components.

SOLUTION TO PROBLEM

[0006] The inventors of the present invention focused their attention on the fact that when yeast is cultured in a tobacco extract liquid to produce useful components from the yeast, many of those useful components remain in the fungal cells of the yeast, resulting in insufficient release of the useful components into the tobacco extract liquid. By improving this aspect did the inventors complete the present invention.

[0007] Specifically, according to one aspect, there is provided a method for producing a tobacco flavor liquid, the method comprising:

extracting, from a tobacco material, a water-soluble component contained in the tobacco material by using an aqueous solvent, thereby obtaining a tobacco extract liquid;
culturing yeast in the tobacco extract liquid, thereby obtaining a yeast-containing culture liquid; and
eluting, from the yeast contained in the yeast-containing culture liquid, a useful component included in fungal cells of the yeast by using an elution solvent including an organic solvent, thereby obtaining a tobacco flavor liquid containing the useful component and the post-culturing tobacco extract liquid.

[0008] According to another aspect, there is provided a tobacco flavor liquid obtainable by the above-mentiond method.

[0009] According to further another aspect, there is provided a tobacco additive comprising:

the tobacco flavor liquid obtainable by the above-mentioned method; and
a tobacco residue obtained when the tobacco extract liquid is obtained in the above-mentioned method.

[0010] According to further another aspect, there is provided a flavor inhaler comprising the above-mentioned tobacco flavor liquid or the above-mentioned tobacco additive.

ADVANTAGEOUS EFFECTS OF INVENTION

[0011] According to the present invention, there is provided a technique relating to a tobacco extract liquid having increased amounts of useful components such as flavor components.

BRIEF DESCRIPTION OF THE DRAWINGS

[0012]

FIG. 1 is a flowchart showing an example of a method for producing a tobacco flavor liquid.
FIG. 2 is a flowchart showing another example of a method for producing a tobacco flavor liquid.
FIG. 3 is a cross-sectional view showing an example of a combustion-type flavor inhaler.
FIG. 4 is a perspective view showing an example of a heating-type flavor inhaler.

FIG. 5 is a diagram showing an internal structure of a tobacco stick.

FIG. 6 is a diagram showing an internal structure of an aerosol-generation device.

FIG. 7 is a microscopic image showing the state of yeast in the elution step (S4) of a method of a first embodiment.

FIG. 8 is a graph showing the concentration of fatty acids in a tobacco flavor liquid.

DETAILED DESCRIPTION

[0013] Hereinafter, the present invention will be described in detail; however, the description below is intended to provide a description of the present invention, and not intended to limit the present invention.

<1. Method for Producing Tobacco flavor liquid>

[0014] A method for producing a tobacco flavor liquid includes:

extracting, from a tobacco material, a water-soluble component contained in the tobacco material by using an aqueous solvent, thereby obtaining a tobacco extract liquid;

culturing yeast in the tobacco extract liquid, thereby obtaining a yeast-containing culture liquid; and

eluting, from the yeast contained in the yeast-containing culture liquid, a useful component included in fungal cells of the yeast by using an elution solvent including an organic solvent, thereby obtaining a tobacco flavor liquid containing the useful component and the post-culturing tobacco extract liquid.

[0015] Since this method includes eluting the useful components produced by the yeast into the tobacco extract liquid, a "tobacco extract liquid having increased amounts of useful components" is obtained. In the present disclosure, the "tobacco extract liquid having increased amounts of useful components such as flavor components" is referred to as a "tobacco flavor liquid".

[0016] In a first embodiment of the above-described method, the obtaining of the tobacco flavor liquid can be performed by:

mixing the yeast-containing culture liquid with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate; and

if necessary, removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate.

In the first embodiment, each of the yeast-containing "useful-component eluate" and the "yeast-free useful-component eluate" corresponds to the tobacco flavor liquid. The method according to the first embodiment will be detailed in <1-1. First Embodiment> below.

[0017] In a second embodiment of the above-described method, the obtaining of the tobacco flavor liquid can be performed by:

separating the yeast-containing culture liquid into the yeast and the tobacco extract liquid;

mixing the separated yeast with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate;

if necessary, removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate; and

mixing the useful-component eluate or the yeast-free useful-component eluate with the separated tobacco extract liquid, thereby obtaining a useful component-containing liquid.

[0018] In the second embodiment, each of the "useful component-containing liquid" prepared without the step of removing the yeast from the useful-component eluate and the "useful component-containing liquid" prepared through the step of removing the yeast from the useful-component eluate corresponds to the tobacco flavor liquid. The method according to the second embodiment will be detailed in <1-2. Second Embodiment> below.

<1-1. First Embodiment>

[0019] According to the first embodiment, a method for producing a tobacco flavor liquid includes:

(S1) extracting, from a tobacco material, a water-soluble component contained in the tobacco material by using an aqueous solvent, thereby obtaining a tobacco extract liquid;

(S2) culturing yeast in the tobacco extract liquid, thereby obtaining a yeast-containing culture liquid;

(S4) mixing the yeast-containing culture liquid with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in fungal cells of the yeast, thereby obtaining a useful-component eluate; and

(S5) if necessary, removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate.

[0020] The method according to the first embodiment is shown in FIG. 1. Hereinafter, the method according to the first embodiment will be described in the order of steps (S1), (S2), (S4), and (S5) with reference to FIG. 1.

As described above, the yeast-removal step (S5) may or may not be performed. That is, the "yeast-free useful-component eluate" obtained in the yeast-removal step (S5) may be used as the tobacco flavor liquid, or the "useful-component eluate" obtained in the elution step (S4) may be used as the tobacco flavor liquid with the yeast remaining therein.

[Extraction Step (Sl)]

[0021] In the extraction step (S1), a water-soluble component contained in a tobacco material is extracted from the tobacco material by using an aqueous solvent, whereby a tobacco extract liquid is obtained. In the extraction step (S1), a tobacco residue is also obtained at the same time as the tobacco extract liquid is obtained (see FIG. 1).

[0022] As the tobacco material, cut tobacco which is ready to be incorporated into a tobacco product, such as a combustion-type or heating-type flavor inhaler, can be used. The "cut tobacco which is ready to be incorporated into a tobacco product" refers to cut tobacco which is ready to be incorporated into a tobacco product through various processes including drying in a farm house, subsequently one to several years of long-term aging in a leaf processing facility, and, subsequently to that, blending and cutting in a manufacturing facility.

[0023] The cut tobacco consists of cut pieces of leaf tobacco. The cut tobacco may be any of the following: cut pieces of stemmed leaves, cut pieces of midrib, cut pieces of reconstituted tobacco (i.e., a tobacco material obtained by processing leaf scraps, cut tobacco scraps, midrib scraps, fine powder, etc., generated in the plant processes into a reusable shape), or a mixture thereof. A pulverized product obtained by pulverizing the cut tobacco may be used for the extraction in order to increase extraction efficiency.

[0024] As the cut tobacco, cut tobacco derived from any tobacco variety can be used. For example, cut tobacco derived from flue-cured tobacco, Burley tobacco, Oriental tobacco or the like can be used. As the cut tobacco, cut tobacco derived from a single variety, or a mixture of cut tobacco derived from different varieties may be used.

[0025] As the aqueous solvent, water or water-containing ethanol can be used. As the water-containing ethanol, a mixture of ethanol and water at a volume ratio of 1 : 1, for example, can be used. The aqueous solvent is generally water, and preferably water having a room temperature (e.g., approximately 20 °C) to a temperature of 70 °C. The aqueous solvent can be used, for example, in an amount of 500 to 5000% by mass with respect to the tobacco material.

[0026] The extraction can be performed by, for example, immersing the tobacco material in warm water having a temperature of 40 to 60 °C for 30 to 180 minutes, or shaking the tobacco material (e.g., 200 rpm) in warm water having a temperature of 40 to 60 °C for 30 to 180 minutes.

[0027] The extraction may also be performed by repeating the extraction multiple times. Specifically, the extraction may be performed by extracting, from the tobacco material, the water-soluble component contained in the tobacco material by using the aqueous solvent, and then placing the resultant tobacco residue in a new aqueous solvent to perform the second extraction, and as necessary, repeating the extraction using a new aqueous solvent.

[0028] A mixture of the tobacco residue and the tobacco extract liquid is obtained by the extraction. The tobacco extract liquid contains a water-soluble component contained in the tobacco material. Examples of the "water-soluble component contained in the tobacco material" include components serving as a nutrient source for yeast (e.g., saccharides, amino acids, proteins, and nutrient salts) and components that contribute to tobacco flavor (e.g., organic acids, foliar resins, terpenoids, and polyphenols).

[0029] After the extraction, the tobacco residue and the tobacco extract liquid are separated, and the tobacco extract liquid is used as a raw material for obtaining a tobacco flavor liquid. On the other hand, the tobacco residue can be used for preparing a tobacco filler by mixing the tobacco residue with the finally obtained tobacco flavor liquid and appropriately processing the resultant mixture. For example, the tobacco residue may be used for preparing a tobacco-molded body such as a sheet tobacco from a mixture obtained by mixing the tobacco residue with the finally obtained tobacco flavor liquid. Alternatively, the tobacco residue may be used for preparing a tobacco powder by mixing the tobacco residue with the finally obtained tobacco flavor liquid and drying and pulverizing the resultant mixture.

[Culture Step (S2)]

[0030] In the culture step (S2), yeast is cultured in the tobacco extract liquid obtained in the extraction step (S1), whereby a yeast-containing culture liquid is obtained (see FIG. 1).

[0031] Any type of yeast can be used as the yeast, provided that it can produce a useful component when cultured in the tobacco extract liquid.

[0032] The term "useful component" as used herein refers to a component which is useful for the finally obtained tobacco flavor liquid. The useful component may be, for example, a component that contributes to the flavor of the tobacco flavor liquid (hereinafter referred to as "a flavor-contributing component"), a component that colors the tobacco flavor liquid (hereinafter referred to as "a coloring component"), or a component that prevents putrefaction or fermentation of the tobacco flavor liquid (hereinafter referred to as "a preservative component").

[0033] The useful component is preferably a flavor-contributing component. The "flavor-contributing component" may be a flavor component that releases a flavor,

or a precursor that is converted into a flavor component when heated or burned in a flavor inhaler.

[0034] Examples of the flavor-contributing component include carotenoids, fatty acids, neutral fats (i.e., glycerin esters of fatty acids), acetic acid esters, fatty acid esters, organic acids, and higher alcohols (e.g., alcohols having 8 to 22 carbon atoms). Examples of the coloring component include carotenoids. Examples of the preservative component include lactic acid, fatty acid glycosides, and benzoic acid.

[0035] Therefore, the yeast known to produce the above-described useful components can be used in the method of the present invention.

[0036] Examples of the yeast that produces carotenoids include: yeast of the genus Rhodotorula, such as Rhodotorula alborubescens, Rhodotorula araucariae, Rhodotorula babjevae, Rhodotorula dairenensis, Rhodotorula diobovata, Rhodotorula evergladensis, Rhodotorula glutinis, Rhodotorula graminis, Rhodotorula kratochvilovae, Rhodotorula mucilaginosa, Rhodotorula ngohengohe, Rhodotorula pacifica, Rhodotorula paludigena, Rhodotorula sinensis, Rhodotorula sphaerocarpa, Rhodotorula taiwanensis, and Rhodotorula toruloides; and yeast of the genus Xanthophyllomyces, such as Xanthophyllomyces australis, Xanthophyllomyces rhodozyma, and Xanthophyllomyces tasmanica.

[0037] Examples of the yeast that produces fatty acids include: yeast of the genus Yarrowia, such as Yarrowia alimentaria, Yarrowia bubula, Yarrowia deformans, Yarrowia divulgata, Yarrowia galli, Yarrowia hollandica, Yarrowia keelungensis, Yarrowia lipolytica, Yarrowia osloensis, Yarrowia parophoni, Yarrowia phangngaensis, Yarrowia porcina, and Yarrowia yakushimensis; and yeast of the genus Lipomyces, such as Lipomyces anomalus, Lipomyces arxii, Lipomyces chichibuensis, Lipomyces doorenjongii, Lipomyces japonicus, Lipomyces kockii, Lipomyces kononenkoae, Lipomyces lipofer, Lipomyces mesembrius, Lipomyces okinawensis, Lipomyces oligophaga, Lipomyces orientalis, Lipomyces smithiae, Lipomyces spencermartinsiae, and Lipomyces starkeyi.

[0038] Examples of the yeast that produces acetic acid esters, fatty acid esters, or higher alcohols include: yeast of the genus Saccharomyces, such as Saccharomyces cerevisiae, Saccharomyces paradoxus, Saccharomyces bayanus, Saccharomyces uvarum, and Saccharomyces arboricola; yeast of the genus Cyberlindnera, such as Cyberlindnera jadinii, Cyberlindnera saturnus, Cyberlindnera fabianii, Cyberlindnera suaveolens, Cyberlindnera americana, and Cyberlindnera xylosilytyca; and yeast of the genus Wickerhamomyces, such as Wickerhamomyces anomalus, Wickerhamomyces ciferri, and Wickerhamomyces canadensis.

[0039] One type of yeast, or two or more types of yeast, may be cultured in the tobacco extract liquid. The yeast may also be recombinant yeast which is genetically modified to increase the production amounts of useful components.

[0040] The conditions for culturing the yeast are not particularly limited. Conditions suitable for the growth of the yeast to be used and the production of the useful components can be appropriately selected. Prior to the culturing, the yeast can be added to the tobacco extract liquid at a concentration of, for example, 10 to 10^8 cells/mL. The culture can be performed at, for example, 10 to 40 °C for, for example, 5 to 168 hours.

[0041] The tobacco extract liquid contains a component serving as a nutrient source for the yeast and a component serving as a raw material of the useful components, and can provide an environment suitable for the growth of the yeast and the production of the useful components. Thus, it is unnecessary to add an additional component to the tobacco extract liquid. However, the method of the present invention does not exclude the addition of an additional component to the tobacco extract liquid.

[0042] In the present disclosure, a mixture of the yeast and the tobacco extract liquid obtained after the yeast is cultured in the tobacco extract liquid is referred to as "a yeast-containing culture liquid". In the yeast-containing culture liquid, the amounts of useful components produced by the yeast are increased as compared with the "mixture of the yeast and the tobacco extract liquid prior to the culture". Also, in the yeast-containing culture liquid, the amounts of substances consumed by the yeast for its own growth and for the production of the useful components are reduced, as compared with the "mixture of the yeast and the tobacco extract liquid prior to the culture".

[Elution Step (S4)]

[0043] In the elution step (S4), the yeast-containing culture liquid obtained in the culture step (S2) is mixed with an elution solvent including an organic solvent, and a useful component included in the fungal cells of the yeast contained in the resultant mixture is eluted from the yeast into the liquid portion of the mixture. Thereby, a useful-component eluate is obtained (see FIG. 1).

[0044] Herein, the mixture that is obtained after the elution step is completed and contains the yeast and the elution solvent is referred to as "a useful-component eluate".

[0045] An elution solvent including an organic solvent can be used as the elution solvent. The elution solvent may be an organic solvent itself or a mixture of an organic solvent and water. The elution solvent is preferably an elution solvent including an organic solvent miscible with water, and more preferably an elution solvent including an alcohol miscible with water. That is, a preferred elution solvent is an alcohol miscible with water or its water-containing alcohol (i.e., said alcohol with water contained therein).

[0046] The organic solvent included in the elution solvent is preferably an organic solvent having an SP value of 10 to 14.5, and more preferably an alcohol having an SP value of 10 to 14.5. Examples of the organic solvent

included in the elution solvent include ethanol, isopropanol, methanol, and butanol. The organic solvent included in the elution solvent is more preferably ethanol or isopropanol, and most preferably ethanol. That is, the most preferred elution solvent is ethanol or water-containing ethanol.

[0047] The SP value refers to a value of a Hildebrand solubility parameter. The higher the SP value of the solvent, the higher the miscibility with water. The SP value is known for various solvents. For example, the SP value of ethanol is 12.7, the SP value of isopropanol is 11.5, the SP value of methanol is 14.5, and the SP value of butanol is 11.4.

[0048] One kind of organic solvent, or a mixture of two or more kinds of organic solvents, may be used as the organic solvent included in the elution solvent.

[0049] The elution solvent can be added to the yeast-containing culture liquid in such an amount that the concentration of the organic solvent in the mixed solution of the yeast-containing culture liquid and the elution solvent becomes, for example, 50% by volume or more, preferably 50 to 95% by volume, and more preferably 60 to 95% by volume. The concentration of the organic solvent can be appropriately adjusted in consideration of the efficiency of eluting the useful component. Herein, the "mixed solution of the yeast-containing culture liquid and the elution solvent" refers to a mixed solution of the liquid portion of the yeast-containing culture liquid (i.e., the tobacco extract liquid after the culture) and the elution solvent, and does not contain yeast.

[0050] The elution solvent can be added in an amount of, for example, 100 to 900% by volume with respect to the yeast-containing culture liquid. The amount of the elution solvent added can be appropriately adjusted in consideration of the efficiency of eluting the useful component.

[0051] For example, if ethanol or water-containing ethanol is used as the elution solvent to elute fatty acids as useful components, the ethanol can be added to the yeast-containing culture liquid in such an amount that the concentration of the ethanol in the mixed solution of the yeast-containing culture liquid and the elution solvent becomes, for example, 50% by volume or more, preferably 50 to 90% by volume, more preferably 60 to 90% by volume, still more preferably 60 to 80% by volume, and most preferably 70% by volume.

[0052] For example, if ethanol or water-containing ethanol is used as the elution solvent to elute carotenoids as useful components, the ethanol can be added to the yeast-containing culture liquid in such an amount that the concentration of the ethanol in the mixed solution of the yeast-containing culture liquid and the elution solvent becomes, for example, 50% by volume or more, preferably 50 to 90% by volume, more preferably 60 to 90% by volume, still more preferably 70 to 90% by volume, yet more preferably 80 to 90% by volume, and most preferably 90% by volume.

[0053] In the elution step (S4), after the yeast-containing culture liquid is mixed with the elution solvent, a useful component included in the fungal cells of the yeast contained in the resultant mixture is eluted from the yeast into the liquid portion of the mixture. The elution can be performed by stirring the mixture of the yeast-containing culture liquid and the elution solvent for a predetermined period of time, while heating the mixture if necessary.

[0054] The elution is preferably performed while stirring the mixture from the standpoint of the efficiency of eluting the useful components; however, it may be performed while allowing the mixture to stand. In the case of stirring the mixture, the stirring rate can be, for example, 60 to 300 rpm.

[0055] If heating promotes the elution of the useful components, the elution may be performed while heating; and if heating does not show the effect of promoting the elution by heating, the elution may be performed at room temperature (for example, 15 to 25 °C) without heating.

[0056] The elution temperature and the elution time can be appropriately adjusted in consideration of the efficiency of eluting the useful component.

[0057] For example, if ethanol or water-containing ethanol is used as the elution solvent to elute fatty acids as useful components, the elution can be performed by stirring the mixture at room temperature (for example, 15 to 25 °C) for 5 to 60 minutes.

[0058] For example, if ethanol or water-containing ethanol is used as the elution solvent to elute carotenoids as useful components, the elution can be performed by stirring the mixture for 15 to 60 minutes while heating the mixture to 80 to 95 °C.

[Yeast-removal step (S5)]

[0059] In the yeast-removal step (S5), the yeast is removed from the useful-component eluate obtained in the elution step (S4), whereby a yeast-free useful-component eluate can be obtained (see FIG. 1). As described above, the yeast-removal step (S5) may or may not be performed.

[0060] The yeast can be removed by a general method such as pressure filtration, filtration under reduced pressure, gravity filtration, centrifugation, or decantation. Removing the yeast from the useful-component eluate can eliminate the possibility of the composition of the tobacco flavor liquid changing due to the metabolism of the yeast, which is preferable in terms of the quality stability of the tobacco flavor liquid.

[0061] If the yeast-removal step (S5) is performed, the "yeast-free useful-component eluate" obtained corresponds to the tobacco flavor liquid. On the other hand, if the yeast-removal step (S5) is not performed, the "useful-component eluate" corresponds to the tobacco flavor liquid, and the yeast remains therein.

[0062] The above-described method may further include removing the organic solvent from the tobacco flavor liquid after obtaining the tobacco flavor liquid. The removal of the organic solvent can be performed by a

general method such as concentration under reduced pressure, concentration under normal pressure, or spray drying.

[0063] The method of the first embodiment excels in the aspect of easy removal of the yeast in the yeast-removal step (S5), as the yeast is easily precipitated in the elution step (S4). This excellence effect is demonstrated in Example 1 described later. Hereinafter, this excellence effect will be concretely described. When an elution solvent including an organic solvent is added to the yeast-containing culture liquid in the elution step (S4), the polarity of the liquid portion (i.e., the tobacco extract liquid after the culture) of the yeast-containing culture liquid changes, causing the water-soluble protein dissolved in the tobacco extract liquid to become insoluble. The presence of the insolubilized protein promotes aggregation of the microorganisms included in the tobacco extract liquid (i.e., the added yeast as well as the yeast, mold, etc., that are originally included in the tobacco extract liquid). This aggregation phenomenon occurs even under the condition where only the fungal cells of the microorganisms exist, but occurs more significantly under the condition where the microorganisms coexist with a protein. Thus, in the method of the first embodiment, a composite of the aggregated microorganisms and the protein can be easily precipitated by an increase in the mass and can form a pellet. In this manner, the method of the first embodiment enables the added yeast to easily form a pellet with the aid of the water-soluble protein contained in the tobacco extract liquid and can easily remove the yeast from the tobacco extract liquid by removing the pellet.

<1-2. Second Embodiment>

[0064] In the first embodiment, a useful component is eluted by culturing yeast in a tobacco extract liquid and mixing the obtained yeast-containing culture liquid with an elution solvent. However, a useful component can also be eluted by separating the yeast from the yeast-containing culture liquid and mixing the separated yeast with an elution solvent.

[0065] Specifically, according to the second embodiment, a method for producing a tobacco flavor liquid includes:

(S1) extracting, from a tobacco material, a water-soluble component contained in the tobacco material by using an aqueous solvent, thereby obtaining a tobacco extract liquid;
(S2) culturing yeast in the tobacco extract liquid, thereby obtaining a yeast-containing culture liquid;
(S3) separating the yeast-containing culture liquid into the yeast and the tobacco extract liquid;
(S4) mixing the separated yeast with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in fungal cells of the yeast, thereby obtaining a useful-component eluate;
(S5) if necessary, removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate; and
(S6) mixing the useful-component eluate or the yeast-free useful-component eluate with the separated tobacco extract liquid, thereby obtaining a useful component-containing liquid.

[0066] The method according to the second embodiment is shown in FIG. 2. Hereinafter, the method according to the second embodiment will be described in the order of steps (S1) to (S6) with reference to FIG. 2. The description below omits description of the same elements as those of the method according to the first embodiment and provides description of the elements differing from those of the method according to the first embodiment.

[0067] As described above, the yeast-removal step (S5) may or may not be performed. Specifically, if the yeast-removal step (S5) is performed, the "yeast-free useful-component eluate" is mixed with the "separated tobacco extract liquid" in the mixing step (S6), whereby a "yeast-free useful component-containing liquid" is obtained; thus, this "yeast-free useful component-containing liquid" can be used as the tobacco flavor liquid. On the other hand, if the yeast-removal step (S5) is not performed, the "useful-component eluate" is mixed with the "separated tobacco extract liquid" in the mixing step (S6), whereby a "useful component-containing liquid" is obtained with the yeast remaining therein; thus, this "useful component-containing liquid" can be used as the tobacco flavor liquid.

[Extraction Step (S1)]

[0068] In the extraction step (S1), a water-soluble component contained in a tobacco material is extracted from the tobacco material by using an aqueous solvent, whereby a tobacco extract liquid is obtained. In the extraction step (S1), a tobacco residue is also obtained at the same time as the tobacco extract liquid is obtained (see FIG. 2). The extraction step (S1) can be performed in the same manner as the extraction step (S1) described for the method of the first embodiment.

[Culture Step (S2)]

[0069] In the culture step (S2), yeast is cultured in the tobacco extract liquid obtained in the extraction step (S1), whereby a yeast-containing culture liquid is obtained (see FIG. 2). The culture step (S2) can be performed in the same manner as the culture step (S2) described for the method of the first embodiment.

[Separation Step (S3)]

[0070] After the culture step (S2) is completed, the

yeast-containing culture liquid is separated into the yeast and the tobacco extract liquid (see FIG. 2). The separation step (S3) can be performed by a general method such as pressure filtration, filtration under reduced pressure, gravity filtration, centrifugation, or decantation.

[Elution Step (S4)]

[0071] In the elution step (S4), the yeast separated in the separation step (S3) is mixed with an elution solvent including an organic solvent, and a useful component included in the fungal cells of the yeast contained in the resultant mixture is eluted from the yeast into a liquid portion of the mixture, whereby a useful-component eluate is obtained (see FIG. 2).

[0072] The same elution solvent as that described for the method of the first embodiment can be used as the elution solvent. Since the first embodiment does not include the separation step (S3), the elution solvent is mixed with the yeast-containing culture liquid (i.e., a liquid). On the other hand, in the second embodiment, the elution solvent is mixed with the yeast (i.e., a solid). Thus, in the second embodiment, the concentration of the organic solvent in the elution solvent is maintained without the organic solvent being diluted, even after the elution solvent is mixed with the yeast.

[0073] In the second embodiment, the concentration of the organic solvent in the elution solvent is, for example, 50% by volume or more, preferably 50 to 100% by volume, and more preferably 60 to 100% by volume. Likewise, the concentration of the organic solvent in the mixture of the yeast and the elution solvent is, for example, 50% by volume or more, preferably 50 to 100% by volume, and more preferably 60 to 100% by volume. The concentration of the organic solvent can be appropriately adjusted in consideration of the efficiency of eluting the useful component.

[0074] The elution solvent can be added to the separated yeast in an amount of, for example, 35 to 100% by volume with respect to the amount of the yeast-containing culture liquid from which the yeast is derived. The amount of the elution solvent added can be appropriately adjusted in consideration of the efficiency of eluting the useful component.

[0075] For example, if ethanol or water-containing ethanol is used as the elution solvent to elute fatty acids as useful components, the concentration of the ethanol in the elution solvent (i.e., the concentration of the ethanol in the mixture of the yeast and the elution solvent) is, for example, 50% by volume or more, preferably 50 to 100% by volume, more preferably 60 to 90% by volume, still more preferably 60 to 80% by volume, and most preferably 70% by volume.

[0076] For example, if ethanol or water-containing ethanol is used as the elution solvent to elute carotenoids as useful components, the concentration of the ethanol in the elution solvent (i.e. the concentration of the ethanol in the mixture of the yeast and the elution solvent) is, for example, 50% by volume or more, preferably 50 to 100% by volume, more preferably 60 to 100% by volume, still more preferably 70 to 100% by volume, yet more preferably 80 to 100% by volume, and most preferably 90 to 100% by volume.

[0077] As for the other elution conditions, the same conditions as those for elution described for the method of the first embodiment can be employed.

[Yeast-removal step (S5)]

[0078] In the yeast-removal step (S5), the yeast is removed from the useful-component eluate obtained in the elution step (S4), whereby a yeast-free useful-component eluate can be obtained (see FIG. 2). As described above, the yeast-removal step (S5) may or may not be performed. The yeast-removal step (S5) can be performed in the same manner as the yeast-removal step (S5) described for the method of the first embodiment.

[Mixing Step (S6)]

[0079] If the yeast-removal step (S5) is performed, the yeast-free useful-component eluate obtained in the yeast-removal step (S5) is mixed with the tobacco extract liquid separated in the separation step (S3), whereby a yeast-free useful component-containing liquid is obtained (see FIG. 2). The "yeast-free useful component-containing liquid" obtained corresponds to the tobacco flavor liquid.

[0080] On the other hand, if the yeast-removal step (S5) is not performed, the useful-component eluate obtained in the elution step (S4) is mixed with the tobacco extract liquid separated in the separation step (S3), whereby a useful component-containing liquid is obtained. The "useful component-containing liquid" obtained corresponds to the tobacco flavor liquid, and the yeast remains therein.

[0081] The above-described method, like the method of the first embodiment, may further include removing the organic solvent from the tobacco flavor liquid after obtaining the tobacco flavor liquid.

<1-3. Effects>

[0082] Culturing yeast in a tobacco extract liquid and eluting useful components such as flavor components from the fungal cells of the yeast into the tobacco extract liquid according to the present invention can produce a tobacco extract liquid having increased amounts of useful components such as flavor components (referred to as a "tobacco flavor liquid" in the present disclosure).

[0083] Since the method of the present invention includes the step of eluting a useful component included in the fungal cells of the yeast into the tobacco extract liquid, the finally obtained tobacco flavor liquid can contain a large amount of useful component. In addition, the method of the present invention is a simple method as it

enables elution of a useful component included in the fungal cells of the yeast without breaking that fungal cells.

<2. Tobacco flavor liquid>

[0084] According to another aspect, there is provided a tobacco flavor liquid obtainable by the "method for producing a tobacco flavor liquid" described above.

[0085] As described above, the tobacco flavor liquid may be any of the following:

(i) the "useful-component eluate" obtained in the elution step (S4) of the method of the first embodiment;
(ii) the "yeast-free useful-component eluate" obtained in the yeast-removal step (S5) of the method of the first embodiment;
(iii) the "useful component-containing liquid" obtained when the mixing step (S6) is performed without performing the yeast-removal step (S5) in the method of the second embodiment; and
(iv) the "yeast-free useful component-containing liquid" obtained when the mixing step (S6) is performed after the yeast-removal step (S5) is performed in the method of the second embodiment.

[0086] Thus, the tobacco flavor liquid produced by the above-described "method for producing a tobacco flavor liquid" includes the above four types of products as specific examples.

[0087] Since the tobacco flavor liquid is produced by the above-described "method for producing a tobacco flavor liquid", it can contain large amounts of useful components, as described above. Thus, if this tobacco flavor liquid is incorporated into a flavor inhaler, the effects of the useful components can be remarkably exhibited in the flavor inhaler. For example, if the useful components are flavor-contributing components, the tobacco flavor liquid can contain large amounts of flavor-contributing components and thus provide a user with an enhanced flavor when incorporated into a tobacco product such as a flavor inhaler.

[0088] The tobacco flavor liquid produced by the above-described "method for producing a tobacco flavor liquid" can be incorporated into a tobacco product such as a flavor inhaler according to a known technique. Examples of the use of the tobacco flavor liquid will be described below.

[0089] For example, the tobacco flavor liquid can be used by adding the tobacco flavor liquid to a tobacco material (e.g., stemmed leaves or leaf tobacco) and drying the resultant mixture.

[0090] Alternatively, the tobacco flavor liquid can be added to a tobacco residue obtained in the extraction step (S1) described above, and a tobacco-molded body such as a sheet tobacco or tobacco granules can be prepared from the resultant mixture and used as a tobacco flavor source of a tobacco product.

[0091] Alternatively, the tobacco flavor liquid can be used by adding the tobacco flavor liquid to a tobacco residue obtained in the extraction step (S1) described above, drying and pulverizing the resultant mixture to prepare a tobacco powder, and adding the tobacco powder to a tobacco material (e.g., stemmed leaves or leaf tobacco).

[0092] Alternatively, the tobacco flavor liquid can be used by adding the tobacco flavor liquid to a tobacco residue obtained in the extraction step (S1) described above, drying and pulverizing the resultant mixture to prepare a tobacco powder, suspending the tobacco powder in water to prepare a tobacco slurry, and adding the tobacco slurry to a tobacco material (e.g., stemmed leaves or leaf tobacco).

[0093] Alternatively, the tobacco flavor liquid can be encapsulated according to a known technique, and the resultant flavor capsules can be incorporated into a filter portion of a tobacco product.

<3. Tobacco Additive>

[0094] As described above, the tobacco flavor liquid may be used in combination with the tobacco residue obtained in the extraction step (S1) described above. Thus, according to another aspect, there is provided a tobacco additive containing:

the tobacco flavor liquid obtainable by the above-described "method for producing a tobacco flavor liquid"; and
a tobacco residue obtained when the tobacco extract liquid is obtained in the above-described "method for producing a tobacco flavor liquid".

[0095] Specific examples of the tobacco additive will be described below.

[0096] For example, the tobacco additive may be a product obtained by drying a mixture of the tobacco residue obtained in the extraction step (S1) and the tobacco flavor liquid. This product can be used as a tobacco flavor source of a tobacco product.

[0097] Alternatively, the tobacco additive may be a tobacco-molded body obtained by molding a mixture of the tobacco residue obtained in the extraction step (S 1) and the tobacco flavor liquid into a specific shape such as a sheet shape or a granular shape. The tobacco-molded body can be used as a tobacco flavor source of a tobacco product.

[0098] Alternatively, the tobacco additive may be a tobacco powder obtained by drying a mixture of the tobacco residue obtained in the extraction step (S1) and the tobacco flavor liquid and then pulverizing the dried mixture into powder form. By adding the tobacco powder to a tobacco material (e.g., stemmed leaves or leaf tobacco), the flavor of the tobacco material can be enhanced.

[0099] Alternatively, the tobacco additive may be a tobacco slurry obtained by drying a mixture of the tobacco residue obtained in the extraction step (S1) and the to-

bacco flavor liquid, pulverizing the dried mixture into powder form, and suspending the resultant powder in water. By adding the tobacco slurry to a tobacco material (e.g., stemmed leaves or leaf tobacco), the flavor of the tobacco material can be enhanced.

[0100] The tobacco additive may contain additives such as a binder, a pH adjuster, a preservative, and an antioxidant, as necessary.

<4. Flavor Inhaler>

[0101] The above-described "tobacco flavor liquid" or the above-described "tobacco additive" can be incorporated into any tobacco product. As a typical example, the above-described "tobacco flavor liquid" or the above-described "tobacco additive" can be incorporated into a flavor inhaler such as a combustion-type flavor inhaler or a heating-type flavor inhaler. Specifically, according to another aspect, there is provided a flavor inhaler which includes the above-described "tobacco flavor liquid" or the above-described "tobacco additive".

[0102] The above-described "tobacco flavor liquid" or the above-described "tobacco additive" can be incorporated into any position of the tobacco product, provided that a user can enjoy an enhanced flavor when using the tobacco product.

[0103] Examples of the combustion-type flavor inhaler include a cigarette, a pipe, a kiseru (i.e., traditional Japanese pipe for fine cut tobacco), a cigar, and a cigarillo. An example of the combustion-type flavor inhaler, that is, a cigarette having a typical structure, is shown in FIG. 3.

[0104] A combustion-type flavor inhaler 1 shown in FIG. 3 includes:

- a tobacco rod 2 that includes a tobacco filler 2a and cigarette paper 2b wrapped around the tobacco filler 2a;
- a filter 3 that includes a filter material 3a and a plug wrapper 3b wrapped around the filter material 3a; and
- tipping paper 4 wrapped over the tobacco rod 2 and the filter 3 so as to connect the tobacco rod 2 and the filter 3.

[0105] The tobacco rod 2 includes a tobacco filler 2a such as cut tobacco or a tobacco-molded body. The tobacco rod can have, for example, a diameter of 5 to 10 mm and a length of 40 to 80 mm, as in the case of an ordinary cigarette.

[0106] The filter 3 is a filter made of a single filter material 3a, that is, a so-called plain filter. The filter material 3a can be made of a filter material such as acetate tow, as in the case of an ordinary cigarette. The filter 3 has almost the same diameter as that of the tobacco rod 2, and may have a length of, for example, 15 to 40 mm, as in the case of an ordinary cigarette. The plug wrapper 3b may have a thickness of 10 to 100 μm. The plug wrapper 3b may or may not be air-permeable; however, it is common to use air-permeable paper.

[0107] The tipping paper 4 is adhered with an adhesive so as to cover the entire plug wrapper 3b and a part of the cigarette paper 2b. The tipping paper 4 may have, for example, a length (width) of 20 to 50 mm in the axial direction of the tobacco rod and a thickness of 10 to 100 μm. As in the case of an ordinary cigarette, the tipping paper 4 may have a number of small openings for ventilation (ventilation holes) provided in a single line, in multiple lines, or irregularly along the circumferential direction of the cigarette.

[0108] In the case of the combustion-type flavor inhaler 1 shown in FIG. 3, the above-described "tobacco flavor liquid" and the above-described "tobacco additive" can be incorporated into, for example, the tobacco filler 2a or the filter material 3 a.

[0109] Next, an example of the heating-type flavor inhaler will be described. Examples of the heating-type flavor inhaler include:

- a carbon heat source-type inhaler that heats a tobacco filler with the combustion heat of a carbon heat source (see, for example, WO 2006/073065);
- an electric heating-type inhaler having a tobacco stick containing a tobacco filler and a heating device for electrically heating the tobacco stick (see, for example, WO 2010/110226); and
- a liquid atomizing-type inhaler in which a liquid aerosol source is heated by a heater to generate aerosol and a flavor derived from a tobacco filler is inhaled together with the aerosol (see, for example, WO 2015/046385).

[0110] Hereinafter, an example of the heating-type flavor inhaler will be described with reference to FIGS. 4 to 6. FIG. 4 is a perspective view showing an example of a heating-type flavor inhaler. FIG. 5 is a diagram showing an internal structure of a tobacco stick. FIG. 6 is a diagram showing an internal structure of an aerosol-generation device.

[0111] As shown in FIG. 4, a heating-type flavor inhaler 100 includes:

- a tobacco stick 110 including a tobacco filler and an aerosol source; and
- an aerosol-generation device 120 to which the tobacco stick 110 is detachably attached, and which heats the tobacco stick 110 to generate aerosol from the aerosol source and release a flavor component from the tobacco filler by an action of the aerosol.

[0112] The tobacco stick 110 is a replaceable cartridge and has a columnar shape extending along the longitudinal direction. The tobacco stick 110 is configured to generate aerosol and a flavor component by being heated while being inserted into the aerosol-generation device 120.

[0113] As shown in FIG. 5, the tobacco stick 110 includes a base portion 11A including a filler 111 and first cigarette paper 112 wrapped around the filler 111, and a mouthpiece portion 11B forming an end opposite to the base portion 11A. The base portion 11A and the mouthpiece portion 11B are connected by second cigarette paper 113.

[0114] The mouthpiece portion 11B includes a paper tube portion 114, a filter portion 115, and a hollow segment portion 116 disposed between the paper tube portion 114 and the filter portion 115. The paper tube portion 114 is a paper tube formed by winding paper in a cylindrical shape, and has a hollow inside. The filter portion 115 includes a filter material such as acetate tow. The hollow segment portion 116 includes a filling layer having one or more hollow channels. The filter material of the filter portion 115 and the filling layer of the hollow segment portion 116 are connected by being covered with a plug wrapper 117. The filling layer is formed of fibers and has a high filling density of fibers; therefore, during inhalation, air or aerosol flows only through the hollow channel and hardly flows through the filling layer. In the tobacco stick 110, when the decrease of aerosol components through filtration in the filter portion 115 is desired to be diminished, it is effective to shorten the length of the filter portion 115 and replace it with the hollow segment portion 116 in order to increase a delivery amount of aerosol.

[0115] Although the mouthpiece portion 11B is composed of three segments, the mouthpiece portion 11B may be composed of one or two segments, or may be composed of four or more segments. For example, the hollow segment portion 116 may be omitted, and the paper tube portion 114 and the filter portion 115 may be disposed adjacent to each other to form the mouthpiece portion 11B.

[0116] The longitudinal length of the tobacco stick 110 is preferably 40 to 90 mm, more preferably 50 to 75 mm, and still more preferably 50 to 60 mm. The circumference of the tobacco stick 110 is preferably 15 to 25 mm, more preferably 17 to 24 mm, and still more preferably 20 to 23 mm. In addition, in the longitudinal direction of the tobacco stick 110, the base portion 11A may have a length of 20 mm, the paper tube portion 114 may have a length of 20 mm, the hollow segment portion 116 may have a length of 8 mm, and the filter portion 115 may have a length of 7 mm, and the lengths of these individual segments can be changed as appropriate according to production suitability, required quality, and the like.

[0117] The filler 111 includes a tobacco filler and an aerosol source. The aerosol source is heated at a predetermined temperature to generate aerosol. The aerosol source may be, for example, glycerin, propylene glycol, triacetin, 1,3-butanediol, and a mixture thereof. The content of the aerosol source in the filler 111 is not particularly limited, and from the viewpoint of generating a sufficient amount of aerosol and providing a good smoking flavor, the content is usually 5% by mass or more, preferably 10% by mass or more, and usually 50% by mass or less, preferably 20% by mass or less.

[0118] The tobacco filler is, for example, in the form of cut tobacco or in the form of a tobacco-molded body, as described above. If the tobacco filler is in the form of cut tobacco, it may be in the form of cut tobacco obtained by cutting leaf tobacco (i.e., aged tobacco leaves) into widths of, for example, 0.8 to 1.2 mm. Alternatively, if the tobacco filler is in the form of sheet tobacco, it may be in the form of elongated sheet tobacco obtained by cutting sheet tobacco into widths of, for example, 0.8 to 1.2 mm, or may be in the form of corrugated sheet tobacco obtained by gathering sheet tobacco without cutting it.

[0119] If the base portion 11A has a circumference of 22 mm and a length of 20 mm, the content of the filler 111 in the tobacco stick 110 is, for example, 200 to 400 mg, and preferably 250 to 320 mg. The moisture content of the filler 111 is, for example, 8 to 18% by mass, and preferably 10 to 16% by mass. Such a moisture content suppresses generation of a stain on the wrapping paper and improves roll-up machinability during manufacture of the base portion 11A.

[0120] For the first cigarette paper 112, the second cigarette paper 113, and the plug wrapper 117, the same cigarette paper, tipping paper, and plug wrapper as those used in a cigarette can be used.

[0121] In the case of the tobacco stick 110 shown in FIG. 5, the above-described "tobacco flavor liquid" and the above-described "tobacco additive" can be incorporated into, for example, the filler 111 or the filter material of the filter portion 115.

[0122] As shown in FIG. 6, the aerosol-generation device 120 includes an insertion hole 130 into which the tobacco stick 110 can be inserted. That is, the aerosol-generation device 120 includes an inner tubular member 132 constituting the insertion hole 130. The inner tubular member 132 may be formed of a heat conductive member such as aluminum or stainless steel (SUS).

[0123] Further, the aerosol-generation device 120 may include a lid portion 140 that closes the insertion hole 130. The lid portion 140 is configured to be slidable between a state where the insertion hole 130 is closed and a state where the insertion hole 130 is exposed (see FIG. 4).

[0124] The aerosol-generation device 120 may include an air flow path 160 communicating with the insertion hole 130. One end of the air flow path 160 is connected to the insertion hole 130, while the other end of the air flow path 160 communicates with the outside (outside air) of the aerosol-generation device 120 at a portion different from the insertion hole 130.

[0125] The aerosol-generation device 120 may include a lid portion 170 that covers an end portion of the air flow path 160 on the side communicating with the outside air. The lid portion 170 can cover the end portion of the air flow path 160 on the side communicating with the outside air, or can expose the air flow path 160.

[0126] The lid portion 170 does not air-tightly close the air flow path 160 even in a state of covering the air flow

path 160. That is, even in a state where the lid portion 170 covers the air flow path 160, the outside air can flow into the air flow path 160 via the vicinity of the lid portion 170.

[0127] In a state where the tobacco stick 110 is inserted into the aerosol-generation device 120, the user holds, in the mouth, one end portion of the tobacco stick 110, specifically, the mouthpiece portion 11B shown in FIG. 5, and performs an inhalation action. The outside air flows into the air flow path 160 through the user's inhalation action. The air flowing into the air flow path 160 passes through the tobacco stick 110 in the insertion hole 130 and is guided into an oral cavity of the user.

[0128] The aerosol-generation device 120 may include a temperature sensor in the air flow path 160 or on an outer surface of a wall portion constituting the air flow path 160. The temperature sensor may be, for example, a thermistor, a thermocouple, or the like. When the user inhales with the mouthpiece portion 11B of the tobacco stick 110, the internal temperature of the air flow path 160 or the temperature of the wall portion constituting the air flow path 160 decreases because of the influence of the air flowing through the air flow path 160 from the lid portion 170 side toward a heater 30 side. The temperature sensor can detect the user's inhalation action by measuring this temperature decrease.

[0129] The aerosol-generation device 120 includes a battery 10, a control unit 20, and a heater 30. The battery 10 stores electric power for use in the aerosol-generation device 120. The battery 10 may be a chargeable and dischargeable secondary battery. The battery 10 may be, for example, a lithium ion battery.

[0130] The heater 30 may be provided around the inner tubular member 132. The space accommodating the heater 30 and the space accommodating the battery 10 may be separated from each other by a partition wall 180. This can prevent the air heated by the heater 30 from flowing into the space accommodating the battery 10. Therefore, an increase in the temperature of the battery 10 can be suppressed.

[0131] The heater 30 preferably has a tubular shape capable of heating the outer periphery of the columnar tobacco stick 110. The heater 30 may be, for example, a film heater. The film heater may include a pair of film-like substrates and a resistance heating element sandwiched between the pair of substrates. The film-like substrate is preferably made of a material excellent in heat resistance and electrical insulating properties, and is typically made of polyimide. The resistance heating element is preferably made of one or two or more metal materials such as copper, nickel alloy, chromium alloy, stainless steel, and platinum rhodium, and may be formed of, for example, a base material made of stainless steel. Further, in order to connect the resistance heating element to a power source by a flexible printed circuit (FPC), copper plating may be applied to a connection portion and a lead portion thereof.

[0132] Preferably, a heat-shrinkable tube may be provided outside the heater 30. The heat-shrinkable tube is a tube that shrinks in a radial direction through heat, and is made of, for example, a thermoplastic elastomer. The heater 30 is pressed against the inner tubular member 132 by the contraction action of the heat-shrinkable tube. This increases the adhesion between the heater 30 and the inner tubular member 132, thereby increasing conduction of the heat from the heater 30 to the tobacco stick 110 via the inner tubular member 132.

[0133] The aerosol-generation device 120 may include a tubular thermal insulator on the outer side of the heater 30 in the radial direction, preferably on the outer side of the heat-shrinkable tube. The thermal insulator may serve to prevent the outer surface of the housing of the aerosol-generation device 120 from reaching an excessively high temperature by blocking the heat of the heater 30. The thermal insulator may be made of an aerogel such as a silica aerogel, a carbon aerogel, or an alumina aerogel. The aerogel as a thermal insulator may typically be a silica aerogel having high thermal insulation performance and relatively low manufacturing costs. However, the thermal insulator may be a fiber-based thermal insulator such as glass wool or rock wool, or a foam-based thermal insulator such as urethane foam or phenolic foam. Alternatively, the thermal insulator may be a vacuum thermal insulator.

[0134] The thermal insulator may be provided between the inner tubular member 132 facing the tobacco stick 110 and an outer tubular member 134 outside the thermal insulator. The outer tubular member 134 may be formed of a heat conductive member such as aluminum or stainless steel (SUS). It is preferable that the thermal insulator be provided in the sealed space.

[0135] The control unit 20 may include a control board, a CPU, a memory, and the like. The aerosol-generation device 120 may include a notification unit for notifying the user of various kinds of information under the control of the control unit 20. The notification unit may be, for example, a light emitting element such as an LED, a vibration element, or a combination thereof.

[0136] Upon detecting an activation request from the user, the control unit 20 starts supplying power from the battery 10 to the heater 30. The activation request from the user is made by, for example, an operation of a push button or a slide switch by the user, or an inhalation action of the user. The activation request from the user may be made by pressing a push button 150. More specifically, the activation request from the user may be made by pressing the push button 150 in a state where the lid portion 140 is opened. Alternatively, the activation request from the user may be made by detection of an inhalation action of the user. The user's inhalation action can be detected by, for example, such a temperature sensor as described above.

<5. Preferred Embodiments>

[0137] Hereinafter, preferred embodiments will be de-

scribed.

[A1] A method for producing a tobacco flavor liquid, the method including:

extracting, from a tobacco material, a water-soluble component contained in the tobacco material by using an aqueous solvent, thereby obtaining a tobacco extract liquid;
culturing yeast in the tobacco extract liquid, thereby obtaining a yeast-containing culture liquid; and
eluting, from the yeast contained in the yeast-containing culture liquid, a useful component included in fungal cells of the yeast by using an elution solvent including an organic solvent, thereby obtaining a tobacco flavor liquid containing the useful component and the post-culturing tobacco extract liquid.

[A2] The method according to [A1], wherein the obtaining of the tobacco flavor liquid is performed by mixing the yeast-containing culture liquid with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate.

[A3] The method according to [A1], wherein the obtaining of the tobacco flavor liquid is performed by:

mixing the yeast-containing culture liquid with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate; and
removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate.

[A4] The method according to [A1], wherein the obtaining of the tobacco flavor liquid is performed by:

separating the yeast-containing culture liquid into the yeast and the tobacco extract liquid;
mixing the separated yeast with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate; and
mixing the useful-component eluate with the separated tobacco extract liquid, thereby obtaining a useful component-containing liquid.

[A5] The method according to [A1], wherein the obtaining of the tobacco flavor liquid is performed by:

separating the yeast-containing culture liquid into the yeast and the tobacco extract liquid;
mixing the separated yeast with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate; and
removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate; and
mixing the yeast-free useful-component eluate with the separated tobacco extract liquid, thereby obtaining a yeast-free useful component-containing liquid.

[A6] The method according to [A2] or [A3], wherein the elution is performed by stirring a mixture of the yeast-containing culture liquid and the elution solvent.

[A7] The method according to [A2] or [A3], wherein the elution is performed by heating a mixture of the yeast-containing culture liquid and the elution solvent.

[A8] The method according to [A2], [A3], [A6] or [A7], wherein the elution solvent is added to the yeast-containing culture liquid in such an amount that the concentration of the organic solvent in a mixed solution of the yeast-containing culture liqiud and the elution solvent becomes 50% by volume or more, preferably 50 to 95% by volume, and more preferably 60 to 95% by volume.

[A9] The method according to [A4] or [A5], wherein the elution is performed by stirring a mixture of the yeast and the elution solvent.

[A10] The method according to [A4] or [A5], wherein the elution is performed by heating a mixture of the yeast and the elution solvent.

[A11] The method according to [A4], [A5], [A9] or [A10], wherein the elution solvent is added to the yeast in such an amount that the concentration of the organic solvent in a mixture of the yeast and the elution solvent becomes 50% by volume or more, preferably 50 to 100% by volume, and more preferably 60 to 100% by volume.

[A12] The method according to any one of [A1] to [A11], wherein the tobacco material is cut tobacco.

[A13] The method according to any one of [A1] to [A12], wherein the aqueous solvent is water or water-containing ethanol, preferably water, more preferably water having a temperature of 20 to 70 °C.

[A14] The method according to any one of [A1] to [A13], wherein the yeast is at least one kind selected from the group consisting of yeast of the genus Rho-

dotorula, yeast of the genus Xanthophyllomyces, yeast of the genus Yarrowia, yeast of the genus Lipomyces, yeast of the genus Saccharomyces, yeast of the genus Cyberlindnera, and yeast of the genus Wickerhamomyces.

[A15] The method according to any one of [A1] to [A14], wherein the elution solvent is an organic solvent or a mixture of an organic solvent and water.

[A16] The method according to any one of [A1] to [A15], wherein the organic solvent is an organic solvent miscible with water, preferably an alcohol miscible with water.

[A17] The method according to any one of [A1] to [A16], wherein the elution solvent is an alcohol miscible with water or its water-containing alcohol (i.e., said alcohol with water contained therein).

[A18] The method according to any one of [A1] to [A17], wherein the organic solvent is an organic solvent having an SP value of 10 to 14.5, preferably an alcohol having an SP value of 10 to 14.5, more preferably ethanol, isopropanol, methanol, or butanol, still more preferably ethanol or isopropanol, and most preferably ethanol.

[A19] The method according to any one of [A1] to [A18], wherein the elution solvent is ethanol or water-containing ethanol.

[A20] The method according to any one of [A1] to [A19], further including removing the organic solvent from the tobacco flavor liquid.

[A21] The method according to any one of [A1] to [A20], wherein the useful component is a flavor-contributing component.

[A22] The method according to any one of [A1] to [A21], wherein the flavor-contributing component is at least one kind selected from the group consisting of carotenoids, fatty acids, neutral fats (i.e., glycerin esters of fatty acids), acetic acid esters, fatty acid esters, organic acids, and higher alcohols (e.g., alcohols having 8 to 22 carbon atoms).

[B1] A tobacco flavor liquid obtainable by the method according to any one of [A1] to [A22].

[B2] The tobacco flavor liquid according to [B1], wherein the tobacco flavor liquid is the useful-component eluate defined in [A2],

[B3] The tobacco flavor liquid according to [B1], wherein the tobacco flavor liquid is the yeast-free useful-component eluate defined in [A3].

[B4] The tobacco flavor liquid according to [B1], wherein the tobacco flavor liquid is the useful component-containing liquid defined in [A4].

[B5] The tobacco flavor liquid according to [B1], wherein the tobacco flavor liquid is the yeast-free useful component-containing liquid defined in [A5],

[C1] A tobacco additive including:

the tobacco flavor liquid obtainable by the method according to any one of [A1] to [A22]; and
a tobacco residue obtained when the tobacco extract liquid is obtained in the method according to any one of [A1] to [A22].

[C2] The tobacco additive according to [C1], which is a product obtained by drying a mixture of:

the tobacco flavor liquid obtainable by the method according to any one of [A1] to [A22]; and
the tobacco residue obtained when the tobacco extract liquid is obtained in the method according to any one of [A1] to [A22].

[C3] The tobacco additive according to [C1], which is a tobacco-molded body obtained by molding a mixture of:

the tobacco flavor liquid obtainable by the method according to any one of [A1] to [A22]; and
the tobacco residue obtained when the tobacco extract liquid is obtained in the method according to any one of [A1] to [A22].

[C4] The tobacco additive according to [C1], which is a tobacco powder obtained by:

drying a mixture of:

the tobacco flavor liquid obtainable by the method according to any one of [A1] to [A22]; and
the tobacco residue obtained when the tobacco extract liquid is obtained in the method according to any one of [A1] to [A22]; and

pulverizing the dried mixture into powder form.

[C5] The tobacco additive according to [C1], which is a tobacco slurry obtained by:

drying a mixture of:

the tobacco flavor liquid obtainable by the method according to any one of [A1] to [A22]; and
the tobacco residue obtained when the tobacco extract liquid is obtained in the method according to any one of [A1] to [A22]; and

pulverizing the dried mixture into powder form; and
suspending the resultant powder in water.

[D1] A flavor inhaler including the tobacco flavor liquid according to any one of [B1] to [B5].

[D2] A flavor inhaler including the tobacco additive according to any one of [C1] to [C5].

[D3] The flavor inhaler according to [D1] or [D2], wherein the flavor inhaler is a combustion-type flavor

inhaler.

[D4] The flavor inhaler according to [D1] or [D2], wherein the flavor inhaler is a heating-type flavor inhaler.

[EXAMPLES]

[Example 1]

[0138] Example 1 confirmed that yeast was precipitated in the elution step (S4) of the method of the first embodiment.

1-1. Method

[0139] According to the method of the first embodiment, the extraction step (S1), the culture step (S2), and the addition of an elution solvent in the elution step (S4) were performed.

Extraction step (S1)

[0140] Flue-cured leaf tobacco was pulverized and used as a "tobacco material". The cut pieces of the flue-cured leaf tobacco (100 g) were pulverized to a size of 100 μm or less by a pulverizer, 600 mL of water having a temperature of 60 °C was added thereto, and a shaking process (200 rpm, 2 hours) was performed. In this manner, a water-soluble component contained in the leaf tobacco was extracted. Subsequently, solid-liquid separation was performed by filtering. Thereby, a tobacco extract liquid and a tobacco residue were obtained.

Culture step (S2)

[0141] Yeast (Yarrowia lipolytica) of the genus Yarrowia was added to 3 mL of the obtained tobacco extract liquid at a concentration of 10^5 cells/mL, and then cultured in the tobacco extract liquid. When culturing the yeast, shaking culture (240 rpm) was performed at 28 °C for 24 hours under aerobic conditions. The "mixture of the yeast and the tobacco extract liquid" obtained after the culture is referred to as a "yeast-containing culture liquid".

Addition of an elution solvent in the elution step (S4)

[0142] After 24 hours had elapsed since the start of the culture, 7 mL of ethanol was added as an elution solvent to 3 mL of the yeast-containing culture liquid. Specifically, ethanol was added in such an amount that the concentration of the ethanol in the mixed solution of the yeast-containing culture liquid and the ethanol would become 70% by volume.

[0143] As a comparative example, ethanol was added as an elution solvent to the yeast in a conventional yeast growth medium. Specifically, the elution solvent was added in such an amount that the concentration of the ethanol in the mixed solution of the yeast growth medium and the ethanol would become 70% by volume.

1-2. Results

[0144] FIG. 7 shows the state of the yeast after the addition of the elution solvent. In FIG. 7, the right photograph shows an example of the present invention, and the left photograph shows a comparative example. In the example of the present invention, the water-soluble protein in the tobacco extract liquid was insolubilized, whereby an aggregated precipitate consisting of the insolubilized protein and the yeast was formed and pelletized. On the other hand, in the comparative example, the yeast did not form an aggregated precipitate.

[0145] These results demonstrate that since the method of the first embodiment allows the yeast cells to form an aggregated precipitate with the insolubilized protein in the elution step (S4), the yeast can be easily removed from the tobacco extract liquid in the subsequent yeast-removal step (S5).

[Example 2]

[0146] In Example 2, a tobacco flavor liquid was prepared using fatty acid-producing yeast according to the method of the first embodiment, and it was confirmed that the amounts of fatty acids in the tobacco flavor liquid were increased.

2-1. Method

Extraction step (S1)

[0147] A tobacco extract liquid and a tobacco residue were obtained according to the same method as that employed in the extraction step (S1) described in Example 1.

Culture step (S2)

[0148] Yeast (Yarrowia lipolytica) of the genus Yarrowia was added to 3 mL of the obtained tobacco extract liquid at a concentration of 10^5 cells/mL, and then cultured in the tobacco extract liquid. When culturing the yeast, shaking culture (240 rpm) was performed at 28 °C for 48 hours under aerobic conditions. The "mixture of the yeast and the tobacco extract liquid" obtained after the culture is referred to as a "yeast-containing culture liquid".

Elution step (S4)

[0149] After 48 hours had elapsed since the start of the culture, any one of ethanol, water-containing ethanol, or water was added as an elution solvent to 3 mL of the yeast-containing culture liquid. Specifically, the elution solvent was added in such an amount that the concentration of the ethanol in the mixed solution of the yeast-

containing culture liquid and the elution solvent would become 0% by volume, 50% by volume, 70% by volume, or 85% by volume.

[0150] The resultant mixture was stirred (120 rpm) at room temperature (25 °C) for 30 minutes. Thereby, the fatty acids produced by the yeast were eluted into the liquid portion of the mixture, and a "useful-component eluate" was consequently obtained.

Yeast-removal step (S5)

[0151] The yeast was removed from the useful-component eluate obtained, whereby a "yeast-free useful-component eluate", that is, a tobacco flavor liquid, was obtained.

Measurement of fatty acids

[0152] The concentrations of the fatty acids in the obtained tobacco flavor liquid were measured using GC-MSD. On the other hand, as Control 1, the concentrations of the fatty acids were likewise measured for the yeast before being cultured in the tobacco extract liquid. As Control 2, instead of performing the elution step (S4), the yeast was crushed with a bead homogenizer to elute the fatty acids contained in the fungal cells of the yeast, and the concentrations of the eluted fatty acids were likewise measured.

2-2. Results

[0153] FIG. 8 shows the results of measuring the concentrations of the fatty acids. The results shown in FIG. 8 demonstrate that culturing fatty acid-producing yeast in a tobacco extract liquid and eluting fatty acid components from the fungal cells of the yeast into the tobacco extract liquid according to the method of the present invention can produce a tobacco extract liquid having increased amounts of fatty acid components (i.e., a "tobacco flavor liquid").

[0154] The analysis of the fatty acids produced by the yeast revealed many of them to be oleic acid and linoleic acid. Also, when the concentration of the ethanol as the elution solvent was 50% by volume or more, the efficiency of eluting the fatty acids was high, and when the concentration of the ethanol as the elution solvent was 70% by volume, the efficiency of eluting the fatty acids was highest.

[0155] In addition, the concentrations of the fatty acids contained in the tobacco flavor liquid obtained according to the method of the present invention were equivalent to the concentrations of the fatty acids measured in Control 2 (bead crushing). This result indicates that the method of the present invention can elute the fatty acids contained in the fungal cells of the yeast in a simple way without crushing that fungal cells.

[Example 3]

[0156] In Example 3, a tobacco flavor liquid was prepared using carotenoid-producing yeast according to the method of the second embodiment, and it was confirmed that the amounts of carotenoids in the tobacco flavor liquid were increased.

3-1. Method

Extraction step (S1)

[0157] A tobacco extract liquid and a tobacco residue were obtained according to the same method as that employed in the extraction step (S1) described in Example 1.

Culture step (S2)

[0158] Yeast (Rhodotorula toruloides) of the genus Rhodotorula was added to 100 mL of the obtained tobacco extract liquid at a concentration of 10^5 cells/mL, and then cultured in the tobacco extract liquid. When culturing the yeast, shaking culture (240 rpm) was performed at 28 °C for 144 hours under aerobic conditions. The "mixture of the yeast and the tobacco extract liquid" obtained after the culture is referred to as a "yeast-containing culture liquid".

Separation step (S3)

[0159] After 144 hours had elapsed since the start of the culture, the yeast-containing culture liquid was separated into the tobacco extract liquid and a precipitate of the yeast through centrifugation.

Elution step (S4)

[0160] Any one of ethanol, water-containing ethanol, or water in an amount of 4 mL was added as an elution solvent to the precipitate of the yeast separated from 8 mL of the yeast-containing culture liquid. The concentration of the added ethanol was 0% by volume, 70% by volume, or 100% by volume.

[0161] The resultant mixture was heated at 90 °C for 1 hour. Thereby, the carotenoids produced by the yeast were eluted into the liquid portion of the mixture, whereby a "useful-component eluate" was obtained.

Yeast-removal step (S5)

[0162] The yeast was removed from the useful-component eluate obtained, whereby a "yeast-free useful-component eluate" was obtained.

Mixing step (S6)

[0163] The yeast-free useful-component eluate obtained was mixed with the tobacco extract liquid sepa-

rated in the separation step (S3), whereby a "yeast-free useful component-containing liquid", that is, a tobacco flavor liquid, was obtained.

Detection of carotenoids

[0164] The carotenoids in the obtained tobacco flavor liquid were detected by thin-film chromatography. Specifically, the tobacco flavor liquid was concentrated, the resultant concentrated liquid was dissolved in a mixture of acetone and hexane (acetone : hexane = 3 : 7 (v/v)), and the sample obtained was analyzed by thin-film chromatography.

[0165] On the other hand, as a control, carotenoids were likewise detected for the yeast before being cultured in the tobacco extract liquid.

3-2. Results

[0166] When the elution step was performed using an elution solvent including ethanol, the tobacco flavor liquid obtained exhibited a yellow or orange color due to the presence of the carotenoids produced by the yeast. On the other hand, when the elution step was performed using an elution solvent that did not include ethanol, the tobacco flavor liquid obtained was colorless.

[0167] In addition, according to the result of the thin-film chromatography, in the case of the control or in the case of performing the elution step using an elution solvent that did not include ethanol, no carotenoid was detected, whereas in the case of performing the elution step using an elution solvent including ethanol, torularhodin, xanthophyll, torulene, and β-carotene were detected as carotenoids.

[0168] When the concentration of the ethanol as the elution solvent was 70% by volume or more, the efficiency of eluting the carotenoids was high. The elution was performed not only at 90 °C for 1 hour but also at 80 °C for 1 hour. In the case where the elution temperature was 80 °C, carotenoids were also detected, as in the case where the elution temperature was 90 °C.

[0169] These results demonstrate that culturing carotenoid-producing yeast in a tobacco extract liquid and eluting carotenoid components from the fungal cells of the yeast into the tobacco extract liquid according to the method of the present invention can produce a tobacco extract liquid having increased amounts of carotenoid components (i.e., a "tobacco flavor liquid").

[0170] These results also demonstrate that the tobacco flavor liquid obtained contains sufficient amounts of carotenoids. Carotenoids are flavor-contributing components and also coloring components. Thus, this tobacco flavor liquid can provide a user with an enhanced flavor when used in a tobacco product such as a flavor inhaler. This tobacco flavor liquid is also useful as a colored tobacco extract liquid.

[Example 4]

[0171] In Example 4, the tobacco flavor liquid prepared in Example 2 was incorporated into a cigarette, and sensory evaluation was carried out.

4-1. Method

Preparation of cigarettes

[0172] The "tobacco flavor liquid" prepared in Example 2 was mixed with the "tobacco residue" obtained in the extraction step (S1) of Example 2. The resultant mixture was dried in a dryer until it became powdery to thereby obtain a tobacco powder. Next, the tobacco powder was suspended in sixfold weight of water to thereby obtain a tobacco slurry.

[0173] The tobacco slurry was added to "cut tobacco ready to be incorporated into a tobacco product (hereinafter referred to as "cut tobacco")". Specifically, the tobacco slurry was added to the cut tobacco such that the tobacco powder in the tobacco slurry was added in an amount of 2% by weight with respect to the cut tobacco. The cut tobacco flavored with the tobacco slurry was conditioned at 22 °C and 60% RH for 2 days.

[0174] The cigarette shown in FIG. 3 (referred to as "cigarette A") was prepared using the flavored cut tobacco as a tobacco filler. As a control, a cigarette (referred to as "cigarette B") was likewise prepared using unflavored cut tobacco.

Evaluation method

[0175] A panel of seven experts smoked the two types of cigarettes.

4-2. Results

[0176] As compared with the cigarette B, the cigarette A was thick in smoke quality and was differentiated. The cigarette A had a neutral impression on the whole, and the smoke quality was thick but mild. This result indicates that the tobacco flavor liquid of the present invention, when incorporated into a flavor inhaler, can contribute to tobacco flavor and provide a user with an enhanced flavor.

Claims

1. A method for producing a tobacco flavor liquid, the method comprising:

   extracting, from a tobacco material, a water-soluble component contained in the tobacco material by using an aqueous solvent, thereby obtaining a tobacco extract liquid;
   culturing yeast in the tobacco extract liquid,

thereby obtaining a yeast-containing culture liquid; and
eluting, from the yeast contained in the yeast-containing culture liquid, a useful component included in fungal cells of the yeast by using an elution solvent including an organic solvent, thereby obtaining a tobacco flavor liquid containing the useful component and the post-culturing tobacco extract liquid.

2. The method according to claim 1, wherein the obtaining of the tobacco flavor liquid is performed by:
mixing the yeast-containing culture liquid with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate.

3. The method according to claim 1, wherein the obtaining of the tobacco flavor liquid is performed by:

mixing the yeast-containing culture liquid with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate; and
removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate.

4. The method according to claim 1, wherein the obtaining of the tobacco flavor liquid is performed by:

separating the yeast-containing culture liquid into the yeast and the tobacco extract liquid;
mixing the separated yeast with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate; and
mixing the useful-component eluate with the separated tobacco extract liquid, thereby obtaining a useful component-containing liquid.

5. The method according to claim 1, wherein the obtaining of the tobacco flavor liquid is performed by:

separating the yeast-containing culture liquid into the yeast and the tobacco extract liquid;
mixing the separated yeast with an elution solvent including an organic solvent, and eluting, from the yeast contained in the resultant mixture into a liquid portion of the mixture, a useful component included in the fungal cells of the yeast, thereby obtaining a useful-component eluate; and
removing the yeast from the useful-component eluate, thereby obtaining a yeast-free useful-component eluate; and
mixing the yeast-free useful-component eluate with the separated tobacco extract liquid, thereby obtaining a yeast-free useful component-containing liquid.

6. The method according to any one of claims 1 to 5, wherein the organic solvent is an organic solvent miscible with water.

7. The method according to any one of claims 1 to 6, further comprising removing the organic solvent from the tobacco flavor liquid.

8. The method according to any one of claims 1 to 7, wherein the useful component is a flavor-contributing component.

9. A tobacco flavor liquid obtainable by the method according to any one of claims 1 to 8.

10. A tobacco additive comprising:

the tobacco flavor liquid obtainable by the method according to any one of claims 1 to 8; and
a tobacco residue obtained when the tobacco extract liquid is obtained in the method according to any one of claims 1 to 8.

11. A flavor inhaler comprising the tobacco flavor liquid according to claim 9 or the tobacco additive according to claim 10.

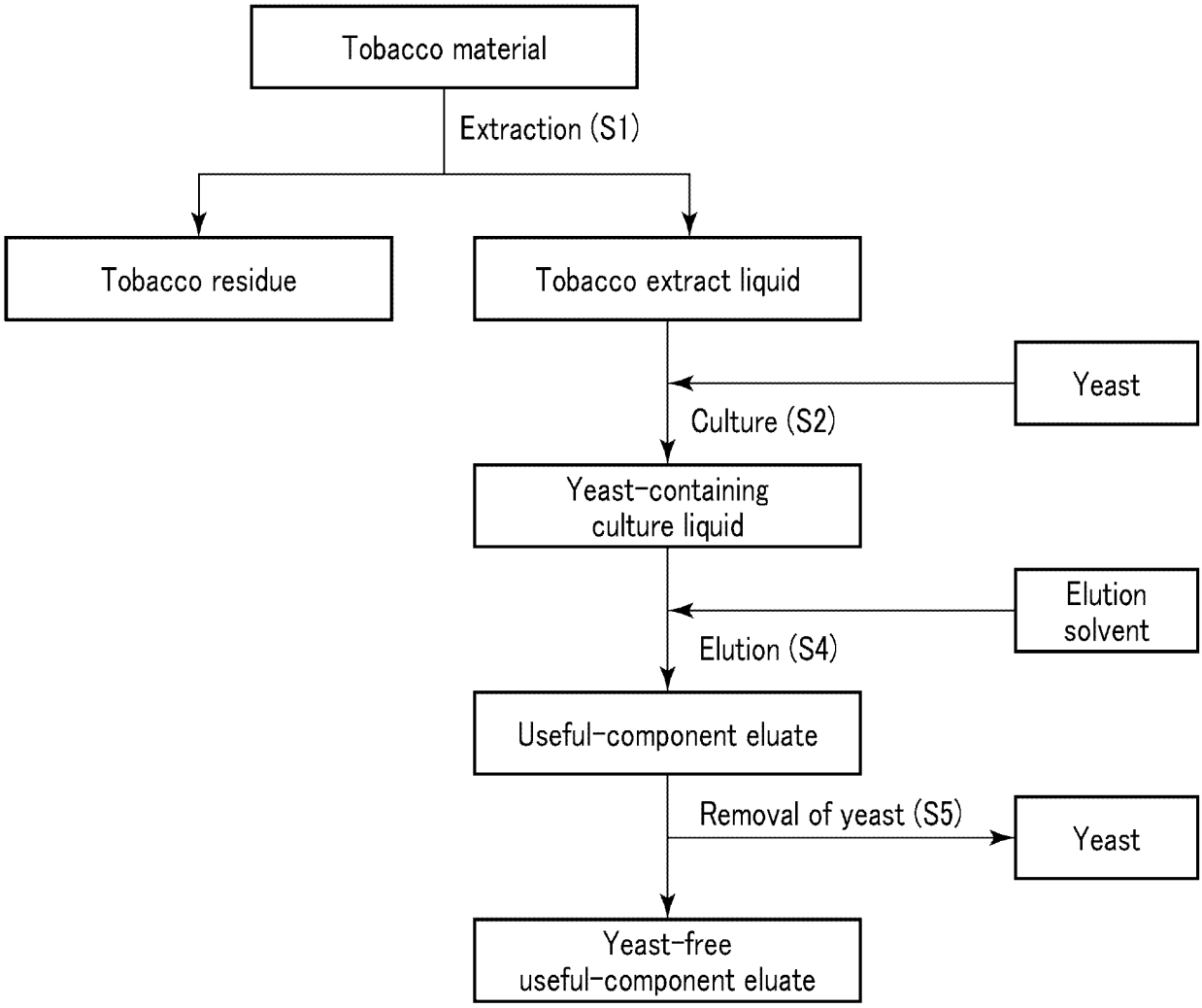
Tobacco material
Extraction (S1)
Tobacco residue
Tobacco extract liquid
Yeast
Culture (S2)
Yeast-containing culture liquid
Elution solvent
Elution (S4)
Useful-component eluate
Removal of yeast (S5)
Yeast
Yeast-free useful-component eluate

FIG. 1

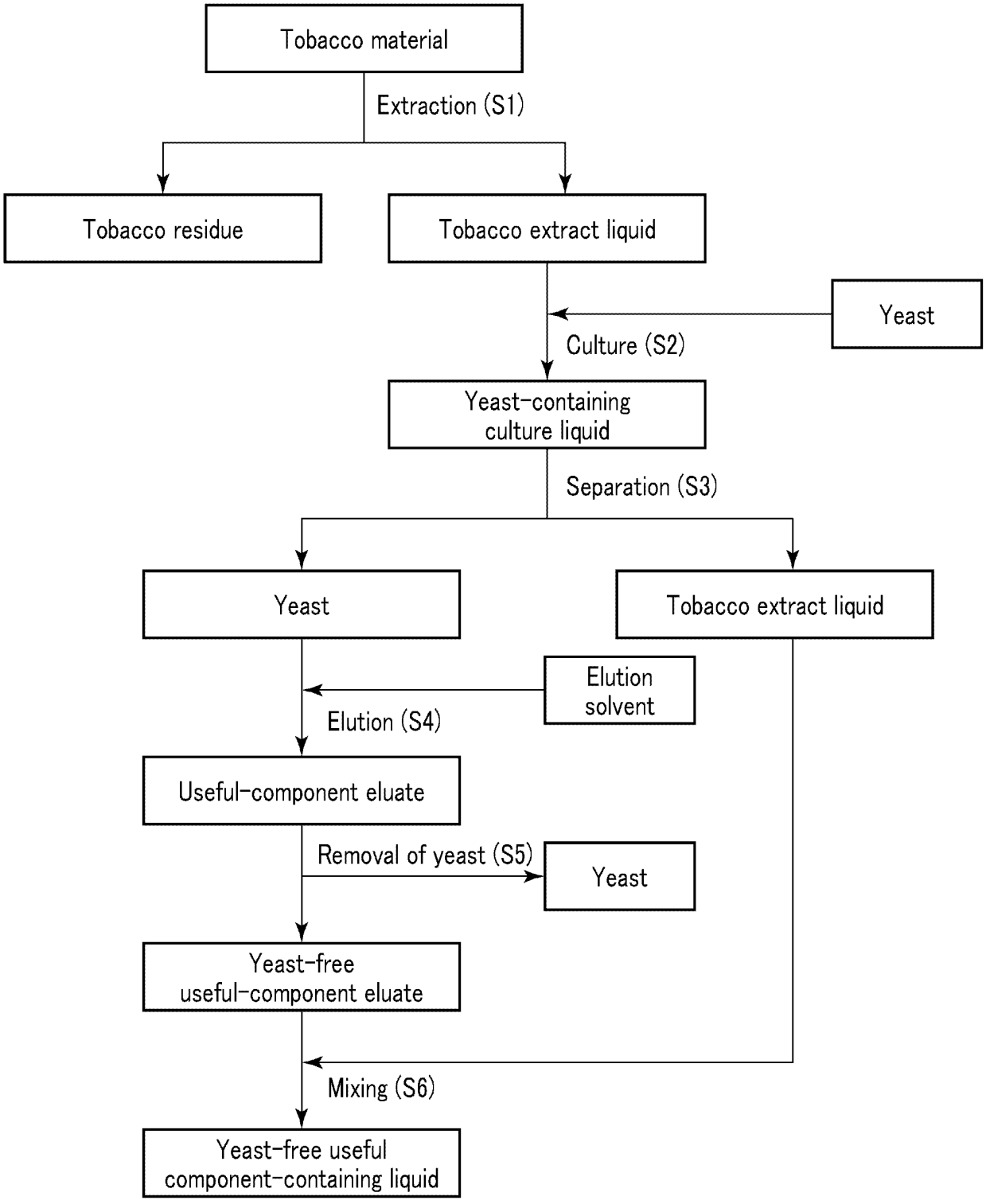
Tobacco material
Extraction (S1)
Tobacco residue
Tobacco extract liquid
Yeast
Culture (S2)
Yeast-containing culture liquid
Separation (S3)
Yeast
Tobacco extract liquid
Elution solvent
Elution (S4)
Useful-component eluate
Removal of yeast (S5)
Yeast
Yeast-free useful-component eluate
Mixing (S6)
Yeast-free useful component-containing liquid

FIG. 2

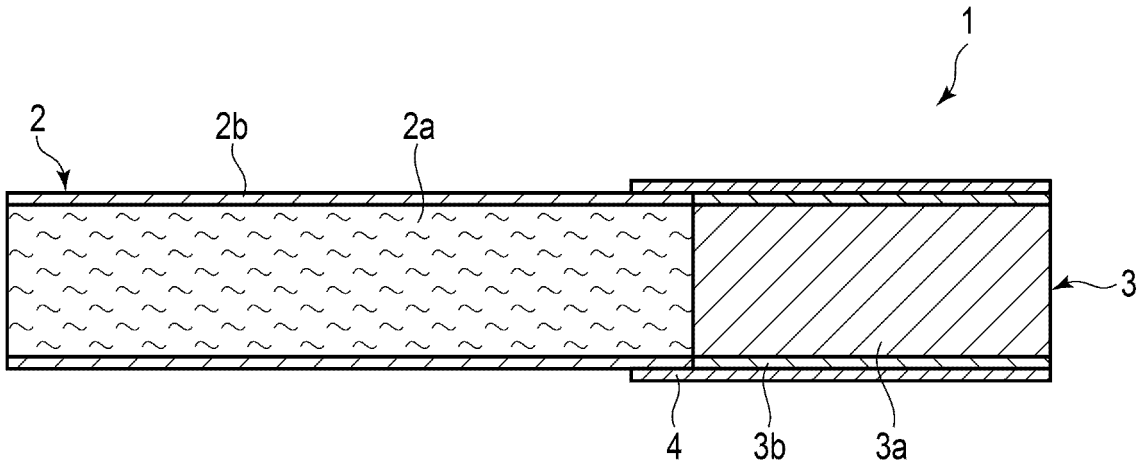

FIG. 3

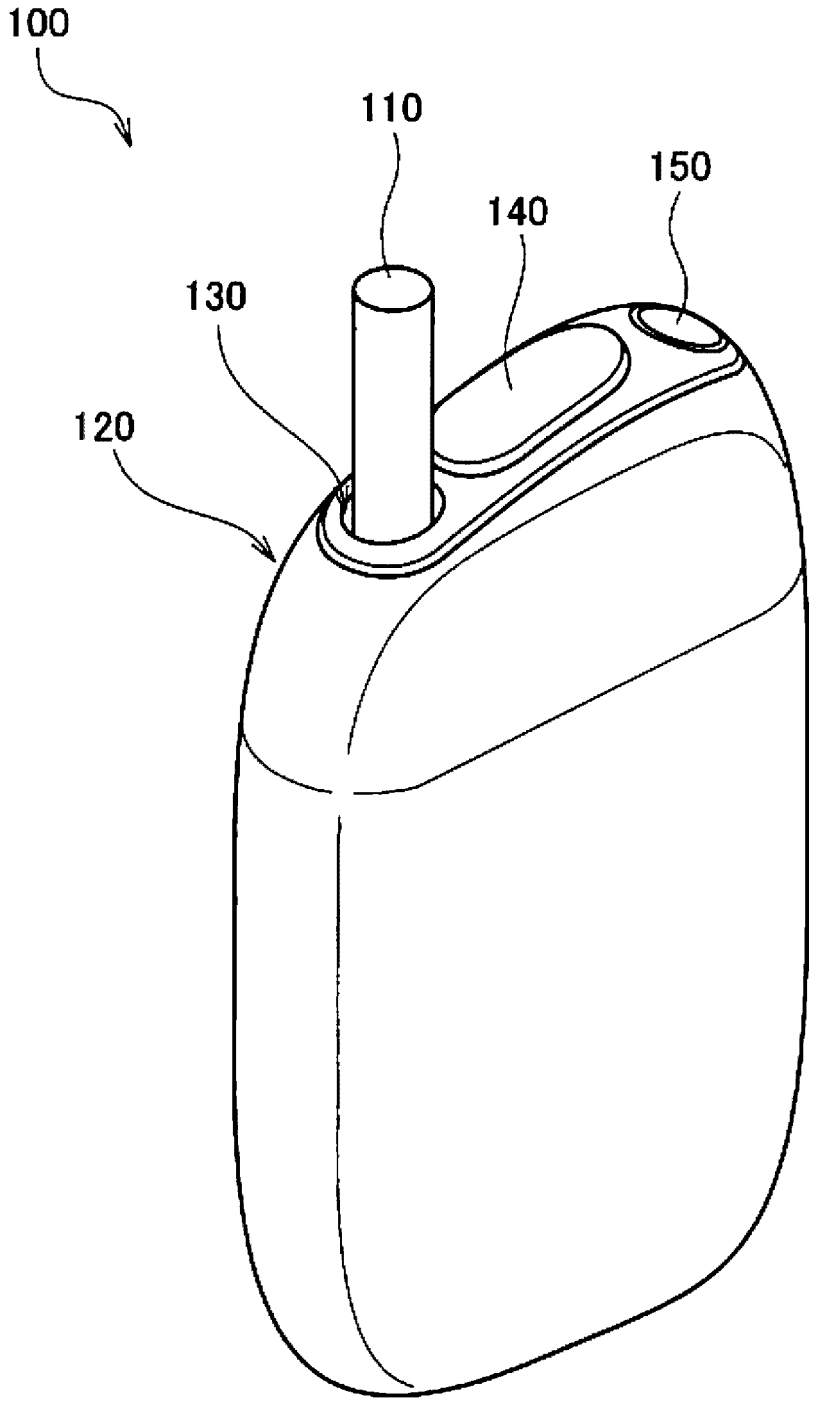

FIG. 4

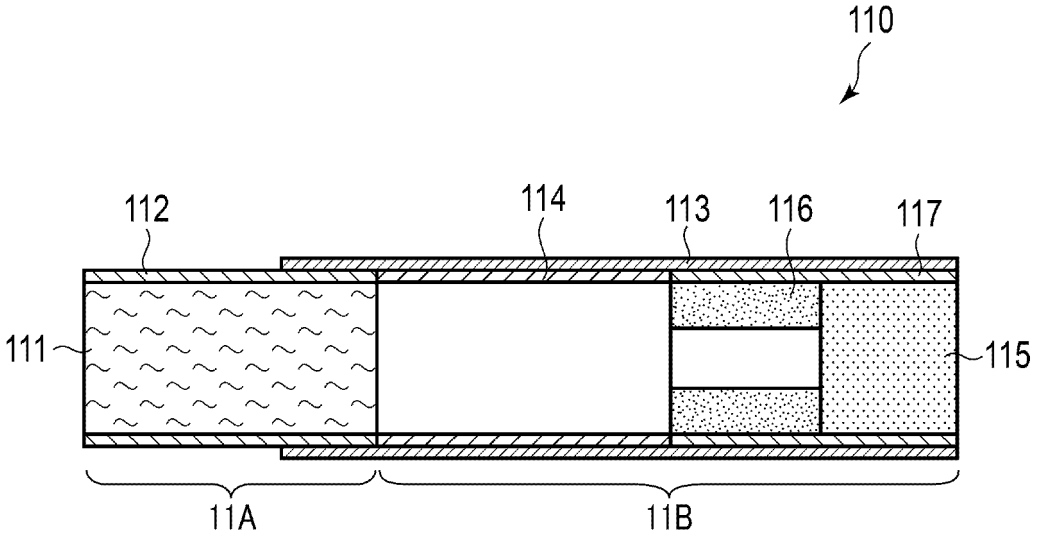

FIG. 5

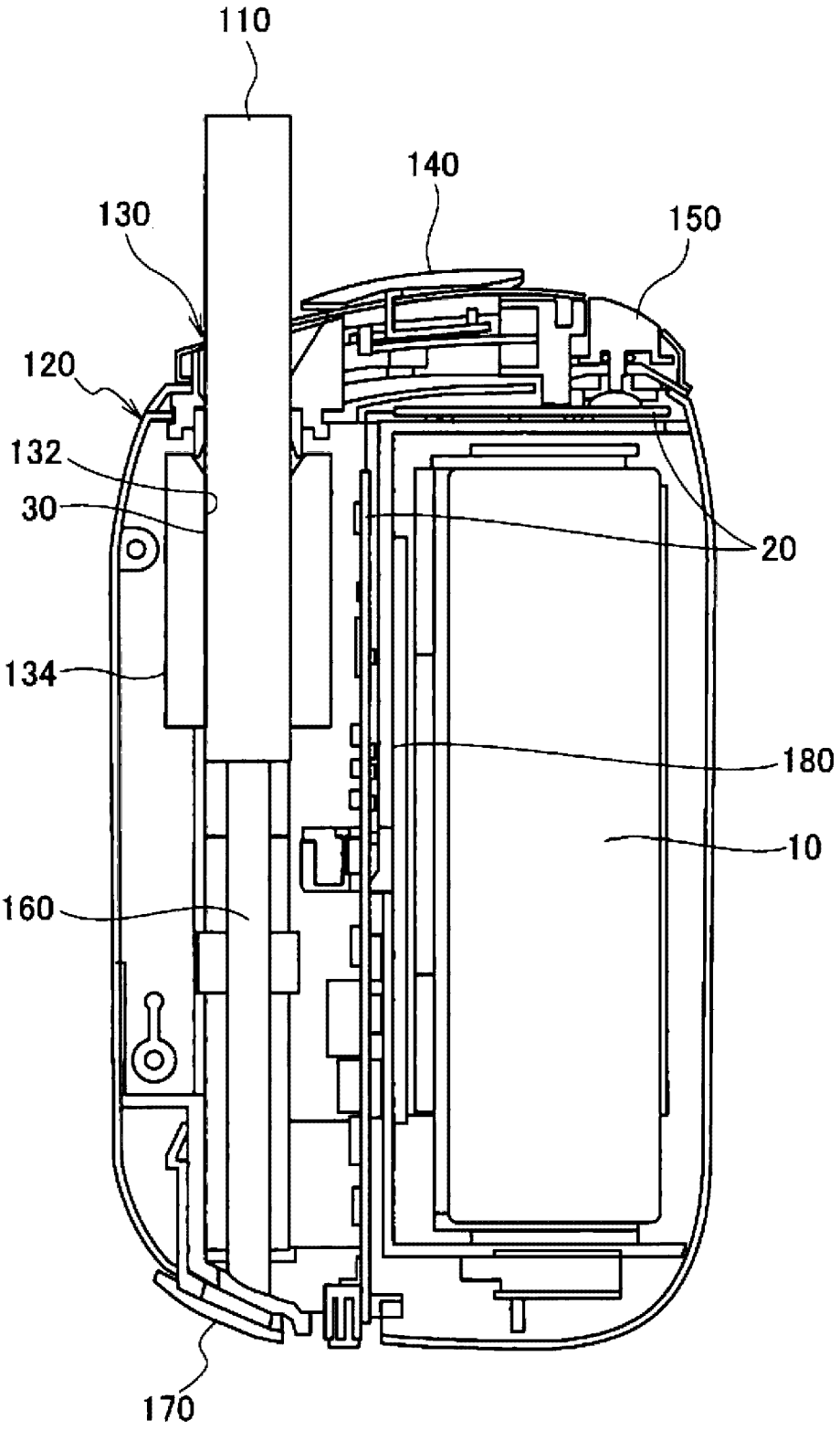

FIG. 6

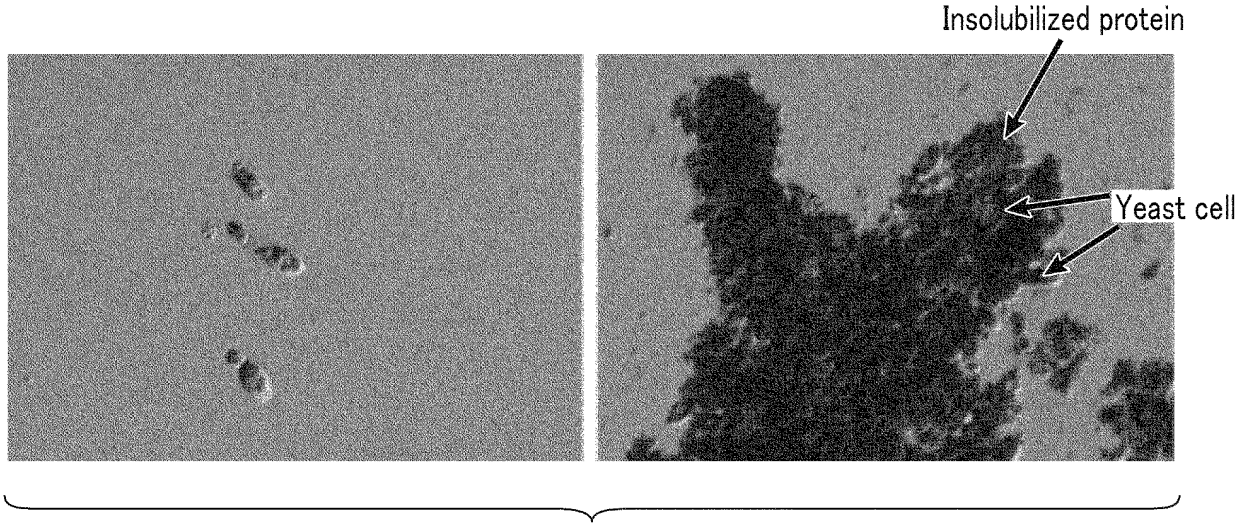
Insolubilized protein
Yeast cell

FIG. 7

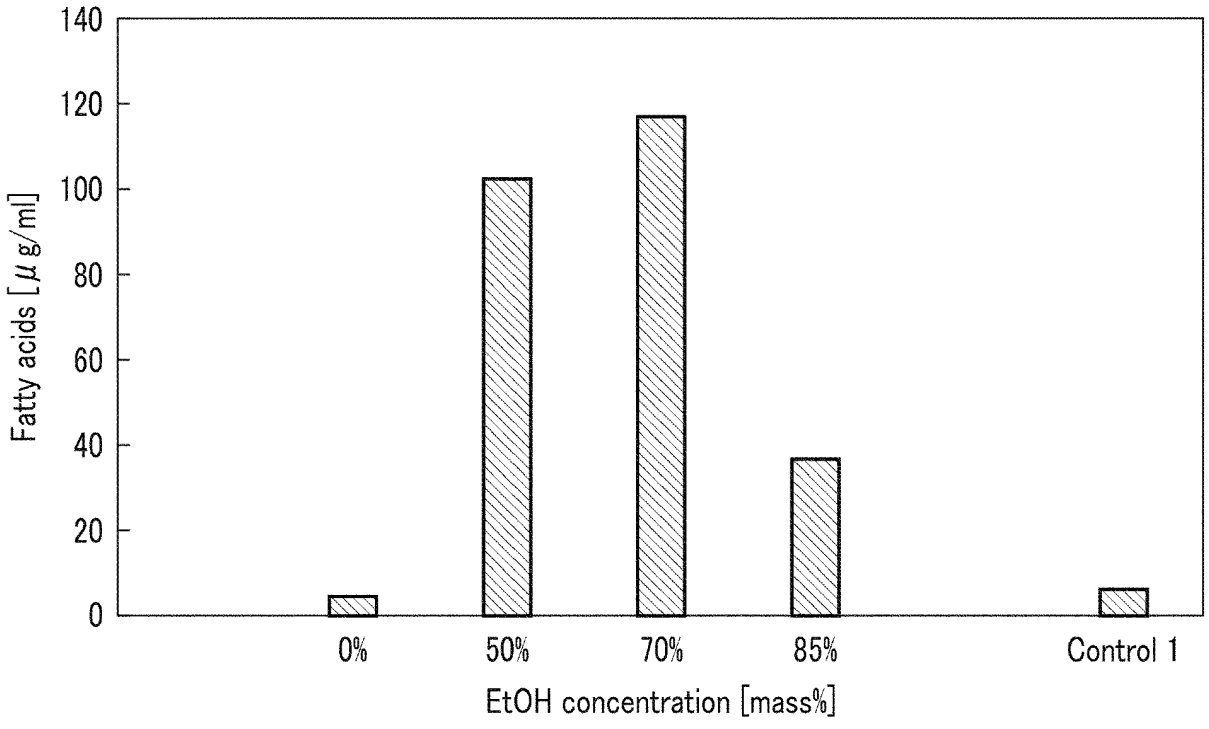
140
120
100
80
60
40
20
0
Fatty acids [μg/ml]
0%
50%
70%
85%
Control 1
EtOH concentration [mass%]

FIG. 8

INTERNATIONAL SEARCH REPORT

International application No.
PCT/JP2021/000222

A. CLASSIFICATION OF SUBJECT MATTER
Int.Cl. A24B15/167(2020.01)i, A24B15/24(2006.01)i
FI: A24B15/24, A24B15/167

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
Int.Cl. A24B15/167, A24B15/24

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
Published examined utility model applications of Japan 1922-1996
Published unexamined utility model applications of Japan 1971-2021
Registered utility model specifications of Japan 1996-2021
Published registered utility model applications of Japan 1994-2021

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	CN 106190558 A (CHINA TOBACCO JIANGXI INDUSTRIAL LLC) 07 December 2016 (2016-12-07), particularly, paragraphs [0026]-[0028]	1-2, 6-11
A	entire text, all drawings	3-5
Y	US 4895175 A (LTR INDUSTRIES) 23 January 1990 (1990-01-23), particularly, claim 1	1-2, 6-11
A	entire text, all drawings	3-5

☐ Further documents are listed in the continuation of Box C. ☒ See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
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"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
22 February 2021	09 March 2021

Name and mailing address of the ISA/	Authorized officer
Japan Patent Office 3-4-3, Kasumigaseki, Chiyoda-ku, Tokyo 100-8915, Japan	Telephone No.

Form PCT/ISA/210 (second sheet) (January 2015)

INTERNATIONAL SEARCH REPORT
Information on patent family members

International application No.
PCT/JP2021/000222

CN 106190558 A	07 December 2016	(Family: none)	
US 4895175 A	23 January 1990	EP 242271 A1	
		FR 2596621 A1	

REFERENCES CITED IN THE DESCRIPTION

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- WO 2006073065 A **[0109]**
- WO 2010110226 A **[0109]**
- WO 2015046385 A **[0109]**