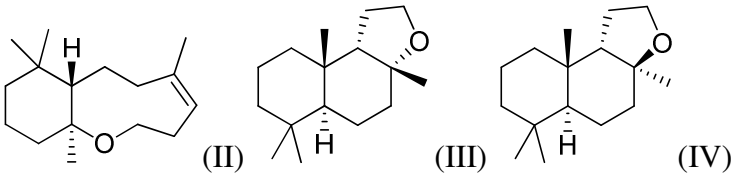
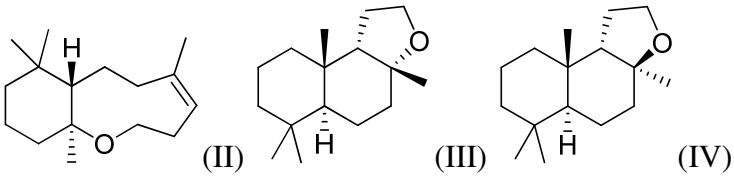
Claims

1. A method of isolating and purifying (-)-Ambrox comprising the step of selectively crystallizing (-)-Ambrox from a reaction mixture comprising one or more of the compounds (II), (III) and (IV)



2. A method of improving or enhancing the odour of (-)-Ambrox comprising the step of separating (-)-Ambrox from a mixture comprising one or more of the compounds (II), (III) or (IV)



by selective crystallization of (-)-Ambrox from the mixture, such that after the step of separating, (-)-Ambrox contains none, or only olfactory acceptable amounts, of the compounds (II), (III) or (IV).

4. The method according to claim 1, wherein the reaction mixture is free, or is substantially free, of homofarnesol.

5. The method according to claim 1, wherein the crystallizing solvent is selected from the group consisting of water, methanol, acetone, petroleum ether, hexane, t-butyl methyl ether, THF and ethyl acetate ethanol, toluene and mixtures thereof.

6. The method according to claim 5, wherein the crystallizing solvent is an ethanol water mixture.

7. The method according to claim 1, wherein the reaction mixture is formed as a result of an enzyme-catalyzed cyclization of homofarnesol comprising a mixture of 7E,3E and 7E,3Z homofarnesol geometric isomers of homofarnesol, wherein the reaction is carried out in the presence of a recombinant microorganism expressing the gene encoding the enzyme.

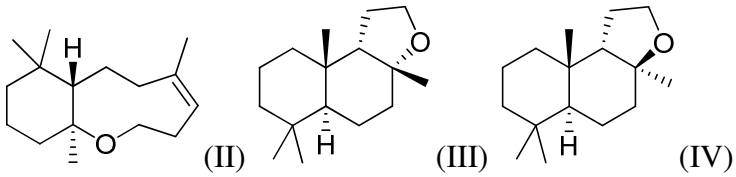
8. The method according to claim 7, wherein the reaction mixture of 7E,3E and 7E,3Z homofarnesol is enriched in the 7E,3E geometric isomer.

9. The method according to claim 7, wherein the reaction mixture of 7E,3E and 7E,3Z homofarnesol consists of 7E,3E and 7E,3Z homofarnesol and no other geometric isomers of homofarnesol.

10. The method according to claim 7, wherein the weight ratio of the 7E,3E isomer to 7E,3Z isomer is at least 80:20.

11. The method according to claim 7, wherein the enzyme is a wild-type squalene hopene cyclase or a variant of the wild-type squalene hopene cyclase.

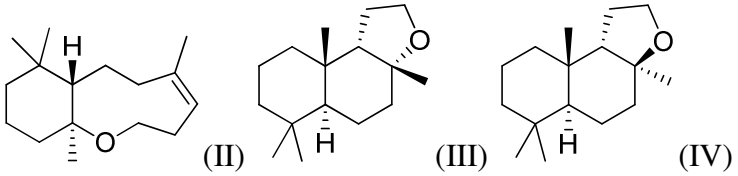
12. 12. A perfume ingredient consisting of (-)-Ambrox and olfactory acceptable amounts of one or more of the compounds (II), (III) or (IV).



(II) (III) (IV)

13. The perfume ingredient according to claim 12, comprising crystalline (-)-Ambrox.

14. A perfume composition comprising (-)-Ambrox and at least one other perfume ingredient, wherein said perfume composition contains olfactory acceptable amounts of one or more of the compounds (II), (III) or (IV).



(II) (III) (IV)

15. The method according to claim 3, wherein the reaction mixture is free, or is substantially free, of homofarnesol.

16. The method according to claim 3, wherein the crystallizing solvent is selected from the group consisting of water, methanol, acetone, petroleum ether, hexane, t-butyl methyl ether, THF and ethyl acetate ethanol, toluene and mixtures thereof.

17. The method according to claim 16, wherein the crystallizing solvent is an ethanol water mixture.

18. The method according to claim 3, wherein the mixture is formed as a result of an enzyme-catalyzed cyclization of homofarnesol comprising a mixture of 7E,3E and 7E,3Z homofarnesol geometric isomers of homofarnesol, wherein the reaction is carried out in the presence of a recombinant microorganism expressing the gene encoding the enzyme.

19. The method according to claim 18, wherein the reaction mixture of 7E,3E and 7E,3Z homofarnesol is enriched in the 7E,3E geometric isomer.

20. The method according to claim 18, wherein the reaction mixture of 7E,3E and 7E,3Z homofarnesol consists of 7E,3E and 7E,3Z homofarnesol and no other geometric isomers of homofarnesol.

21. The method according to claim 18, wherein the weight ratio of the 7E,3E isomer to 7E,3Z isomer is at least 80:20.

22. The method according to claim 18, wherein the enzyme is a wild-type squalene hopene cyclase or a variant of the wild-type squalene hopene cyclase.

23. The method according to claim 10, wherein the weight ratio of the 7E,3E isomer to 7E,3Z isomer is at least 90:10.

24. The method according to claim 10, wherein the weight ratio of the 7E,3E isomer to 7E,3Z isomer is at least 95:5.