CLAIMS

1. Method for treating a biological material, comprising the steps of:

   i) using biological material, and

   ii) contacting the biological material with a first non-aqueous composition comprising:
   (a1) 10 to 90 vol.% methanol, and
   (a2) at least one additional additive not comprising chloroform and/or trichloroethane, and

   iii) transferring the biological material into a second composition (B) comprising up to 99 vol.% ethanol.

2. Method according to claim 1,wherein the contacting of the biological material with the composition(s) is effected at a temperature in a range -80 °C to +80 °C.

3. Method according to claim 1 or 2, wherein the biological material, after having been contacted with the composition in step ii), is stored after step iii) at a temperature in a range of from –80°C to +80°C.

4. Method according to any of claims 1 to 3, wherein transfer step iii) is carried out by taking out the biological material from the composition of step ii) and immersing the material in composition (B), or by combining the composition of step ii) containing the biological material with composition (B).

5. Method according to any of claims 1 to 4, wherein the method comprises, an

iv) additional processing step

selected from manual processing of the biological material, processing of the biological material by microwave energy or processing of the biological material by any tissue processor device.

6. Method according to any of claims 1 to 5, wherein the biological material comprises cells or tissue.

7. Method according to claim 6, wherein the biological material contains organisms, isolated cells, organella, bacteria, fungi or parts of fungi, viruses, viroids, prions, tissue, tissue fragments, tissue sections, body fluids, natural, optionally isolated proteins, synthetic or modified proteins, natural, optionally isolated nucleic acids, synthetic or modified nucleic acids, other biomolecules such as, lipids, carbohydrates, metabolic products and metabolites, plants or parts of plants, faecal matter, swabs, aspirates, food samples, environmental samples, forensic samples.

8. Method according to any of claims 1 to 7, wherein the biological material is fresh or frozen biological material.

9. Method according to any of claims 1 to 8, further comprising : infiltration and/or embedding the material after step iii) in an embedding material (C), preferably selected from paraffin, mineral oil, non-water soluble waxes, celloidin, polyethylene glycols, polyvinyl alcohol, agar, gelatine, nitrocelluloses, methacrylate resins, epoxy resins, or other plastic media.

10. Method according to any of claims 1 to 9, further comprising:
storing the treated biological material,

isolation of at least one of the biological components originally contained in the sample,

analysis of at least one of the biological components in or of the biological material,

wherein the biological components comprise nucleic acids, proteins,peptides and/or peptide nucleic acids,

and/or

histological analysis of the biological material in particular comprising staining, microscopy, dissection, hybridisation and/or immuno-histo chemistry.

11. Method according to any of claims 1 to 10, wherein component (a2) is selected from the group comprising one or more detergents, one or more inhibitors that inhibit the degradation of nucleic acids and/or proteins, one or more viscosity regulators, one or more dyes, one or more buffer compounds, one or more preservatives, one or more complexants, one or more reducing agents, one or more substances that improve the permeability of cells, one or more chaotropic substances, one or more fixatives, one or more additional solvents that are different from methanol as well as mixtures of at least two of these additives.

12. Method according to claim 11, wherein component (a2) is selected from C_2 to C_{12} polyols, particularly diols and/or triols like 1,3-butanediol, 1,4-butanediol, 1,3-propanediol, 1,2-propanediol, 3-methyl-1,3,5-pentanetriol, 1,2,6-hexanetriol, glycerine, glycol; PEG and DEGMEA.

13. Method according to claim 1 wherein the first non-aqueous composition further comprises (a3) an acid.

14. Method according to claim 13, wherein component (a3) comprises formic acid, acetic acid, propionic acid and mixtures thereof.

15. Method according to claim 1 or 2, wherein the biological material, after having been contacted with the composition in step ii), is stored after step ii) at a temperature in a range of from -80°C to +80°C.

16. Method according to any of claims 1 to 12, wherein component (a2) is present in an amount of 1 to 50%.

17. Non-aqueous composition (A) for preservation of biological material, usable in a method according to any of claims 1 to 15 comprising

(α1) 10 to less than 80 vol.%, methanol, and

(α2) at least one additional additive, which either is not comprising chloroform and/or trichloroethane and/or formalin,
or which is selected from C_2 to C_{12} polyols, particularly diols and/or triols like 1,3-butanediol, 1,4-butanediol, 1,3-propanediol, 1,2-propanediol, 3-methyl-1,3,5-pentanetriol, 1,2,6-hexanetriol, glycerine, glycol, PEG and DEGMEA, and

(α3) an acid.

18. Composition according to claim 17, wherein component (α1) is present in an amount of 20% to less than 80 vol.%, or from 30 to less than 80% or from 50 to less than 80 vol.%.

19. Composition according to claims 17 or 18, wherein component (α2) or (a2) is present in an amount of 1 to 50 %.

20. Composition according to one of claims 17 to 19, wherein component (α3) comprises an organic acid.

21. A kit, comprising

(b1) a composition usable as the first composition in step ii) in the method of any of claims 17 to 20, and

(b2) composition (B) comprising up to 99 vol.% ethanol.

22. A kit, comprising

(b1) composition (A) of claim 17;

(b2) composition (B) or without composition (B); and

(b3) embedding material (C) and/or reagents for the analysis of bio-molecules in or of a biological material or for the analysis of the morphology of a biological material.

23. Use of a composition (A) according to any of claims 17 to 20 or of the kit according to claims 21 or 22 for the treatment of a biological material.

24. Use of a composition (A) according to any of claims 17 to 20 or of the kit according to claims 21 or 22 in a method according to any of the claims 1 to 16.

25. Method for the production of a treated biological material, wherein a composition (A) according to any of claims 17 to 20 or a kit according to claims 21 or 22 is used.

26. Method for the analysis of a biological material outside of a living body, wherein a method according to any of claims 1 to 16, a composition (A) according to any of claims 17 to 20 or a kit according to claims 21 or 22 is used.

27. Treated biological material, obtained by a method according to any of claims 1 to 16 or by contacting a biological material with a composition (A), as is defined in any of claims 17 to 20.

28. Method according to claim 2 further comprising storing the composition within the temperature range.

29. Composition according to claim 20 wherein the organic acid is a weak organic acid selected from one of formic acid, acetic acid, propionic acid and mixtures thereof.

CLAIMS

1. Method for treating a biological material, wherein the method comprises the steps of

   i) providing a biological material, and

   ii) contacting the biological material with a first non-aqueous composition comprising:

   (a1) 10 to 90 vol.% methanol, and

   (a2) at least one additional additive not comprising chloroform and/or trichloroethane, and

   (a3) an acid

   iii) transferring the biological material of step ii) into a second composition (B) comprising ethanol, wherein the ethanol content is up to 99 vol.% ethanol.

2. Method according to claim 1, wherein the contacting of the biological material with the composition(s) is effected at a temperature in a range -80 °C to +80 °C and optionally the composition further is stored as well within said temperature range.

3. Method according to claim 1 or 2, wherein the biological material, after having been contacted with the composition in step ii), is stored after step ii) or preferably after step iii) at a temperature in a range of from -80°C to +80°C.

4. Method according to any of claims 1 to 3, wherein transfer step iii) is carried out by taking out the biological material from the composition of step ii) and immersing the material in composition (B), or by combining the composition of step ii) containing the biological material with composition (B).

5. Method according to any of claims 1 to 4, wherein the method comprises, an

iv) additional processing step selected from manual processing of the biological material, processing of the biological material by microwave energy or processing of the biological material by any tissue processor device.

6. Method according to any of claims 1 to 5, wherein the biological material comprises cells or tissue.

7. Method according to claim 6, wherein the biological material contains organisms, isolated cells, organella, bacteria, fungi or parts of fungi, viruses, viroids, prions, tissue, tissue fragments, tissue sections, body fluids, natural, optionally isolated proteins, synthetic or modified proteins, natural, optionally isolated nucleic acids, synthetic or modified nucleic acids, other biomolecules such as, lipids, carbohydrates, metabolic products and metabolites, plants or parts of plants, faecal matter, swabs, aspirates, food samples, environmental samples, forensic samples.

8. Method according to any of claims 1 to 7, wherein the biological material is fresh or frozen biological material.

9. Method according to any of claims 1 to 8, further comprising: infiltration and/or embedding the material after step iii) in an embedding material (C), preferably selected from paraffin, mineral oil, non-water soluble waxes, celloidin, polyethylene glycols, polyvinyl alcohol, agar, gelatine, nitrocelluloses, methacrylate resins, epoxy resins, or other plastic media.

10. Method according to any of claims 1 to 8, further comprising :
storing the treated biological material,
isolation of at least one of the biological components originally contained in the sample,
analysis of at least one of the biological components in or of the biological material,
wherein the biological components comprise nucleic acids, proteins, peptides and/or peptide nucleic acids,

and/or

histological analysis of the biological material in particular comprising staining, microscopy, dissection, hybridisation and/or immuno-histo chemistry.

11. Method according to any of claims 1 to 10, wherein component (a2) is selected from the group comprising one or more detergents, one or more inhibitors that inhibit the degradation of nucleic acids and/or proteins, one or more viscosity regulators, one or more dyes, one or more buffer compounds, one or more preservatives, one or more complexants, one or more reducing agents, one or more substances that improve the permeability of cells, one or more chaotropic substances, one or more fixatives, one or more additional solvents that are different from methanol as well as mixtures of at least two of these additives.

12. Method according to claim 11, wherein component (a2) is selected from C_2 to C_{12} polyols, particularly diols and/or triols like 1,3-butanediol, 1,4-butanediol, 1,3-propanediol, 1,2-propanediol, 3-methyl-1,3,5-pentanetriol, 1,2,6-hexanetriol, glycerine, glycol; PEG and DEGMEA.

13. Non-aqueous composition (A) for preservation of biological material, usable in a method according to any of claims 1 to 12 but with no adding of crosslinking substances comprising:

(α1) 10 to less than 80 vol.%, methanol, and

(α2) at least one additional additive, which either is not comprising chloroform and/or trichloroethane and /or formalin,

or which is selected from C_2 to C_{12} polyols, particularly diols and/or triols like 1,3-butanediol, 1,4-butanediol, 1,3-propanediol, 1,2-propanediol, 3-methyl-1,3,5-pentanetriol, 1,2,6-hexanetriol, glycerine, glycol; PEG and DEGMEA, and

(α3) an acid.

14. Composition according to claim 13, wherein component (α1) is present in an amount of 20 % to less than 80 vol.%, preferably from 30 to less than 80% and most preferably from 50 to less than 80 vol.%.

15. Composition according to claims 13 or 14 or method according to any of claims 1 to 12, wherein component (α2) or (a2) is present in an amount of 1 to 50 %.

16. Composition or method according to any of claims 1 to 15, wherein component (α3) or (a3) is at least one organic acid, preferably a weak organic acid, more preferably selected from formic acid, acetic acid and propionic acid or mixture thereof.

17. A kit, comprising

(b1) a first non-aqueous composition according to claim 1, when used in step ii) in the method of any of claims 1 to 12, and

(b2) a non-aqueous composition comprising ethanol, wherein the ethanol content is up to 99% when used as composition (B) in step iii) in the method of any one of claims 1 to 12, and optionally

(b3) embedding material (C) and/or reagents for the analysis of biomolecules in or of a biological material or for the analysis of the morphology of a biological material.

18. A kit, comprising

(b1) composition (A) as claimed in claim 13 and at least one of:

(b2) non-aqueous composition comprising ethanol, wherein the ethanol content is up to 99% when used as composition (B) in step iii) in the method of any one of claims 1 to 12, and

(b3) embedding material (C) and/or reagents for the analysis of biomolecules in or of a biological material or for the analysis of the morphology of a biological material.

19. Use of a composition (A) according to any of claims 13 to 16 or of the kit according to claims 17 or 18 for the treatment of a biological material.

20. Use of a composition (A) according to any of claims 13 to 16 or of the kit according to claims 17 or 18 in a method according to any of the claims 1 to 12, 15 or16.

21. Method for the production of a treated biological material, wherein a composition (A) according to any of claims 13 to 16 or a kit according to claims 17 or 18 is used.

22. Method for the analysis of a biological material outside of a living body, wherein a method according to any one of claims 1 to 12, 15 or 16, a composition (A) according to any of claims 13 to 16 or a kit according to claims 17 or 18 is used.

23. Treated biological material, obtained by a method according to any of claims 1 to 12, 15 or 16 or by contacting a biological material with a composition (A), as is defined in any of claims 13 to 16.