Claims:

1. A cardiac organoid comprising a heart tissue with at least one inner cavity or a central chamber, wherein the cardiac organoid comprises at least two different heart tissues selected from left ventricle tissue, right ventricle tissue, atrial tissue, outflow tract tissue, atrioventricular canal tissue, sinoatrial node tissue, and atrioventricular node tissue, wherein the central chamber can be shared by at least two different heart tissues, and wherein the at least two different heart tissues comprise a electrophysiological or calcium signaling connection and/or ability to propagate a tissue contraction;
and wherein either i) the cardiac organoid has the central chamber and said central chamber is shared by at least two different heart tissues, or ii) the cardiac organoid has at least two different heart tissues with the ability to propagate a contraction with a beating behaviour that starts in one of the different heart tissues and propagates or projects into a neighbouring heart tissue, or both i) and ii).

2. The cardiac organoid of claim 1, wherein
the left ventricle tissue comprises at least 60% cardiac cells selected from cardiomyocytes, endocardial cells and epicardial cells;
the right ventricle tissue comprises at least 60% cardiomyocytes;
the atrial tissue comprises at least 60% cardiomyocytes;
the outflow tract tissue comprises at least 60% cardiomyocytes;
the atrioventricular canal tissue comprises at least 60% cardiomyocytes;
the sinoatrial node tissue comprises at least 60% cardiomyocytes; and/or
atrioventricular node tissue comprises at least 60% cardiomyocytes.

3. The cardiac organoid of claim 1 or 2, wherein the inner cavity or central chamber is completely surrounded by tissue selected from left ventricle tissue, right ventricle tissue, atrial tissue, outflow tract tissue, atrioventricular canal tissue, sinoatrial node tissue, or atrioventricular node tissue; and/or wherein the volume of the inner cavity or central chamber

is not leading into a major blood vessel.

4. The cardiac organoid of any one of claims 1 to 3 having a size in its largest dimension of 0.3 mm to 50 mm.

5. The cardiac organoid of any one of claims 1 to 4, wherein left ventricle tissue cells express one or more expression markers selected from NPPA, IRX4 and HEY2; and/or left ventricle tissue cells lack expression of one or more expression markers selected from NR2F2, TBX2 and TBX3;
right ventricle tissue cells express one or more expression markers selected from NPPA, IRX1, IRX2 and PRDX1; and/or right ventricle tissue cells lack expression of one or more expression markers selected from NR2F2, TBX2, and WNT5A;
atrial tissue cells express one or more expression markers selected from NPPA, NR2F1, NR2F2 and HEY1; and/or atrial tissue cells lack expression of one or more expression markers selected from IRX1, IRX4 and HEY2;
outflow tract tissue cells express one or more expression markers selected from WNT5A, MSX1, BMP4, WNT11 and RSPO3; and/or outflow tract tissue cells lack expression of one or more expression markers selected from TBX3, NR2F1 and NPPA;
atrioventricular canal tissue cells express one or more expression markers selected from TBX2, MSX2 and RSPO3; and/or atrioventricular canal tissue cells lack expression of one or more expression markers selected from IRX1, IRX4 and NPPA;
sinoatrial node tissue cells express one or more expression markers selected from SHOX2, TBX3, HCN4, ISL1 and GJC1; and/or sinoatrial node tissue cells lack expression of one or more expression markers selected from NKX2.5, IRX1, IRX4 and NPPA; and/or
atrioventricular node tissue cells express one or more expression markers selected from TBX3, TBX5, KCNE1, HCN4 and GJC1; and/or atrioventricular node tissue cells lack expression of one or more expression markers selected from RSPO3, MSX2, IRX4 and NPPA.

6. The cardiac organoid of any one of claims 1 to 5, wherein the size of the inner cavity or central chamber at its largest dimension is at least 30% of the size of the cardiac organoid at

its largest dimension.

7. A method to generate a cardiac organoid of any one of claims 1 to 6 comprising generating at least two different heart tissues in vitro, wherein the different heart tissues are selected from left ventricle progenitor first heart field tissue, right ventricle/outflow tract progenitor anterior second heart field tissue, right ventricle progenitor anterior second heart field tissue, atrial progenitor posterior second heart field tissue, outflow tract progenitor anterior second heart field tissue, atrioventricular canal progenitor posterior second heart field tissue, sinoatrial node progenitor posterior second heart field tissue, and atrioventricular node tissue, and fusing the at least two heart tissues,
culturing the fused tissue model and letting calcium signaling connection, ability to propagate a tissue contraction and/or a central chamber between the different heart tissues form.

8. The method of claim 7, wherein the different heart tissues have been cultured and differentiated from a pluripotent cell and wherein the fusion is at culture day 1 to 7 from a pluripotent stage; preferably right ventricle/outflow tract progenitor anterior second heart field tissue, right ventricle progenitor second heart field tissue, atrial progenitor second heart field tissue, atrioventricular progenitor second heart field canal tissue, sinoatrial node progenitor second heart field tissue, and/or atrioventricular node tissue is fused at culture day 2 to 5; or preferably left ventricle progenitor first heart field tissue is fused when expressing the expression marker TBX5 and/or HAND1; preferably right ventricle/outflow tract progenitor anterior second heart field tissue or right ventricle progenitor anterior second heart field tissue is fused when expressing the expression marker TBX1, FOXC1 and/or FOXC2; preferably atrial progenitor posterior second heart field tissue is fused when expressing the expression marker HOXB1, TBX5 and/or OSR1; preferably atrioventricular canal progenitor posterior second heart field tissue is fused when expressing the expression marker TBX3, FOXF1 and/or HOXB1; preferably sinoatrial node progenitor posterior second heart field tissue is fused when expressing the expression marker SHOX2, TBX3, HCN4, ISL1 and/or

GJC1; and/or preferably atrioventricular node tissue is fused when expressing the expression marker TBX3, TBX5, KCNE1, HCN4 and/or GJC1.

9. The method of claim 7 or 8,

wherein one of the at least two different heart tissues is left ventricle progenitor first heart field tissue and generating left ventricle progenitor first heart field tissue comprises differentiating mesoderm cells into left ventricular precursor cells in a medium comprising a bone morphogenic protein, preferably BMP4, a fibroblast growth factor, preferably FGF2, insulin, a Wnt inhibitor, preferably Wnt-C59 or IWP2, and retinoic acid, with the retinoic acid having a concentration of 5 nM to 100 nM, in the medium;

wherein one of the at least two different heart tissues is right ventricle/outflow tract progenitor anterior second heart field tissue and generating right ventricle/outflow tract progenitor anterior second heart field tissue comprises differentiating mesoderm cells into right ventricular and/or outflow tract precursor cells in a medium comprising a TGF-beta inhibitor, preferably SB 431542, and a Wnt inhibitor, preferably Wnt-C59 or XAV-939;

wherein one of the at least two different heart tissues is outflow tract progenitor anterior second heart field tissue and generating outflow tract progenitor second heart field tissue comprises differentiating mesoderm cells into outflow tract tissue precursor cells in a medium comprising a TGF-beta inhibitor, preferably SB 431542, and a Wnt inhibitor, preferably Wnt-C59 or XAV-939;

wherein one of the at least two different heart tissues is atrial progenitor posterior second heart field tissue and generating atrial progenitor posterior second heart field tissue comprises differentiating mesoderm cells into atrial tissue precursor cells in a medium comprising a TGF-beta inhibitor, preferably SB 431542, a Wnt inhibitor, preferably Wnt-C59 or XAV-939, and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM; and/or

wherein one of the at least two different heart tissues is atrioventricular canal progenitor second heart field tissue and generating atrioventricular canal progenitor posterior second

heart field tissue comprises differentiating mesoderm cells into atrioventricular canal tissue precursor cells in a medium comprising a TGF-beta inhibitor, preferably SB 431542, a Wnt inhibitor, preferably Wnt-C59 or XAV-939, a bone morphogenic protein, preferably BMP4 and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM;

wherein one of the at least two different heart tissues is sinoatrial node progenitor posterior second heart field tissue and generating sinoatrial node progenitor posterior second heart field tissue comprises differentiating mesoderm cells into sinoatrial node tissue precursor cells in a medium comprising a TGF-beta inhibitor, preferably SB 431542, a bone morphogenic protein, preferably BMP4 and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM; and/or

wherein one of the at least two different heart tissues is atrioventricular node tissue and generating atrioventricular node tissue comprises differentiating mesoderm cells into atrioventricular canal tissue precursor cells in a medium comprising a TGF-beta inhibitor, preferably SB 431542, a Wnt inhibitor, preferably Wnt-C59 or XAV-939, a bone morphogenic protein, preferably BMP4 and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM; and wherein atrioventricular canal progenitor posterior second heart field tissue is further differentiated into atrioventricular node tissue by further maturing in a medium containing an activator of sonic hedgehog signaling and/or a BMP.

10. The method of any one of claims 7 to 9, wherein fusing the at least two heart tissues comprises culturing in a medium comprising a Wnt inhibitor, preferably Wnt-C59, a bone morphogenic protein, preferably BMP4, a fibroblast growth factor, preferably FGF2, insulin, and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM, especially preferred 500 nM.

11. The method of any one of claims 7 to 10 for screening or testing a candidate compound on its effects on heart development and/or functionality comprising generating a cardiac organoid according to any one of claims 7 to 10 while treating the cells with the candidate compound and comparing development of the

cardiac organoid with development and/or or functionality of a cardiac organoid that was not treated with the candidate compound.

12. A method of observing the effects of suppressed, mutated or overexpressed genes during on heart development comprising generating a cardiac organoid according to any one of claims 7 to 10 wherein the cells have a supressed or mutated candidate gene or overexpress a candidate gene and comparing development of the cardiac organoid with development of a cardiac organoid that was not generated with a supressed, mutated or overexpressed gene.

13. A method of screening or testing a candidate compound on its effects on heart functionality comprising treating a cardiac organoid according to any one of 1 to 6 with the candidate compound and comparing with a functionality of a cardiac organoid that was not treated with the candidate compound.

14. Use of a cell, preferably cardiomyocyte, from a cardiac organoid of any one of claims 1 to 6, in the manufacture of a medicament for treating a heart injury in a patient, wherein the cell is to be transplanted to the patient.

15. Use of a cell culture medium comprising
a) a bone morphogenic protein, preferably BMP4, a fibroblast growth factor, preferably FGF2, insulin, a Wnt inhibitor, preferably Wnt-C59 or XAV-939, and retinoic acid, with the retinoic acid having a concentration of less than 100 nM;
b) a TGF-beta inhibitor, preferably SB 431542, and a Wnt inhibitor, preferably Wnt-C59 or XAV-939;
c) a Wnt inhibitor, preferably Wnt-C59 or XAV-939, a bone morphogenic protein, preferably BMP4, a fibroblast growth factor, preferably FGF2, insulin, and retinoic acid, preferably with the retinoic acid having a concentration of 50 nM to 500 nM in the medium;
d) a TGF-beta inhibitor, preferably SB 431542, and a Wnt inhibitor, preferably Wnt-C59 or XAV-939;
e) a Wnt inhibitor, preferably Wnt-C59 or XAV-939, a bone morphogenic protein, preferably BMP4, a fibroblast growth factor, preferably FGF2, insulin, and the medium lacking retinoic acid;

f) a TGF-beta inhibitor, preferably SB 431542, a Wnt inhibitor, preferably Wnt-C59 or XAV-939, and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM;
g) a Wnt inhibitor, preferably Wnt-C59 or XAV-939, a bone morphogenic protein, preferably BMP4, a fibroblast growth factor, preferably FGF2, insulin, and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM;
h) a TGF-beta inhibitor, preferably SB 431542, a Wnt inhibitor, preferably Wnt-C59 or XAV-939, a bone morphogenic protein, preferably BMP4 and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM;
i) activin and CHIR99021; preferably the activin is at a concentration of 1 ng/mL to 8 ng/mL, preferably about 5 ng/mL, and/or the CHIR99021 is at a concentration of 1 µM to 6 µM, preferably about 3 µM; or preferably the activin is at a concentration of 30 ng/mL to 100 ng/mL, preferably about 50 ng/mL, and/or the CHIR99021 is at a concentration of 2 µM to 8 µM, preferably about 4 µM; and/or preferably the activin is at a concentration of 6 ng/mL to 30 ng/mL, preferably about 10 ng/mL, and/or the CHIR99021 is at a concentration of 0.4 µM to 4 µM, preferably about 2 µM; and/or preferably the activin is at a concentration of 30 ng/mL to 100 ng/mL, preferably about 50 ng/mL, and/or the CHIR99021 is at a concentration of 0.1 µM to 4 µM, preferably about 1 µM; or
j) a Wnt inhibitor, preferably Wnt-C59, a bone morphogenic protein, preferably BMP4, a fibroblast growth factor, preferably FGF2, insulin, and retinoic acid, preferably the retinoic acid is in a concentration of 300 nM to 800 nM, especially preferred 500 nM;
in a method according to any one of claims 7 to 13.

16. A cell, preferably cardiomyocyte, from a cardiac organoid of any one of claims 1 to 6, for use in therapy.