TURUN YLIOPISTO
Lääketieteellinen tiedekunta

SANTTU TIKKALA: Katsaus tukikudospankin toiminnasta vuosien 2014 ja 2020 välillä Turussa, Suomessa

Syventävien opintojen kirjallinen työ
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Luu- ja tukikudospankin tehtävänä on kerätä, testata, säilyttää ja tarjota luu- ja tukikudossiirteitä, joita tarvitaan ortopedisissä toimenpiteissä. Kudossiirteet kerätään eläviltä ja kuolleilta kudosluovuttajilta steriileissä leikkaussaliolosuhteissa siirteiden bakteerikontaminaation välttämiseksi. Kuolleet kudosluovuttajat valitaan teho-osastolla hoidettujen monielinluovuttajien joukosta, ja tiukkoja poissulkukriteerejä noudatetaan tarttuvien tautien siirtymisen estämiseksi. Luu- ja tukikudospankki on toiminut Turussa, Suomessa vuodesta 1972 lähtien, ja viimeinen katsaus sen toiminnasta on vuodelta 2003. Tämän tutkimuksen tarkoituksena oli tehdä katsaus VSSHP Tyks Orto kudospankin toiminnasta Turussa, Suomessa vuosien 2014 ja 2020 välillä, sekä analysoida kuolleilta kudosluovuttajilta kerättyjen kudossiirteiden määrä, tyypit ja bakteerikontaminaatioaste. Lisäksi tutkittiin mahdollisia kudosluovuttajaan liittyviä tekijöitä, jotka aiheuttavat kudossiirrännäisten bakteerikontaminaatiota sekä jääkö potentiaalisia kudosluovuttajia hyödyntämättä monielinluovuttajien joukosta.

Retrospektiivinen katsaus suoritettiin vuosien 2014 ja 2020 välillä VSSHP Tyks Orto kudospankissa hyödynnetyistä monikudosluovuttajista. Aineisto monikudosluovuttajilta kerätyistä luu- ja tukikudossiirteistä kerättiin ja esitettiin tutkimuksessa.

Tutkimusaineistossa oli 28 monikudosluovuttajaa, joilta kerättiin yhteensä 636 luu- ja tukikudossiirrettä. Kudossiirteiden bakteerikontaminaatioaste oli 2.5 %, mikä on huomattavasti matalampi verrattuna aiempaan kansainväliseen lähdeaineistoon, jossa kontaminaatioaste on vaihdellut 10 ja 52 %:n välillä. Pidempi keskimääräinen hoitoaika ja korkeampia C-reaktiivisen proteiinin tasoja mitattiin kudosluovuttajilla, joilta löytyi positiivisia bakteerinäytteitä, mutta lisää tutkimusta suuremmalla aineistolla tarvitaan tämän havaitun korrelaation osoittamiseksi. Lisäksi havaitsimme, että joitain potentiaalisia kudosluovuttajia jäi hyödyntämättä monielinluovuttajien joukosta.

Avainsanat: Tukikudospankki, Tukikudossiirre, Kudosluovuttaja, Bakteerikontaminaatio

Original article

Experience with the tissue bank service in years 2014 and 2020 in Turku, Finland

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Abstract

Purpose of a musculoskeletal tissue bank is to collect, test, store and provide musculoskeletal tissue allografts required in orthopedic procedures. Allografts are harvested from living and cadaver donors under sterile conditions to avoid allograft bacterial contamination. Cadaver musculoskeletal tissue donors are selected among multiorgan donors, and strict exclusion criteria are followed to prevent transmission of infectious diseases. Tissue banking in Turku, Finland began in 1972, and tissue bank service was last time reviewed in 2003. This study aimed to review operation of VSSHP Tyks Orto tissue bank in Turku, Finland between year 2014 and 2020, and to analyze number, types and contamination rate of allografts harvested from cadaver donors. Potential donor relating factors causing allograft bacterial contamination and whether potential musculoskeletal tissue donors were missed among multiorgan donors were also studied.

A retrospective review of cadaver musculoskeletal tissue donors utilized in VSSHP Tyks Orto tissue bank during study period was conducted, and data concerning harvested allografts was collected and presented.

Total number of 28 cadaver donors were utilized and 636 allografts were harvested between year 2014 and 2020. Allograft bacterial contamination rate was 2.5 %, which is substantially lower when compared to previous international literature, where contamination rate has varied between 10 and 52 %. Longer average treatment time and higher C-reactive protein levels were recorded in donors with positive bacterial cultures, but more research with higher donor volume is required to verify this correlation. We also discovered that some suitable musculoskeletal tissue donors were missed among multiorgan donors.

Keywords: Tissue bank, Musculoskeletal tissue allograft, Tissue donor, Bone transplantation, Bacterial contamination

Introduction

Utilization of musculoskeletal tissue allografts is common in orthopedic surgery and need for allografts is constantly growing (Amini et al., 2012). Allografts are harvested from living and cadaver donors, and harvested allografts are stored in tissue banks. Tissue banks are responsible for collecting, testing, storage and distribution of allografts. Allografts are noted to be suitable material in reconstructive orthopedic surgery, but risk for infection due to transmissible diseases and allograft contamination exist (Khan et al., 2005; Tomford, 1995). Strict donor selection and serological screening tests together with aseptic allograft procurement methods and microbiological sampling of allografts are followed to provide high quality allografts and to prevent infections in allograft recipients.

Musculoskeletal tissue banking for clinical purpose in Turku, Finland began in 1972, and first allograft transplantation took place in 1973 (Virolainen et al., 2003). At first only bone allografts were stored, and ligament grafts were next added to selection. Previously only Aho et al. (1998) and Virolainen et al. (2003) have reviewed tissue bank service in Turku. Since then, operation of the tissue bank has developed markedly, and today stored tissues include long bones, tendons, meniscus and fresh osteochondral allografts. In addition to progression in clinical practices, administrational changes have also been made: in January 29th 2014 the Finnish Red Cross gave up administration of the tissue bank and The Hospital District of Southwest Finland (VSSHP) took charge. Emerged organization is now called VSSHP Tyks Orto tissue bank.

The aim of this study was review VSSHP Tyks Orto tissue bank function between years 2014 and 2020. Number of harvested musculoskeletal tissue allografts and allograft bacterial contamination rate were reported, documenting possible changes in numbers of harvested allografts and bacterial contamination rates in relation to previous reviews on tissue bank service in Turku, Finland. Bacterial contamination rate was also compared to previous international literature. Special interest was focused on cadaver musculoskeletal tissue donors, aiming to study possible donor relating factors causing allograft bacterial contamination, and whether some potential musculoskeletal tissue donors were missed among multiorgan donors.

Materials and methods

A retrospective review of all cadaver multiorgan donors treated at Turku University Hospital (TYKS) intensive care unit (ICU) between January 1st 2014 and December 31st 2020 was

conducted. Medical history of all donors was studied, and donor related factors were analyzed: age, sex, cause of death, time treated in ICU, antibiotic therapy during treatment in ICU, highest C-reactive protein (CRP) value during treatment period and positive bacterial growth in tissue samples. Data concerning allografts harvested from cadaver donors was collected from Tissue Db (BCB Medical Ltd) database. Since our aim was 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 in Turku (Dnro TO1/022/2020).

Operating principle, administration and control

Operation of VSSHP Tyks Orto tissue bank is carried on as activity owned and coordinated by VSSHP. Tissue bank is responsible for collection, testing, packaging and distribution of musculoskeletal allografts. Primary objective of VSSHP Tyks Orto tissue bank is to secure sufficiency of safe allografts for hospitals in 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 and working principles follow assumed agreement of the donor and permission from next of kin. Operational guidelines are described in the quality manual of the tissue bank. Hospitals operating under VSSHP Tyks Orto tissue bank have also their own directives that have to be in line with the quality manual. Finnish medicine agency (FIMEA) is the authority supervising tissue banking in Finland. Authoritative dictates concerning operation of VSSHP Tyks Orto tissue bank are following: Law on the use of human organs, tissues and cells for medical purposes 101/2001 (2001), Ordinance of the Ministry of Social Affairs and Health on the use of human organs, tissues and cells for medical purposes 1302/2007 (2007), Order 3/2014 Tissue bank services, Fimea (2014), European Council directive 2004/23/EC (2004), European Commission Directive 2006/17/EC (2006) and European Commission Directive 2006/86/EC (2006). There are also documents created by European Association of Tissue Banks (EATB), Council of Europe and Finnish Orthopedic Association witch guide operation of the tissue bank. (For more detail see Supplement 1.)

Donor selection, tissue retrieval and microbiological testing

Musculoskeletal tissue donors are selected from brain-dead multiorgan donors. After amendment to a law in 2010, agreement based on assumption that when alive, donor would not have objected to the procedure is followed. Assumed agreement of the donor and permission for donation are always

verified from next of kin. Significant attention is paid to donor criteria due to risk of transmissible diseases (Tomford, 1995), and contraindication list is strict for musculoskeletal tissue donation (Table 1). Cadaver musculoskeletal tissue donors are typically previously healthy person aged 16-65 who have died of a sudden cerebral death. The medical history of potential donor is carefully studied. Suitability of cadaver donor is evaluated by orthopedic surgeon performing tissue procurement together with tissue banking coordinator. Contraindication list is gone through using medical records of the donor and possible hemodilution is noticed. Blood samples are screened from every donor for presence of transmissible diseases. Serological screening tests are made to detect HIV (S-HIVAgAb, P-HIV1Nh), hepatitis B (S-HBsAg, S-HBcAb, S-HBVNhO), hepatitis C (S-HCVAb, P-HCVNhO) and Treponema pallidum (S-TrpaAb). Positive test result leads to discarding of all harvested allografts. Donor's suitability for tissue donation is documented in the medical records. Harvesting of allografts is performed less than 12 hours after expiration of donor's blood circulation.

Harvesting and handling of the allografts is performed aseptically in an operating room by experienced surgical team. If signs of contamination appear in any stage of harvesting proses, possibly contaminated graft is immediately rejected. Grafts are harvested in a predetermined order, one graft at a time. Soft tissues are removed from around the grafts and bones are separated in parts when needed. Bone marrow is scooped empty, and grafts are washed with pressure lavage using sterile saline solution. After washing, microbiological samples are taken from every graft. Three tissue fragment samples are taken with a surgical bone biter or tweezers from each allograft. Samples are cultured for aerobic and anaerobic bacterial growth. Cultures are also made for fungal growth. Microbiological testing is executed in qualified noted and accredited laboratories using validated methods. Graft is approved only if all cultures are negative.

Storage and record-keeping

After microbiological sampling, grafts are packed in 2-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 to avoid external contamination. Each package is labelled and marked to identify the donor, type and size of tissue, blood type of the donor, date of harvesting and expiration day of the graft. Allografts are frozen within 12 hours after harvesting, in practice freezing is done immediately after packaging. Fresh osteoarticular grafts are stored in antibiotic solution in refrigerator temperature (+2 - +8 °C) no more than 21 days.

Frozen allografts are stored at temperature of -75 °C, and temperature is not allowed to rise over -40 °C during storage. Freezers have local and wireless alarm systems for temperature rise and freezers are connected to emergency power source.

Data concerning used allografts is preserved for 30 years after clinical usage of the graft. All data related to donation of allograft is preserved. There is a computerized database and a manual record for record-keeping. Record-keeping is essential for traceability of the allografts, donors and receivers of the grafts. Every graft is marked with SEC (Single European Code) number and tissue identification number to make identification of allografts easier and to protect anonymity of each individual donor. Grafts are identified using the identification number, so donor of the graft is not recognizable while handling and using allografts. By entering the identification number to the database, all information concerning graft and donor is available if necessary.

Clinical use of allografts

Classical sites for usage of allografts are bone defects caused by bone tumor or trauma, spinal fusion surgery and filling of a cavity in revision arthroplasties. Tendon grafts are used for example in reconstructive or reinsertion surgery of ligaments and tendons in upper and lower limbs.

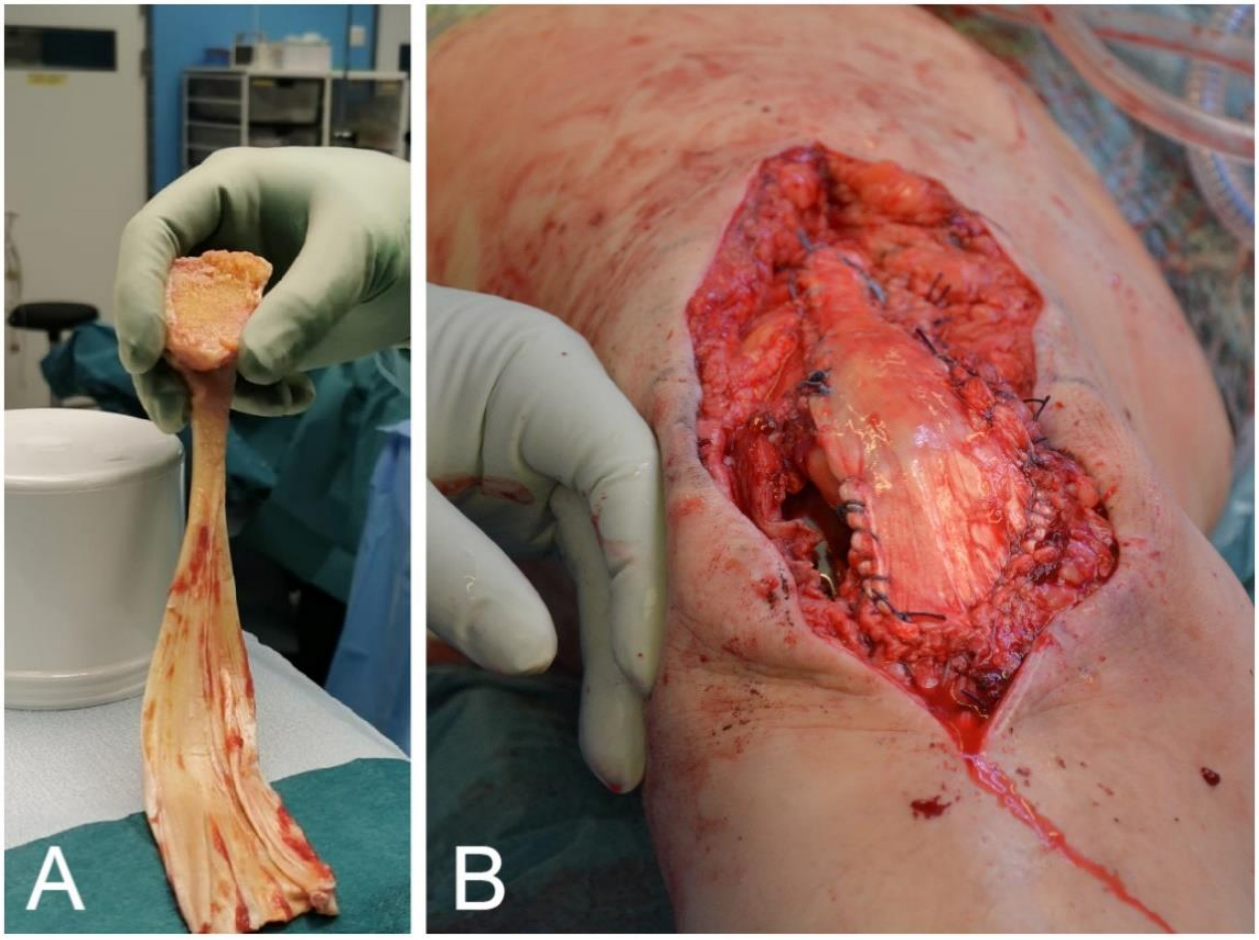

Figure 1. A) Detached achilles tendon allograft. B) Achilles tendon allograft used in knee surgery.

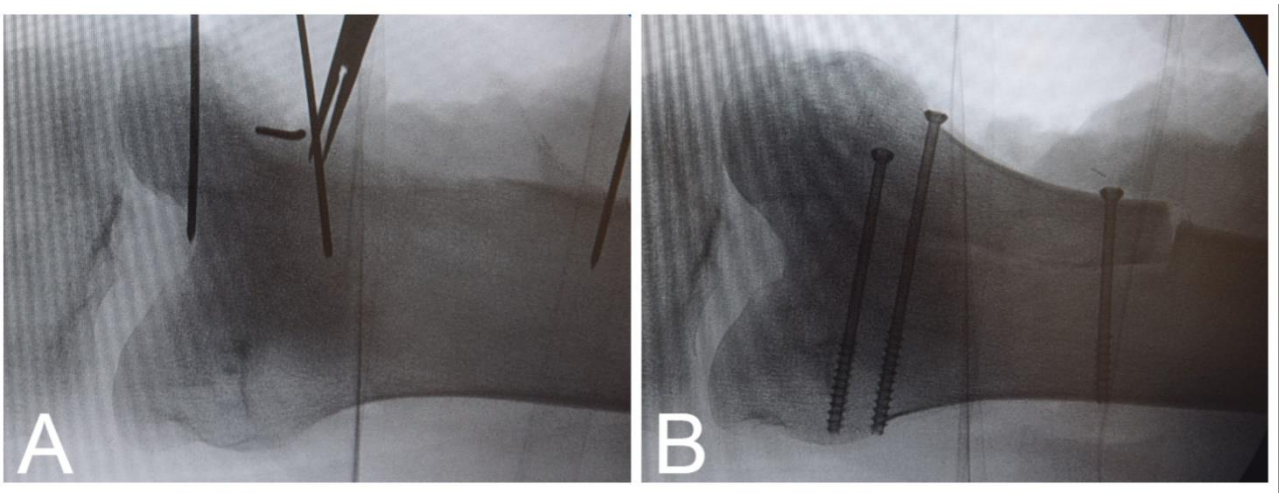

Figure 2. A) Bone defect in distal femur. B) Bone defect of distal femur filled with bone allograft.

Results

During the study period of 7 years from 2014 to 2020, 28 musculoskeletal tissue donors were utilized and total of 636 allografts were harvested from donors in VSSHP Tyks Orto tissue bank. Most frequently harvested allografts were femur, tibia and tibialis anterior and posterior tendons. The detailed information regarding donors and tissue allografts are presented in table 2. Number of harvested allografts per donor varied from 10 to 34 grafts and average number of harvested allografts was 23. Yearly number of harvested allografts varied from 22 to 138 allografts and mean number of harvested allografts was 91 grafts per year. Number of donors varied from 2 to 6 per year (Diagram 1).

Total number of discarded allografts was 49, covering 7.7 % of all harvested grafts. Causes for discarding were: 21 (3.3 %) outdated 5 years storage time, 16 (2.5 %) positive bacterial cultures, 5 (0.8 %) grafts were melted for usage but were not used during operation, 4 (0.6%) melted during storage, 2 (0.3 %) were discarded because of poor quality of the graft and 1 (0.16 %) wrong size of graft.

At least one bacterial culture was positive in 16 allografts. Two different bacterial species were identified in three grafts, so in total 19 positive bacterial cultures were found. Bacterial contamination was the cause for discarding in 32.7 % of all discarded cases. Total of 7 different bacterial species were identified and bacteria were divided to low and high virulent. Low virulent Staphylococcus species were cultured most frequently. High virulent pathogens were found in two samples, both taken from same donor. Species and numbers of bacteria are summarized in table 3.

Positive serological screening test for viral diseases was not detected in any of the donors during study period.

Musculoskeletal tissue donors were divided in two 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 and number of contaminated allografts varied from 1 to 5 per donor. All contaminated allografts were discarded, and rest were banked in normal manner. When comparing results between the two groups, mean treatment time in ICU (5 vs 2.8 days), incidence of elevated CRP levels (100 vs 55 %), average of highest CRP-value (146 vs 53 mg/l) and use of combination antibiotic therapy during treatment (66 vs 50 %) were all higher in group of donors with positive tissue bacterial cultures.

Both tissue bacterial samples containing high virulent pathogens were found in one donor. The same donor also had the longest treatment time of 11 days in ICU and highest number of positive tissue bacterial cultures. The second longest treatment time in ICU was 6 day, and positive low

virulent bacterial sample was found from this donor. Notably high CRP values of 194 and 270 mg/ml respectively were measured from the two donors. Longest treatment time in donors with no positive bacterial cultures was 5 days and highest CRP value was 244 mg/ml. In the group of donors with no positive bacterial cultures, average of highest CRP value increased during each additional day in ICU. Treatment time and highest CRP value of each donor is presented in table 5.

61 multiorgan donors were treated in ICU during study period and 33 (54 %) of these were excluded from musculoskeletal tissue donation. Exclusion criteria were met in 28 (82 %) donors: 11 (33 %) were aged over 65 years, 8 (24 %) had history of alcohol abuse and 5 (15 %) had multiple injury trauma. Other exclusion criteria were diabetes mellitus, rheumatoid arthritis and septic infection once (3 %) each. In 6 (18 %) cases exclusion criterion for musculoskeletal tissue donation was not identified. Donor was once (3 %) missed because surgical team was not at disposal. In 3 (9 %) cases it was assumed that donor was missed because the information from ICU did not reach the tissue banking coordinator and orthopedic in charge of tissue procurement. In 2 (6 %) cases there was no certainty if the information did not reach the tissue banking coordinator or if surgical team was not at disposal.

Discussion

This study analyzed operation of VSSHP Tyks Orto tissue bank in Turku, Finland with special interest in possible changes in number of harvested allografts and bacterial contamination rates compared to previous studies regarding tissue bank service in Turku. Study goals were also to discover factors effecting tissue bacterial contamination and weather cadaver multiorgan donors suitable for musculoskeletal tissue donors were missed.

Aho et al. (1998) reported an increase of 205 % in harvested long bones from 1984-1989 to 1990-1995 in the Turku Bone Bank. At the time total number of harvested allografts was just over 100 (approximately 17 grafts yearly). Virolainen et al. (2003) reported total number of 375 harvested allografts from 98 musculoskeletal tissue donors in 30 years period (mean 3.8 grafts per donor) from 1972 to 2003. Result of average 23 harvested allografts per donor and 91 allografts yearly in this study indicates that utilization of individual donor in the tissue bank service has improved significantly during the last 20 years, and increased numbers of harvested allografts reflect the constantly growing need for musculoskeletal allografts. The growing need for allografts has been evident since the beginning of tissue bank service in Turku (Aho et al., 1998). In the study period

between years 2014 and 2020, number of harvested allografts varies yearly mainly due to changes in the prevalence of cadaver donors.

Total discarding ratio of allografts was 24 % between year 1972 and 1995 in the Turku Bone Bank. Bacterial contamination rate was 8 %, discarding because of technical failure 13 % and 3 % of grafts outdated 5 years storage time. (Aho et al., 1998) In our material bacterial contamination rate of 2.5 % is notably lower and different type of technical failures leading to discarding of allograft have also decreased notably. Incidence of allograft outdating the 5-year storage time was identical in both studies, possibly marking that ratio between need and supply of allografts has remained stable.

Baseri et al. (2021) published a meta-analysis including 17 studies and 19 805 bone allografts regarding the incidence of bacterial contamination rates in musculoskeletal tissue allografts from 2000 to 2021. In their analysis overall bacterial contamination rate was 19.9 % among cadaver donors. In Europe, bacterial contamination rate of allografts was 14.3 %. The lowest contamination rate was reported in Australia with 5.2 %. This was considered to be explained by higher hygienic standards of tissue banks in Australia. The higher contamination rates in Europe remain unclear. Contamination rates reported in other studies consisting only cadaver donors ranges from 10 to 52 % (Table 6) (Bohatyrewicz et al., 2006; Ibrahim et al., 2004; Ilays et al., 2021; Ivory & Thomas, 1993; Journeaux et al., 1999; J. W. Liu et al., 2002; Naves et al., 2018; Paolin et al., 2017; Schubert et al., 2012; Terzaghi et al., 2015; Vehmeyer et al., 2002; Viñuela-Prieto et al., 2019). The result of 2.5 % bacterial contamination rate in VSSHP Tyks Orto tissue bank is low when compared to previous literature.

It has been noted that screening methods between swab and fragment samples results in different contamination rates and no international guidelines for screening methods exist (Baseri et al., 2021; M. Y. J. Hirn et al., 2001; Veen et al., 1994). That together with different procurement methods makes comparison of bacterial contamination rates between studies difficult. Swab samples are only able to reach microbes from the surface of the allograft and swab is considered to be more exposed for external contamination. Fragment samples in the other hand reaches deeper into the graft revealing microbes growing deeper from the surface, but fragment samples cover only small area of the graft. Various decontamination methods from irradiation and antibiotic solutions to mechanical lavage are used for disinfection of allografts during procurement process (Mohr et al., 2016). Low-pressure pulse lavage with sterile saline solution has been shown to be an effective disinfection method (M. Hirn et al., 2004; M. Y. J. Hirn et al., 2001; Salmela et al., 2002). Swab samples were used for microbiological screening and allografts were immersed in antibiotic solution during

procurement process priorly in tissue banking procedure in Turku (Aho et al., 1998; Virolainen et al., 2003). The study findings suggest that currently followed methods of pulse lavage with sterile saline and fragment sampling show good results regarding allograft sterility. We agree with author Baseri on the issue that tissue sampling process should be standardized.

Bacteria identified from microbiological cultures were divided to low and high virulence in study of Deijkers et al. (1997), and same division was followed by Naves et al. (2018) and Schubert et al. (2012). Low pathogenic microbes are considered to be skin commensals representing external contamination during procurement process and rarely causing clinical infection in receiver. Our study findings corresponded with findings of authors Deijkers and Schubert, suggesting that majority of contaminations resulted from external contamination during procurement process.

Length of stay in ICU has been associated to the risk of allograft bacterial contamination. Longer stay increases the likelihood of contamination. (Naves et al., 2018; Schubert et al., 2012). Naves et al. studied if number of leucocytes during treatment period in ICU predicts tissue contamination and did not find number of leucocytes relevant for predicting contamination rate. In TYKS ICU each donor was intubated and treated with standard broad spectrum antibiotic therapy (Meropenem 1 g i.v. 3 times a day) during the whole treatment period. With certain donors, combination antibiotic therapy (usually with Cefuroxime 1.5 g i.v. 3 times a day) was used. In the present study, mean treatment time in ICU was longer in donors with positive tissue bacterial cultures and combination antibiotic therapy was used more frequently. Number of leucocytes were not recorded but the average of highest CRP value was higher in donors with positive bacterial cultures. CRP levels were lower in patients with no positive tissue bacterial cultures and with short treatment period in ICU. Like number of leucocytes, high CRP level indirectly indicates inflammation reaction and possible bacterial infection in patient. Cut-off value for elevated CRP level was set at 40 mg/ml (Hogarth et al., 1997; A. Liu et al., 2010). It seems that long stay in ICU together with high CRP-values during treatment could be a risk factor for allograft bacterial contamination, but more research is required.

Interestingly, in the present study, both high virulent bacteria samples were found in the same donor, and the same donor had notably longer treatment time in ICU than other donors. The donor also had the highest number of positive tissue bacterial samples and high CRP value during treatment. These findings could suggest that factors considered above, especially notably long treatment time in ICU could be a risk factor for tissue contamination with high virulent bacteria, possibly not originated from external contamination. However, conclusions cannot be made because all high virulent bacterial findings were from a single study donor.

Study donor with the second longest treatment period had also a positive tissue bacterial sample. The two donors with longest treatment times had high CRP levels during treatment. However, in group of donors with no positive tissue bacterial samples, high CRP value was also measured from donor with long treatment time. Positive tissue bacterial samples were also found in donors with shorter treatment period and lower CRP levels. The findings are not unambiguous, and potential external contamination during procurement process hinders evaluation of these factors.

High experience of the surgical team reduces the risk of bacterial contamination during procurement process (Naves et al., 2018; Schubert et al., 2012). Large number of surgical team members is also associated to the risk of contamination, growing with every extra staff member in operating room (Deijkers et al., 1997; Segur et al., 2000). A small surgical team performs all allograft procurements in VSSHP Tyks Orto tissue bank. Surgical team consist of 2 to 3 nurses and 2 surgeons, so total number of surgical team members in operating room is 4-5 person at a time. Only experienced orthopedic surgeons with special training perform allograft procurements from musculoskeletal tissue donors. Operating room nurses attending allograft procurement belong to tissue bank staff and they have been given special training regarding aseptic working methods during tissue procurement process. Written instructions are created to guide working principles in operating room. Operating room is sealed during procurement process to prevent unnecessary passage of persons and microbes into the room. These arrangements ensure that surgical process is performed effectively, aseptically and without unnecessary action, minimizing possibility for external contamination of allografts. We believe that high hygienic standards followed in our practice, together with accurate patient selection and effective operation of highly experienced and trained surgical team are factors affecting low incidence of bacterial contamination rate in VSSHP Tyks Orto tissue bank.

There has been doubt that in some cases information about a potential musculoskeletal tissue donor in TYKS ICU does not reach tissue banking coordinator and orthopedic in charge of tissue procurement, leading to miss a suitable donor. Despite careful study, exclusion criterion for musculoskeletal tissue donation was not noticed in medical records of 6 donors. It is not possible to say for sure if there was an exclusion criterion that was not mentioned in medical records. It seems that at least three patients were missed due to lack of communication. Two other cases took place at holiday times, and there is no certainty if donor was missed due to lack of communication or if surgical team was not at disposal. Once surgical team was not at disposal due to holiday time. Lack of communication between departments of ICU and tissue bank in Turku has been noted before, and the study findings confirm that some suitable candidates for musculoskeletal tissue donation are

missed among the organ donors. Moreover, although small number of surgical team members improves efficiency in allograft procurement process and reduce allograft contamination, all members of the team may not be available especially during holiday times. Because of small number of potential musculoskeletal tissue donors, it is essential that any suitable donor does not remain unnoticed. Actions have already been made to reduce this possibility. Electronic instructions have been created and regular training is given to staff in ICU that in future suitable musculoskeletal tissue donors will not remain unutilized.

We acknowledge that this study has some limitations. The number of study donors was small and hence, statistical analyzes could not be made and statistically significant results could not be presented. Encouraging results were found in correlation between long stay in ICU and high CRP levels to possibility for tissue bacterial contamination, but study findings focused on a small group of donors reducing reliability of the results. Research with higher donor volume must be conducted to verify the findings of this study. Because of retrospective nature of the study, details regarding study donors based only on documents in medical records. For that reason, if exclusion criterion was not documented in the medical records, we could not be sure if there still was some undocumented exclusion criterion for musculoskeletal tissue donation. Comprehensive documentation is executed during treatment in ICU, and we believe that no significant details remained undocumented. Therefore, we assume that in those cases where documented exclusion criterion for musculoskeletal tissue donation was not found in medical records, exclusion criterion did not exist.

Conclusion

Based on this review, operation of VSSHP Tyks Orto tissue bank is in accordance with directives and guidelines established by national and European authorities and EATB concerning operation of musculoskeletal tissue bank. Number of harvested musculoskeletal allografts have increased markedly during last decades, and percentage of allograft bacterial contamination has decreased simultaneously in the tissue bank service in Turku, Finland. Bacterial contamination rate in VSSHP Tyks Orto tissue bank is low when compared to previous international literature. Long treatment period in ICU together with high CRP levels could be a risk factor for donor tissue bacterial contamination, but more research is needed. Some potential musculoskeletal tissue donors were missed among multiorgan donors, and actions have already been made that in future suitable musculoskeletal tissue donors will not remain unutilized.

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 for bones and tendons
Under 40 for meniscus and FOCA-grafts
Exclusion criteria
Multiple injury trauma or vast tissue damage
Hemodilution over 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 immunosupressive therapy
Chronic neurological disease
Use of human origin pituitary hormone
Previously received tissue transplant
Born in foreigen country or foreigen descent
Vaccinated with live-attenuated vaccine within 6 weeks
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)

Table 2. Donor and tissue details

	N	
Donors	28	
Male (%)	13	(46)
Female (%)	15	(54)
Mean age (years)	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 (sturt, 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)
Meniskus	1	(0.2)
Quadriceps tendon	1	(0.2)
Other	38	(6.0)
Total	636	(100)

Diagram 1. Number of harvested allografts and donors yearly

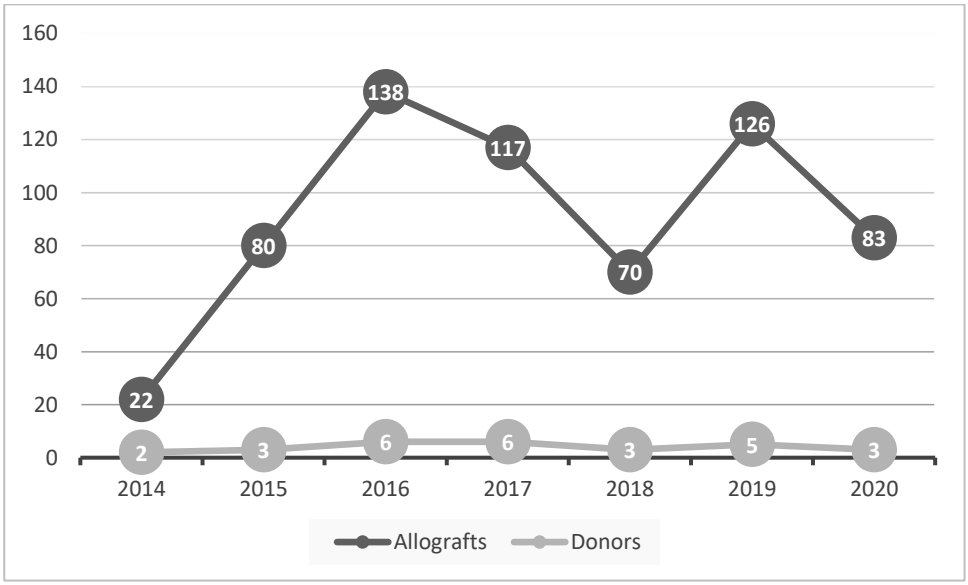

Table 3. Species and number of cultured bacteria from harvested allografts in VSSHP Tyks Orto tissue bank 2014-2020

		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

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

	N	
Donors with positive bacterial tissue cultures (%)	6	(21)
Mean treatment time in ICU (days)	5	(range 2-11)
Elevated CRP value (> 40 mg/ml) during treatment (%)	6	(100)
Average of highest CRP value (mg/ml)	146	(range 45-270)
Combination antibiotic therapy during treatment (%)	4	(66)
Traumatic cause of death (%)	1	(17)
Donors with no positive tissue bacterial cultures (%)	22	(79)
Mean treatment time in ICU (days)	2.8	(range 1-5)
Elevated CRP value (> 40 mg/ml) during treatment (%)	12	(55)
Average of highest CRP value (mg/ml)	53	(range 1-244)
Combination antibiotic therapy during treatment (%)	11	(50)
Traumatic cause of death (%)	4	(18)

Table 5. Characteristics of each musculoskeletal tissue donor utilized in VSSHP Tyks Orto tissue bank between year 2014 and 2020

Study patient no.	Treatment time in ICU (days)	Highest CRP value (mg/ml)	Use of combination antibiotic therapy	Number 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

Table 6. Contamination rates of musculoskeletal allografts harvested from cadaver donors in literature

First author	N (contaminated/total)	%
Paolin (2017)	5 211/10 035	52
Vehmeyer (2002)	2 546/5 710	45
Ibrahim (2004)	120/437	27
Journeaux (1999)	65/272	24
Terzaghi (2015)	635/2 778	23
Ilays (2021)	115/506	22.7
Viñuela-Prieto (2019)	227/1 162	19.5
Ivory (1993)	4/22	18
Naves (2018)	218/1 271	17.1
Liu (2002)	25/201	12.4
Bohatyrewicz (2006)	45/424	10.7
Schubert (2012)	365/3 612	10.1

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Supplement 1. List of authoritative dictates concerning operation of VSSHP Tyks Orto tissue bank

Law on the use of human organs, tissues and cells for medical purposes 101/2001. Government Regulation on the use of human organs, tissues and cells for medical purposes 594/2001

https://www.finlex.fi/fi/laki/ajantasa/2001/20010594

Law on the use of human organs, tissues and cells for medical purposes, proposal of the Board of Directors 54/2018

https://www.eduskunta.fi/FI/vaski/KasittelytiedotValtiopaivaasia/Sivut/HE_54+2018.aspx

Ordinance of the Ministry of Social Affairs and Health on the use of human organs, tissues and cells for medical purposes 1302/2007

https://finlex.fi/fi/laki/ajantasa/2007/20071302

Order 3/2014 Tissue bank services, Fimea

https://www.fimea.fi/documents/160140/744738/25766_Maarays_3_2014.pdf

Directive 2004/23/EC of the European Parliament and of the Council of 31 March 2004 on setting standards of quality and safety for the donation, procurement, testing, processing, preservation, storage and distribution of human tissues and cells

https://eur-lex.europa.eu/legal-content/EN/ALL/?uri=CELEX%3A32004L0023

Commission Directive 2006/17/EC of 8 February 2006 implementing Directive 2004/23/EC of the European Parliament and of the Council as regards certain technical requirements for the donation, procurement and testing of human tissues and cells

https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX%3A32006L0017

Commission Directive 2006/86/EC of 24 October 2006 implementing Directive 2004/23/EC of the European Parliament and of the Council as regards traceability requirements, notification of serious adverse reactions and events and certain technical requirements for the coding, processing, preservation, storage and distribution of human tissues and cells

https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX%3A32006L0086

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