## Claims

1. Use of a peptide and/or peptidomimetic, capable of self-assembling and forming a hydrogel in a physiologically compatible salt solution, having the general formula:

$$Z_a\text{-}(X)_b\text{-}(Y)_c\text{-}Z'_d$$

wherein

***Z*** is an N-terminal protecting group;

**a** is 0 or 1, preferably 1;

**X** is, at each occurrence, independently selected from the group consisting of aliphatic amino acids and aliphatic amino acid derivatives, and wherein the overall hydrophobicity decreases from N- to C-terminus;

**b** is an integer selected from 1 to 7;

**Y** is selected from the group consisting of basic polar amino acids and basic polar amino acid derivatives;

**c** is 1 or 2;

***Z'*** is a C-terminal polar head group; and

**d** is 1,

and **b** + **c** is at least 3,

in biofabrication.

2. The use according to claim 1, wherein said aliphatic amino acids and aliphatic amino acid derivatives, and said polar amino acids and polar amino acid derivatives are either D-amino acids or L-amino acids, optionally selected from the group consisting of alanine (Ala, A), homoallylglycine, homopropargylglycine, isoleucine (Ile, I), norleucine, leucine (Leu, L), valine (Val, V) and glycine (Gly, G), preferably from the group consisting of alanine (Ala, A), isoleucine (Ile, I), leucine (Leu, L), valine (Val, V) and glycine (Gly, G, wherein optionally all or a portion of said aliphatic amino acids are arranged in an order of decreasing amino acid size in the direction from N- to C-terminus, wherein the size of the aliphatic amino acids is defined as I = L > V > A > G.

3. The use according to claim 1 or 2, wherein said aliphatic amino acids have a sequence selected from

LIVAG (SEQ ID NO: 1),
ILVAG (SEQ ID NO: 2),
LIVAA (SEQ ID NO: 3),
LAVAG (SEQ ID NO: 4),
AIVAG (SEQ ID NO: 5)
GIVAG (SEQ ID NO: 6)
VIVAG (SEQ ID NO: 7)
ALVAG (SEQ ID NO: 8)
GLVAG (SEQ ID NO: 9)
VLVAG (SEQ ID NO: 10)
IVAG (SEQ ID NO: 11)
LIVA (SEQ ID NO: 12)
LIVG (SEQ ID NO: 13)
IVA (SEQ ID NO: 47) and
IV (SEQ ID NO: 48),

wherein, optionally, there is an A preceding such sequence at the N-terminus.

4. The use according to any one of the foregoing claims, wherein said basic polar amino acids or basic polar amino acid derivatives are selected from the group consisting of ornithine (Orn), lysine (Lys, K), 2,4-diaminobutyric acid (Dab), and 2,3-diaminopropionic acid (Dap),.

5. The use according to any one of the foregoing claims, wherein **c** is 2 and said polar amino acids are identical amino acids,
or wherein **c** is 1 and said polar polar amino acid comprises any one of lysine, ornithine, 2,4-diaminobutyric acid (Dab) and 2,3-diaminopropionic acid (Dap), wherein optionally $(\mathbf{Y})_c$ has a sequence selected from Lys, Orn, Dab, Cys-Lys, Cys-Orn, Cys-Dab, Cys-Dap, Lys-Lys, Lys-Orn, Lys-Dab, Lys-Dap, Ser-Lys, Ser-Orn, Ser-Dab, Ser-Dap, Orn-Lys, Orn-Orn, Orn-Dab, Orn-Dap, Dab-Lys, Dab-Orn, Dab-Dab, Dab-Dap, Dap-Lys, Dap-Orn, Dap-Dab, Dap-Dap.

6. The use according to any one of the foregoing claims, wherein $(\mathbf{X})_b$-$(\mathbf{Y})_c$ has a sequence selected from the group consisting of

LIVAGK (SEQ ID NO: 20),

ILVAGK (SEQ ID NO. 21),
AIVAGK (SEQ ID NO: 24),
IVAK (SEQ ID NO: 28),
IIIK (SEQ ID NO: 30),
LIVAGOrn (SEQ ID NO: 31),
ILVAGOrn (SEQ ID NO: 32),
AIVAGOrn (SEQ ID NO: 33),
LIVAGDab (SEQ ID NO: 34),
ILVAGDab (SEQ ID NO: 35),
AIVAGDab (SEQ ID NO: 36),
LIVAGDap (SEQ ID NO: 37),
ILVAGDap (SEQ ID NO: 38),
AIVAGDap (SEQ ID NO: 39),
IVOrn (SEQ ID NO: 63),
IVDab (SEQ ID NO: 64),
IVDap (SEQ ID NO: 65),
IVK (SEQ ID NO: 66),
VIK (SEQ ID NO: 67),
VIOrn (SEQ ID NO: 68),
VIDab (SEQ ID NO: 69), and
VIDap (SEQ ID NO: 70),
.

7. The use according to any one of the foregoing claims, wherein **a** is 1 and said N-terminal protecting group ***Z*** has the general formula –C(O)–R, wherein R is selected from the group consisting of H, unsubstituted or substituted alkyls, and unsubstituted or substituted aryls,. wherein R is preferably selected from the group consisting of methyl, ethyl, propyl, isopropyl, butyl and isobutyl, wherein optionally said N-terminal protecting group ***Z*** is an acetyl group.

8. The use according to any one of claims 1 to 7, wherein said N-terminal protecting group ***Z*** is a peptidomimetic molecule, including natural and synthetic amino acid derivatives, wherein the N-terminus of said peptidomimetic molecule may be modified with a functional group selected from the group consisting of carboxylic acid, amide, alcohol, aldehyde, amine, imine, nitrile, an urea analog, phosphate, carbonate, sulfate, nitrate, maleimide, vinyl sulfone, azide,

alkyne, alkene, carbohydrate, imide, peroxide, ester, aryl, ketone, sulphite, nitrite, phosphonate, and silane.

9. The use according to any one of the foregoing claims, wherein said C-terminal polar head group ***Z'*** is selected from

(a) polar functional groups, comprising

-COOH, -COOR, -COR, -CONHR or -CONRR' with R and R' being selected from the group consisting of H, unsubstituted or substituted alkyls, and unsubstituted or substituted aryls,

$-NH_2$, -OH, -SH, -CHO, maleimide, imidoester, carbodiimide ester, isocyanate;

(b) small molecules,

comprising sugars, alcohols, hydroxy acids, amino acids, vitamins, biotin, L-Dopa, thyroxine;

(c) linkers terminating in a polar functional group, comprising

ethylenediamine, PEG, carbodiimide ester, imidoester;

(d) linkers coupled to small molecules or vitamins, comprising such as biotin, sugars, hydroxy acids,

wherein the polar head group ***Z'*** is preferably an amide group,

wherein optionally the polar functional group(s) can be used for chemical conjugation or coupling of at least one compound selected from

(a) bioactive molecules or moieties,

comprising growth factors, cytokines, lipids, cell receptor ligands, hormones, prodrugs, drugs, vitamins, antigens, antibodies, antibody fragments, oligonucleotides (including but not limited to DNA, messenger RNA, short hairpin RNA, small interfering RNA, microRNA, peptide nucleic acids, aptamers), saccharides;

(b) label(s), dye(s),

comprising imaging contrast agents;

(c) pathogens,

comprising viruses, bacteria and parasites;

(d) micro- and nanoparticles

or (e) combinations thereof

wherein said chemical conjugation can be carried out before or after self-assembly of the peptide and/or peptidomimetic.

10. The use according to any one of the foregoing claims, wherein said C-terminal polar head group ***Z'*** is a peptidomimetic molecule, including natural and synthetic amino acid derivatives, wherein the C-terminus of said peptidomimetic molecule may be modified with a functional group selected from the group consisting of carboxylic acid, amide, alcohol, aldehyde, amine, imine, nitrile, an urea analog, phosphate, carbonate, sulfate, nitrate, maleimide, vinyl sulfone, azide, alkyne, alkene, carbohydrate, imide, peroxide, ester, aryl, ketone, sulphite, nitrite, phosphonate, and silane.

11. The use according to any one of the foregoing claims, comprising a conformational change of the peptide(s) and/or peptidomimetic(s) during self-assembly,
preferably a conformational change from a random coil conformation to a helical intermediate structure to a final beta turn or cross beta conformation,
wherein, preferably, the conformational change is dependent on the peptide concentration, ionic environment, pH and temperature.

12. The use according to any one of the foregoing claims, wherein at least one peptide and/or peptidomimetic as defined in any one of claims 1 to 10 forms a hydrogel, or wherein different peptide(s) and/or peptidomimetic(s) as defined in any one of claims 1 to 10 are used to form the hydrogel.

13. The use according to any one of the foregoing claims, comprising stimuli-responsive gelation of at least one peptide and/or peptidomimetic as defined in any one of claims 1 to 10, wherein said stimulus/stimuli or gelation condition(s) is/are the physiologically compatible salt solution and pH, or temperature, wherein optionally the physiologically compatible salt solution comprises 0.9% saline and PBS and/or gelation is enhanced at a pH above physiological pH, preferably pH 7 to 10.

14. The use according to claim 12 or 13, wherein the gelation condition(s) (in particular pH, salt concentration and/or temperature) influence the properties of the hydrogel obtained, such as its mechanical stiffness, rigidity, porosity.

15. The use according to any one of the foregoing claims, wherein at least one peptide and/or peptidomimetic as defined in any one of claims 1 to 10 is dissolved in water and subsequently the physiologically compatible salt solution is added wherein the solution obtained can be dispensed through needles and print heads prior to or after addition of the physiologically compatible salt solution.

16. The use according to any one of the foregoing claims, comprising conjugation or coupling of further compound(s) to the peptides and/or peptidomimetic, preferably to the polar functional group(s), post-assembly, wherein said further compound(s) are selected from

(a) bioactive molecules or moieties,

comprising growth factors, cytokines, lipids, cell receptor ligands, hormones, prodrugs, drugs, vitamins, antigens, antibodies, antibody fragments, oligonucleotides (including but not limited to DNA, messenger RNA, short hairpin RNA, small interfering RNA, microRNA, peptide nucleic acids, aptamers), saccharides;

(b) label(s), dye(s),

comprisingimaging contrast agents;

(c) pathogens,

comprisingviruses, bacteria and parasites;

(d) micro- and nanoparticles

or (e) combinations thereof.

17. The use according to any of the foregoing claims, wherein the peptide and/or peptidomimetic is present at a concentration in the range of from 0.1 % to 30 % (w/w), preferably 0.1 % to 20 % (w/w), more preferably 0.1 % to 10 % (w/w), more preferably 0.1 % to 5 % (w/w), even more preferably 0.1 % to 3 % (w/w), with respect to the total weight of said hydrogel.

18. The use according to any one of the foregoing claims, comprising the addition or mixing of cells prior or during gelation, which are encapsulated by the hydrogel,
preferably comprising the addition of further compound(s) prior or during gelation, which are co-encapsulated by the hydrogel,
optionally the cells are added onto a printed hydrogel after gelation,
wherein said cells comprise any one of:

(a) stem cells (adult, progenitor, embryonic and induced pluripotent stem cells);
(b) transdifferentiated progenitor cells; or
(c) primary cells (isolated from patients); or
(d) cell lines (such as epithelial, neuronal, hematopoietic and cancer cells).

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19. The use according to claim 18, comprising (1) the addition or mixing of cells prior or during gelation, which are encapsulated by the hydrogel,
and (2) subsequently comprising the addition of cells onto the printed hydrogel,wherein said cells of (1) and (2) are the same or different,
preferably comprising:

(a) stem cells (adult, progenitor, embryonic and induced pluripotent stem cells),
(b) transdifferentiated progenitor cells, or
(c) primary cells (isolated from patients) or
(d) cell lines (such as epithelial, neuronal, hematopoietic and cancer cells).

20. The use according to any one of the foregoing claims, comprising the addition of cross-linkers to the peptide(s) and/or peptidomimetic(s),wherein said cross-linkers preferably include short linkers, linear and branched polymers, polymers conjugated with bioactive molecules or moieties.

21. A method of preparing a hydrogel, the method comprising dissolving at least one peptide and/or peptidomimetic as defined in any one of claims 1 to 10 in an aqueous solution and subsequently adding a physiologically compatible salt solution.

22. The method according to claim 21, comprising stimuli-responsive gelation of the at least one peptide and/or peptidomimetic as defined in any one of claims 1 to 10,
wherein said stimulus/stimuli or gelation condition(s) is/are the physiologically compatible salt solution and pH, or temperature, wherein optionally the physiologically compatible salt solution comprises PBS or 0.9% saline and PBS and gelation is enhanced at a pH above physiological pH, preferably pH 7 to 10.

23. The method according to claim 21 or 22, wherein the dissolved peptide and/or peptidomimetic is further warmed or heated, wherein the temperature is in the range from 20 °C to 90 °C, preferably from about 30 °C to 70 °C, more preferably from about 37 °C to 70 °C.

24. The method according to any of claims 21 to 23, wherein the at least one peptide and/or peptidomimetic is dissolved at a concentration from 0.01 μg/ml to 100 mg/ml, preferably at a concentration from 1 mg/ml to 50 mg/ml, more preferably at a concentration from about 1 mg/ml to about 20 mg/ml.

25. A method of preparing continuous fibres, the method comprising

(a) dissolving at least one peptide and/or peptidomimetic as defined in any one of claims 1 to 10 in an aqueous solution, such as water, and

(b) dispensing the solution obtained through needles, print heads, fine tubings and/or microfluidic devices into a buffered solution, such as PBS.

26. The method according to any one of claims 21 to 25, comprising the addition of *further compound(s)* prior to or during gelation/self-assembly, which are encapsulated by the hydrogel,

wherein said further compound(s) can be selected from

(a) bioactive molecules or moieties,

comprising growth factors, cytokines, lipids, cell receptor ligands, hormones, prodrugs, drugs, vitamins, antigens, antibodies, antibody fragments, oligonucleotides (including but not limited to DNA, messenger RNA, short hairpin RNA, small interfering RNA, microRNA, peptide nucleic acids, aptamers), saccharides;

(b) label(s), dye(s),

comprising imaging contrast agents;

(c) pathogens,

comprising viruses, bacteria and parasites;

(d) quantum dots, nano- and microparticles; or

(e) combinations thereof.

27. The method according to any one of claims 21 to 26, comprising the addition or mixing of *cells* prior or during gelation/self-assembly, which are encapsulated by the hydrogel, preferably comprising the addition of further compound(s) prior or during gelation preferably those defined in claim 26, which are co-encapsulated by the hydrogel,
optionally the cells are added onto the printed hydrogel after gelation,
wherein said cells comprise and one of:

(a) stem cells (mesenchymal, adult, progenitor, embryonic and induced pluripotent stem cells),
(b) transdifferentiated progenitor cells or
(c) primary cells isolated from patient samples (fibroblasts, nucleus pulposus) or
(d) cell lines (preferably epithelial, neuronal, hematopoietic and cancer cells).

28. The method according to claim 27, comprising the following steps:
(1) the addition or mixing of cells prior or during gelation, which are encapsulated by the hydrogel, and
(2) subsequently the addition of cells onto the printed hydrogel,
wherein said cells of (1) and (2) are the same or different,
and comprise at least one of:
(a) stem cells (adult, progenitor, embryonic and induced pluripotent stem cells),
(b) transdifferentiated progenitor cells, or
(c) primary cells (isolated from patients) or
(d) cell lines (such as epithelial, neuronal, hematopoietic and cancer cells).

29. The method according to any one of claims 21 to 28, comprising the addition of *cross-linkers* to the peptide(s) and/or peptidomimetic(s) prior, during or after gelation/self-assembly, wherein said cross-linkers preferably include short linkers, linear and branched polymers, polymers conjugated with bioactive molecules or moieties preferably those defined in claim 26, wherein, preferably, said cross-linkers interact electrostatically with the peptides and/or peptidomimetic(s) during self-assembly.

30. The method according to any one of claims 21 to 29, comprising the use of different peptide(s) and/or peptidomimetic(s).

31. Use of a hydrogel obtained by a method according to any one of claims 21 to 30 for *in vitro* substrate-mediated gene delivery, wherein oligonucleotides are encapsulated in the hydrogel and cells are co-encapsulated or seeded onto said hydrogel.

32. The use according to any one of claims 1 to 12 or the use of a hydrogel obtained by a method according to any of claims 21 to 30, for obtaining 2D mini-hydrogel arrays, preferably

comprising using printers, pintools and micro-contact printing, preferably comprising printing the 2D mini-hydrogels onto electrical circuits or piezoelectric surfaces that conduct current.

33. The use according to any one of claims 1 to 12 or the use of a hydrogel obtained by a method according to any of claims 21 to 30, in the manufacture of an injectable injectable scaffold, injectable implant and/or implantable scaffold,
for the treatment of degenerative disc disease.

34. The use according to any one of claims 1 to 12 or the use of a hydrogel obtained by a method according to any of claims 21 to 30, comprising bioprinting, such as 3D microdroplet printing, and biomoulding, preferably for obtaining 3D organoid structures or 3D macromolecular biological constructs, optionally comprising the use of moulds preferably silicone to pattern the hydrogels in 3D.

35. The use according to claim 34, for obtaining multi-cellular constructs,
which comprise different cells/cell types,which preferably comprise co-encapsulated further compound(s) preferably those defined in claim 24 and/or cross-linkers preferably defined in claim 26, wherein optionally the 3D cellular constructs or scaffolds comprise encapsulated cells and/or cells deposited or printed onto the surface of the printed/fabricated scaffold.

36. A method for obtaining a multi-cellular construct, comprising
(i) preparing a hydrogel by the method according to any one of claims 21 to 30,
comprising the addition or mixing of different cells or cell types prior or during gelation/self-assembly, which are encapsulated by the hydrogel,
wherein said cells comprise any one of:
(a) stem cells (mesenchymal, progenitor, embryonic and induced pluripotent stem cells), or
(b) transdifferentiated progenitor cells; or
(c) primary cells isolated from patient samples (fibroblasts, nucleus pulposus)
preferably comprising the addition of further compound(s) including those defined in claim 26 prior or during gelation, which are co-encapsulated by the hydrogel,
optionally comprising the addition of cross-linkers preferably those defined in claim 29 to the peptide(s) and/or peptidomimetic(s) prior or during gelation/self-assembly,
(ii) obtaining the multi-cellular construct.

37. A method for obtaining a multi-cellular construct, comprising

(i) preparing a hydrogel by the method according to any one of claims 21 to 30, comprising the following steps:

(1) the addition or mixing of cells prior or during gelation, which are encapsulated by the hydrogel, and

(2) subsequently the addition of cells onto the printed hydrogel,

wherein said cells of (1) and (2) are different,

andare selected from:

(a) stem cells (adult, progenitor, embryonic and induced pluripotent stem cells), or

(b) transdifferentiated progenitor cells, and primary cells (isolated from patients) or

(c) cell lines (such as epithelial, neuronal, hematopoietic and cancer cells),

preferably comprising the addition of further compound(s) including those defined in claim 26 prior or during gelation, which are co-encapsulated by the hydrogel,

optionally comprising the addition of cross-linkers preferably those defined in claim 29 to the peptide(s) and/or peptidomimetic(s) prior or during gelation/self-assembly,

(ii) obtaining the multi-cellular construct.

38. The method of claim 36 or 37, wherein the multi-cellular construct obtained is formed in a mould preferably silicone, wherein preferably the multi-cellular construct comprises micro-domains.

39. Use of a 3D biological construct obtained by a method according to any one of claims 21 to 30 or a multi-cellular construct obtained according to any one of claims 36 to 38 as

(a) organoid model for screening biomolecule libraries, studying cell behavior, infectivity of pathogens and disease progression, screening infected patient samples, evaluating drug efficacy and toxicity,

(b) *in vitro* disease model.

40. The use according to any of claims 34, 35 or 39 for

(a) preparation of cell based assays,

preferably for identifying patient specimens, more preferably for identifying patient specimens containing pathogens including dengue, malaria, norovirus, which do not infect primary cells that have lost their native phenotype;

(b) recovery of infected cells to identify and expand pathogen(s) of interest,

preferably for elucidating mechanism(s) of infection and/or enabling the design of molecules that inhibit pathogen infection and/or replication.