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REVIEW ARTICLE

It's the Biology Orthopods! Heralding a Reconstructive Revolution Through Musculoskeletal Tissue Banks (MSTB) in India

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Abstract

Background A tissue bank is an establishment that aids in retrieval, processing, storage, and distribution of human tissue for transplantation. For many years, such banks have been dispensing tissue to orthopaedic surgeons, performing reconstructive surgeries.

Methodology The retrieval, preparation, and delivery of musculoskeletal tissue used for transplantation is an intricate process involving varying practices among different musculoskeletal tissue banks.

Results Musculoskeletal allografts are used in various orthopaedic surgeries ranging from primary bone defects, trauma, and carcinoma to congenital disabilities. Every decade brings in paradigm shifts and new hope for treating challenging cases with the aid of newer devices and materials.

Conclusion This review article outlines various technical, regulatory and quality enhancement steps involved in tissue banking. Also, it discusses the road ahead and the research avenues for developing novel allograft products with the synergy of tissue banks and clinicians.

Keywords Musculoskeletal tissue banks · Orthopaedic surgeons · Graft selection · Tissue engineering · Bone graft · Tissue bank · Tissue sterilization · Allograft · Cryopreservation

Introduction

Every decade bring in paradigm shifts and new hope for treating challenging cases with the aid of newer devices and materials. Orthopaedic surgeons use various reconstructive techniques involving both implants and biological tissue, enabling them to manage complex cases unimaginable in the last century [1–3]. The use of bone grafts dates back many centuries to Indian scientist *Shushrut* who is credited with using autogenous bone grafts. The credit for modern bone grafting and increasing its popularity goes to several clinicians and researchers. One of the key person, Vittorio Putti, was the founder of SICOT (*Societè Internationale de Chirugie Orthopedique et Traumatologi*). Bone allografts during surgical reconstructions are more recent and are credited to William MacEwan and Fred Albee [4].

Currently, there are over a million grafts used in the field of reconstructive surgery. In orthopaedics, these are primarily human tissue allografts. However, some specialities like dentistry use ground bovine bone, while cardiac surgeons use porcine grafts, perhaps a reflection of the unavailability of human grafts [5]. Recently, history was created wherein

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a porcine heart transplant was performed in Baltimore in the US where, in this year alone, a record 3817 Americans received a heart transplants [6]. Other countries, particularly developing countries such as India, are comparatively deficient [6]. Human tissue can be donated as surgical residues from living donors or may be retrieved from deceased donors (Table 1). It is pertinent to note that a single deceased donor can give the gift of allografts to nearly 100 patients. As the awareness for donations increases, the possibility of universal availability of these allografts for use in reconstructive procedures will increase. Parallelly, the surgeons' experience will continue to drive the innovations and improvisation in the sector. Despite its vast population, India lacks severely in terms of availability and use of these allografts. This is likely to change with better techniques and tissue banks and standards across the country.

The present review article outlines various technical, regulatory and quality enhancement steps involved in tissue banking. The paper also discusses the road ahead and the research avenues for developing novel allograft products with the synergy of tissue banks and clinicians.

Classification of Allografts Usage

Anatomical Usage

While some grafts may be used to simulate the actual functional use, for example, Patellar Bone Patella tendon graft for reconstruction during the ACL reconstruction, or cortical strut graft for critical bone defects, others may be used in entirely non-anatomical locations [7]. Examples of these include dermal allografts used for enhancing the rotator cuff repair.

Type of Usage

Grafts can be classified as structural or non-structural depending on the site of use and the function. Structural grafts include tendons used for ligament reconstruction or strut grafts used for limb reconstructions. Examples of non-structural grafts, on the other hand, include a demineralized bone matrix where the properties of the graft are used to enhance the regenerative potential of the host tissue. [8] Sometimes, an anatomically structural graft may be used as a non-structural adjuvant, e.g. the femoral head can be morselized and mixed with bone marrow aspirate to serve as a biologic non-structural graft [9].

Musculoskeletal Allograft Possibilities

1. Bone
(a) Spine
* ACDF spacer
* ALIF Spacer
(b) Foot and Ankle
* Arch Wedge
(c) Joint Replacement:
* Wedge
* Sleeve
* Tibial Cross-section
* HTO Wedges
(d) Universal usage:
* Cancellous block
* Cancellous chips
* Cortical struts,
* Threaded Dowell
* Rib Segment
2. Tendons:
* Bone Patellar Bone Graft
* Tendo Achilles Graft
* Peroneus Graft
* Semi Tendinosus graft
* Gracillis Graft
* Quadricep Tendon Graft
* Tibialis Posterior Graft
* Palmaris Longus Graft

Table 1 Common sites for Musculoskeletal tissue retrieval

Living donors	Deceased donors
Femoral Heads from patients with fractured neck femur or hip arthritis, undergoing hip replacement	Long bones (tibia, fibula, femur, humerus, radius, ulna, ribs)
Bone cuts from patients undergoing total knee replacement (TKR) surgery	Flat bones (calcaneum, iliac crest, hemipelvis, sternum, vertebrae, skull bones, scapula, clavicle, mandible)
Cranioplasty bone flaps were removed during surgery and reused mainly as autografts	Tendons (semitendinosus, gracillis, tendoachilles, quadriceps, patellar tendon, peroneus longus, tibialis posterior, palmaris longus)
–	Cartilages and periarticular tissues (meniscus. acetabular labrum, costal cartilage)
–	Osteochondral allograft from around the joints especially knee and proximal femur

3. Membranes:
* Dermal graft – Unmeshed
* Dermal Graft – Meshed
* Split thickness Skin
* Amnion Graft
* Amnion Powder
* Acellular Peritoneum Matrix
* Fascia Lata
* Pericardium
4. Osteochondral Grafts
* Fresh Frozen Allograft
* Allograft Matrix
5. Osseo inductive materials:
* Demineralized Bone Matrix
* Demineralized Bone Paste
* Demineralized Bone Putty
6. Cardiac Valves
* Mitral
* Aortic
7. Value Added Products: Proprietary Formulations and Research-based products
* Bio-Composites
* Pre-sutured grafts for ACL and PCL
* Cellularised bone graft with viable cells
8. Antibiotic Eluting Allograft

Musculoskeletal Tissue Banks (MSTB)

Tissue banks became popular in the mid-twentieth century with the US Navy Tissue Bank in Bethesda [10]. An astute orthopaedic surgeon, George Hyatt, realized the potential of allograft tissues and envisaged this tissue bank. He developed a system for procuring tissues, especially the bones from cadavers, and employed freeze drying to store them for later usage for reconstruction and regeneration. Allografts processing became refined at the tissue bank and enabled their use in diverse clinical settings based on his pioneering work.

Designing Tissue Bank and Meeting Key Objectives

Different countries use different nomenclatures to identify tissue banks. It is a common term for any establishment that procures, processes and stores the human tissues, including cells, for human application or medical research purposes. The alternative name includes Tissue establishment (when the primary aim is a clinical application) and biobank (when the primary intent is research application) (Table 2). The modern-day processing of tissue is often a combination of physical, chemical and biomechanical strategies to achieve the following key objectives:

Table 2 The function of the bank categorized into ten heads

1. Personnel and organization
2. Premise and physical infrastructure
3 .Equipment and consumable materials
4. Standard operating protocols
5. Documentation, coding and data protection
6. Quality control, audits and improvement
7. Traceability, complaints and recalls
8. Donor retrieval program and third-party agreements
9. Research and innovation
10. Fiscal and continuity planning

1. To reduce the risk of transmitting diseases;
2. To decrease the immunogenic response;
3. To ensure and enhance the optimal physiological graft performance;
4. To physically shape the graft to suit specified requirements;
5. To increase the shelf life of the graft.

Key Processes Involved in MSTB

To achieve the 5-cardinal objectives of MTSB, various techniques and processes are involved. They are described below under headings for each of the objectives. Sometimes, a technique is deployed to achieve dual or multiple purposes.

A. Reducing the Risk of Disease transmission.
B. Enhancing the regeneration potential.
C. Lowering immunogenicity.
D. Optimising Shape and Size for clinical usage.
E. Storage and Preservation.
F. Organisational and Personnel.
G. Adherence to statutory regulation.
H. Research & Innovation.
I. Social and Corporate Responsibilities for MSTB.

A. Reducing Risk of Disease Transmission

Any transplanted tissue carries an inherent risk of disease transmission. However, the modern-day processing methodologies and sterilization techniques have ensured that these inherent risks are reduced to a minimum (Table 3). The American Association of Tissue Banks (AATB), in a 2005

Table 3 Tissue recovery procedure with the specified characteristic

Factor	Low	Risk			High
Duration of exposure of procured tissues/cells during procurement	No exposure (closed system)	≤1 h	1–2 h	2–3 h	≥3 h
No. of personnel present while tissues/cells are exposed to the environment	1persons	2–3 persons	4 persons	5 persons	≥6 persons
Reduction of bioburden during or after procurement	Closed system	Validated antibiotic/substances treatment	Only substances intended to reduce microbiological contamination (e.g. glycerol)	Washing intended to reduce microbiological contamination	No reduction
Reduction of bioburden during processing	Validated sterilisation	Substantial microbial reduction	Limited microbial reduction (e.g. antibiotics)	Washing intended to reduce microbiological contamination	No reduction
The risk that contaminants will not be detected in the tissue or cell due to the limitations of the sampling method	Tissue or cells preserved in culture medium (contamination is visible or revealed during microbiological testing of the medium)	Culture of transport media and/or washing solution	A biopsy of tissue tested from each tissue	Swabbing	No detection method
Route of application	Superficial coverage (e.g. corneas, skin, amniotic membrane) or application in the intra-uterine cavity	Durable implant in a poorly vascularized site	Small durable clinical application in a well-vascularised site	Large durable clinical application in a well-vascularised site	Direct application into the blood-stream (infusion)

survey, has shown that this risk of allograft related infection stands below 0.1% [11]. This was further enhanced by the use of Nucleic Acid Testing (NAT) for certain viruses and the provision of terminal sterilization of the tissues. The key lies in ensuring appropriate avoidance, control and reduction of viral and bacterial bioburden during tissue processing [1].

To reduce the risk of disease transmission the following means are adopted globally (Table 3):

a. Reject and discard tissue suspected of harbouring unacceptable bioburden: This is done by a robust donor screening process that eliminates high-risk individuals. It includes a physical examination, serology/ NAT testing, a review of medical records accompanied with a screening questionnaire about travel history, high-risk sexual behaviour, illicit drug use, and aerobic–anaerobic culture, as mentioned in Fig. 1

The following are contraindicated for Deceased Donors:

1. Local Infection: bacterial, viral, mycotic or parasitic
2. Radiation Exposure
3. Malignancy
4. Exposure to toxic substance; ingestion or local injection
5. Metabolic/ Connective Tissue/ Steroid use/ Poor Nutritional Status.
6. Evidence of significant structural damage to the tissues.

b. Ensuring the processing centre has policies and procedures that ensure no contamination during tissue handling: Aseptic handling during recovery and processing helps prevent any contamination, although by itself they may not eliminate the existing bioburden.
c. Use of disinfectants and/or sterilization techniques to ensure near sterile outputs

Bioburden Reduction Methods

Different steps as stand-alone or in various combinations, may be used to reduce the bioburden. These include:

a. Debridement and removal of bone marrow elements, lipids and low molecular weight proteins. This also reduces immunogenicity;
b. Low dose pre-irradiation (before chemical processing);
c. Physical cleaning with water-jets/ pulsatile lavage, centrifuge, fluid bath rotation and sonication;
d. Enzymatic digestion of cellular material;
e. Use of penetrating chemical agents e.g. supercritical carbon dioxide with chemical activators;
f. Disinfecting with mild chemicals like alcohols, detergents, antibiotic solutions.
g. Use of stronger chemicals like NaOH, Hydrogen Peroxide and Acetone. Mainly used for bone tissues but best avoided for soft tissue allografts like tendons.

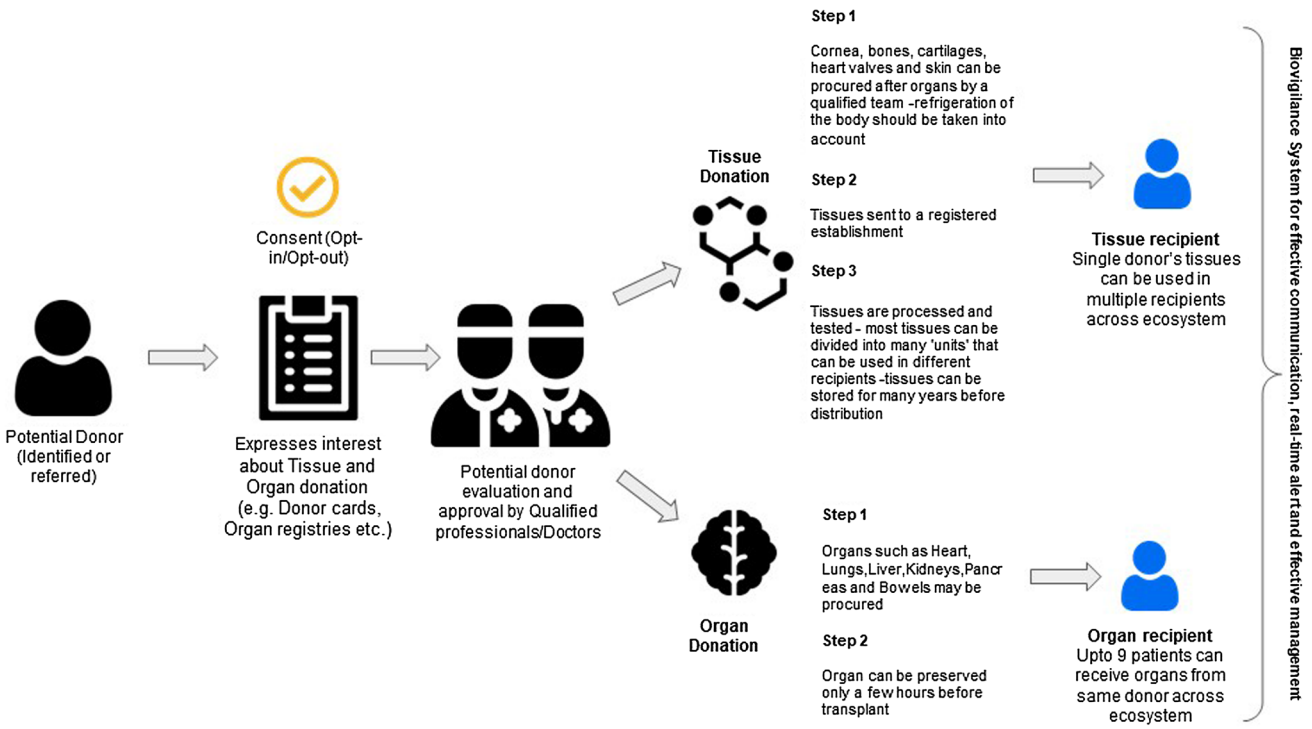

Fig. 1 Graft selection design

Terminal Sterilization Methods

Each of these has its advantages and disadvantages, including the impact on the graft, the cost and the logistics [12].

a. Plasma H_2O_2 [12]
b. Supercritical CO_2 [12]
c. Electron Beam Irradiation [12]
d. Gamma Irradiation [12]

Gamma Irradiation

The use of gamma radiation does not adversely affect the clinical efficacy of grafts, making it the most preferred method of sterilization [13, 14]. There are several advantages to using gamma radiation as a sterilization agent [13]:

1. Its penetrating power enables grafts to be sterilized while being packaged and sealed;
2. The dose delivered is time-dependent requiring only one parameter to be controlled;
3. The products that are irradiated do not become radioactive and can be handled typically.

To maintain the safety and efficacy of the graft, four essential variables need to be considered:

1. Targeted Dose: Depending on bioburden, an irradiation dose of ≥ 25 kGy may be required for sterilization and on a certain occasion, depending on the nature and extent of viral contamination, a dose ≥ 34 kGy may be required for virus inactivation (Ref EDQM 2019).
2. The temperature at irradiation: Irradiating musculoskeletal tissues in the frozen state retains the direct effects of gamma radiation sterilization (breaking covalent bonds by high energy gamma rays) while minimizing the secondary effect of the process (generation of free radicals), thereby reducing damage to the tissue. However, it may also protect micro-organisms [13, 15].
3. Reducing the bioburden before irradiation: It is important to note that a SAL of 10^{-6} can only be achieved if the grafts have a bioburden of less than 1000 CFU per graft. Thus care should be taken to reduce contamination through stringent donor screening, environmental control, aseptic techniques, and decontamination procedures before terminal sterilization [15, 16].

It is important to note that self-contained dry source storage gamma irradiators known as Gamma Chambers or cells used at many research centres, and blood banks are not usually suitable for sterilising grafts as their chambers are too small to accommodate the needs of tissue banks. In India, grafts may be sterilised at Gamma Radiation Processing Plants set up by the Board of Radiation and Isotope Technology (BRIT), an independent Industrial Unit of the Department of Atomic Energy (DAE), Government of India [17].

Sterility Assurance Level (SAL)

While no process can guarantee absolute sterility, the Sterility Assurance Level, commonly referred to as SAL, indicates the degree or level of assurance of sterility expected and achieved through a validated sterilization manoeuvre. It is important to note here that this label may not necessarily mean that the product is sterile but that a sample of the batch of the product was culture-negative during testing. Alternatively, if the tissue is subjected to terminal sterilization as discussed below, the sterilization markers are surrogate indicators of tissue sterility [18, 19]. For allografts, the sterilization process must be validated to give a value of SAL for the product to be labelled 'sterile'. Most SAL labelled product means that the likelihood of non-sterility is 1 in 1 million [19, 20].

B. Enhancing the Regeneration Potential

Studies have clearly shown that for bone regeneration, three critical elements are required: Osteogenic, Osteoinductive and Osteoconductive potentials. Autografts have all three but have the disadvantages of being limited in quantity and running the risk of donor site morbidity [2]. To enhance the regenerative potential, there are certain age recommendations and two key critical process described.

Donor Age Limit

This differs for different types, and in the absence of any validation studies, the limits defined are arbitrary and vary from country to country. A general guideline is as follows for bone [21]. The minimum recommended age for both sexes is six years with no upper age limit. The age range recommended for osteoarticular grafts, cartilage, and menisci is 15–45 years and for tendons and fascia lata is 5–65 years.

Two other key elements to help enhance regeneration potential are:

1. Demineralization: It is established that the Bone morphogenic protein is an important and powerful osteoinductive agent. The bone must be demineralized to the right extent to enhance the bioavailability of this growth factor, as both over and under demineralization lower the osteoinductive potential. Demineralization is usually performed using a dilute (0.5 M or 0.6 M) HCl solution.
2. Cellularization of bone allografts: This is a recent concept and relies on the augmentation of the osteo-

genic potential of the tissue by adding the cells that are adipose or amniotic membrane-derived. However, the approach is still debated as there is a lack of clarity and consensus on how these non-bones derived mesenchymal stem cells will behave in the local milieu and that they are in some manner counterintuitive to the process of lowering immunogenicity described below.

C. Lowering Immunogenicity–Decellularisation

G Burwell, an orthopaedic surgeon [22] from Leeds in the year 1960, did several tests and demonstrated the potential role of bone marrow in the generation of the immune response [22]. He also proved that frozen bone performed better than fresh allogeneic bone tissue because of its lowered immunogenicity. His seminal work paved the way for several bone and tissue banking protocols. Friedlaender carried on with this work and concluded that immunogenicity was dependent on processing and storage, with fresh allogenic being the most immunogenic and frozen bone the least. Freeze-dried had intermediate immunogenicity between the above groups.

It is known that the different types of tissues exhibit different immunogenicity, and to be universally biocompatible, the immunogenicity needs to be decreased to an optimal level. This optimal level would mean that the primary character of the tissue, including its mechanical strength, should not get significantly altered. Certain issues like skin are more immunogenic than others, like bone and tendons. While mere storage and cleaning take care of the lower immunogenic tissues, the higher immunogenicity tissue may require additional treatment. This could be the use of chemical washes or mechanical debridement. Glutaraldehyde has been shown to greatly reduce immunogenicity by cross-linking the antigens [23]. However, a downside of this treatment is the tendency of glutaraldehyde to cause allergic reactions by itself. The decellularization process is particularly helpful for dermal tissues and decreases immunogenicity significantly [24]. The decellularized scaffolds can be used for augmenting the rotator cuff and Achilles tendon repairs. The process may involve the use of anionic agents (Sodium dodecyl sulfate, sodium chloride), Alkali compounds (e.g., sodium hydroxide), and oxidizing agents (e.g., hydrogen peroxide) to dissolve and solubilize the cell-based remnants. The treatment with these agents suffers from a distinct drawback—they tend to adversely affect the strength of the graft by simultaneously removing the collagen and glycosaminoglycans. Thus, their use must be balanced with the requirement of reducing immunogenicity [23, 24].

D. Shaping Allografts for Optimal Usage

While many grafts can be delivered to the operating room without any alteration in shape and size, a few of the grafts need to be customized according to surgical requirements as shown in Fig. 2. This saves the surgeon's effort and valuable operating time. These can be done by simple surgical instruments like osteotomes and chisels or may involve sophisticated machining processes.

Fig. 2 Different shapes of allograft that are contoured as per the need of surgeons

E. Preservation and Storage

The beauty of musculoskeletal tissue banking stems from the fact that unlike organ and composite tissue allografts that need to be transplanted in a few hours, these tissues can be preserved for a very long time running into years [25, 26]. However, to maintain their safety and preserve their efficacy, certain interventions and procedures need to be followed (Table 4). The available methods are:

1. Use of a storage media
2. Refrigeration
3. Cryopreservation
4. Freeze-drying

Key Factors to be considered before choosing optimal preservation and storage method [27–31]:

a. Cell viability
b. Structural integrity and native properties.
c. Cost efficiency
d. The convenience of storage and logistics

Refrigeration

While being one of the simplest ways to preserve tissue, it does require complex logistical arrangements. These refrigerated tissues have a very short shelf life and are thus classified as 'fresh'. Examples of these fresh tissue grafts include osteochondral allografts that help in cartilage restoration around the knee, ankles and shoulder. They are better than the more rigorously processed tissue as a certain number of viable cells are preserved during the process helping in the rapid regeneration of tissues. It is believed that since these cells can be classified as immune-privileged, the transplantation process does not require the use of any immunosuppressant [27, 29]. The key requirements for these fresh products are aseptic recovery during retrieval, microbial, cultures, debridement, disinfection, sizing and treatment with antibiotics. Once the processing is over, the allograft is stored at 1–10 degree Centigrade in quarantine till the time all the serology and microbiology indicator results confirm the sterility [27, 28]. The time of storage is an important factor in their quality as the viability of cells decrease over time. Usually, these grafts must not be stored beyond 60 days, including the quarantine period. As the availability of these fresh grafts improves, their usage and the clinical acumen related to the use of these products too is improving [27–31].

Frozen and Cryopreserved

While refrigerated tissues have a very short shelf life, the process of freezing and cryopreservation can significantly improve the shelf life, and some grafts can be used years later, having been preserved this way. The major drawback is the loss of cell viability as freezing leads to the formation of ice crystals within the cells or extracellular matrix, resulting in their lysis [31, 32]. It is thus best suited for conditions where the main intent is only structural support, as is the case with structural bone grafts, tendons and the acellular dermal matrix. The workflow typically is pathogen testing, debridement, cleaning, disinfection and decellularisation to remove the cells, marrow, fat and decrease bioburden, followed by freezing and storage. In situations where there is a need to protect some tissue viability, such as cellular bone void fillers, osteochondral grafts, cryopreservation solutions may be used. These enable them to be stored for a prolonged period without cell lysis as there is no formation of ice crystals in the process This happens as the water in the cells are replaced by cryo-Preservant, typically glycerol and dimethyl sulfoxide; the allografts are exposed to these cryo-servants and slowly cool down to the cryogenic temperatures. They can then be stored in ultra-low temperature chambers [32, 33].

Freeze-Dried

As cryopreservation is a complex procedure involving logistics and costs, alternate storage methods like the process of freeze-drying, also known as lyophilisation, was developed. With this technique, the graft may be preserved for many years and then rehydrated when needed. The workflow involves cleaning, processing and then using specialized equipment to reduce the residual moisture to a level where the tissue quality is maintained [33, 34]. This varies from tissue to tissue and needs some degree of expertise in the handling of the graft material. A big advantage of this process is that it obviates the need for a special freezer and specialized shipping conditions [35]. On the other hand, the drawback is the need for gradual rehydration and the fact that the tissue may never get fully rehydrated. Even when fully rehydrated, they may not have native properties, and the resultant graft may become brittle and fragile. These may be a significant impediment in their use as a structural graft where the strength of the graft is a critical consideration [36–38].

Use of Preservants

The two main preservants used are ethanol and glycerol. Ethanol has been used since time immemorial for storage

and preservation. Its use has been varied from museums to palaeologists preserving DNAs. Its preservation property is derived from the fact it can derive the water out of the tissue and cause it to dehydrate. Glycerol is a non-toxic, biodegradable liquid that acts as a humectant and just like ethanol, it is widely used in food and beverages, cosmetics and pharmaceuticals [39, 40]. The advantage of using preservants is that they endow the allograft with optimal storage characteristics, specifically eliminating the need for lengthy thawing and rehydration times which in turn translate into ease of shipping and flexibility in the operating room. An additional advantage is that it reduces wastage as there is no prior thawing required, and the surgeons can decide at the very moment whether or not to use the allograft.

Packaging and Labelling

Both the procured and processed allograft tissue should be inspected, identified, packaged and labelled carefully to avoid mix-ups. The packaging should be done in a manner to enable complete isolation and prevent any possibility of external contamination. The packaging could be sterile or non-sterile. The sterile packing is usually blister packed using the blister sealing machine or pouch packed with a validated vacuum–sealing machine under cleanroom conditions. For non sterile packaging—shrink wrapping, or laminar flow packaging may be done to reduce the microbe counts.

F. Personnel and Organization

Tissue banking is a labour-intensive job that requires a dedicated workforce and a high work ethic. The tissue acceptance, body preparation, documentation and tissue recovery area is open 24 h a day. Monitoring the various pre-set temperatures of the tissues stored in each freezer is an important task. Cryogenic technicians who specialize in dealing with cryogenic malfunctions should be available at short notice. The number of staff will vary depending on the scope and volume of the work being done at the institute. They need to be backed up by a retrieval team comprising orthopaedic surgeons and allied specialists. Typically, the bank will need staff under the following heads:

- Departmental Head
- Tissue Transplant Coordinator
- Processing Manager
- Quality Control Manager
- Laboratory Assistants
- Administrative Staff

Equipment and infrastructure: While there are several major and minor equipment required. Some key ones are summarised in the.

G. Tissue Bank Standards and Guidelines

While the initial allografts that were used in clinical setting underwent minimal manipulation in the form of chemical treatment with mild cleansing agents, the modern tissue bank globally are working in standardised optimal condition. These standards and the adopted techniques vary widely and are country/ region-specific. It is recognized by all the personnel working in the field that there is a need to have a defined methodology that ensures that the graft is safe and consistently effective [39]. There is also a need for ensuring appropriate respect and acknowledgement for the tissue donors and their families. One of the most adopted guidelines belongs to the American Association of Tissue Banks (AATB) which was formed in 1976 [41]. The Association in 1984 issued the very first set of official documents containing the processing standards and formulated the guidelines, including those that involved acceptable time-lapse from mortality to recovery, the storage standards, need and standards of microbiological testing, defining the demineralization methods, storage methodology like freeze-drying etc. The official US government intervention came in through the US food and Drug Administration (FDA) classifying Human Cell and Tissue Products (HCT/Ps) as a stand-alone category separate from other medical devices [41–44]. Most musculoskeletal tissue bank products fall under the "minimal manipulation' category implying that 1. The intended use is homologous: meaning the tissue is used clinically in a manner akin to that intended in the body of origin, and 2. The original relevant characteristic has not been altered. As the tissue banks become more innovative and experiment with combination therapies combining the artificial product with a natural product and enhancing the processing techniques—the definition of minimal manipulation is likely to get tested. For the moment, the standards laid down by the AATB and FDA serves as general guidelines to establish good practices and ensure safe practices all across the globe [45, 46]. In India, the national body (NOTTO), and the regional organization (ROTTO) are mandated to formulate the guidelines for the smooth functioning of tissue banks.

H. Research and Innovation- the Way Forward

Much research and several innovations are happening in the field of musculoskeletal tissue banking and most of these are focused on process improvisation. Beyond these, however, two critical areas of research and innovation leading to value addition are the development of newer bio-composites and the development of antibiotic eluting allografts.

Bio-composites

Bio-composites refer to a unique genre of products wherein the allograft are cross-linked with biodegradable polymers resulting in the enhancement of their structural strength and improvisation in their biomechanical properties and development of favourable immunogenic profile [42]. While several studies are in the preclinical stage in this arena, there is an expectation that the future of orthopaedic traumatology lies in the amalgamation of polymers with the natural grafts [43, 44].

Antibiotic Eluting Allografts

A biodegradable product that elutes antibiotics is likely to solve many problems in reconstruction. Impregnation of cancellous bones with antibiotics to ensure higher local levels of antibiotics that help eliminate biofilms is an attractive proposition [47]. The simplest way to make an antibiotic eluting, allograft is to mix the antibiotic solution or powder with the graft. However, the elution is unpredictable and short term. To circumvent this, various techniques have been devised, and these include iontophoresis, coating the bone with alginate, altering the bone surface with EDTA and addition of a linker and use of additives such as bee wax and glycerin. The choice of antibiotic is also a subject of interest, and amongst various studies, it has been shown that Vancomycin is the least cytotoxic, followed by Amikacin and Tobramycin. The antibiotics that caused the most cell damage are Rifampicin, Colistin, Ciprofloxacillin and Doxycycline [46, 48]. A concentration > 200 µg/ml of these antibiotics negatively impacted the cell numbers in the area [47, 49].

Key Areas for future development in the field of Tissue Banking are:

a. Preserved/Increased cell viability and strength of grafts.
b. Development of efficient and less toxic sterilization process.
c. Increase in shelf life of tissues.
d. Faster, accurate and efficient detection of Microbes.
e. Rapid tests for deceased and living donors.
f. Addition of antimicrobials to reduce chances of infection.
g. Isolating relevant peptides from the grafts that promote regeneration e.g. BMP.
h. Development of expandable allograft to fill the voids.
i. Allografts as a local antibiotic delivery device.
j. Creation of bio-composites using crosslinking methods.

I. Social Activity and Responsibility

The success of the tissue bank is measured by the number of tissues recovered from a deceased donor. The availability of suitable deceased donors is an important limiting factor. P6Ignorance, misconception, fear of mutilation of the body, religious beliefs and grief, etc. are usually the reasons for refusing consent. Massive awareness campaigns about tissue donation and its lifesaving and life enhancing potential should be carried out at regular intervals to encourage active community participation. The involvement of print and electronic media, community leaders, religious preachers, representatives of the people and social organizations can mobilize massive support for tissue donation. Public congratulations, praise for the noble gesture, and public awards to families who have donated tissues from their deceased relatives will go a long way in mobilizing the masses.

Current Situation of MSTBs in INDIA

In India, setting up a tissue bank can be challenging. In addition to financial constraints and inadequate infrastructure and personnel, religious and social restrictions limit tissue donation. A schematic step-by-step flow chart depicting the various steps in obtaining registration by the licensing authority has been added (Fig. 3). Form 14, which is required to apply for the proposal of registration of any tissue bank, is also appended (Supplementary) [52].

The recent establishment of the Regional cum State Organ and Tissue Transplant Organization (ROTTO-SOTTO) by the Ministry of Health and Family Welfare, Government of India, as stipulated by THOTA, 1994 [51]. These government organisations promote and regulate organ and tissue donation from deceased individuals. The functions of these organizations are mentioned in Table 5.

Conclusion

Allografts and tissue bank are likely to become a part of mainstream orthopaedic very soon. The enthusiasm expressed by early adopters from the late eighteenth

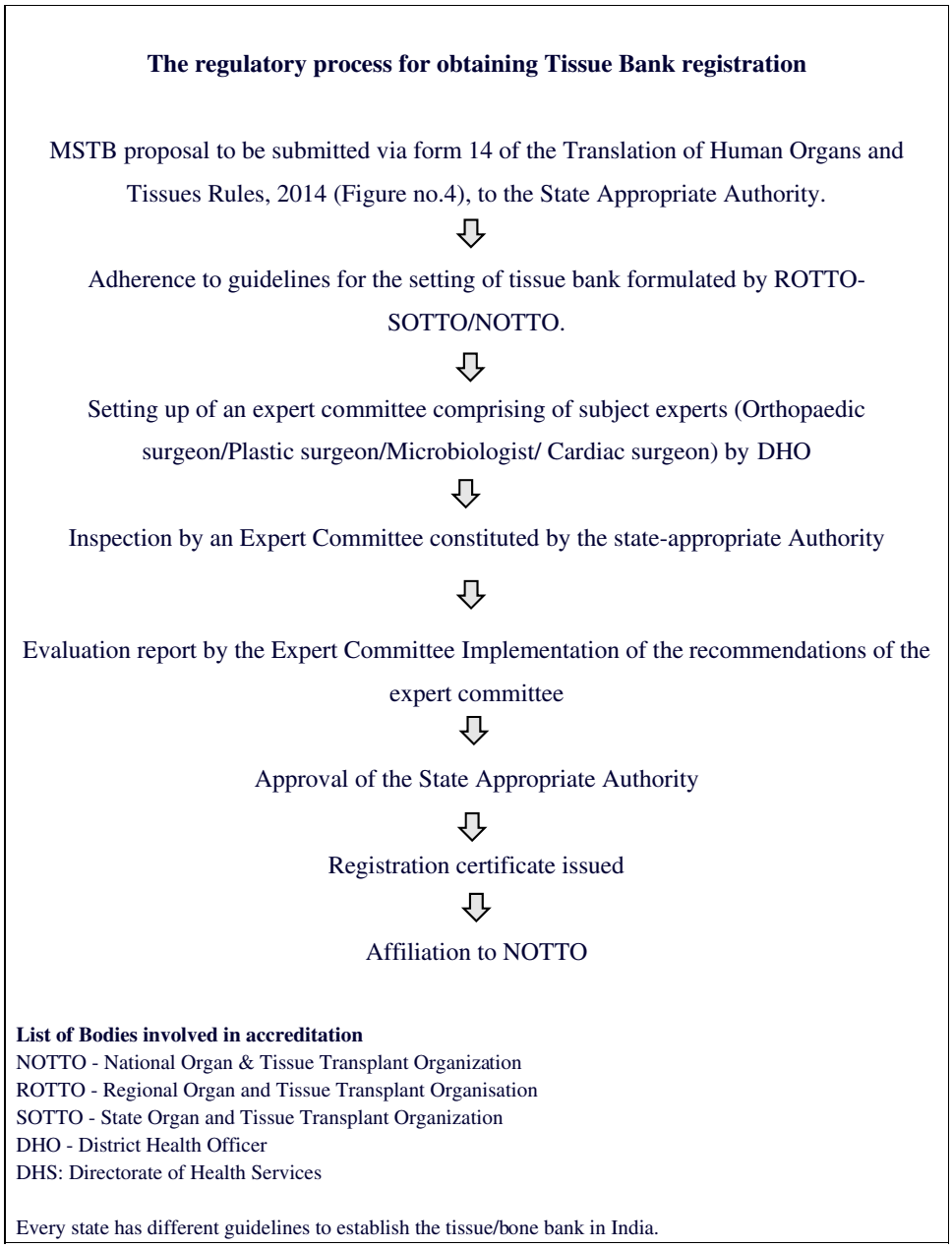

Fig. 3 Steps to establish a tissue/bone bank in Maharsthara state India

century has been transmitted to the early twenty-first century surgeons who find it an important element in their regenerative portfolio. Today the majority of their use is confined to anatomical substitutes in the form of bones, tendons, skin and other soft tissues. With innovation, they have been modified and are currently available as injectable, coatings, putties and cellular therapy besides their original structural configurations. For the field to grow to its full potential—all aspects of banking need improvisation, including procurement, sterilization, processing as well as preservation. The future research enabling biologically interactive constructs and amalgamation of newer technologies of genetic alterations, cell seeding, protein isolation and reconstitution methods are on the horizon. The future for musculoskeletal tissue banking is indeed bright!

Table 4 Allograft preservation and storage methods

Preservation method	Storage temperature	Advantage	Potential disadvantages
Fresh	Refrigerated at 1–10 °C	Viable cells	Complex logistics Limited shelf life
Frozen	Frozen at − 40 to – 80 °C	Fully hydrated	Thaw time
			Need validated and monitored freezer for storage
		Long shelf life	Shipping costs
Cryopreserved	Liquid nitrogen (LN2) or − 80 °C	Cell viability Maintains biomechanical properties	Need validated and monitored freezer or LN2 tanks to store on site
		Long shelf life	Shipping costs
Freeze-dried	Ambient temperature	Easy storage	Altered biochemical properties
		Long shelf life	Long rehydration time
Preserving treated	Ambient temperature		Reaction to preservation solution in susceptible patients
		Fully hydrated	Reaction to preservation solutions in susceptible patients
		Easy storage	Potential tissue alterations if the solution is dehydrating
		Long shelf life	
		More options in the operating room	
		Maintains biochemical properties	
		Easy storage	
		Long shelf life	
	Ambient temperature	More options in the operating room	

Table 5 Role of ROTTO (Regional Organ and Tissue Transplant Organisation) and SOTTO (State Organ and Tissue Transplant Organization)

ROTTO	SOTTO
Monitoring and surveillance of transplantation	Maintaining patient waiting lists for organ transplants
Collection of statistics from SOTTO	Facilitating multi-organ and tissue retrieval from a brain stem death donors
Maintaining data bank including organ transplant follow up	Coordination from procurement of organs from a donor till transplantation into a recipient
Operating various schemes for organ n, donor health check-ups	Matching of organ recipients with donor and organ allocation
Organizing and conduct training programs	Dissemination of information to hospitals, organizations, and individuals
Creating awareness for deceased organ and tissue donation, Dissemination of information	Post-organ transplant patients and liveinge donors' follow-up for assessment of graft rejection, survival rates
Organizing interactive oral meetings and advocacy workshops for the Region and States	Collection of data for ROTTO

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Declarations

Conflict of Interest The authors declare that they have no conflict of interest.

Ethical Approval This article does not contain any studies with human or animal subjects performed by the any of the authors.

Informed Consent For this type of study informed consent is not required.

References

1. Cobb & Douglas, Recommendations for reducing the risk of disease transmission in a swimming pool. Public Health, Reducing Risk of Disease Transmission

2. Koelling, S., & Miosge, N. (2009). Stem cell therapy for cartilage regeneration in osteoarthritis. *Expert Opinion on Biological Therapy, 9*(11), 1399–1405. https://doi.org/10.1517/14712590903246370
3. Amillo, S., Cara, J. A., & Valenti, J. R. (1990). Tissue bank for the musculoskeletal system. Clinical applications. *Revista de Medicina de la Universidad de Navarra, 34*(4), 227–34.
4. Donati, D., Zolezzi, C., Tomba, P., & Viganò, A. (2007). Bone grafting: Historical and conceptual review, starting with an old manuscript by Vittorio Putti. *Acta Orthopaedica*. https://doi.org/10.1080/17453670610013376
5. Beer, A. J., Tauro, T. M., Redondo, M. L., Christian, D. R., Cole, B. J., & Frank, R. M. (2019). Use of allografts in orthopaedic surgery: Safety, procurement, storage, and outcomes. *Orthopaedic Journal of Sports Medicine, 7*(12), 2325967119891435.
6. Deborah Kotz. University of Maryland School of Medicine Faculty Scientists and Clinicians Perform Historic First Successful Transplant of Porcine Heart into Adult Human with End-Stage Heart Disease. University of Maryland School of Medicine 2022
7. Cerulli, G., Placella, G., Sebastiani, E., Tei, M. M., Speziali, A., & Manfreda, F. (2013). ACL reconstruction: Choosing the Graft. *JOINTS, I*(1), 18–24.
8. Noyes, F. R., Butler, D. L., Grood, E. S., Zernicke, R. F., & Hefzy, M. S. (1984). Biomechanical analysis of human ligament grafts used in knee ligament repairs and reconstructions. *Journal of Bone and Joint Surgery. American Volume, 66*(3), 344–352.
9. Roberts, T., & Rosenbaum, A. (2012). Bone grafts, bone substitutes and orthobiologics. *Organogenesis, 8*(4), 114–124. https://doi.org/10.4161/org.23306
10. Strong, D. M. (2000). The US Navy Tissue Bank: 50 Years on the cutting edge. *Cell and Tissue Banking, 1*(1), 9–16. https://doi.org/10.1023/A:1010151928461
11. Moore, M. A., Samsell, B., & McLean, J. (2018). Allograft tissue safety and technology. *Biologics in Orthopaedic Surgery, 2019*, 49–62. https://doi.org/10.1016/B978-0-323-55140-3.00005-9
12. Lambert, B. J., Mendelson, T. A., & Craven, M. D. (2011). Radiation and ethylene oxide terminal sterilization experiences with drug-eluting stent products. *AAPS PharmSciTech, 12*, 1116–1126.
13. Singh, R., Singh, D., & Singh, A. (2016). Radiation sterilization of tissue allografts: A review. *World Journal of Radiology, 8*(4), 355–369.
14. Nguyen, H., Morgan, D. A., & Forwood, M. R. (2007). Sterilization of allograft bone: Effects of gamma irradiation on allograft biology and biomechanics. *Cell and Tissue Banking, 8*, 93–105.
15. Anderson, M. J., Keyak, J. H., & Skinner, H. B. (1992). Compressive mechanical properties of human cancellous bone after gamma irradiation. *Journal of Bone and Joint Surgery American, 74*, 747–752. [PMID: 1624490].
16. Nguyen, H., Morgan, D. A., & Forwood, M. R. (2007). Sterilization of allograft bone: Is 25 kGy the gold standard for gamma irradiation? *Cell and Tissue Banking, 8*, 81–89.
17. Board of Radiation and Isotope Technology (BRIT) harnessing nuclear radiation for mankind. Department of Atomic energy. Government of India. Board of Radiation & Isotope Technologies. https://britatom.gov.in/
18. Food and Drug Administration. Guidance for Industry: Validation of Procedures for Processing of Human Tissue Intended for Transplantation, Center for Biologics Evaluation and Research. Guidance for Industry. March 2002. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/validation-procedures-processing-human-tissues-intended-transplantation
19. LK Archibald, DB Jernigan. Update: Allograft-Associated Bacterial Infections---United States, 2002 MMWR, 2002:51:207–210. https://www.cdc.gov/mmwr/preview/mmwrhtml/mm5110a2.htm
20. Sterilization of health care products — Radiation — Part 1: Requirements for development, validation and routine control of a sterilization process for medical devices ANSI/AAMI/ISO 11137: 2006. Sterilization of health care products. ISO https://www.iso.org/standard/33952.html
21. Hovanyecz, P., Lorenti, A., Lucero, J. M. J., et al. (2015). Living donor bone banking: Processing and discarding—from procurement to therapeutic use. *Cell and Tissue Banking, 16*, 593–603. https://doi.org/10.1007/s10561-015-9507-8
22. Britain British orthopaedic association. Proceedings and reports of universities colleges, councils and associations great spring meeting The Journal of Bone and Joint Surgery 1974
23. Xu, K., Kuntz, L. A., Foehr, P., Kuempel, K., Wagner, A., Tuebel, J., Deimling, C. V., & Burgkart, R. H. (2017). Efficient decellularization for tissue engineering of the tendon-bone interface with preservation of biomechanics. *PLoS One, 12*(2), e0171577.
24. Lovati, A. B., Bottagisio, M., & Moretti, M. (2016). Decellularized and engineered tendons as biological substitutes: A critical review. *Stem Cells International, 2016*, 7276150.
25 Moore, M. A., Samsell, B., & McLean, J. (2019). Allograft tissue safety and technology. *Biologics in Orthopaedic Surgery, 2019*, 49–62.
26. de Roeck, N. J., & Drabu, K. J. (2001). Impaction bone grafting using freeze-dried allograft in revision hip arthroplasty. *Journal of Arthroplasty, 16*(2), 201–206.
27. Garrity, J. T., Stoker, A. M., Sims, H. J., & Cook, J. L. (2012). Improved osteochondral allograft preservation using serum-free media at body temperature. *American Journal of Sports Medicine, 40*(11), 2542–2548.
28. Teng, M. S., Yuen, A. S., & Kim, H. T. (2008). Enhancing osteochondral allograft viability: Effects of storage media composition. *Clinical Orthopaedics and Related Research, 466*(8), 1804–1809.
29. Ball, S. T., Amiel, D., & Williams, S. K. (2004). The effects of storage on fresh human osteochondral allografts. *Clinical Orthopaedics and Related Research, 418*, 246–252.
30. Cook, J. L., Stoker, A. M., & Stannard, J. P. (2014). A novel system improves preservation of osteochondral allografts. *Clinical Orthopaedics and Related Research, 472*(11), 3404–3414.
31. Pegg, D. E. (2015). Principles of cryopreservation. *Methods in Molecular Biology, 1257*, 3–19.
32. Song, Y. C., Khirabadi, B. S., Lightfoot, F., Brockbank, K. G., & Taylor, M. J. (2000). Vitreous cryopreservation maintains the function of vascular grafts. *Nature Biotechnology, 18*(3), 296–299.
33. de Roeck, N. J., & Drabu, K. J. (2001). Impaction bone grafting using freeze-dried allograft in revision hip arthroplasty. *Journal of Arthroplasty, 16*(2), 201–206.
34. Pinkowski, J. L., Reiman, P. R., & Chen, S. L. (1989). Human lymphocyte reaction to freeze-dried allograft and xenograft ligamentous tissue. *American Journal of Sports Medicine, 17*(5), 595–600.
35. Lasanianos, N., Mouzopoulos, G., & Garnavos, C. (2008). The use of freeze-dried cancellous allograft in the management of impacted tibial plateau fractures. *Injury, 39*(10), 1106–1112.
36. Pinkowski, J. L., Reiman, P. R., & Chen, S. L. (1989). Human lymphocyte reaction to freeze-dried allograft and xenograft ligamentous tissue. *American Journal of Sports Medicine, 17*(5), 595–600.
37. Burchardt, H., Glowczewskie, F., & Miller, G. (1987). Freeze-dried segmental fibular allografts in azathioprine-treated dogs. *Clinical Orthopaedics and Related Research, 218*, 259–267.
38. Graham, R. S., Samsell, B. J., & Proffer, A. (2015). Evaluation of glycerol-preserved bone allografts in cervical spine fusion: A

prospective, randomized controlled trial. *Journal of Neurosurgery. Spine, 22*(1), 1–10.
39. Strong, D. M. (1998). The contribution of Kenneth W. Sells to the progress of tissue banking. In G. O. Philips, R. Versen, D. M. Strong, & A. Nather (Eds.), *Advances in tissue banking* (Vol. 2, pp. 1–19).
40. Strong, D. M., & The, U. S. (2000). Navy tissue bank: 50 years on the cutting edge. *Cell and Tissue Banking, 1*, 9–16.
41. Tognetti, L., Pianigiani, E., Ierardi, F., Mariotti, G., Perrotti, R., Di Lonardo, A., et al. (2017). Current insights into skin banking: Storage, preservation and clinical importance of skin allografts. *Journal of Biorepository Science for Applied Medicine, 5*, 41–56.
42 Oyce, M. J. (1998). American Association of Tissue Banks—the present and future. In G. O. Philips, R. Versen, D. M. Strong, & A. Nather (Eds.), *Advances in tissue banking* (Vol. 2, pp. 29–35). Singapore: World Scientific.
43. Song, R., Murphy, M., Li, C., Ting, K., Soo, C., & Zheng, Z. (2018). Current development of biodegradable polymeric materials for biomedical applications. *Drug Design, Development and Therapy, 12*, 3117–3145.
44. Geiger, M., Rh, L., & Friess, W. (2003). Collagen sponges for bone regeneration with rhBMP-2. *Advanced Drug Delivery Reviews, 55*(12), 1613–1629.
45. Jovanovic, S. A., Hunt, D. R., Bernard, G. W., Spiekermann, H., Wozney, J. M., & Wikesjö, U. M. E. (2007). Bone reconstruction following implantation of rhBMP-2 and guided bone regeneration in canine alveolar ridge defects. *Clinical Oral Implants Research, 18*(2), 224–230.
46. Neut, D., van De Belt, H., Stokroos, I., van Horn, J. R., van Der Mei, H. C., & Busscher, H. J. (2001). Biomaterial-associated infection of gentamicin-loaded PMMA beads in orthopaedic revision surgery. *Journal of Antimicrobial Chemotherapy, 47*, 885–891.
47. Nelson, C. L., Jones, R. B., Wingert, N. C., Foltzer, M., & Bowen, T. R. (2014). Sonication of antibiotic spacers predicts failure during two-stage revision for prosthetic knee and hip infections. *Clinical Orthopaedics and Related Research, 472*, 2208–2214.
48. Winkler, H., & Haiden, P. (2017). Allograft bone as antibiotic carrier. *Journal of Bone and Joint Infection, 2*(1), 52–62.
49. Tunney, M. M., Ramage, G., Patrick, S., Nixon, J. R., Murphy, P. G., & Gorman, S. P. (1998). Antimicrobial susceptibility of bacteria isolated from orthopaedic implants following revision hip surgery. *Antimicrobial Agents and Chemotherapy, 42*, 3002–3005.
50. Government of India. The Gazette of India, Extraordinary Part II-Section I. vol. 421994. p. 1-13. 53. https://egazette.nic.in/WriteReadData/1956/E-2176-1956-0055-99246.pdf
51. National organ and tissue transplant organization under the aegis of NOTP directorate general of health services ministry of health and family welfare government of India. Ministry of Health & Family Welfare, Government of India. https://notto.gov.in/download-forms.htm
52. Application for registration of tissue bank other than eye bank. https://notto.gov.in/WriteReadData/Portal/images/FORM14.pdf