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Other: **EPODOC & WPI; Patent Fulltext**

(54) Title of the Invention: **Cartilage plug**
Abstract Title: **Osteochondral plug**

(57) A joint plug 10 for repairing cartilage damage in a joint (figure 1b) has a bone scaffold portion 12 for accommodating bone tissue therein, an overlying cartilage scaffold portion 14 for accommodating cartilage tissue therein, and preferably a permeable membrane portion 18 provided as a surface layer. The membrane may be of polycaprolactone with a maximum pore size of less than 50 microns. The bone scaffold may be seeded with osteoblasts and the cartilage scaffold seeded with chondrocytes to promote integration and repair. Nutrients reach these cells by diffusing through the permeable membrane. The component portions may be provided as a kit of parts. A barrier layer in the bone scaffold may serve to limit the propagation of cartilaginous cells into the bone scaffold. The cartilage scaffold may be coupled to the bone scaffold via legs 16. A patient-specific joint plug may be manufactured by 3D printing based on scan data.

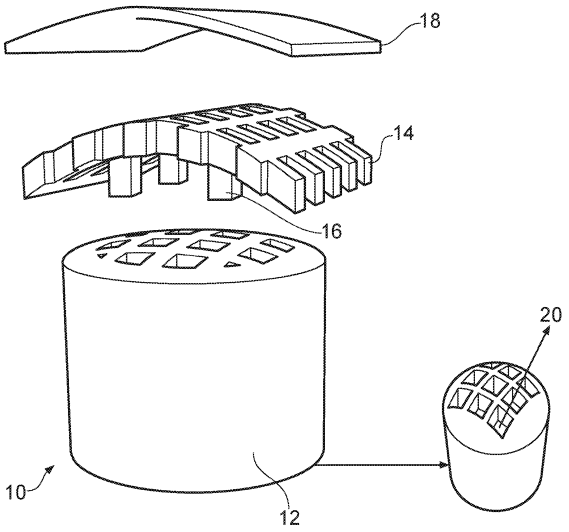

FIG. 1a

GB 2567173 A

At least one drawing originally filed was informal and the print reproduced here is taken from a later filed formal copy.

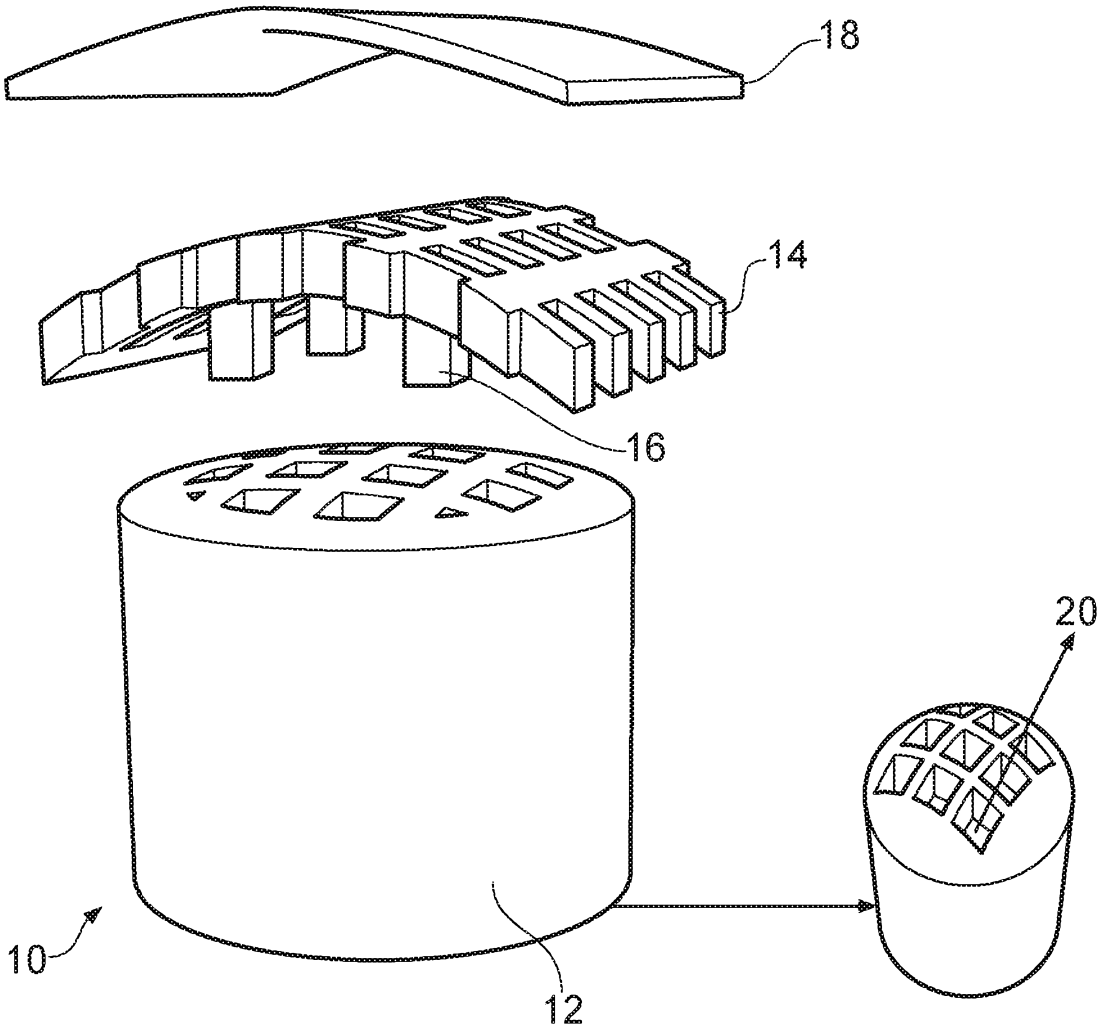

FIG. 1a

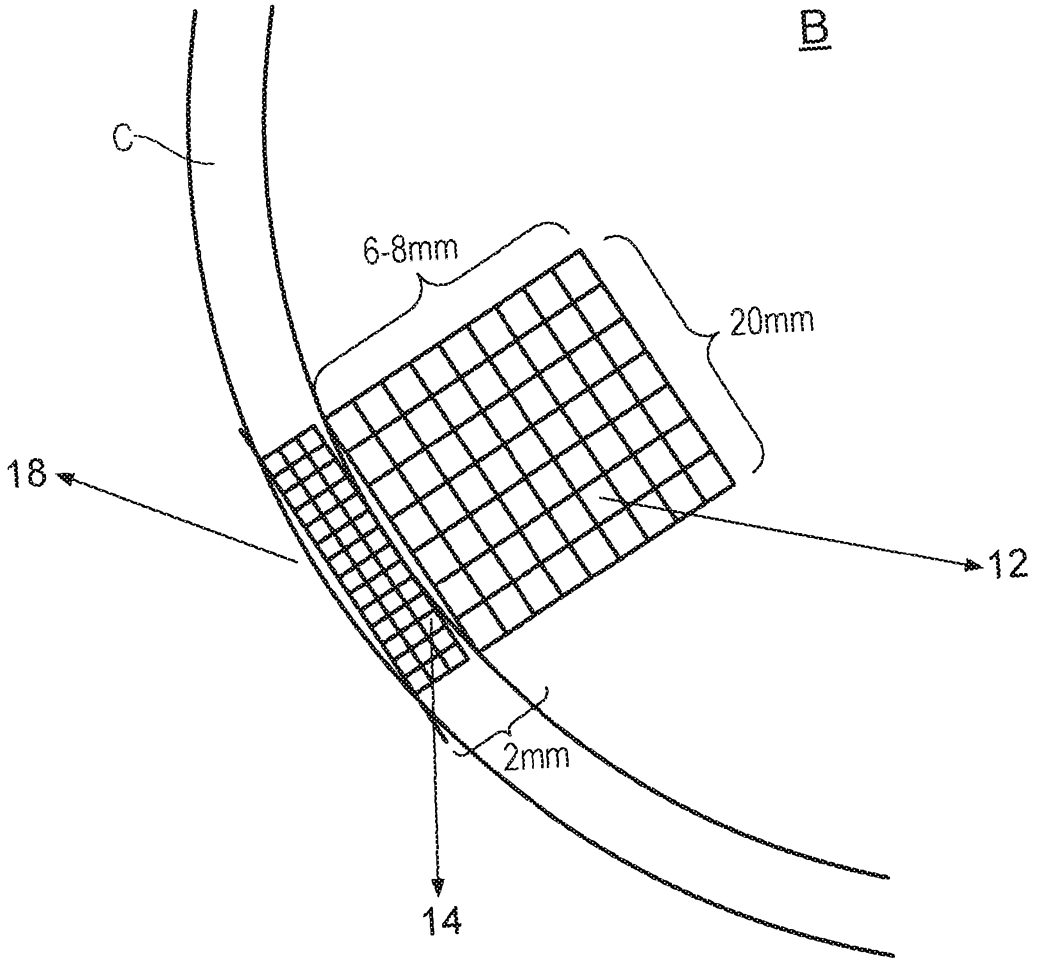

FIG. 1b

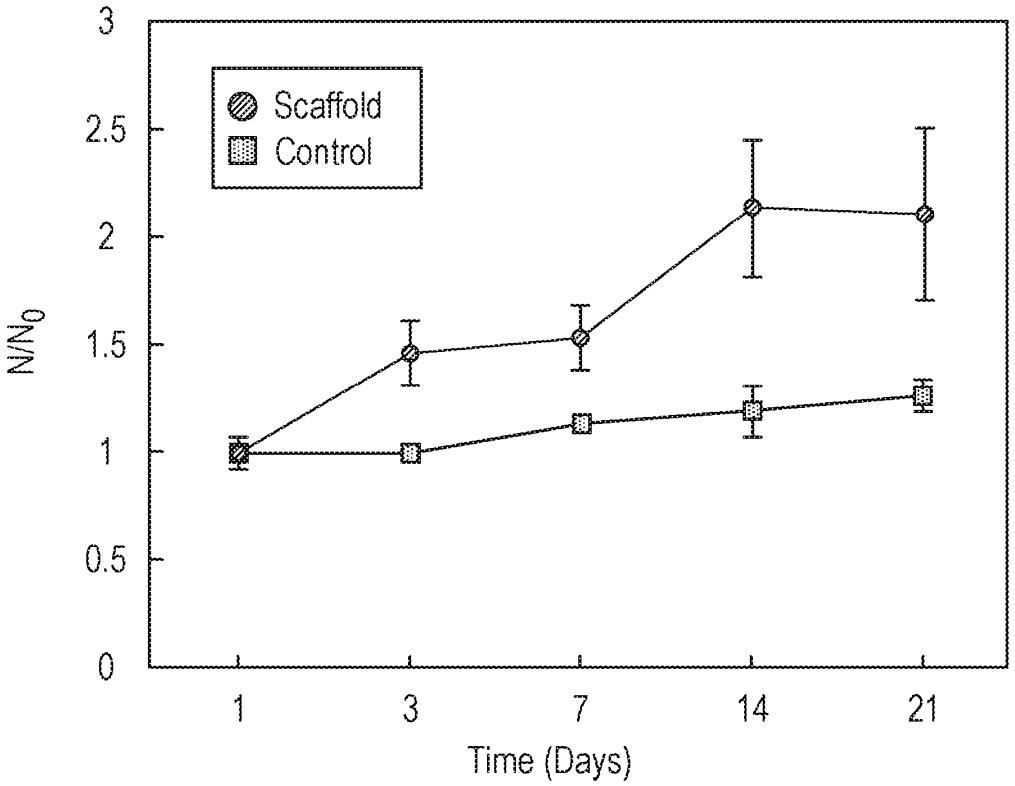

FIG. 2

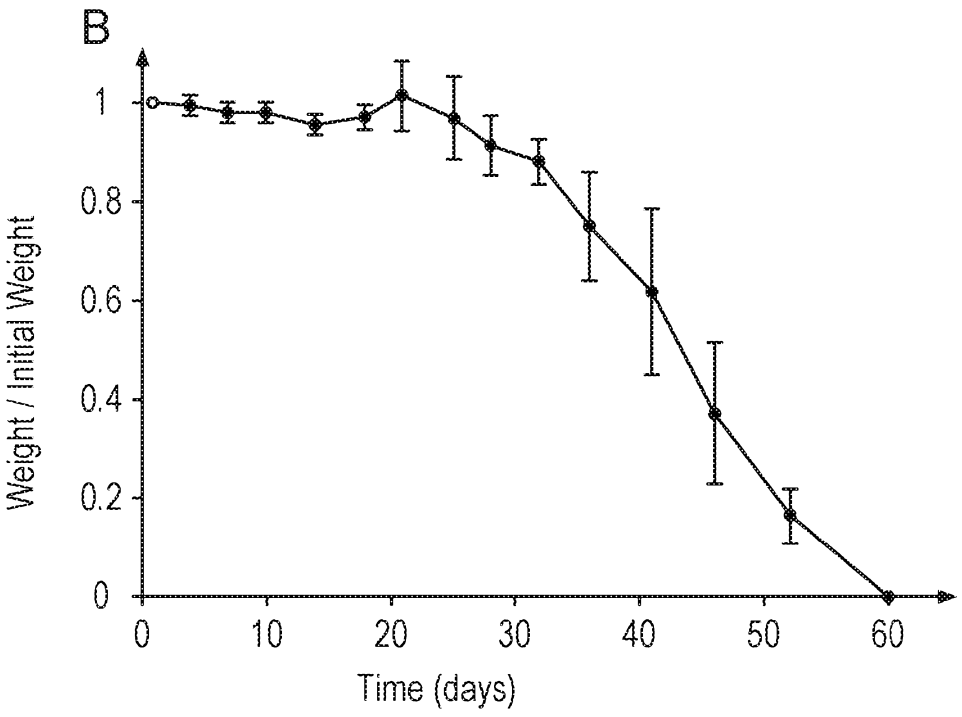

FIG. 3

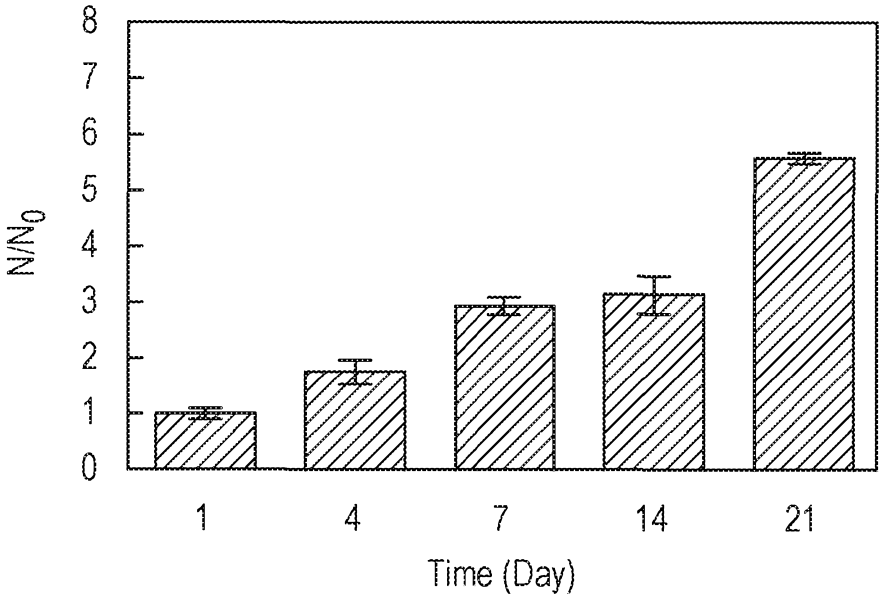

FIG. 4

Cartilage Plug

[0001] This invention relates to a kit of parts for forming a joint plug, for example a knee plug, for use in the treatment of damaged cartilage and an assembled implant for the same.

BACKGROUND

[0002] Cartilage is a thin, elastic tissue that protects the bone of a joint, and allows supple movement of the joint by ensuring that the surfaces within the joint can slide easily over each other. Cartilage tissue lacks a neural network. Accordingly, when cartilage is damaged and partly removed, the joint can become painful as the neural network of the underlying bone becomes exposed. In addition, cartilage has very limited capacity for self-restoration.

[0003] To treat cartilage damage in a joint, surgeons may remove damaged cartilage and underlying bone tissue from the damaged joint, and stimulate growth of tissue in the damaged area by cell therapy or other methods. For example, surgeons may take bone from other parts of the patient's body (autograft) and apply the autograft to the damaged area of the joint (e.g. knee). They may also use allografts. However, using autograft bone can cause morbidity of other parts of the body, while allografts can sometimes transfer disease.

BRIEF DESCRIPTION OF THE DRAWINGS

[0004] Embodiments of the invention are further described below, by way of example, with reference to the accompanying drawings, in which:

[0005] Figure 1a provides a schematic view of the kit according to an embodiment of the present invention;

[0006] Figure 1b provides a schematic view of the kit of Figure 1a assembled as a joint plug and located within a bone layer and cartilage layer of a damaged knee joint;

[0007] Figure 2 is a graph showing the growth of osteoblasts on a bone scaffold portion of a kit according to an embodiment of the present invention;

[0008] Figure 3 is a graph showing the rate of degradation of a cartilage scaffold portion of a kit according to an embodiment of the present invention; and

[0009] Figure 4 is a graph showing the proliferation rate of chondrocytes on a cartilage scaffold portion of a kit according to an embodiment of the present invention.

DESCRIPTION

[0010] In accordance with an aspect of the present invention, there is provided a kit of parts for assembly into a joint plug. The joint plug is for insertion within an opening of a bone layer in a joint and through a cartilage layer on the bone layer. The kit of parts comprises a bone scaffold portion for accommodating bone tissue therein. The bone scaffold portion is configured for insertion into the opening of the bone layer. The kit of parts further comprises a cartilage scaffold portion for accommodating cartilage tissue therein. The cartilage scaffold portion is configured to be positioned over the bone scaffold portion. The kit of parts may further comprise a permeable membrane portion configured to be provided over the cartilage scaffold portion.

[0011] Viewed from another aspect, there is provided a joint plug for insertion within an opening of a bone layer in a joint and through a cartilage layer on the bone layer. The joint plug is for treatment of damaged cartilage. The joint plug comprises a bone scaffold for accommodating bone tissue therein, said bone scaffold portion configured for insertion into the opening of the bone layer. The joint plug further comprises a cartilage scaffold portion for accommodating cartilage tissue therein, said cartilage scaffold portion being configured to be positioned over the bone scaffold portion. The joint plug further comprises a permeable membrane portion configured to be provided over the cartilage scaffold portion.

[0012] The joint plug may comprise any of the features of the kit of parts described herein.

[0013] The kit of parts can be assembled to form a joint plug for the treatment of damaged cartilage in a joint. By providing osteoblasts in the bone scaffold portion, bone tissue can grow in the bone scaffold portion when the joint plug is inserted within the opening of the bone layer. The bone scaffold may be configured to accommodate osteoblasts as they propagate to form bone tissue. For example, the bone scaffold portion may be provided with openings or pores for accommodating osteoblasts. Accordingly, once positioned within the bone layer, the scaffold can facilitate growth of bone tissue within the bone layer. Thus, bone tissue can grow into the bone layer of the joint, so as to secure the joint plug in place.

[0014] The cartilage scaffold portion is configured to accommodate cartilage tissue. For example, the cartilage scaffold portion may comprise openings or pores for accommodating chondrocytes. Thus, when positioned over the bone scaffold portion, chondrocytes present in the cartilage scaffold portion can propagate and form a cartilage layer over the bone. The cartilage scaffold portion may facilitate cartilage growth within the cartilage layer and retain chondrocytes within the layer, improving cell differentiation between the bone layer and the cartilage layer.

[0015] The permeable membrane portion may be positioned over the cartilage scaffold portion to help to retain cartilage tissue within the cartilage layer, while allowing nutrients to diffuse through the membrane portion into the cartilage scaffold portion to facilitate cell propagation and growth.

[0016] The kit of parts of the present disclosure may be assembled into a joint plug that can be used for joint repair. Advantageously, the joint plug can be used to support bone tissue growth and cartilage growth, while maintaining good cell differentiation between the bone and cartilage layers. In some embodiments, the kit of parts may also simplify the surgical procedure employed for joint repair, as the bone scaffold portion and the cartilage scaffold portion may be seeded with cells *ex-vivo* and the seeded portions inserted into the joint, for example, together as a unit. In some embodiments, the permeable membrane portion may be placed over the cartilage scaffold portion before insertion, again potentially simplifying the surgical procedure as the assembled plug may be inserted into the joint.

[0017] The joint plug may be a knee plug. In alternative embodiments, the joint plug may be for use at the elbow, ankle, hips, wrists or other joints of the body. Preferably, the joint plug is a knee plug or ankle plug.

[0018] There is also provided a bone scaffold portion for the kit of parts for assembly into the joint plug as described herein, the bone scaffold portion for accommodating bone tissue therein and for having a cartilage scaffold portion of the joint plug provided thereon.

[0019] There is further provided a cartilage scaffold portion for the kit of parts for assembly into the joint plug as described hereinbefore, the cartilage scaffold portion for accommodating cartilage tissue therein and configured to be provided on a bone scaffold portion of the joint plug.

[0020] Viewed from another aspect, there is also provided a method of forming the bone scaffold portion for the kit of parts for assembly into the joint plug. The method comprises receiving sensor data from a sensor device. The sensor data is indicative of the shape of the opening of the bone layer into which the bone scaffold portion will be inserted. The method further comprises determining a bone scaffold portion shape based on the obtained sensor data, and forming the bone scaffold portion having the bone scaffold portion shape using a three-dimensional (3D) printing device.

[0021] Thus, bone scaffold portions for joint plugs can be manufactured to be personalised and accurate for the precise shape of a joint, and in particular an opening in the bone layer of the joint. During the treatment of a joint (e.g. a knee), an area of the joint is typically surgically removed to form an opening in the bone layer. The kit of parts in accordance with the present invention may be formed with a controlled structure. In

particular, the bone scaffold layer and/or the cartilage scaffold layer may be formed using 3D printing, such that they correspond to the size of the opening in the bone layer in which they are to be inserted. Thus, the present invention allows for personalised treatment of an individual joint, which can allow for more effective treatment of the damaged joint and allow for easier insertion into the joint.

[0022] 3D printing also provides controllability of the size of the pores within the bone scaffold layer and/or the cartilage scaffold layer. This is important for the growth of new tissues such as bone tissue and cartilage tissue. Specifically, the pore size of the bone scaffold layer may be tailored to accommodate osteoblasts and other cells for bone tissue growth. The pore size of the cartilage scaffold layer may be tailored to support chondrocytes.

[0023] The method may further comprise seeding the bone scaffold portion with bone cells.

[0024] The method may further comprise determining a cartilage scaffold portion shape based on the obtained sensor data, and forming the cartilage scaffold portion having the cartilage scaffold portion shape using the or a further three-dimensional (3D) printing device.

[0025] Viewed from another aspect, there is provided a method of preparing a joint plug for insertion into an opening in a bone layer of a body. The joint plug is for treatment of a damaged cartilage layer covering the bone layer. The join plug comprises a bone scaffold portion for accommodating bone tissue therein, said bone scaffold portion being configured for insertion into the opening of the bone layer. The joint plug further comprises a cartilage scaffold portion for accommodating cartilage tissue therein, said cartilage scaffold portion being configured to be positioned over the bone scaffold portion. The joint plug yet further comprises a permeable membrane portion configured to be provided over the cartilage scaffold portion. The method comprises: seeding the bone scaffold portion with bone tissue; seeding the cartilage scaffold portion with cartilage tissue; and assembling the joint plug by providing the cartilage scaffold portion over the bone scaffold portion and providing the permeable membrane portion over the cartilage scaffold portion.

[0026] Thus, the cartilage scaffold portion is provided between the bone scaffold portion and the permeable membrane portion in the assembled joint plug, which is also a seeded joint plug and ready for insertion into the body for treatment of damaged cartilage in or around a joint.

[0027] The joint plug may be assembled after seeding of the bone scaffold portion with bone tissue. Alternatively, the joint plug may be assembled before seeding of the bone

scaffold portion with bone tissue. In examples, the bone scaffold portion may be seeded with bone tissue as part of assembly of the joint plug.

[0028] The joint plug may be assembled after seeding of the cartilage scaffold portion with cartilage tissue. Alternatively, the joint plug may be assembled before seeding of the cartilage scaffold portion with cartilage tissue. In examples, the cartilage scaffold portion may be seeded with cartilage tissue as part of assembly of the joint plug.

[0029] The kit of parts may further comprise bone scaffold engagement means for mounting the cartilage scaffold portion on the bone scaffold portion. Thus, the cartilage scaffold portion may be secured to the bone scaffold portion. This can restrict the position of the bone scaffold portion relative to the cartilage scaffold portion, which, in turn, can facilitate cell differentiation, as bone tissue growth and cartilage growth may be substantially constrained in their respective parts. Engagement between the cartilage scaffold portion and the bone scaffold portion may also allow the two portions to be coupled together prior to insertion into the joint. This may simplify the surgical procedure.

[0030] The kit of parts may further comprise membrane portion engagement means for mounting the permeable membrane portion on the cartilage scaffold portion. Thus, the permeable membrane portion may be secured to the cartilage scaffold portion. The juxtaposition of the membrane portion relative to the cartilage scaffold portion can help to prevent migration of chondrocytes away from the cartilage layer, keeping the chondrocytes in place within the cartilage scaffold portion. The membrane may also facilitate sliding of the joint, for example, while the cartilage layer is still undergoing repair. Engagement between the membrane portion and the cartilage scaffold portion may also allow the two portions to be coupled together prior to insertion into the joint. This may, in some circumstances, simplify the surgical procedure, particularly if the bone scaffold portion is also coupled to the cartilage layer as the assembled plug may be inserted as an integral unit.

[0031] The kit of parts may further comprise a barrier portion to be provided in the bone scaffold portion to substantially prevent passage of cartilage tissue from the cartilage scaffold portion beyond the barrier portion within the bone scaffold portion. The barrier portion may comprise pores that are sized to prevent the passage of cartilage tissue or chondrocytes form the cartilage layer into the bone scaffold portion. In one embodiment, the barrier portion is devoid of pores. The barrier portion may be integral with the bone scaffold portion or may be a separate layer.

[0032] The barrier portion may be configured to be provided less than 500 micrometres from an interface between the cartilage scaffold portion and the bone scaffold portion.

[0033] The barrier portion may be provided by a portion of the bone scaffold portion. The barrier portion may be provided by a layer of the bone scaffold portion.

Bone scaffold portion

[0034] The bone scaffold portion may be formed from a biodegradable material. The biodegradable material may be configured to degrade in less than 80 days, or less than 65 days, for example in less than 50 days. Alternatively, the bone scaffold may be formed from a suitable material that is not degradable.

[0035] The bone scaffold portion may be formed from a ceramic or composite material. In one embodiment, the bone scaffold portion may be formed from at least one of the following materials: bioactive glasses, biodegradable magnesium alloys, biocompatible ceramics, biodegradable synthetic polymers, and biodegradable natural polymers. Examples of biocompatible ceramics include β-tri-calcium phosphate, tricalcium phosphate, aluminium oxide, calcium oxide, tribasic calcium phosphate, zirconium oxide, bioactive hydroxyapatite, and pyrolitic carbon ((LTI)Pyrolitic carbon). The bone scaffold portion may be in the form of a composite.

[0036] Examples of biodegrable synthetic polymers include poly(α-esters), polyurethanes, poly(ester amides), poly(ortho esters), poly(ortho esters), polyanhydrides, poly(anhydride-co-imide), cross-linked polyanhydrides, poly(propylene fumarate), pseudo poly(amino acid), poly(alkyl cyanoacrylates), polyphosphazenes and polyphosphoesters. Examples of poly(α-esters) include polyglycolide, polylactides, poly(lactide-co-glycolide), polydioxanone, polycaprolactone, poly(trimethylene carbonate) and bacterial polyesters. In another embodiment, the porous film may be formed from a synthetic material selected from proteins and poly(amino acids), and polysaccharides. Examples of proteins and poly(amino acids) include collagen, poly(amino acids), elastin, elastin-like peptides, albumin and fibrin. The polysaccharide may be of human origin or of non-human origin.

[0037] In a preferred embodiment, the bone scaffold portion may be formed from a mixture of tricalcium phosphate (TCP) and hydroxyapatite (HA). Preferably, the tricalcium phosphate is sintered tricalcium phosphate. The ratio of TCP to HA may be 2:1 to 10:1, preferably, 3:1 to 6:1, for example 4:1 to 5:1. The mixture of TCP and HA may additionally include, for example, sodium tripolyphosphate and /or carboxymethylcellulose.

[0038] The bone scaffold portion can be formed from a paste material suitable to be formed by a 3D printing process, which enables direct matching of the shape of the bone scaffold portion to the opening in the bone layer. Accordingly, the present disclosure may also provide a 3D printing ink composition comprising tricalcium phosphate (TCP), hydroxyapatite (HA) and a liquid carrier. The ratio of TCP to HA may be 2:1 to 10:1,

preferably, 3:1 to 6:1, for example 4:1 to 5:1. The ink may additionally include, for example, sodium tripolyphosphate and /or carboxymethylcellulose.

[0039] The bone scaffold portion comprises bone tissue accommodation means to accommodate bone tissue therein. The bone scaffold portion may comprise pores having a large pore size. In one embodiment, the pores have a diameter of between 300 μm – 1mm, preferably between 400 – 800 μm, for example between 500 – 700 μm. The pores may be regularly shaped. For example, the pores may define chambers or tunnels extending through at least a portion of the bone scaffold portion. The bone scaffold portion may take the form of a matrix. In one embodiment, the bone scaffold portion takes the form of a matrix comprising a plurality of pores. The pores may be regularly spaced and, in some embodiments, take the form of regularly spaced chambers or tunnels extending through at least part of the bone scaffold portion.

[0040] The bone scaffold portion may be configured to support the cartilage scaffold portion thereon. In some embodiments, the bone scaffold portion may include engagement means for mounting the cartilage scaffold portion on the bone scaffold portion. The bone scaffold engagement means may comprise one or more protrusions for mechanical engagement between the bone scaffold portion and the cartilage scaffold portion when the kit of parts is assembled into the joint plug. The one or more protrusions may be one or more legs extending between the bone scaffold portion and the cartilage scaffold portion when the kit of parts is assembled into the joint plug. The one or more legs may extend from the cartilage scaffold portion within the bone scaffold portion when the kit of parts is assembled into the joint plug to mount the cartilage scaffold portion to the bone scaffold portion. The one or more protrusions may be a plurality of protrusions.

[0041] Alternatively, the bone scaffold portion may be coupled to the cartilage scaffold portion by other means, for example, by an adhesive, stitching or stapling.

[0042] Alternatively, the bone scaffold portion may be in the form of granules or a powder matrix that supports bone tissue growth.

Cartilage scaffold portion

[0043] The cartilage scaffold portion may be formed from a biodegradable material. The biodegradable material may be configured to degrade in less than 80 days, or less than 65 days, for example in less than 50 days. Alternatively, the cartilage scaffold may be formed from a suitable material that is not degradable.

[0044] The cartilage scaffold portion may be formed from a hydrogel, biodegradable polymer, or a composite material. In one embodiment, the cartilage scaffold portion may be formed from a synthetic material selected from poly(α-esters), polyurethanes, poly(ester

amides), poly(ortho esters), poly(ortho esters), polyanhydrides, poly(anhydride-co-imide), cross-linked polyanhydrides, poly(propylene fumarate), pseudo poly(amino acid), poly(alkyl cyanoacrylates), polyphosphazenes and polyphosphoesters. Examples of poly(α-esters) include polyglycolide, polylactides, poly(lactide-co-glycolide), polydioxanone, polycaprolactone, poly(trimethylene carbonate) and bacterial polyesters. In another embodiment, the cartilage scaffold portion may be formed from a synthetic material selected from proteins and poly(amino acids), and polysaccharides. Examples of proteins and poly(amino acids) include collagen, poly(amino acids), elastin, elastin-like peptides, albumin and fibrin. The polysaccharide may be of human origin or of non-human origin.

[0045] In one embodiment, the cartilage scaffold portion comprises at least one of gelatin, elastin and sodium hyaluronate. In one embodiment, the cartilage scaffold portion comprises gelatin. In another embodiment, the cartilage scaffold portion comprises gelatin and elastin. In a yet another embodiment, the cartilage scaffold portion comprises gelatin, elastin and sodium hyaluronate. The ratio of gelatin to elastin may be 1:1 to 10:1, preferably 2:1 to 8:1, for example, 3:1 to 4:1. The ratio of gelatin to sodium hyaluronate may be 10:1 to 30:1, for example 12:1 to 20:1.

[0046] As well as having the appropriate mechanical and biocompatible properties for supporting cartilage tissue growth in a joint, it may, in certain embodiments, be desirable for the material to be printable e.g. by 3D printing.

[0047] In one embodiment, the cartilage scaffold portion may be in the form of a composite comprising two or more of said synthetic or natural materials.

[0048] Any suitable method may be used to attach the cartilage scaffold portion to the bone scaffold portion and/or the membrane portion. The cartilage scaffold portion may comprise one or more legs or protrusions that extend from the cartilage scaffold portion to mechanically interlock with the bond scaffold portion. In another embodiment, the cartilage scaffold portion may be suturable or stitchable to at least one of the bone scaffold portion and the membrane portion. In another embodiment, chemical methods may be used to attach the cartilage scaffold portion to the bone scaffold portion and/or the membrane portion. Alternatively, the cartilage scaffold portion is positioned on top of the bone scaffold portion.

[0049] The cartilage scaffold portion comprises cartilage tissue accommodation means to accommodate cartilage tissue therein. The cartilage tissue accommodation means may comprise openings or pores. The openings may have a diameter of between 300 – 600 μm, preferably 400 – 500 μm. The cartilage tissue accommodation means may be seeded with chondrocytes. The cartilage tissue accommodation means may be seeded with chondrocytes prior to assembly of the joint plug. The pore size and pore profile of the

cartilage scaffold portion may be tailored to accommodate chondrocytes and cartilage growth. In some embodiments, the pore size adjacent the bone scaffold portion may be tailored so as to reduce the risk of chondrocytes falling into the bone scaffold portion. For instance, the pore size at the interface with the bone scaffold portion may be smaller than the pore size in the remainder of the cartilage scaffold portion.

[0050] In one embodiment, the cartilage scaffold portion is formed as a substrate having one or more openings defined therein. The cartilage tissue accommodation means is provided by a plurality of openings defined within the substrate. The substrate can be substantially planar. The substrate may be a single layer or multiple layer substrate. The substrate may be irregular or regular in form. The substrate may include through-voids and/or internal voids.

[0051] In one embodiment, the cartilage scaffold portion takes the form of a lattice, wherein the lattice has a regularly repeating shape. The lattice may have openings provided at regular intervals.

[0052] The growth of the cartilage tissue accommodated within the cartilage scaffold portion can be encouraged or improved by nutrients provided through the permeable membrane portion.

[0053] The cartilage scaffold portion may be for accommodating chondrocytes therein. In this way, it will be understood that chondrocytes are to be understood as a form of cartilage tissue. Indeed, the term cartilage tissue should be understood to mean any biological material naturally present in cartilage in the body, or which results in the formation of cartilage.

[0054] The kit of parts may further comprise a source of chondrocytes. The source of chondrocytes may be provided in the cartilage scaffold portion.

[0055] Permeable membrane portion

[0056] The membrane portion is configured to be provided over the cartilage scaffold portion. The membrane portion may comprise a porous film. Thus, the permeable membrane portion may be permeable due to a plurality of through-holes defined within the permeable membrane portion. Preferably, the membrane portion comprises small pores for nutrient and gas delivery, for example for delivery to the propagating cells within the cartilage scaffold portion and/or bone scaffold portion. The maximum pore diameter may be less than 100 μm. Thus, the passage of cartilage tissue from the cartilage scaffold portion out of the joint plug through the permeable membrane portion can be substantially reduced or prevented. A maximum pore size of the porous film may be less than 50 micrometres or less than 25 μm. In one embodiment, the membrane portion comprises

pores having a diameter of between 5-25 μm. In a preferred embodiment, the pore diameter may be less than 15 micrometres, for example, the pore size may be about 10 μm. As mentioned above, he membrane portion allows retention of the growing cartilage within the cartilage layer.

[0057] Preferably, the membrane portion is suturable or stitchable. For example, the membrane portion may be stitched to the surrounding joint tissue. In a further example, the membrane portion may be sutured or stitched to the cartilage scaffold portion.

[0058] The membrane portion may be formed from a hydrogel, biodegradable polymer, or a composite material.

[0059] The permeable membrane portion may be formed from a biodegradable material. Thus, the permeable membrane portion may be arranged to dissolve or otherwise degrade in the body in time. In examples, the permeable membrane portion may be formed to degrade naturally in the body within less than 80 days.

[0060] In one embodiment, the membrane portion may be formed from a synthetic material selected from poly(α-esters), polyurethanes, poly(ester amides), poly(ortho esters), poly(ortho esters), polyanhydrides, poly(anhydride-co-imide), cross-linked polyanhydrides, poly(propylene fumarate), pseudo poly(amino acid), poly(alkyl cyanoacrylates), polyphosphazenes and polyphosphoesters. Examples of poly(α-esters) include polyglycolide, polylactides, poly(lactide-co-glycolide), polydioxanone, polycaprolactone, poly(trimethylene carbonate) and bacterial polyesters. In another embodiment, the porous film may be formed from a synthetic material selected from proteins and poly(amino acids), and polysaccharides. Examples of proteins, poly(amino acids) and polysaccharides include polyvinyl alcohol, collagen, poly(amino acids), gelatin, alginate, elastin, elastin-like peptides, albumin and fibrin. The polysaccharide may be of human origin or of non-human origin.

[0061] In one embodiment, the membrane portion may be in the form of a composite comprising two or more of said synthetic or natural materials.

[0062] In a preferred embodiment, the membrane portion comprises polycaprolactone. Thus, the permeable membrane portion may be biocompatible, mechanically strong, porous and substantially degradable.

[0063] The membrane portion may comprise membrane portion engagement means for mounting the membrane portion on the cartilage portion. The membrane portion may be provided with an inner surface for facing the cartilage scaffold portion when the membrane portion is provided on the cartilage scaffold portion, and wherein the inner surface of the membrane portion is hydrophilic. The membrane portion may be provided with an outer

surface for facing away from the cartilage scaffold portion when the membrane portion is provided on the cartilage scaffold portion, wherein the outer surface of the membrane portion is hydrophobic. Thus, the cartilage tissue is substantially prevented from passing from the cartilage scaffold portion out of the joint plug through the permeable membrane portion.

[0064] The hydrophobicity of the membrane's surface may be altered by surface treatment. For example, in the case of a polycaprolactone film, the hydrophobicity may be controlled by surface treatment with a hydroxide, for example, sodium hydroxide.

Barrier Portion

[0065] The kit of parts may further comprise a barrier portion. The barrier portion may be provided within the bone scaffold portion and may be integral with it. The barrier portion may be provided at a distance of less than 500 μm microns from the top of the bone portion, preferably less than 250 μm, for example less than 100 μm from the top of the bone portion. In one embodiment, the barrier portion is provided by a layer of the bone scaffold portion. The barrier portion may substantially prevent passage of cartilage tissue from the cartilage scaffold portion beyond the barrier portion within the bone scaffold portion. Some chondrocytes and osteoblasts can still interact at the interface between the bone portion and scaffold portion.

[0066] Alternatively, the pore size of the pores within the bone scaffold portion may be smaller at the top of the bone scaffold portion, i.e. in the part of the bone scaffold portion that is nearest to the cartilage scaffold portion. In one embodiment, the pore size may be reduced in the region of less than 500 μm microns from the top of the bone portion, preferably less than 250 μm, for example less than 100 μm from the top of the bone portion. In one embodiment, the pore size is reduced within at least one layer of the bone scaffold portion. The maximum diameter of the pores within the at least one layer may be less than 100 μm, or less than 50 μm, or less than 25 μm. In one embodiment, the pore diameter is substantially zero.

Preparation Methods

[0067] Each of the bone scaffold portion, cartilage scaffold portion and porous membrane may be formed by any suitable method. Examples of suitable methods include injection molding, freeze drying, solvent casting, particulate leaching, gas foaming, porogen leaching, self-assembly methods, phase separation, 3D printing (for example rapid prototyping), melt molding, fiber bonding, fiber mesh, membrane lamination, and molding.

[0068] In a preferred embodiment, the bone scaffold portion is formed using a 3D printing method. The method for forming the bond scaffold portion using 3D printing may comprise a first step of receiving sensor data from a sensor device, the sensor data being indicative of the shape of the opening of the bone layer into which the bond scaffold portion will be inserted. The shape of the bone scaffold portion may then be determined based on the obtained sensor data. A bone scaffold portion having the shape determined using the obtained sensor data may then be formed using a 3D printing device.

[0069] In a preferred embodiment, the cartilage portion is formed by a 3D printing method. The cartilage portion may be formed by the same 3D printing method described for forming the bone scaffold portion.

[0070] In a preferred embodiment, the porous membrane is formed using a moulding method.

[0071] Aspects of the present disclosure will now be described by way of example with reference to Figures 1a and 1b. Figure 1a provides a schematic view of the kit according to an embodiment of the present invention. Figure 1b provides a schematic view of the kit of Figure 1a assembled as a joint plug and located within a bone layer and cartilage layer of a damaged knee joint.

[0072] Figure 1a shows a kit 10 of parts for assembly into a joint plug. The kit 10 comprises a bone scaffold portion 12 for accommodating bone tissue therein. The bone scaffold portion 12 is configured for insertion into an opening of the bone layer. The bone scaffold portion 12 comprises openings for accommodating osteoblasts and for growth of bone tissue. The openings may be regularly spaced and may define tunnels or longitudinal chambers that extend through the length of the bone scaffold portion 12.

[0073] The kit 10 further comprises a cartilage scaffold portion 14 for accommodating cartilage tissue therein. The cartilage scaffold portion 14 takes the form of a lattice having openings for accommodating chondrocytes and supporting the growth of cartilage tissue. The cartilage scaffold portion 14 comprises protrusions 16 for securement of the cartilage scaffold portion 14 to the bone scaffold portion 12. The protrusions 16 can be received within openings in the bone scaffold portion 12, thus securing the cartilage scaffold portion 14 to the bone scaffold portion 12.

[0074] The kit 10 of parts further comprises a permeable membrane portion 18 configured to be provided over the cartilage scaffold portion. The kit 10 of parts may further comprise optional barrier layer 20.

[0075] Figure 1b shows the kit 10 of Figure 1a mounted within a joint. As shown in the Figure, the bone scaffold portion 12 is received within a bone layer (B), while the cartilage

scaffold portion 14 is received within a cartilage layer (C). The permeable membrane portion 18 is positioned over the cartilage scaffold portion 14 and permits the flow of nutrients to the underlying cells in the cartilage and bone tissue.

Examples

Materials

[0076] Tricalcium phosphate (TCP), hydroxyapatite (HA), polycaprolactone (PCL, Mn 80000), polyethylene glycol (PEG, Mn 2000) and gelatin (Type A, from porcine skin) were purchased from Sigma (USA). Carboxymethylcellulose (CMC), sodium tripolyphosphate (TPP), 1-Ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC) and N-Hydroxysuccinimide (NHS) were purchased from Alfa Aesar (USA). Elastin with a molecular weight of 60 KDa (Elastin-Soluble, No. ES12) was purchased from Elastin Products Company, Inc. Sodium Hyaluronate (Research Grade, 500-749KDa) was obtained from Lifecore Biomedical. All the solvents were of reagent grade.

3D-printing of the bone and cartilage parts

[0077] A printable paste composed of TCP, HA, CMC and TPP was used to print the bone portion. To prepare the ink, 12 g TCP, 3 g HA, 0.5 g TPP and 0.075 g CMC were added to 5.75 ml water and homogenized at 2000 rpm for 2 min using a centrifugal mixer (Thinky, USA). Prior to printing, the viscosity and applied stress was measured as a function of shear rate at different temperatures. For this purpose, a shear rheometer (Kinexus, Malvern, UK) with a stainless steel parallel-plate geometry of 20 mm in diameter and a Peltier temperature control was used. The viscometry of the samples was conducted, applying a gap distance of 0.5 mm at temperatures in decreasing 2°C increments, ranging from 28°C to 20°C. The shear rate varied logarithmically in ramp mode from 0 to 50 s^{-1} and then back to 0 s^{-1}. Finally, the ink was loaded into standard Nordson cartridges and printed using the parameters presented in Table 1.

[0078] *Table 1: Applied parameters to 3D-print the bone part of the plug.*

Parameter	Value
Dimensions	20 × 20 × 7.5 mm with a solid rod in the middle (D = 4 mm)
Cartridge Temperature	22°C
Platform Temperature	22°C
Pressure	2 bars
Speed	4 mm/s

Distance between strands	2 mm
Slicing	300 µm
Nozzle diameter	410 µm

[0079] The resulted constructs were allowed to air-dry overnight before being transferred to a furnace. The samples were heated up to 600°C with a rate of 3°C/min and held at this temperature for 2 h to eliminate the organic additives. The temperature was then raised to 1100°C at a rate of 5°C/min and kept at this temperature for 4 h to ensure complete sintering of the ceramic scaffolds.

[0080] To print the cartilage portion, a solution of gelatin/elastin/sodium hyaluronate in DI water was prepared and used as the ink. The final concentration of gelatin, elastin and sodium hyaluronate was adjusted to 8%, 2% and 0.5% w/v, respectively. The ink was loaded into standard Nordson cartridges and printed using the parameters listed in Table 2.

[0081] *Table 2: Applied parameters to 3D-print the cartilage part of the plug.*

Parameter	Value
Dimensions	20 × 20 × 2 mm
Cartridge Temperature	31°C
Platform Temperature	12°C
Pressure	2 bars
Speed	15 mm/s
Distance between strands	1.5 mm
Total number of layers	8
Nozzle diameter	410 µm

[0082] The printed constructs were then cross-linked using a solution of 6 mg/ml EDC and 0.75 mg/ml NHS in 70% v/v ethanol for 4 h. To remove the residual cross-linker, the constructs were washed carefully through soaking in a large amount of DI water. The prepared constructs were stored in pure ethanol inside a -20°C freezer to be used after rehydration, when needed.

Fabrication and surface treatment of barrier film

[0083] In order to be used as a physical barrier, porous PCL films were prepared using the combination of film casting and sacrificial material leaching methods. Briefly, 1 g PCL was dissolved in 15 ml 2,2,2-Trifluoroethanol, and varying amounts of PEG (0, 0.2, 0.4, 1 and 1.5 g) were added to the solution. After complete dissolution, the solution was casted and the solvent was allowed to evaporate overnight. The obtained films were soaked in water to eliminate PEG, whereas the porous films were achieved by sacrificing PEG. In other words, the PEG is dispersed in the polymer and then removed (i.e. sacrificed) to form pores.

[0084] To treat the surface of the film that faced toward the cells, the prepared films were allowed to float on 10% w/v NaOH overnight. The films were finally rinsed with copious amount of water to remove residual NaOH.

Characterization and biological evaluation of the bone part

Mechanical properties

[0085] The mechanical properties of the bone portion were measured using an Electromechanical Precision Universal Tester (AGS-X 5 kN, Shimadzu, Japan). The samples were tested in compression mode using a 5 kN load cell and crosshead speed of 1 mm/min.

Osteoblast attachment and proliferation

[0086] Human osteoblasts (HOB, Cell Applications, USA) were cultured under standard aseptic conditions. The cells were cultured in standard flasks and nourished with Dulbecco Modified Eagle Medium (DMEM) supplemented with 10% v/v fetal bovine serum (FBS), 100 U/mL penicillin, 100 µg/mL streptomycin and 0.25 µg/mL amphotericin every 2 days until a confluency of 90% was reached. The cells were trypsinized using TrypLE (Gibco, US) and sub-cultured. The cells of the third passage were used to seed the scaffolds at a density of 2500 cells/mm^2.

[0087] Cell attachment to the scaffolds was evaluated using scanning electron microscopy (SEM, JEOL JSM6510). At certain time intervals (i.e. 3, 7, 14 and 21 days), the constructs were taken out, washed with PBS and fixed using Karnovsky's fixative (composed of Paraformaldehyde-Glutaraldehyde) for 2 hours. The samples were then fixed using 1% w/v Osmium Tetroxide solution, dehydrated using ascending ethanol series (30, 50, 75, 95 and 100% v/v) and left overnight to air dry at room temperature. After being sputter-coated with gold, the samples were imaged using back-scattering and secondary electron modes at different magnifications.

[0088] The capability of the prepared scaffolds to induce cell proliferation was evaluated using prestoblue assay. At certain time intervals (i.e. 3, 7, 14 and 21 days), the cell culture

media was replaced with 10% v/v prestoblue reagent (life technologies, USA) in phenol red free DMEM and incubated at 37°C and 5% CO_2 for 1.5 h. The fluorescence intensity was recorded at the excitation/emission wavelengths of 540/590 nm using a micro-plate reader (Synergy HTX, BioTek, USA).

Characterization and biological evaluation of the cartilage part

Degradation

[0089] Sample degradation rate was measured by monitoring the weight of samples over time. Samples were immersed in PBS and kept at 37°C in a shaker incubator (IKA KS 3000) for 60 days. At certain times, samples were taken out, weighed and returned to the container. The ratio of the recorded weight to the initial weight at each time point was reported as a function of time.

Chondrocyte attachment and proliferation

[0090] Normal human chondrocytes (Cell Applications Inc, USA) were cultured and sub-cultured under standard aseptic condition. At the confluency of 90%, chondrocytes were trypsinized and suspended in fetal bovine serum (FBS, Sigma-Aldrich, USA). Scaffolds were disinfected using 70% ethanol, washed and seeded with a density of 3×10^5 cells/scaffold in 12-well culture plates. The scaffolds were submerged after 30 min and incubated at 37°C, 5% CO_2, using a chondrocyte growth medium (Cell Applications Inc, USA). The media were changed every other day.

[0091] Prestoblue assay was used to measure the proliferation rate of chondrocytes on the scaffold. At certain time intervals (i.e. 4, 7, 14, 21 days), the media of the wells were replaced with 10% v/v prestoblue. The plates were then incubated for 1.5h at 37°C and 5% CO_2. The fluorescence intensity was measured at an excitation wavelength of 540 nm and emission wavelength of 590 nm. The same procedure was performed on the first day, after complete attachment of the cells to the scaffolds. The number of cells at each time point (N) to the initial number of the cells (N_0) was calculated by dividing the corresponding intensity value by the absorbance value of the first day.

[0092] Scanning electron microscopy was used to investigate cell attachment. Scaffold-cell complexes were washed using PBS and immersed in Karnovsky's fixative for 1.5 h. The complexes were then submerged in 1% w/v osmium tetroxide for 1.5 h. Ethanol series (30, 50, 75, 95 and 100% v/v) were used to dehydrate the samples. The samples were then air-dried and sputter-coated with gold. SEM imaging was performed at accelerating voltages between 1-5 kV with different magnifications.

[0093] At the mentioned intervals considered for proliferation rate measurement (i.e. 7, 14, 21 days), scaffolds were fixed in formalin solution (10%, neutral buffered) for H&E

staining. The cells on the scaffold were fixed overnight and stained using Eosin and Hematoxylin. The color was adjusted using 1% v/v acidic alcohol, ethanol and bluing agent, according to standard histology protocols. Laser microscopy (Lext, Olympus) and fluorescence microscopy (Evos FI, Life Technologies) were employed for monitoring cell growth on the scaffold and extracellular matrix (ECM) secretion by chondrocytes.

Characterization of the barrier film

Scanning Electron Microscopy (SEM)

[0094] To investigate the morphology of porous PCL film, scanning electron microscopy (SEM, JEOL-JSM6510, Japan) was employed. Samples were sputter-coated with gold and imaging was carried out at accelerating voltage of 3kV at various magnifications.

Mechanical properties and suture retention strength

[0095] Tensile strength of the films was examined using the Electromechanical Precision Universal Tester. The samples were fixed into a screw flat tensile grip and underwent the tensile test using a 5 kN load cell and crosshead speed of 1 mm/min. To investigate the effect the addition of PEG and the resulting porosity had on tensile strength, dense PCL films were also prepared using solvent-casting method and tested under the same condition.

[0096] To measure the suture retention strength, a steel wire of 0.15 mm diameter was used, by which the effects of suture materials on force-displacement curves is minimized. The steel wire was passed through a pinhole created in the film to form a loop and fixed to the tensile testing machine, such that the distance from the grip was 10 mm. The other edge of the film was fixed to the inferior screw flat tensile grip. The steel wire was pulled at a rate of 1 mm/min until the film was completely torn.

Wettability and permeability

[0097] The wettability of the films was evaluated through contact angle measurement. A 5 µl drop of DI water was placed on either side of the samples, and the image was captured using a Dino-lite digital microscope camera. Each image was then processed to determine the contact angle.

[0098] Fluorescein sodium salt was selected as the model probe to quantify the permeability of the films. The film (D=10 mm) was fixed between two reservoirs filled with 4 mg/ml of solution or pure water. The entire system was fixed in a clamp and placed at 37°C. At certain time intervals, 100 µl of the solution was taken out from the low concentration side, transferred to a 96-well plate and the fluorescence intensity was recorded at excitation/emission wavelengths of 540/590 nm using a micro-plate reader.

The diffusion kinetics were reported as the variation in the ratio of concentration at each specific time to the equilibrium concentration over time.

Results

Characterization of the bone portion

[0099] The results of the TCP-HA paste viscometry show that the paste has flow properties that are suitable for 3-D printing. The morphology of the printed and sintered scaffolds also showed that the scaffold portions have desirable mechanical properties.

[00100] The proliferation rate of osteoblasts on the TCP-HA scaffolds, as well as the control group (cell culture plate), is shown in Figure 2. The results are reported as the variation of N/N_0 over time, which is the ratio of the number of cells at each time point (N) to the initial number of the cells (N_0). The ratio can be obtained by dividing the corresponding intensity values. The results are compared to a control (standard cell culture plate (control group)).

[00101] As observed, except for the starting point, the number of cells on the scaffold is higher than the control group at all other time points. Furthermore, the cells seem to grow and proliferate faster on the scaffold as the slope of the TCP-HA graph increases.

[00102] Fluorescence and SEM images of the osteoblasts attached on the TCP-HA scaffolds show that the osteoblasts grew within the pores and on the surface of the strands of the 3D-printed scaffold, forming an adherent plexus all over the scaffold. The images show that osteoblasts have anchored their cytoskeletal projections, lamellipodia and filopodia, to the surface features, revealing the bioactive characteristics of the scaffolds.

Characterization of the cartilage portion

[00103] The morphology of the printed scaffold was studied from SEM images. The layers were shown to be interlocking. The average pore size was found to be 892 ± 62 μm. The degradation rate of the cartilage portion is demonstrated in Figure 3. The scaffold weight remained almost constant in the first 30 days, implying the stability of the constructs in this timeframe, but decreased drastically between 30 and 60 days. After 60 days, the scaffolds completely collapsed and disappeared.

[00104] SEM images showed that, after one day of cell seeding, chondrocytes attached perfectly to the surface of gelatin/elastin/sodium hyaluronate construct.

[00105] The proliferation rate of normal human chondrocytes on the scaffolds is illustrated in Figure 4. The results are reported as the variation of N/N_0 over time, which is the ratio of the number of cells at each time point (N) to the initial number of the cells (N_0). The ratio can be obtained by dividing the corresponding intensity values.

[00106] The number of the cells on the scaffold persistently increased from day 1 to day 21. After 21 days, the number of cells was more than 5 times that of the initial number, seeded on the first day.

[00107] Both laser and fluorescence microscopy confirm the attachment, growth, proliferation and ECM secretion over 21 days.

Characterization of the barrier film

[00108] By comparing SEM images, it was observed that the addition of either none or a very small amount of PEG resulted in none or a very small number of pores per unit of surface area. Through employing 1:1 PEG/PCL w/w ratio, more pores were observed on one side. However, water could not pass through the film, even when a high vacuum was applied to the other side as a driving force. While there were adequate pores on one side, the pores seem to be close-ended, as the transportation of water molecules was still not possible. However, increasing the ratio to 3:2 w/w resulted in the formation of some pores on the other side, through which the water molecules could permeate by applying high vacuum. The pores on the optimum porous PCL film were found to be smaller than 10 μm. Such pore size allows diffusion of nutrients, like glucose, but prevents cell migration.

[00109] Table 3 represents the mechanical properties and suture retention strength of the prepared films compared to solid PCL film fabricated with addition of no PEG. The porous PCL film was found to have lower modulus and ultimate strength, but higher elongation at break. Having a porous structure, as well as the plasticizing effect of the residual PEG, might account for lower stiffness and strength, along with more flexibility. The suture retention strength was 9.04 ± 0.92 N for the porous PCL film, which is about half of the corresponding value for solid film.

[00110] Table 3. The mechanical properties and suture retention strength of porous PCL film compared to a solid film.

Parameter	Solid PCL film	Optimized porous PCL film
Young's modulus (MPa)	112.02 ± 3.62	16.91 ± 2.75
Ultimate Strength (MPa)	12.96 ± 0.99	4.91 ± 1.36
Elongation at break %	566.83 ± 71.07	973.02 ± 15.35
Suture Retention Strength (N)	18.04 ± 5.28	9.04 ± 0.92

[00111] The treatment of the surface of the films with NaOH resulted in improved wettability. Floating the films on NaOH solution resulted in a dramatic decrease in contact angle from 76.7 ± 3.3° to 36.4 ± 3.7°. The films were also found to be permeable.

[00112] Throughout the description and claims of this specification, the words "comprise" and "contain" and variations of them mean "including but not limited to", and they are not intended to (and do not) exclude other moieties, additives, components, integers or steps. Throughout the description and claims of this specification, the singular encompasses the plural unless the context otherwise requires. In particular, where the indefinite article is used, the specification is to be understood as contemplating plurality as well as singularity, unless the context requires otherwise.

[00113] Features, integers, characteristics, compounds, chemical moieties or groups described in conjunction with a particular aspect, embodiment or example of the invention are to be understood to be applicable to any other aspect, embodiment or example described herein unless incompatible therewith. All of the features disclosed in this specification (including any accompanying claims, abstract and drawings), and/or all of the steps of any method or process so disclosed, may be combined in any combination, except combinations where at least some of such features and/or steps are mutually exclusive. The invention is not restricted to the details of any foregoing embodiments. The invention extends to any novel one, or any novel combination, of the features disclosed in this specification (including any accompanying claims, abstract and drawings), or to any novel one, or any novel combination, of the steps of any method or process so disclosed.

CLAIMS

1. A kit of parts for assembly into a joint plug, the joint plug for insertion within an opening of a bone layer in a joint and through a cartilage layer on the bone layer, the kit of parts comprising:

a bone scaffold portion for accommodating bone tissue therein, said bone scaffold portion being configured for insertion into the opening of the bone layer;

a cartilage scaffold portion for accommodating cartilage tissue, said cartilage scaffold portion being configured to be positioned over the bone scaffold portion; and, optionally,

a permeable membrane portion configured to be provided over the cartilage scaffold portion.

2. A kit of parts as claimed in claim 1, further comprising bone scaffold engagement means for mounting the cartilage scaffold portion on the bone scaffold portion.

3. A kit of parts as claimed in any preceding claim, further comprising membrane portion engagement means for mounting the permeable membrane portion on the cartilage scaffold portion.

4. A kit of parts as claimed in any preceding claim, wherein a maximum pore size of the permeable membrane portion is less than 50 micrometres.

5. A kit of parts as claimed in any preceding claim, wherein the cartilage scaffold portion comprises a substrate having a plurality of openings, and wherein the cartilage tissue is accommodated by the plurality of openings.

6. A kit of parts as claimed in any preceding claim, wherein at least one of the bone scaffold portion, the cartilage scaffold portion and the permeable membrane portion are formed from a biodegradable material.

7. A kit of parts as claimed in any one of the preceding claims, wherein the permeable membrane portion comprises polycaprolactone.

8. A kit of parts as claimed in any preceding claim, wherein the bone scaffold portion is formed from a ceramic or a composite material.

9. A kit of parts as claimed in any preceding claim, further comprising a barrier portion provided in the bone scaffold portion.

10. A kit of parts as claimed in any preceding claim, wherein the cartilage tissue accommodation further comprises a source of chondrocytes.

11. A kit of parts as claimed in any preceding claim, wherein the joint is a knee joint or ankle joint.

12. A joint plug for insertion within an opening of a bone layer in a joint and through a cartilage layer on the bone layer, for treatment of damaged cartilage, the joint plug comprising:

a bone scaffold portion for accommodating bone tissue therein, said bone scaffold portion configured for insertion into the opening of the bone layer;

a cartilage scaffold portion for accommodating cartilage tissue therein, said cartilage scaffold portion being configured to be positioned over the bone scaffold portion; and

a permeable membrane portion configured to be provided over the cartilage scaffold portion.

13. A bone scaffold portion for the kit of parts for assembly into the joint plug of any of claims 1 to 11, the bone scaffold portion for accommodating bone tissue therein and configured for having a cartilage scaffold portion of the joint plug provided thereon.

14. A cartilage scaffold portion for the kit of parts for assembly into the joint plug of any of claims 1 to 11, the cartilage scaffold portion for accommodating cartilage tissue therein and configured to be provided on a bone scaffold portion of the joint plug.

15. A method of forming the bone scaffold portion for the kit of parts for assembly into the joint plug of any of claims 1 to 11, or the bone scaffold portion of claim 13, the method comprising:

receiving sensor data from a sensor device, the sensor data indicative of the shape of the opening of the bone layer into which the bone scaffold portion will be inserted;

determining a bone scaffold portion shape based on the obtained sensor data; and

forming the bone scaffold portion having the bone scaffold portion shape using a three-dimensional (3D) printing device.

16. The method of claim 15, further comprising forming bone tissue accommodation means of the bone scaffold portion, the bone tissue accommodation means to accommodate bone tissue therein, and seeding the bone tissue accommodation means with bone cells.

17. A method of preparing a joint plug as claimed in claim 12 , the method comprising:

seeding the bone tissue accommodation means of the bone scaffold portion with bone cells;

seeding the cartilage tissue accommodation means of the cartilage scaffold portion with chondrocytes;

assembling the joint plug by providing the cartilage scaffold portion over the bone scaffold portion and providing the membrane portion over the cartilage scaffold portion.

18. A method of preparing a joint plug as claimed in claim 17, wherein the joint plug is assembled after seeding of the bone tissue accommodation means with bone cells.

19. A method of preparing a joint plug as claimed in claim 17 or claim 18, wherein the joint plug is assembled after seeding of the cartilage tissue accommodation means with chondrocytes.

Intellectual Property Office

Application No:	GB1716195.1	**Examiner:**	Andrew Hughes
Claims searched:	1-14, 17-19	**Date of search:**	26 February 2018

Patents Act 1977: Search Report under Section 17

Documents considered to be relevant:

Category	Relevant to claims	Identity of document and passage or figure of particular relevance
X,Y	X:1,2,5,6, 9-14; Y:1,3,4,7, 17-19	WO 2007/046746 A1 (ARTIMPLANT AB) particularly figures 5f and 8
X,Y	X:1,2,5,6, 11,13,14; Y:1,3,4,7, 17-19	WO 2009/155232 A1 (RTI BIOLOGICS, INC.) whole document
X,Y	X: 1,2,5,8,10 ,11,13,14; Y:1,3,4,7, 17-19	WO 2009/036431 A1 (UNIVERSITY OF MISSOURI) whole document
X,Y	X:1,5,6,1 0,11,13,1 4; Y:1,3,4,7, 17-19	US 5876452 A (ATHANASIOU et al.) whole document
X,Y	X:1,2,5,6, 10; Y:1,3,4,7, 17-19	WO 2006/026981 A1 (TECHNISCHE UNIVERSITÄT DRESDEN) whole document
X	12	US 2007/0113951 A1 (HUANG) whole document
Y	1,3,4,7,17 -19	US 2011/0224801 A1 (MANSMANN) particularly paragraphs 15 and 37
Y	1,3,4,7,17 -19	WO 2009/011849 A2 (LIFENET HEALTH) paragraph 88
Y	1,3,4,7,17 -19	WO 2006/045330 A1 (TETEC-TISSUE ENGINEERING TECHNOLOGIES) particularly the second paragraph of page 8 and the second paragraph of page 19

Intellectual Property Office

Y	1,3,4,7,17 -19	EP 1216718 A1 (ETHICON, INC.) paragraphs 18 and 65 and barrier layer 16 in figure 3
Y	1,3,4,7,17 -19	WO 98/08469 A2 (VTS HOLDINGS, LTD.) line 24 of page 3, line 33 of page 4, and figures 2 and 3C
Y	4	WO 03/024463 A1 (RUSH-PRESBYTERIAN-ST. LUKE'S MEDICAL CENTER) page 21
Y	7	WO 2014/016816 A2 (A4TEC) particularly paragraphs 18-20
Y	7	WO 2013/169374 A1 (THE STEVENS INSTITUTE OF TECHNOLOGY) page 10
Y	17-19	WO 2017/041068 A1 (THE TEXAS A & M UNIVERSITY SYSTEM) particularly paragraphs 4, 10, 11, 13 and 17
A	15,16	US 2017/0172743 A1 (BONUTTI) especially paragraphs 4, 25, 27, 31, 32 and figures 2-4

Categories:

X	Document indicating lack of novelty or inventive step	A	Document indicating technological background and/or state of the art.
Y	Document indicating lack of inventive step if combined with one or more other documents of same category.	P	Document published on or after the declared priority date but before the filing date of this invention.
&	Member of the same patent family	E	Patent document published on or after, but with priority date earlier than, the filing date of this application.

Field of Search:

Search of GB, EP, WO & US patent documents classified in the following areas of the UKC^{X} :

Worldwide search of patent documents classified in the following areas of the IPC

A61F

The following online and other databases have been used in the preparation of this search report

EPODOC & WPI; Patent Fulltext

International Classification:

Subclass	Subgroup	Valid From
A61F	0002/30	01/01/2006
A61F	0002/28	01/01/2006