Experience with Tissue Bank Services in 2014 and 2020 in Turku, Finland

Santtu Tikkala[a*], Kari Tirkkonen[b], Elina Ekman[b], and Kaisa Lehtimäki[b]

[a]University of Turku, Turku, Finland; and [b]Department of Orthopaedic Surgery, Turku University Hospital, Turku, Finland

ABSTRACT

Background. The objective of a musculoskeletal tissue bank is to collect, test, store, and provide musculoskeletal tissue allografts required in orthopedic procedures. Strict exclusion criteria are followed when selecting suitable cadaver musculoskeletal tissue donors, and the allografts are procured under sterile conditions to avoid bacterial contamination. Tissue banking in Turku, Finland, began in 1972, and tissue bank services were last reviewed in 2003. This study aimed to review the operation of the musculoskeletal tissue bank in Turku, Finland, between 2014 and 2020 and to analyze the number, types, and contamination rate of the allografts procured from the cadaver donors. Potential donor-related factors causing bacterial contamination of the allografts and whether potential musculoskeletal tissue donors were overlooked among multiorgan donors were also studied.

Methods. A retrospective review of all cadaver musculoskeletal tissue donors used in the Hospital District of Southwest Finland Tyks Orto Musculoskeletal Tissue Bank during the study period was conducted, and data on the procured allograft was collected and presented. The donors were selected among patients treated in the intensive care unit (ICU) of Turku University Hospital (TYKS).

Results. A total of 28 cadaver donors were used, and 636 allografts were procured between 2014 and 2020. The bacterial contamination rate was 2.5%, which was lower than that in the previous international literature. The median treatment time in the ICU was significantly longer, and the median value of the highest C-reactive protein level was significantly higher in the group of donors with positive allograft bacterial cultures.

Conclusions. The bacterial contamination rate in the tissue bank was low on an international scale. Some suitable musculoskeletal tissue donors were overlooked among multiorgan donors.

THE use of musculoskeletal tissue allografts is common in orthopedic surgery, and the need for allografts is constantly growing [1−3]. Allografts are procured from living and cadaver donors, and procured allografts are stored in tissue banks. Tissue banks are responsible for collecting, testing, storing, and distributing allografts. Allografts are considered suitable materials in reconstructive orthopedic surgery, but the risk of infection due to transmissible diseases and allograft contamination exists [4,5]. Strict donor selection and serologic screening tests, together with aseptic procurement methods and microbiological sampling of allografts, are followed to provide high-quality allografts and prevent infections in allograft recipients.

Musculoskeletal tissue banking for clinical purposes in Turku, Finland, began in 1972, and the first allograft transplantation took place in 1973 [6]. Initially, only bone allografts were stored, and ligament grafts were eventually added to the selection. Aho et al [7] and Virolainen et al [6] have previously reviewed tissue bank services in Turku. Since then, the operation of tissue banks has developed markedly. Currently, stored tissues include long bones, tendons, the meniscus, and fresh osteochondral allografts. Administration changes have also been made. On January 29, 2014, the Finnish Red Cross gave up the administration of tissue banks, and the Hospital District of Southwest Finland (VSSHP) took charge. This organization is now called the VSSHP Tyks Orto Musculoskeletal Tissue Bank.

*Address correspondence to Santtu Tikkala, MD, University of Turku, Kiinanmyllykatu 4-8, Turku, Finland, 20014. E-mail: saakti@utu.fi

0041-1345/20
https://doi.org/10.1016/j.transproceed.2023.09.030

This study aimed to review the operation of VSSHP Tyks Orto musculoskeletal tissue bank between 2014 and 2020. The number of procured musculoskeletal tissue allografts and the allograft bacterial contamination rate were reported. The results were compared to previous reviews of tissue bank services in Turku, Finland, and the bacterial contamination rate was also compared to previous international literature. Special interest was focused on cadaver musculoskeletal tissue donors to examine possible donor-related factors causing bacterial contamination of the allografts and whether some potential musculoskeletal tissue donors were overlooked among multiorgan donors.

MATERIALS AND METHODS

A retrospective review of all cadaver multiorgan donors treated in the intensive care unit (ICU) of the Turku University Hospital (TYKS) between January 1, 2014, and December 31, 2020 was conducted. The medical history of each donor was studied, and the following donor-related factors were analyzed: age, sex assigned at birth, cause of death, time treated in the ICU, antibiotic therapy during treatment in the ICU, the highest C-reactive protein (CRP) level during treatment period, and positive bacterial growth in tissue samples. Data on the allografts procured from cadaver donors were collected from the Tissue DB database (BCB Medical Ltd). As we aimed to study cadaver donors, femoral heads collected from living donors were not included in our data. The study was approved by the Regional Ethical Review Board of Turku (Dnro TO1/022/2020).

Operating Principles, Administration, and Control

The primary objective of the VSSHP Tyks Orto musculoskeletal tissue bank is to secure the sufficiency of safe allografts for hospitals in the Southwest Finland region. Allografts are also sent to other regions in Finland when needed. Ethical principles are highly maintained. Tissue donation is free of charge, the working principles follow the assumed agreement of the donor, and permission from the next of kin is always requested. The Finnish Medicines Agency is the authority supervising tissue banking in Finland. Operational guidelines are described in the quality manual of the tissue bank, and the operating principles comply with national and European legislation. Guidelines and documents created by the European Association of Tissue Banks, the Council of Europe, and the Finnish Orthopedic Association concerning the operation of the tissue bank are also included.

Donor Selection, Tissue Retrieval, and Microbiological Testing

Musculoskeletal tissue donors are selected amongst brain-dead multiorgan donors. Special attention is paid to the donor criteria because of the risk of transmissible diseases [5], and the contraindication list is strict for musculoskeletal tissue donation (Table 1). Cadaver musculoskeletal tissue donors are typically previously healthy persons aged 16 to 65 years who have died of a sudden cerebral death. The suitability of the cadaver donor is evaluated by an orthopedic surgeon performing tissue procurement together with a tissue banking coordinator. The contraindication list is examined utilizing the medical records and physical examination of the donor, and a possible hemodilution is observed. A donor's suitability for tissue donation is documented in the medical records. Blood samples from each donor are screened for the presence of transmissible diseases. Serologic screening tests are performed to detect the human immunodeficiency virus (S-HIVAgAb and P-HIV1Nh), hepatitis B (S-HBsAg, S-HBcAb, and S-HBVNhO), hepatitis C (S-HCVAb and P-HCVNhO), and *Treponema pallidum* (S-TrpaAb). A positive test result leads to the discarding of all procured allografts. The procurement of allografts is performed less than 12 hours after the expiration of the donor's blood circulation.

The procurement and handling of the allografts are performed aseptically in an operating room by a surgical team. If signs of contamination appear at any stage of the procurement process, a possibly contaminated graft is immediately rejected. The grafts are procured in a predetermined order, one graft at a time. Soft tissues are removed from around the grafts, and bones are separated into parts when needed. Bone marrow is scooped empty, and the grafts are washed with a pressure lavage using a sterile saline solution. Microbiological samples are taken after the washing. Three tissue biopsy samples are taken from each allograft. The samples are cultured for both aerobic and anaerobic bacterial and fungal growth. Microbiological testing is executed in qualified, noted, and accredited laboratories using validated methods. The grafts are approved only if all cultures are negative.

Table 1. Inclusion and Exclusion Criteria for Musculoskeletal Tissue Donation

Inclusion criteria
Assumed agreement of the donor and permission of the next of kin
Age:
Under 65 y for bones and tendons
Under 40 y for meniscus and FOCA-grafts
Exclusion criteria
Multiple injury trauma or a vast tissue damage
Hemodilution over a half of total plasma volume
Generalized infection: sepsis, tuberculosis, systemic viral, fungal or parasite infection
Unclear cause of death or unknown disease
Previous or active HBV or HCV infection
Syphilis
HIV or HTLV infection
Skin jaundice
Previous or active malignancy
Osteoporosis
Chronic autoimmune disease
Long term use of corticosteroids or other immunosuppressive therapy
Chronic neurologic disease
Use of human origin pituitary hormone
Previously received tissue transplant
Born in foreign country or foreign descent
Vaccinated with live-attenuated vaccine within 6 wk
Carriage of resistant bacterium
Local infection in tissue procurement area
Risk group of HIV or hepatitis
Abnormal sexual history
History of drug or alcohol abuse
Exposure to chemical or heavy metal
Intoxication
Imperfect skin at procurement area: damaged or infected, large tattoos, suspicious moles, needle pricks (other than hospital origin)
Positive test for COVID-19 (since 2019)

HBV, hepatitis B virus; HCV, hepatitis C virus; HTLV, human T-lymphotropic virus type.

Storage and record keeping. After microbiological sampling, the grafts are packed in two-fold sterile plastic packages or plastic jars. Each graft is packed straight after sampling, and no more than one graft is processed at a time. Each package is labeled and marked to identify the donor and type and size of the allograft, blood type of the donor, date of procurement, and expiration date of the allograft. The allografts are frozen within 12 hours after procurement. In practice, freezing is done immediately after packaging. Frozen allografts are stored at a temperature of -75°C. Fresh osteoarticular grafts are stored in an antibiotic solution at refrigerator temperature (2-8°C) for no more than 21 days.

Data on the used allografts are preserved for 30 years after clinical usage of the graft. All data related to the donation of allografts are preserved. Each graft is marked with a tissue identification number. By entering the identification number into the database, all information concerning the allograft and the donor is available, if necessary.

Clinical use of allografts. The classical sites for the usage of allografts are bone defects caused by bone tumors or trauma, spinal fusion surgery, and filling of a cavity in revision arthroplasties. Tendon grafts are used in reconstructive or reinsertion surgery of ligaments and tendons in the upper and lower limbs (Figs 1 and 2).

Statistical Analysis

Variables were summarized with descriptive statistics. For analysis, the donors were categorized into two groups: positive tissue bacterial cultures and no positive tissue bacterial cultures. Associations between the bacterial contamination and the variables were studied one by one with the Kruskal−Wallis test and Fisher exact test (for categorical variables).

The normality of variables was evaluated visually and tested with the Shapiro−Wilk test. Due to the non-normality of continuous variables, nonparametric methods were used. All tests were performed as two-sided with a significance level set at $P < .05$. The analyses were performed using the SAS System, version 9.4 for Windows (SAS Institute Inc).

RESULTS

During the seven-year study period from 2014 to 2020, a total of 28 musculoskeletal tissue donors were used, and 636 allografts were procured in the VSSHP Tyks Orto tissue bank. The most frequently procured allografts were the femur, tibia, and tibialis anterior and posterior tendons. Detailed information on the donors and tissue allografts is presented in Table 2. The number of procured allografts per donor varied from 10 to 34 grafts, and the mean number of procured allografts was 23. The annual number of procured allografts varied from 22 to 138,

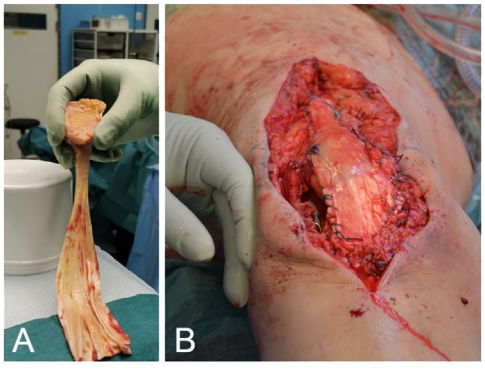

Fig 1. (A) A detached Achilles tendon allograft. **(B)** Achillies tendon allograft used in a knee surgery.

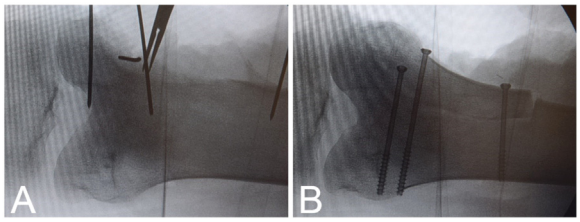

Fig 2. (A) A bone defect in the distal femur. **(B)** Bone defect of the distal femur filled with a bone allograft.

Table 2. Donor and Tissue Details

Donor details	N	
Donors	28	
Male (%)	13	(46)
Female (%)	15	(54)
Mean age (y)	51.4	(Range 16-65)
Cause of death (%)		
Subarachnoid hemorrhage	19	(67.8)
Subdural hematoma	3	(10.7)
Intracerebral hemorrhage	3	(10.7)
Crush injury of skull	1	(3.6)
Ischemic stroke	1	(3.6)
Hypoxic-ischemic brain injury	1	(3.6)
Traumatic cause of death	5	(17.9)
Distribution of allografts	**N**	
Total	636	
Used (%)	370	(58)
Sent to other organizations (%)	51	(8)
Discarded (%)	49	(8)
In store (%)	166	(26)
Types and numbers of allografts (%)	**N**	
Femur (strut, diaphysis or distal)	110	(17.3)
Tibia (distal or diaphysis)	83	(13.1)
Tibialis anterior tendon	56	(8.8)
Tibialis posterior tendon	56	(8.8)
Achilles tendon	55	(8.7)
Patella (Bone-tendon-bone)	46	(7.2)
Humerus (distal, diaphysis or proximal)	32	(5.0)
Fibula	29	(4.6)
Extensor Digitorum Longus	25	(3.9)
Semitendinosus tendon	24	(3.8)
Radius	16	(2.5)
Flexor hallucis longus tendon	13	(2.0)
Gracilis tendon	13	(2.0)
Extensor hallucis longus tendon	11	(1.7)
Flexor hallucis longus tendon	11	(1.7)
Flexor digitorum longus tendon	6	(0.9)
Ulna	5	(0.8)
FOCA	5	(0.8)
Meniscus	1	(0.2)
Quadriceps tendon	1	(0.2)
Other	38	(6.0)
Total	636	(100)

FOCA, Fresh osteochondral allograft.

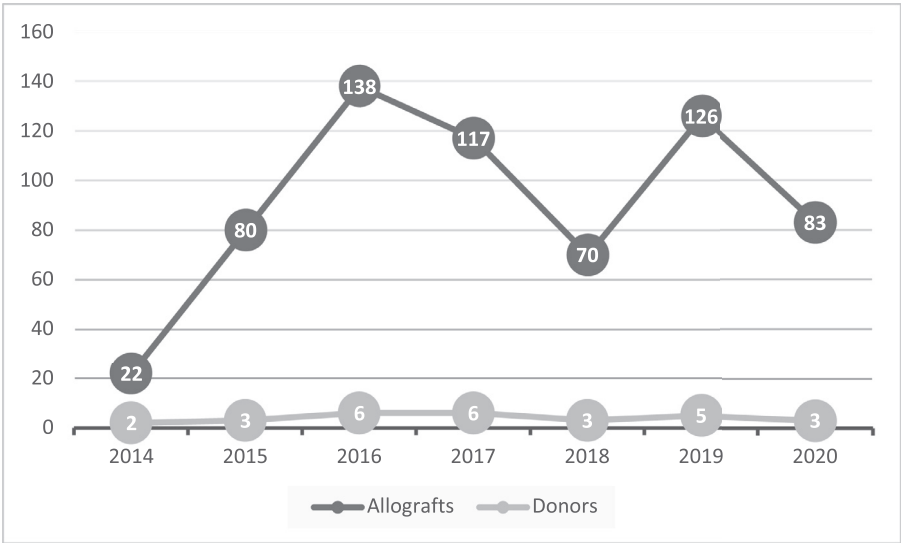

Fig 3. The number of harvested allografts and used cadaver donors yearly.

and the mean number of procured allografts was 91 grafts per year. The number of donors varied from 2 to 6 per year (Fig 3).

The total number of discarded allografts was 49, accounting for 7.7% of all procured grafts. The causes for discarding were as follows: 21 (3.3%) outdated the five-year storage time, 16 (2.5%) were positive for bacterial cultures, 5 (0.8%) grafts were melted for usage but were not used during the operation, 4 (0.6%) melted during storage, 2 (0.3%) were discarded because of poor graft quality, and 1 (0.16%) had an incorrect graft size.

At least one bacterial culture was positive in 16 allografts. Two different bacterial species were identified in 3 grafts; thus, in total, 19 positive bacterial cultures were found. Bacterial contamination was the cause of discarding in 32.7% of all discarded cases. Seven different bacterial species were identified, and the bacteria were divided into low and high virulence. Low-virulence Staphylococcus species were most frequently cultured. High-virulence pathogens were found in 2 samples, both taken from the same donor. The species and numbers of bacteria are summarized in Table 3.

Table 3. The Species and Number of Cultured Bacteria from Harvested Allografts in the VSSHP Tyks Orto Tissue Bank 2014-2020

Virulence	Species	N	%
Low virulent			
	Staph. Epidermidis	8	42.0
	Staph. Capitis	3	15.8
	Propionibacterium Acnes	3	15.8
	Staph. Caprae	2	10.5
	Staph. Hominis	1	5.3
	Total	17	89.4
High virulent			
	Enterococcus faecalis	1	5.3
	Parabacteroides distasonis	1	5.3
	Total	2	10.6
All bacteria		19	100

Staph, Staphylococcus; VSSHP, Hospital District of Southwest Finland.

Positive serologic screening tests were not detected in any donors during the study period.

The donors were divided into 2 groups based on positive and negative tissue bacterial sample findings. The summary statistics for both groups are presented in Table 4. At least one positive bacterial culture was found in 6 (21%) donors, whereas no positive bacterial cultures were found in 22 (79%) donors. The number of contaminated allografts varied from 1 to 5 per donor. In comparing the results between the two groups, the median treatment time in the ICU (4.0 days vs 2.5 days) (95% CI 1.6−8.4 vs 95% CI 2.3−3.3, P = .04) and the median value of the highest CRP level during the treatment period in the ICU (158 mg/l vs 25 mg/l) (95% CI 54−237 vs 95% CI 25-82, P = .01) were significantly higher in the group of donors with positive tissue bacterial cultures. A combination antibiotic therapy during treatment in the ICU was used more frequently in the group of donors with positive tissue bacterial cultures (67% vs 50%), but the difference was not significant (P = .65). Correlation between CRP level and bacterial contamination was statistically significant when the CRP level was >103 mg/l (P = 0.038).

Bacterial samples containing high-virulence pathogens were found in one donor. The same donor also had the longest treatment time of 11 days in the ICU and the highest number of positive tissue bacterial cultures. The second-longest treatment time in the ICU was 6 days, and a positive low-virulence bacterial sample was found from this donor. Notably high CRP levels of 194 and 270 mg/l, respectively, were found in the two donors. The longest treatment time in the group of donors without positive bacterial cultures was 5 days, and the highest CRP level was 244 mg/l. In the group of donors without positive bacterial cultures, the average of the highest CRP level increased during each additional day in the ICU. The treatment time and the highest CRP level of each donor are presented in Table 5.

A total of 61 multiorgan donors were treated in the ICU during the study period, and 33 (54%) of these donors were excluded from the musculoskeletal tissue donation. The exclusion criteria were met in 27 (82%) donors: 11 (33%) were over

Table 4. Summary statistic of the donors with and without positive tissue bacterial cultures

Donor statistics	N	
Donors with positive tissue bacterial cultures (%)	6	(21)
Median treatment time in the ICU (d)	4.0	(Range 2-11)
Elevated CRP level (≥45 mg/l) during treatment (%)	6	(100)
Median value of the highest CRP level (mg/l)	158	(Range 45-270)
Combination antibiotic therapy during treatment (%)	4	(66)
Traumatic cause of death (%)	1	(17)
Donors without positive tissue bacterial cultures (%)	22	(79)
Median treatment time in the ICU (d)	2.5	(Range 1-5)
Elevated CRP level (≥45 mg/l) during treatment (%)	9	(41)
Median value of highest CRP level (mg/l)	25	(Range 1-244)
Combination antibiotic therapy during treatment (%)	11	(50)
Traumatic cause of death (%)	4	(18)

CRP, C-reactive protein; ICU, intensive care unit.

65 years of age, 8 (24%) had a history of alcohol abuse, and 5 (15%) had multiple injury trauma. Other exclusion criteria were diabetes mellitus, rheumatoid arthritis, and septic infection once (3%). The exclusion criteria for musculoskeletal tissue donation were not identified in 6 cases (18%). A donor was overlooked once (3%) because the surgical team was not available. In 3 (9%) cases, the donors were overlooked because information from the ICU did not reach the tissue bank coordinator or the orthopedic surgeon in charge of tissue procurement. In 2 (6%) cases, whether the information did not reach the tissue banking coordinator or whether the surgical team was unavailable was uncertain.

Table 5. Characteristics of Each Musculoskeletal Tissue Donor Used in the VSSHP Tyks Orto Tissue Bank Between 2014 and 2020

Study patient no.	Treatment time in ICU (d)	Highest CRP level (mg/l)	Use of combination antibiotic therapy	No. of positive tissue bacterial cultures
15	11	194	Yes	6
23	6	270	Yes	1
20	4	52	No	1
14	4	45	Yes	2
10	3	180	Yes	5
6	2	135	No	4
11	5	244	No	0
5	5	71	Yes	0
17	4	103	Yes	0
28	4	75	Yes	0
24	4	71	Yes	0
8	3	135	No	0
18	3	134	Yes	0
27	3	103	Yes	0
12	3	27	No	0
9	3	25	Yes	0
21	3	1	No	0
1	2	108	No	0
16	2	25	Yes	0
7	2	11	No	0
2	2	10	No	0
13	2	10	Yes	0
26	2	8	Yes	0
4	2	4	No	0
25	2	4	Yes	0
22	2	2	No	0
19	2	1	No	0
3	1	2	No	0

CRP, C-reactive protein; ICU, intensive care unit; VSSHP, Hospital District of Southwest Finland.

DISCUSSION

This study analyzed the operation of the VSSHP Tyks Orto tissue bank in Turku, Finland, with a special interest in possible changes in the number of procured allografts and bacterial contamination rates compared to previous studies on the tissue bank service in Turku. The study also aimed to discover factors affecting tissue bacterial contamination and determine whether cadaver multiorgan donors suitable as musculoskeletal tissue donors were overlooked.

Aho et al reported an increase of 205% in procured long bones from 1984 to 1989 to 1990 to 1995 at the Turku Bone Bank [7]. Virolainen et al reported 375 procured allografts from 98 musculoskeletal tissue donors over a 30-year period (a mean of 3.8 grafts per donor) from 1972 to 2003 [6]. The average number of 23 procured allografts per donor and 91 allografts annually found in this study indicate that the use of individual donors in the tissue bank service has improved markedly during the last 20 years. Moreover, the increased number of procured allografts reflects the constantly growing need for musculoskeletal allografts. In the study period from 2014 to 2020, the number of procured allografts varied annually, mainly due to changes in the prevalence of cadaver donors.

The total discarding ratio of allografts was 24% from 1972 to 1995 at the Turku Bone Bank, and the bacterial contamination rate was 8.4% [7]. From 1972 to 2003, the total discarding rate was 9.9%, and the bacterial contamination rate was 6.6% [6]. The bacterial contamination rate of 2.5% in the current study was notably lower, and different types of technical failures leading to discard allografts also decreased. The incidence of allografts outdating the five-year storage time was identical in the studies, possibly indicating that the ratio of supply to demand of the allografts remained stable.

Baseri et al published a meta-analysis, including 17 studies and 19,805 bone allografts, regarding bacterial contamination rates in musculoskeletal tissue allografts from 2000 to 2021 [8]. In their analysis, the overall bacterial contamination rate was 19.9% among cadaver donors. In Europe, the bacterial contamination rate of allografts was 14.3%. The lowest contamination rate was reported in Australia, at 5.2%. This was explained by the higher hygienic standards of tissue banks in Australia. The higher contamination rates in Europe remained unclear. The contamination rates reported in other studies consisting only of cadaver donors were 10% to 52% (Table 6). The 2.5% bacterial contamination rate in the VSSHP Tyks Orto tissue bank was lower than that in the previous literature.

The screening methods for swabs and biopsy samples result in different contamination rates, and no unambiguous international guidelines for screening methods exist [8−10]. This, together with the different procurement methods, makes the comparison of bacterial contamination rates between studies difficult. Various decontamination methods, from irradiation and antibiotic solutions to mechanical lavage, are used to disinfect allografts during the procurement process [11]. Low-pressure pulse lavage with a sterile saline solution has been shown to be an effective disinfection method [10,12,13]. Swab samples were used for microbiological screening, and the allografts were immersed in an antibiotic solution during the previously followed procurement process in the tissue banking procedure in Turku [6,7]. The findings suggest that the currently followed methods of pulse lavage with sterile saline and biopsy sampling show good results for allograft sterility. We agree with Baseri on the issue of standardizing the tissue sampling process.

Table 6. Contamination Rates of Musculoskeletal Allografts Harvested from Cadaver Donors in Literature

First author	N (contaminated/total)	%
Paolin [32]	5 211/10 035	52
Vehmeyer [33]	2 546/5 710	45
Ibrahim [36]	120/437	27
Journeuax [37]	65/272	24
Terzaghi [26]	635/2 778	23
Ilays [38]	115/506	22.7
Viñuela-Prieto [39]	227/1 162	19.5
Ivory [40]	4/22	18
Naves [15]	218/1 271	17.1
Liu [41]	25/201	12.4
Bohatyrewicz [42]	45/424	10.7
Schubert [16]	365/3 612	10.1

Bacteria identified from the microbiological cultures were divided into low and high virulence in the study of Deijkers et al [14], and the same grouping was followed by Naves et al [15] and Schubert et al [16]. Low-pathogenic microbes are considered skin commensals, representing external contamination during the procurement process, and they rarely cause clinical infection in the receiver. Our study findings correspond with those of Deijkers and Schubert, suggesting that most contaminations result from external contamination during the procurement process.

The treatment time in the ICU was significantly longer in the group of donors with positive bacterial cultures. Also, the only donor in this study possessing high-virulence bacterial samples had a notably longer treatment time in the ICU than the rest of the donors. Length of stay in the ICU has been associated with the risk of bacterial contamination, and a longer stay increases the likelihood of contamination [15,16]. Caution was suggested regarding selecting donors with long treatment times in the ICU.

Naves et al analyzed whether the number of leukocytes during the treatment period in the ICU predicts tissue contamination and did not find the number of leukocytes relevant for predicting tissue contamination rate [15]. We did not find previous studies regarding the correlation between measured CRP levels from donors during treatment in the ICU and tissue bacterial contamination. C-reactive protein is a widely used biomarker to detect bacterial infection in ill patients. Elevated CRP levels predict bacterial infection, but other inflammatory responses can also elevate CRP levels [17−20]. The predictive cut-off value for CRP level to diagnose sepsis in critically ill patients in the ICU surroundings has been studied before, but the optimal cut-off value is yet to be established. A cut-off value for CRP levels between 50 mg/l and 100 mg/l has been considered reasonable sensitivity and specificity vice [21−24]. The

risk for nosocomial bacterial infection during treatment in the ICU is high [25]. It is considered that ICU surroundings and procedures, such as various catheterizations, tracheal intubation, and mechanical ventilation, promote colonization with highly virulent bacteria and that these pathogens can spread via the hematogenous route to the procured tissues [15,26]. Notably elevated CRP levels are also associated with bacteremia [27−30]. Deijkers et al found a strong correlation between a positive blood culture and the contamination of allograft with highly virulent bacteria [14]. Terzaghi et al, surprisingly, did not find a correlation between positivity of blood culture and increased risk of allograft contamination [26]. In this study, a statistically significant correlation between CRP level and bacterial contamination was found when the CRP level was higher than 103 mg/l.

However, most of the bacterial contaminations were caused by low-virulent bacteria that are considered to represent external contamination during the procurement process. The only donor in the present study with high virulent bacterial contamination had a highly elevated CRP level of 194 mg/l. We believe that a possible cut-off value for CRP level to predict allograft contamination with endogenous high-virulent bacteria could be even higher than that found in this study. However, because all the high-virulence bacterial contaminations were found from a single study donor, conclusions cannot be drawn based on this study. More research with a higher volume of donors and allografts containing high-virulent bacteria is needed to study this topic. We believe that markedly elevated CRP levels and the previously mentioned long treatment times in the ICU could be risk factors for allograft contamination, with high-virulent bacteria originating from endogenous contamination.

The study donor with the second-longest treatment period also had a positive, though low-virulent, bacterial sample and a high CRP level of 270 mg/l during treatment in the ICU. However, a high CRP level of 244 mg/l was found in the donor with the longest treatment time in the group of donors without positive tissue bacterial samples (Table 5). Positive low-virulent bacterial cultures were also found in donors with shorter treatment periods and lower CRP levels, yet each donor with positive bacterial contamination had at least a moderately elevated CRP level (≥45 mg/l). The findings are unclear, and potential external contamination during the procurement process hinders the evaluation of these factors. Therefore, the potential association between long treatment time, high CRP level, and allograft bacterial contamination should probably be limited to only high-virulent bacteria expected to originate from endogenous contamination.

A combination antibiotic therapy was used more frequently in the group of donors with positive bacterial contaminations, but the difference was not significant. The absence of antibiotic therapy has been associated with an increased risk of allograft bacterial contamination, but a combination antibiotic therapy was not found to be relevant regarding contamination [15]. Prophylactic antibiotic therapy was recommended, with no need for combination antibiotic therapy. In the TYKS ICU, each donor was treated with standard broad-spectrum antibiotic therapy (Meropenem 1 g intravenously 3 times a day) during the entire treatment period. With certain donors, a combination antibiotic therapy (usually with Cefuroxime 1.5 g intravenously 3 times a day) was used when the clinical situation of the donor evaluated by the ICU physician required it. It seems that the use of prophylactic antibiotic therapy should be recommended during treatment, but combination therapy does not provide additional protection against allograft bacterial contamination.

The high experience of the surgical team reduces the risk of bacterial contamination during the procurement process [15,16]. The large number of surgical team members is also associated with the risk of contamination, increasing with every extra staff member in the operating room [14,26,31−33]. A small surgical team performs all allograft procurements in the VSSHP Tyks Orto tissue bank. The surgical team consists of 2 to 3 nurses and 2 surgeons; therefore, the total number of surgical team members in the operating room is 4 to 5 persons at a time. Only trained orthopedic surgeons perform the procurement process. Surgical team members are familiar with the procurement process and have been given special training on aseptic working methods during the musculoskeletal tissue procurement process from cadaver donors. Written instructions are established to guide the working principles in the operating room, and the operating room is sealed during the procurement process to prevent unnecessary passage of persons and microbes into the room. These arrangements ensure that the surgical process is performed effectively, aseptically, and without unnecessary action, minimizing the possibility of external contamination of the allografts during the procurement process. We believe that the high hygienic standards followed in our practice, accurate patient selection, and effective operation of a well-trained surgical team are factors affecting the low incidence of bacterial contamination rate in the VSSHP Tyks Orto tissue bank.

Overlooking potential organ donors is a previously recognized issue [34,35], and less than half of the organ donors treated in the TYKS ICU were also used as musculoskeletal tissue donors. In some cases, there were doubts that information about a potential musculoskeletal tissue donor in the TYKS ICU did not reach the tissue banking coordinator or the orthopedic surgeon in charge of tissue procurement, leading to the overlooking of suitable donors. Despite careful study, the exclusion criteria for musculoskeletal tissue donation were not observed in the medical records of 6 donors. It is impossible to say for certain whether an exclusion criterion was not mentioned in the medical records. It seems that at least 3 patients were overlooked due to a lack of communication. As 2 other cases took place during the holiday, it was uncertain whether the donor was overlooked due to a lack of communication or the surgical team being unavailable. One time, the surgical team was not available due to the holiday season. A lack of communication between the ICU and the tissue bank in TYKS was previously noted, and the study findings confirmed that some suitable candidates for musculoskeletal tissue donation were overlooked among organ donors.

Moreover, although the small number of surgical team members improves efficiency in the allograft procurement process

and reduces allograft contamination, all team members may not be available, especially during holidays. Owing to the small number of potential musculoskeletal tissue donors, any suitable donor should not be overlooked. Actions have already been taken to reduce this possibility. Electronic instructions have been created, and regular training is given to the ICU staff so that suitable musculoskeletal tissue donors will not remain unused in the future.

We acknowledge that this study has some limitations. The number of study donors was small, thus reducing the reliability and significance of the study results. Encouraging results were found in the correlation between a long stay in the ICU and high CRP level and the possibility of tissue bacterial contamination. However, the main findings of this study focused only on a small group of study donors. Research with a higher donor volume must be conducted to verify the findings of this study. Because of the retrospective nature of this study, the details of the study donors were based only on their medical records. Comprehensive documentation is integrated into medical records during treatment time in the ICU, and we believe that no significant details remained undocumented.

CONCLUSIONS

This review indicates that the operation of the VSSHP Tyks Orto musculoskeletal tissue bank conforms to the directives and guidelines established by national and European authorities and the European Association of Tissue Banks. The number of procured musculoskeletal allografts has increased markedly during the last decades, and the percentage of allograft bacterial contamination has decreased simultaneously. The bacterial contamination rate of 2.5% in allografts procured from cadaver donors was lower than that found in the previous international literature. Median treatment time in the ICU was significantly longer, and the median value of the highest CRP level during the treatment period was significantly higher in the group of donors with positive allograft bacterial contaminations. A long treatment period in the ICU and a high CRP level could be risk factors for allograft contamination with high-virulent bacteria, but more research is needed on this topic. Overlooking potential organ donors is a known issue, and some potential musculoskeletal tissue donors were overlooked among multiorgan donors treated in the TYKS ICU.

DATA AVAILABILITY

Data will be made available on request.

DECLARATION OF COMPETING INTEREST

The authors have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

REFERENCES

[1] Amini AR, Laurencin CT, Nukavarapu SP. Bone tissue engineering: recent advances and challenges. Crit Rev Biomed Eng 2012;40:363.

[2] Greenwald AS, Boden SD, Goldberg VM, Khan Y, Laurencin CT, Rosier RN. Bone-graft substitutes: facts, fictions, and applications. J Bone Joint Surg 2001;83:98–103.

[3] Kurtz S, Ong K, Lau E, Mowat F, Halpern M. Projections of primary and revision hip and knee arthroplasty in the United States from 2005 to 2030. J Bone Joint Surg 2007;89:780–5.

[4] Khan SN, Cammisa EP, Sandhu HS, Diwan AD, Girardi FP, Lane JM. The biology of bone grafting. J Am Acad Orthop Surg 2005;13:77–86.

[5] Tomford WW. Transmission of disease through transplantation of musculoskeletal allografts. J Bone Joint Surg 1995;77:1742–54.

[6] Virolainen P, Heikkilä J, Hirn M, Aro HT, Aho AJ. 30 years of bone banking at Turku bone bank. Cell Tissue Bank 2003;4:43–8.

[7] Aho AJ, Hirn M, Aro HT, Heikkilä JT, Meurman O. Bone bank service in Finland. Acta Orthop Scand 1998;69:559–65.

[8] Baseri N, Meysamie A, Campanile F, Hamidieh AA, Jafarian A. Bacterial contamination of bone allografts in the tissue banks: a systematic review and meta-analysis. J Hosp Infect 2022;123:156–73.

[9] Veen MR, Bloem RM, Petit PL. Sensitivity and negative predictive value of swab cultures in musculoskeletal allograft procurement. Clin Orthop Relat Res 1994;300:259–63.

[10] Hirn MYJ, Salmela PM, Vuento RE. High-pressure saline washing of allografts reduces bacterial contamination. Acta Orthop Scand 2001;72:83–5.

[11] Mohr JW, Germain M, Winters M, Fraser S, Duong A, Garibaldi A, Simunovic N, Alsop D, Dao D, Bessemer R, Ayeni OR. Disinfection of human musculoskeletal allografts in tissue banking: a systematic review. Cell Tissue Bank 2016;17:573–84.

[12] Salmela PM, Hirn MYJ, Vuento RE. The real contamination of femoral head allografts washed with pulse lavage. Acta Orthop Scand 2002;73:317–20.

[13] Hirn M, Laitinen M, Pirkkalainen S, Cefuroxime Vuento R. rifampicin and pulse lavage in decontamination of allograft bone. J Hosp Infect 2004;56:198–201.

[14] Deijkers RLM, Bloem RM, Petit PLC, Brand R, Vehmeyer SBW, Veen MR. Contamination of bone allografts: analysis of incidence and predisposing factors. J Bone Joint Surg 1997;79:161–6.

[15] Naves GG, Silva AF, Antebi U, Cristovam PC, Honda EK, Guimarães RP. Analysis of potential contamination factors in musculoskeletal tissues. Cell Tissue Bank 2018;19:659–66.

[16] Schubert T, Bigaré E, VanIsacker T, Gigi J, Delloye C, Cornu O. Analysis of predisposing factors for contamination of bone and tendon allografts. Cell Tissue Bank 2012;13:421–9.

[17] Clyne B, Olshaker JS. The C-reactive protein. J Emerg Med 1999;17:1019–25.

[18] Sproston NR, Ashworth JJ. Role of C-Reactive protein at sites of inflammation and infection. Front Immunol 2018;9:754.

[19] Simon L, Gauvin F, Amre DK, Saint-Louis P, Lacroix J. Serum procalcitonin and C-reactive protein levels as markers of bacterial infection: a systematic review and meta-analysis. Clin Infec Dis 2004;39:206–17.

[20] Hu L, Shi Q, Shi M, Liu R, Wang C. Diagnostic value of PCT and CRP for detecting serious bacterial infections in patients with fever of unknown origin: a systematic review and meta-analysis. Appl Immunohistochem Mol Morphol 2017;25:e61–9.

[21] Póvoa P. C-reactive protein as an indicator of sepsis. Intensive Care Med 1998;24:1052–6.

[22] Ugarte H, Silva E, Mercan D, De Mendonça A, Vincent JL. Procalcitonin used as a marker of infection in the intensive care unit. Crit Care Med 1999;27:498–504.

[23] Mitaka C. Clinical laboratory differentiation of infectious versus non-infectious systemic inflammatory response syndrome. Clin Chim Acta 2005;351:17–29.

[24] Tian T, Wei B, Wang J. Study of C-reactive protein, procalcitonin, and immunocyte ratios in 194 patients with sepsis. BMC Emerg Med 2021;21:81.

[25] Ozer B, Akkurt CO, Duran N, Onlen Y, Savas L, Turhanoglu S. Evaluation of nosocomial infections and risk factors in critically ill patients. Med Sci Monit 2011;17:18.

[26] Terzaghi C, Longo A, Legnani C, Bernasconi DP, Faré M. Incidence of bacterial contamination and predisposing factors during bone and tendon allograft procurement. Cell Tissue Bank 2015;16:151–7.

[27] Bell K, Wattie M, Byth K, Silvestrini R, Clark P, Stachowski E, et al. Procalcitonin: a marker of bacteraemia in SIRS. Anaesth Intensive Care 2003;31:629–36.

[28] De La Torre-Prados MV, García de la Torre A, Cámara Sola E, Puerto Morlán A, Tsvetanova Spasova T, Nuevo Ortega P, et al. Etiology of bacteremias associated with c-reactive protein, procalcitonin and lactate levels. Intensive Care Med 2015;3:1.

[29] Póvoa P, Coelho L, Almeida E, Fernanades A, Mealha R, Moreira P, et al. Pilot study evaluating C-reactive protein levels in the assessment of response to treatment of severe bloodstream infection. Clin Infect Dis 2005;40:1855–7.

[30] Vandijck DM, Hoste EA, Blot SI, Depuydt PO, Peleman RA, Decruyenaere JM. Dynamics of C-reactive protein and white blood cell count in critically ill patients with nosocomial Gram positive vs. Gram negative bacteremia: a historical cohort study. BMC Infect Dis 2007;7:106.

[31] Segur JM, Suso S, García S, Combalía A, Fariñas O, Llovera A. The procurement team as a factor of bone allograft contamination. Cell Tissue Bank 2000;1:117–9.

[32] Paolin A, Trojan D, Petit P, Coato P, Rigoli R. Evaluation of allograft contamination and decontamination at the Treviso Tissue Bank Foundation: a retrospective study of 11,129 tissues. PLoS One 2017;12.

[33] Vehmeyer S, Wolkenfelt J, Deijkers R, Petit P, Brand R, Bloem R. Bacterial contamination in postmortem bone donors. Acta Orthop Scand 2002;73:678–83.

[34] Rakhra SS, Opdam HI, Gladkis L, Arcia B, Ma Fink, Kanellis J, et al. Untapped potential in Australian hospitals for organ donation after circulatory death. Med J Aust 2017;207:294–301.

[35] Kutsogiannis DJ, Asthana S, Townsend DR, Singh G, Karvellas CJ. The incidence of potential missed organ donors in intensive care units and emergency rooms: a retrospective cohort. Intensive Care Med 2013;39:1452–9.

[36] Ibrahim T, Stafford H, Esler CNA, Power RA. Cadaveric allograft microbiology. International Orthop 2004;28:315–8.

[37] Journeaux SF, Johnson N, Bryce SL, Friedman SJ, Sommerville SMM, Morgan DAF. Bacterial contamination rates during bone allograft retrieval. J Arthroplasty 1999;14:677–81.

[38] Ilays I, Alsakran SA, Fallatah AB, Alyateem M, Al-Mohrej OA. The contamination of allografts in multi-organ donors: a bone bank experience. Cell Tissue Bank 2021;22:499–504.

[39] Viñuela-Prieto JM, Soria-García AM, González-Romero M, Candel FJ. Bacterial contamination rate and associated factors during bone and tendon allograft procurement from Spanish donors: exploring the contamination patterns. J Hosp Infect 2019;102:287–94.

[40] Ivory JP, Thomas IH. Audit of a bone bank. J Bone Joint Surg 1993;75:355–7.

[41] Liu JW, Chao LH, Su LH, Wang JW, Wang CJ. Experience with a bone bank operation and allograft bone infection in recipients at a medical centre in southern Taiwan. J Hosp Infec 2002;50:293–7.

[42] Bohatyrewicz A, Bohatyrewicz R, Klek R, Kaminski A, Dobiecki K, Bialecki P, et al. Factors determining the contamination of bone tissue procured from cadaveric and multiorgan donors. Transplant Proc 2006;38:301–4.