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BIOMATERIALS

A survey on the usage of decellularized tissues in orthopaedic clinical trials

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Aims

Orthopaedic surgery requires grafts with sufficient mechanical strength. For this purpose, decellularized tissue is an available option that lacks the complications of autologous tissue. However, it is not widely used in orthopaedic surgeries. This study investigated clinical trials of the use of decellularized tissue grafts in orthopaedic surgery.

Methods

Using the ClinicalTrials.gov (CTG) and the International Clinical Trials Registry Platform (ICTRP) databases, we comprehensively surveyed clinical trials of decellularized tissue use in orthopaedic surgeries registered before 1 September 2022. We evaluated the clinical results, tissue processing methods, and commercial availability of the identified products using academic literature databases and manufacturers' websites.

Results

We initially identified 4,402 clinical trials, 27 of which were eligible for inclusion and analysis, including nine shoulder surgery trials, eight knee surgery trials, two ankle surgery trials, two hand surgery trials, and six peripheral nerve graft trials. Nine of the trials were completed. We identified only one product that will be commercially available for use in knee surgery with significant mechanical load resistance. Peracetic acid and gamma irradiation were frequently used for sterilization.

Conclusion

Despite the demand for decellularized tissue, few decellularized tissue products are currently commercially available, particularly for the knee joint. To be viable in orthopaedic surgery, decellularized tissue must exhibit biocompatibility and mechanical strength, and these requirements are challenging for the clinical application of decellularized tissue. However, the variety of available decellularized products has recently increased. Therefore, decellularized grafts may become a promising option in orthopaedic surgery.

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Keywords: Decellularized tissue, Clinical trials, Orthopedic surgery, Autologous tissue harvesting

Article focus

- Although autografts are frequently used in orthopaedic surgery, decellularized allografts/xenografts have attracted attention because of the limited harvest volume and the risk of donor site morbidity for autografts.
- The extent to which decellularized tissue grafts are used in orthopaedic surgery is unclear; therefore, this study comprehensively investigated their use in orthopaedic surgery clinical trials.
- We hypothesized that few commercially available decellularized products would be identified in orthopaedic clinical trials.

Key messages

- There have been 27 trials investigating the use of decellularized tissue in orthopaedic surgery, nine of which were completed, and 16 product names were identified.
- We identified one decellularized product that will complete at least one clinical trial with favourable outcomes for knee

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joint grafts requiring mechanical load resistance that makes clinical use difficult.
- The variety of decellularized products used in orthopaedic surgery requiring mechanical strength and biocompatibility has recently increased, suggesting that they could be a promising option.

Strengths and limitations

- This is the first study to comprehensively investigate the clinical use of orthopaedic decellularized grafts.
- We comprehensively surveyed the tissue processing methods, clinical use, and outcomes of decellularized products in trials found on the CTG and ICTRP databases.
- This study may have omitted products and studies not registered on the CTG and ICTRP databases.

Introduction

The tissues treated in orthopaedic surgery are mainly those of the musculoskeletal system, including bone, cartilage, muscles, ligaments, and tendons. Autologous and allogenic orthopaedic tissue transplantations are effective treatments when the volume of these tissues is reduced by injury or degenerative disease.[1] While autologous tissue is biocompatible, there is a limited harvest volume and a risk of donor site morbidity. Allografts avoid these problems but have their own disadvantages, including lower biocompatibility and potential disease transmission.[2,3] To overcome the limitations of autografts and allografts, research has endeavoured, through in vitro tissue engineering, to create artificial grafts able to perform some of the biological functions of real tissue.[4] This is achieved using artificial biological scaffolds. Such scaffolds require a 3D microenvironment able to maintain the growth of seed cells and cytokines. A well-constructed scaffold is a prerequisite for good biocompatibility when the graft is implanted.[5] The biomaterials used to construct artificial biological scaffolds are generally composed of one or two extracellular matrix (ECM) components, such as collagen, fibronectin, and hyaluronic acid. These are selected and constructed to mimic the natural ECM. However, these scaffolds cannot reproduce the dynamic complexity of the natural microenvironment or perform the range of functions of natural ECMs.[6] The ECM structure differs significantly in different types of tissue, even within the same individual. Conversely, the ECM structure of a given type of tissue is conserved across different species.[7] Therefore, xenotransplantation using natural tissue from other species, in which the ECM shares the structure of equivalent human ECM, has attracted attention. Xenografts are more available than allografts because of their animal origin. However, there is a risk of rejection by xenogeneic antigens[8] and disruption of the ECM structure by tissue processing methods.[9] Therefore, the use of xenogeneic bone in orthopaedic surgery is not recommended because of poor biocompatibility and poor clinical results compared to those achieved using allogenic bone. Because of these disadvantages, xenogeneic bone tissue is seldom commercially available.[10] A further issue with xenotransplantation is the difficulty of both removing cellular components and preserving the ECM, as one tends to preclude the other.[11] However, if such limitations can be overcome, then xenotransplantation would certainly be a desirable option in orthopedics, a field in which reconstructive and reparative surgeries are common. In recent years, the US Food and Drug Administration (FDA) approved the use of decellularized and antigenically weakened ECM from allogenic or xenogeneic tissues or organs.[12] Although a few reports have described the use of decellularized grafts in orthopaedic surgery,[11] there have been no comprehensive investigations into the clinical use of orthopaedic decellularized grafts. Therefore, this study surveyed and analyzed the clinical applications and indications of decellularized tissues in orthopaedic surgery and the decellularization and sterilization methods used in each product. We hypothesized that not many decellularized products that had passed clinical trials would be commercially available in orthopaedic surgery.

Methods

Ethical approval. No ethical approval or consent was required for this study because data were obtained from internet databases and involved no humans or animals.

Clinical trial search methods. Using the two leading databases of the clinical trial, ClinicalTrials.gov (CTG), a registry of clinical trials maintained by the National Library of Medicine of the USA National Institutes of Health, and the International Clinical Trials Registry Platform (ICTRP), maintained by the World Health Organization, we thoroughly surveyed clinical trials in orthopaedics that used decellularized tissue. We searched for trials registered by 1 September 2022, using the search terms "decellularized OR decellularization OR acellular OR acellularization OR xenograft OR allograft". The status of extracted clinical trials and details of decellularized products used in trials were reviewed by 10 October 2022.

Trial selection and data extraction. The study included clinical trials that assessed the use of allogenic or xenogeneic decellularized tissue for orthopaedic grafting purposes. Studies with a status of 'not yet recruiting', those unrelated to orthopaedics, those concerned with the treatment of skin ulcers using decellularized tissue-derived dressings, those in which decellularized tissue transfer was not performed, and those in which decellularized tissues such as injectable or wrapping materials that do not require mechanical strength were excluded. Two independent reviewers (MI and JI) assessed the trials returned in our search for eligibility. The senior author (KI) was asked to provide a final decision in cases of disagreement.

Scrutiny of the medical products used in the clinical trials. The diseases or parts of the body to which any decellularized tissue was applied, the status of the clinical trials, the name of the tissue product, the type of derived tissue, and the clinical results of the application of each

Table I. Overview of nine clinical trials for the treatment of shoulder rotator cuff injuries using decellularized tissue.

Trial no.	Condition	Clinical trial name	Length	Status	Product name	Source tissue	Country	Hospital/ Company	Clinical reports	Decellularization method/ Sterilization method
S1	Rotator cuff tear	Use of graft jacket for rotator cuff repair	2007 to 2011	Completed	GRAFTJACKET allograft	Human dermis	USA	Southern California Orthopaedic Institute/Wright Medical	Barber,[13] Gupta[14]	N/A; tissue underwent no general sterilization and freezing[15]
S2	Rotator cuff tear	Outcome evaluation of allograft scaffold augmentation for arthroscopic repair of full thickness of rotator cuff tear	2013 to 2016	Completed	CGDerm	Human dermis	South Korea	Hallym University Medical Center Kangnam/CGBio	Lee[16]	N/A
S3	Rotator cuff tear	Clinical outcomes and structural integrity of arthroscopic superior capsular reconstruction using cryopreserved acellular dermal matrix with increased elasticity and thickness in patients with irreparable rotator cuff tear	2019 to 2020	Completed	CGDerm	Human dermis	South Korea	Ewha Womans University Seoul Hospital/CGBio	Shin[17]	N/A
S4	Rotator cuff tear	Acellular dermis in rotator cuff repair	2013 to 2021	Completed	N/A	Human dermis	UK	The Royal Orthopaedic Hospital NHS Trust	N/A	N/A
S5	Rotator cuff tear	Allograft reconstruction of massive rotator cuff tears vs partial repair alone	2015 to 2021	Completed	Allopatch HD	Human dermis	Canada	Nova Scotia Health Authority/ MTF Sports Medicine	Agrawal[18]	N/A; harvested by sterile techniques; tissue underwent no general sterilization[19]
S6	Rotator cuff tear	Arthroscopic superior capsular reconstruction – study of different types of grafts	2018 to 2022	Active, not recruiting	N/A	Pig dermis, bovine pericardium, allogenic/ autologous femoral fascia, allogeneic Achilles tendon	Portugal	Hospital de Egas Moniz, Centro Hospitalar Lisboa Ocidental, General Hospital of Santo António, etc.	N/A	N/A
S7	Rotator cuff tear	Massive rotator cuff tear reconstruction	2018 to 2023	Recruiting	GRAFTJACKET allograft	Human dermis	Canada	Nova Scotia Health Authority/ Wright Medical	Barber,[13] Gupta[14]	N/A; tissue underwent no general sterilization and freezing[15]
S8	Rotator cuff tear	Comparison of partial rotator cuff repair vs superior capsular reconstruction for irreparable rotator cuff tears	2020 to 2024	Recruiting	N/A	Human dermis	USA	University Hospitals Cleveland Medical Centre, Lake Health, Midwest Orthopaedics at Rush	N/A	N/A
S9	Rotator cuff tear	An outcomes study utilizing Allomend (R) HD for superior capsular reconstruction	2021 to 2025	Recruiting	AlloMend	Human dermis	USA	Western Orthopaedics Research and Education Foundation/ AlloSource	N/A	Detergents or enzymes/electron irradiation not required[20]

N/A, not available.

product were carefully scrutinized and recorded using the PubMed (Medline), Web of Science, Cochrane, and EMBASE databases, as well as the websites of the companies responsible for the sale and manufacture of the relevant medical technology. If still unsure, we emailed the clinical trial or manufacturer's representative if the product name was known, or the clinical trial representative if the product name was unsure. 'Not available (N/A)' was assigned (Tables I to IV) when the representatives refused information, or there was no response to our email from the representatives, and further information collection was not possible. Because this study was intended as an overview of the decellularized products used in trials extracted from the database, no quantitative evaluation was performed and no statistical analysis was required.

Results

Extracted clinical trials. We identified 4,402 clinical studies (CTG: 2,811, ICTRP: 1,591). From these, 1,022 duplicates were removed, resulting in 3,380 trials being screened. After screening, 85 trials with a status of "not yet recruiting", 2,919 trials unrelated to orthopaedics, 61

Table II. Overview of eight clinical trials for the treatment of knee disorders using decellularized tissue.

Trial no.	Condition	Clinical trial name	Length	Status	Product name	Source tissue	Country	Hospital/Company	Clinical reports	Decellularization method/sterilization method
K1	Cartilage injury	Chondrofix osteochondral allograft prospective study	2011 to 2014	Terminated (inadequate enrolment and decreased need for clinical data to support product)	Chondrofix	Human osteochondral plug	USA	Plancher Orthopaedics and Sports Medicine, Cartilage Repair Centre, Brigham and Women's Hospital, Orthopaedic Research Foundation, etc/Zimmer Biomet	Farr[21]	N/A; gamma irradiation, freezing[22]
K2	Cartilage injury	Second line treatment of knee osteochondral lesion with treated osteochondral graft	2016 to 2021	Terminated (low accrual rate)	OD-PHOENIX	Human osteochondral plug	France	TBF Genie Tissulaire	N/A	N/A; gamma irradiation (25 to 32 kGy), freezing[23]
K3	Cartilage injury	Clinical study of decalcification bone scaffold for cartilage lesions of the knee	2018 to 2023	Recruiting	N/A	Decalcification bone	China	Peking University Third Hospital	N/A	N/A
K4	Cartilage injury	Clinical trial to evaluate the efficacy and safety of MegaCarti in knee cartilage defects	2020 to 2027	Active, not recruiting	MegaCarti	Human cartilage	South Korea	Yonsei University Health System, Gangnam Severance Hospital/ L&C Bio	N/A	N/A; gamma irradiation[24]
K5	ACL injury	Safety and efficacy study of the Z-Lig medical device compared to allograft	2010 to ?	Unknown status	Z-Lig	Porcine bone-patellar tendon	Belgium, Italy, Netherlands, South Africa, Spain	Ghent University Hospital, Aarhus University Hospital, Istituti Ortopedici Rizzoli, etc/Aperion Biologics	Zaffagnini[25]	Enzymes and α-galactosidase/electron irradiation[26]
K6	ACL injury	OrthoPure XT pilot clinical study	2018 to 2020	Withdrawn (unspecified business decision/strategic reason)	OrthoPure XT	Porcine tendon	USA	St. Joseph's Outpatient Surgery Centre, OrthoIndy Hospital South, Jewish Hospital/ TRX Orthopedics	Hunt[27]	0.1% sodium dodecyl sulfate and protease inhibitor[28]/ antibiotics, peracetic acid[29]
K7	ACL injury	Clinical evaluation of dCELL ACL scaffold for reconstruction of the anterior cruciate ligament	2015 to 2021	Active, not recruiting	dCELL ACL scaffold	Porcine tendon	Poland, Spain, UK	Robert Jones and Agnes Hunt Hospital NHS Foundation Trust, Hospital Universitari de Bellvitage, Klinika Chirurgii Endoskopowej Sp. z o.o. etc/Tissue Regenix	Hunt[27]	0.1% sodium dodecyl sulfate and protease inhibitor[28]/ antibiotics, peracetic acid[29]
K8	Meniscus injury	Clinical evaluation of dCELL meniscus for partial arthroplasty of the meniscus	2014 to 2018	Suspended	dCELL meniscus	Porcine meniscus	Poland, UK	"Ortotrauma" Spółka Z Ograniczoną, The Hillingdon Hospitals NHS Foundation Trust, the Robert Jones & Agnes Hunt Orthopaedic Hospital NHS Foundation Trust/ Tissue Regenix	N/A	0.1% sodium dodecyl sulfate and protease inhibitor;[28,30] N/A

ACL, anterior cruciate ligament; N/A, not available.

Table III. Overview of four clinical trials for the treatment of ankle and hand disorder using decellularized tissue.

Trial no.	Condition	Clinical trial name	Length	Status	Product name	Source tissue	Country	Hospital/ Company	Clinical reports	Decellularization method/ sterilization method
AH1	Ankle: osteochondral lesions of the talus	OD-PHOENIX in talus osteochondral lesion	2017 to 2021	Terminated (low accrual rate)	OD-PHOENIX	Human osteochondral plug	Belgium, Israel	AZ Monica, Poriya Medical Centre/TBF Genie Tissulaire	N/A	N/A; gamma irradiation (25 to 32 kGy), freezing[23]
AH2	Ankle: lateral collateral ligament instability of the ankle	The comparison of clinical and radiological outcomes between modified Broström operation and lateral ankle ligament reconstruction using acellular dermal matrix augmentation in chronic lateral ankle instability: A 5 year randomized controlled trial	2020 to 2025	Recruiting	SureDerm	Human dermis	South Korea	Wonkwang University Hospital/ Hans Biomed Corporation	N/A	Enzymes, EDTA/ freeze-dried[31]
AH3	Hand: CM arthritis of the thumb	Use of FlexHD as Post Trapeziectomy Spacer	2013 to 2017	Completed	FlexHD	Human dermis	USA	Cedars-Sinai Department of Hand Surgery/ Ethicon	Yao[32]	Non-ionic surfactant (Triton X-100)/ alcohol, peracetic acid[33]
AH4	Hand: STT arthritis	Use of a treated, devitalized and sterile meniscus segment (MENISC-T) in the treatment of STT osteoarthritis	2021 to 2023	Recruiting	MENISC-T	Human meniscus	France	Institut Chirurgical de la Main et du Membre Supérieur/TBF Genie Tissulaire	N/A	N/A

CM, carpometacarpal; EDTA, ethylenediaminetetraacetic acid; N/A, not available; STT, scaphotrapeziotrapezoid.

trials related to the treatment of skin ulcers using decellularized tissue-derived dressings, and 281 trials that did not use decellularized medical devices were excluded. Furthermore, from the remaining 34 clinical trials, seven trials were excluded: four using injectable agents, two using membranes to prevent adhesion, and one using a matrix that facilitates bone regeneration; thus, 27 clinical trials were finally eligible for inclusion in our study (Figure 1).

Target disease or body part and clinical trial status. Of the 27 trials included in our review, nine (S1 to 9) were related to shoulder injuries, including studies of rotator cuff patch grafting[40] and superior capsule reconstruction (SCR)[41] for rotator cuff injuries (Table I); eight (K1 to 8) were related to knee injuries, including four studies on grafts for osteochondral injury, three on grafts for anterior cruciate ligament (ACL) reconstruction, and one on grafts for meniscal transplantation (Table II); and four trials (AH1 to 4) were related to ankle or hand disorders, including one study on graft for osteochondral injury of the ankle, one on graft for lateral ligament reconstruction of the ankle, one on graft for void-filling after trapeziectomy, and one on graft for scaphotrapeziotrapezoid osteoarthritis (Table III). The remaining six trials (N1 to N6) were on nerve grafts for peripheral nerve injury of the upper limb (Table IV). Five trials (S1 to S5) in the shoulder field, one trial (AH3) in the hand field, and three trials (N2 to N4) in the nerve field were completed (Tables I to IV).

Decellularized tissue products used and the type of derived tissue. We identified 16 decellularized tissue product names used in 27 clinical trials: GRAFTJACKET (Wright Medical, USA), CGDerm (CGBio, South Korea),[16] Allopatch HD (MTF Biologics, USA), AlloMend (AlloSource, USA),[20] Chondrofix (Zimmer Biomet, USA),[21] OD-PHOENIX (TBF Genie Tissulaire, France),[23] Z-lig (APEIRON Biologics, Austria),[25] MegaCarti (L&C Bio, South Korea),[24] dCELL ACL Scaffold (Tissue Regenix, UK),[27,28,30] OrthoPure XT (Tissue Regenix),[27,28,30] dCELL Meniscus (Tissue Regenix),[28,30] SureDerm (Hans Biomed Corporation, South Korea),[31] FlexHD (Ethicon, USA),[32] MENISC-T (TBF Genie Tissulair), Avance Nerve Graft (Axogen, USA),[34] and NerVFIX (TBF Genie Tissulair).[38] GRAFTJACKET, CGDerm, and OD-PHOENIX were used in two trials (S1 and S7, S2 to S3, and K2 and AH1, respectively), and Avance in three trials (N1, N4, and N5). The product names used in six trials (S4, S6, S8, K3, and N2 to N3) were not provided. Of the 16 products, Chondrofix, OD-PHOENIX, and Z-lig contained bone tissues, and the rest were sourced from the soft-tissue or cartilage tissue alone.

Clinical results, decellularization methods, and sterilization methods. Clinical results were presented for 16 trials: S1 to S3, S5, and S7;[13,16–18] K1, K5 to K7;[21,25,27] AH3,[32] and N1 to N6.[34,36–38] The decellularization methods were provided in nine products in 12 trials (S9, K5 to K8, AH2 and AH3, and N1 to N5). AlloMend was treated without detergents or enzymes.[20] Z-lig was treated with enzymes for decellularization and with α-galactosidase for α-galactose removal.[26] The remaining seven products were decellularized with sodium dodecyl sulphate, protease inhibitors, enzymes, ethylenediaminetetraacetic acid, Triton X-200,

Table IV. Overview of five clinical trials for the treatment of peripheral nerve injury of the upper limbs using decellularized tissue.

Trial no.	Condition	Clinical trial name	Length	Status	Product name	Source tissue	Country	Hospital/ Company	Clinical reports	Decellularization method/sterilization method
N1	Peripheral nerve injury of upper limb	A comparative post-marketing study of commercially available peripheral nerve gap repair options	2009 to 2014	Terminated (transitioning from 361 HCT/P Tissue to a Biological (BLA))	Avance nerve graft	Human peripheral nerve	USA	Georgia Hand, Shoulder and Elbow, Indiana Hand Centre, University of Kentucky/Axogen Corporation	Safa[34]	50 mM phosphate + 100 nM Na, 0.14% Triton X-200, 0.6 mM sulfobetaine-16/gamma irradiation[35]
N2	Peripheral nerve injury of upper limb	Human acellular nerve graft for repair of peripheral nerve defects: a prospective, multicentre clinical study	2009 to 2017	Completed	N/A	Human peripheral nerve	China	The Sixth Affiliated Hospital of Shanghai Jiao Tong University /Guangzhou Zhongda Medical Devices Company	Zhu[36]	46 mM Triton X-100, 96 mM sodium deoxycholate;[36] N/A
N3	Peripheral nerve injury of upper limb	Human acellular nerve graft for repair of pure sensory nerve defects: a prospective, multicentre clinical study	2009 to 2017	Completed	N/A	Human peripheral nerve	China	The First Affiliated Hospital of Sun Yat-sen University/ Guangzhou Zhongda Medical Devices Company	Zhu[36]	46 mM Triton X-100, 96 mM sodium deoxycholate;[36] N/A
N4	Peripheral nerve injury of upper limb	Comparison of processed nerve allograft and collagen nerve cuffs for peripheral nerve repair (RECON)	2015 to 2021	Completed	Avance nerve graft	Human peripheral nerve	USA	Virginia Commonwealth University Medical Centre, University of Pennsylvania, University of Florida/Axogen Corporation	Axogen[37]	50 mM phosphate + 100 nM Na, 0.14% Triton X-200, 0.6 mM sulfobetaine-16/gamma irradiation[35]
N5	Peripheral nerve injury of upper limb	BMAC nerve allograft study	2017 to 2021	Unknown status (Previously: active, not recruiting)	Avance nerve graft	Human peripheral nerve	USA	Brooke Army Medical Centre, Walter Reed National Military Medical Centre, Curtis National Hand Centre at MedStar Union Memorial Hospital/Axogen Corporation	Safa[34]	50 mM phosphate + 100 nM Na, 0.14% Triton X-200, 0.6 mM sulfobetaine-16/gamma irradiation[35]
N6	Peripheral nerve injury of upper limb	Use of a nerve regeneration conduit (NerVFIX) in the treatment of nerve section of the wrist	2020 to 2023	Recruiting	NerVFIX	Human artery or vein from umbilical cord	France	Clinique de la Main - Nantes Atlantique, Institut Chirurgical de la Main et du Membre Supérieur/TBF Genie Tissulaire	Barnouin[38]	N/A; gamma irradiation (25 to 32 kGy), freezing[39]

BLA, biologic license application; BMAC, bone marrow aspirate concentrate; CM, carpometacarpal; HCT/P, human cell & tissue products; N/A, not available.

phosphate, sulfobetaine-16, and Triton X-100.[28,30,31,33,35,36] The sterilization methods were provided in 13 products in 17 trials (S1, S5, S7, S9, K1, K2, K4 to K7, AH1 to AH3, and N1, N4 to N6). Allopatch HD and GRAFTJACKET were only decellularized after the tissue had been harvested in a sterile manner.[15,19] The remaining 11 products were sterilized using gamma irradiation and freezing,[21–23,39] electron beam irradiation,[20,26] antibiotics and peracetic acid (animal study),[29] gamma radiation,[24,35] freezing,[31] and alcohol and peracetic acid[33] (Tables I to IV).

Discussion

The most important findings of this study were, firstly, that there have been 27 orthopaedic clinical trials using decellularized tissue and only nine of these were completed. Secondly, decellularized tissue has been used for only a few sites and conditions in orthopaedics. The first of these findings verified our hypothesis.

Rotator cuff injuries were the most common condition treated in the trials surveyed. In this context, decellularized tissue was used as a patch, as an augmentation for rotator cuff repair, and as a graft for SCR.[41] In general, repairs for small rotator cuff tears have good outcomes; however, massive tears, which account for 40% of rotator cuff tears, display poorer outcomes with primary repair alone.[42] Following debridement and primary repair, the treatment options for massive rotator cuff tears include: reverse shoulder arthroplasty; transfer of the latissimus dorsi, pectoralis major, and trapezius muscles; and SCR.[41,42] However, these methods do not provide anatomical repair or reconstruction. No improvement was reported in patient symptoms after the anatomical repair of a two-tendon rotator cuff tear using decellularized porcine small intestine mucosa tissue as a patch.[43] The poor results of this procedure may be explained by the fact that the small intestine mucosa is an inappropriate graft because its mechanical strength is inferior to that of the rotator cuff.[44] Lee et al[16] reported retear rates of 9.1% in the group with CGDerm as a patch augmentation for repair of large rotator cuff tears and 38.1% in

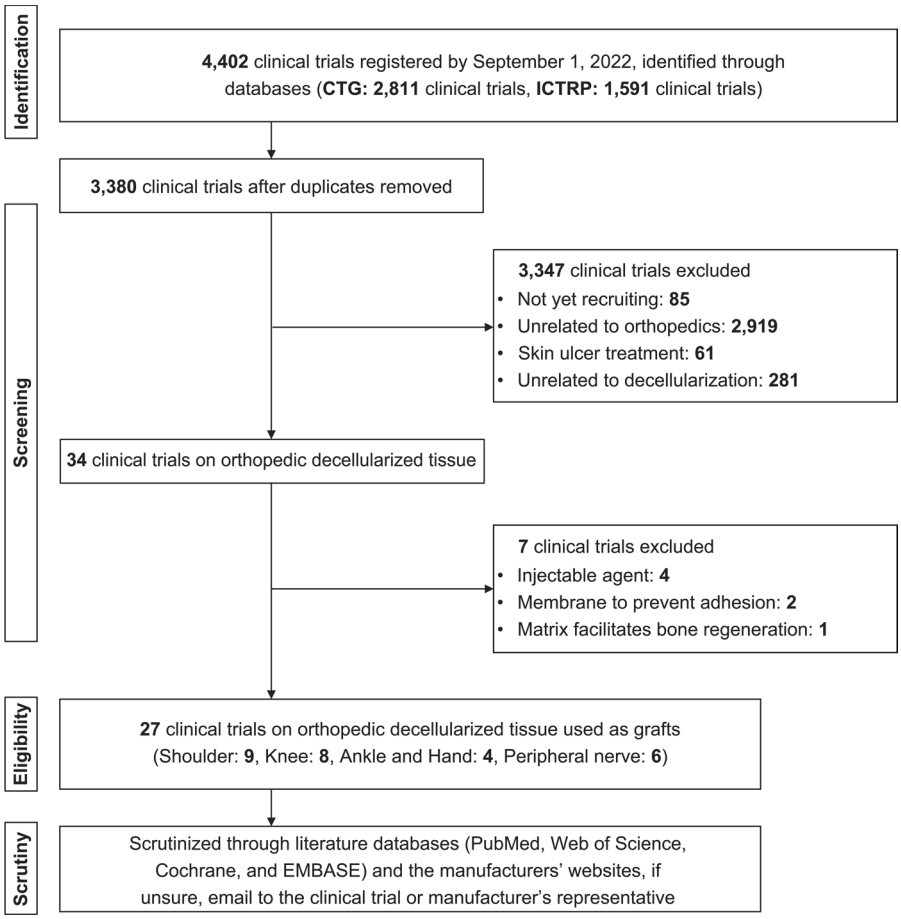

Fig. 1

Flowchart of the clinical trial search process and the identification of decellularized products used in trials. We identified 4,402 trials initially and eventually reviewed a total of 27 trials: nine in the shoulder field, eight in the knee field, four in the ankle and hand field, and six in the peripheral nerve field. CTG, ClinicalTrials.gov; ICTRP, International Clinical Trials Registry Platform.

the repair-alone group ($p = 0.034$). Shin et al[17] reported a visual analogue scale for pain improvement from 4.0 to 0.7 ($p < 0.001$) after SCR with CGDerm. Agrawal[18] used Allopatch HD for patch repair of massive rotator cuff tears. On MRI, performed a median of 16.8 months after the procedure, they found that 85.7% (12/14 patients) of the repaired rotator cuffs were intact, indicating good clinical outcomes. Barber et al[13] conducted a prospective randomized study on 22 patients treated with GRAFTJACKET augmentation for massive rotator cuff tears and 22 without augmentation. MRI performed a mean of 14.5 months after surgery found that the repaired rotator cuff was intact in 85% of patients in the GRAFTJACKET group versus 40% of those in the non-augmentation group ($p < 0.01$). In a prospective observational study with a mean follow-up of three years, all 24 patients with massive rotator cuff tears treated with GRAFTJACKET displayed significant improvements in pain, range of motion, and muscle strength.[14] FDA has approved GRAFTJACKET for use as reinforcement for rotator cuffs, ligaments, and joint capsules,[45] but not for use as a graft bridge for rotator cuff tears with gaps larger than 1 cm.[14] This prohibition of the use of GRAFTJACKET as a bridge is likely because the strength of the dermis comprising the product is inferior to that of the rotator cuff. However, an increasing number of completed clinical trials in recent years and good clinical outcomes suggest that decellularized tissues will be a promising device for rotator cuff treatment.

Chondrofix, a decellularized human osteochondral plug currently unavailable, produced poor clinical results of a high failure rate of 72% (23/32 knees) at a two-year follow-up.[21] OD-PHOENIX is derived from the human osteochondral plug; however, the clinical trial has been terminated because of the low accrual rate. We could identify incompleted trials in the osteochondral injury field. Z-lig is a decellularized porcine bone-patellar tendon graft. It has been used with just three patients but no failure was observed at two-year follow-up evaluations.[25] Z-lig is the world's first artificial biological device to receive the CE marking, which was awarded in 2014, as a graft for ACL revision or multiple ligament reconstruction. However, this product is no longer on the market, suggesting that problems may have been

identified. OrthoPure XT and dCELL ACL scaffold are identical in terms of indications, tissue derivation, and manufacturer and appear to be identical products. The K6 trial (withdrawn due to business decision) appears to have been USA-based, presumably to obtain FDA approval and USA market access. The K7 trial appears to have been Europe-based, presumably to obtain CE mark approval and European market access. OrthoPure XT was reported to have received CE marking on 1 June 2020, for its use in ACL revision grafts or multiple ligament reconstruction surgeries. Neil Hunt,[27] the principal investigator of the K7 trial, reported favourable clinical outcomes after 48 months of primary ACL reconstruction using OrthoPure XT at the European Society of Sports Traumatology, Knee Surgery & Arthroscopy in 2022. The status of the dCELL meniscus trial on partial meniscus reconstruction using decellularized porcine meniscus-derived was "suspended". We could identify incomplete trials in the knee field, including the dCELL ACL Scaffold trial. However, it would be changed to "completed". Studies using bovine bone-patellar tendon grafts for ACL reconstruction were conducted in the 1980s using a technique whereby glutaraldehyde cross-linked porcine heart valves were transplanted into humans. However, the large amounts of glutaraldehyde used for cross-linking resulted in poor biocompatibility of the tissue, inappropriate biomechanical properties, and a lack of integration with the host tissue. Hence, the clinical results of bovine tendon tissue use in humans were poor.[46] Therefore, the fact that OrthoPure XT is about to enter the market with distribution agreements in the UK and Italy is a significant step forward.[47,48] However, the practical application of decellularized tissues for meniscus disorders is not yet available. As mentioned, there is a trade-off between ECM maintenance and adequate decellularization when developing these products.[11] The challenge of maintaining both graft strength and biocompatibility is likely to be greater in tissue for the knee, due to the heavier load on the graft than occurs elsewhere on the body.[49]

In terms of the ankle joint, the AH1 trial using OD-PHOENIX for osteochondral lesions of the talus bone was terminated for the same reason as the K2 trial; however, the AH2 trial using SureDerm to augment the lateral ligament is ongoing, and the product is expected to be put into practical use. Yao et al[32] evaluated the clinical outcomes of FlexHD following its use as a void-filling spacer after trapeziectomy for carpometacarpal (CM) arthropathy. At six-month follow-ups, no complications were observed, and patients reported improvements in pain and grip strength compared to preoperative levels ($p < 0.01$). The AH3 trial is also in progress, which studies the placement of a decellularized meniscus segment at the joint interposition for scaphotrapeziotrapezoid arthropathy.

Safa et al[34] conducted a trial of Avance Nerve Graft and found that, in 624 nerve repairs with various gaps between transects, the mean meaningful recovery rate was 82% (91% for gaps smaller than 15 mm and 69% for gaps of 50 to 70 mm), which was comparable to results previously reported for autologous nerve grafts. It was also recently reported that the group using Avance Nerve Graft showed significantly better recovery than those using conduits for digital nerve injury (N5).[37] Other decellularized allogenic nerves and umbilical cord vessels (NerVFIX) have been used as scaffolds for peripheral nerve repair, yielding favourable results.[36,38] One of the reasons for the good clinical results in the hand and peripheral nerve field may be partially explained by the fact that these grafts need less mechanical strength than those applied to the knee.

An issue with the decellularized products identified in the current study was the use of dermal tissue patches for rotator cuff repair. Because the dermis is weaker than the rotator cuff,[44] these patches may not be suitable for rotator cuff repair, as the rotator cuff is subject to relatively high loads. However, the technology to decellularize thicker tissue more suitable for rotator cuff repair, while maintaining both the tissue strength and the ECM, has not yet been developed.[50] Stronger and thicker grafts more suitable for the rotator cuff may provide a more "anatomical" repair than SCR for large tears. We also identified some issues with the processing of tissues in the studies evaluated. Glutaraldehyde is used as a chemical cross-linking agent to stabilize tissue strength. However, its use compromises the integrity of collagen fibres and inhibits the infiltration of host cells into the graft.[51] Gamma irradiation sterilization destroys collagen fibres and impairs tissue regeneration.[52] Sterilization using peracetic acid, which is highly acidic and a strong oxidant, can affect the physical and chemical properties of the tissue.[53]

As previously mentioned, decellularized tissue used in orthopaedic surgeries requires both the biocompatibility necessary in other medical fields and the endurance of high mechanical loads. This latter requirement is specific to orthopaedics, making its clinical application in this field particularly difficult. Overcoming this issue will lead to the widespread use of decellularized tissue, which is highly demanded in orthopaedics. Therefore, development of decellularized tissues that are biocompatible and endure high mechanical loads is in high demand.

This study had several limitations. First, because the survey was conducted using the CTG and ICTRP databases, it may not have included clinical trials or products that were not registered on these databases. However, these databases reflect registries of 18 countries and appear to be highly comprehensive. Second, other tissue products may have unpublished tissue processing methods. However, this is unavoidable considering the risk of product features and specifications being leaked to the public. Third, there was little information available on the approval of the 16 products by relevant medical authorities; only Z-lig and OrthoPure XT have achieved CE marking.

In conclusion, of the 27 clinical trials of decellularized tissue in orthopaedics identified by a comprehensive survey, nine were completed. Despite the demand

for decellularized tissue, not many decellularized tissue products are commercially available now, particularly for the knee joint. However, the variety of available decellularized products, including OrthoPure XT for knee surgery, has recently increased. Therefore, decellularized grafts may become a promising option in orthopaedic surgery.

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