Medtronic

Mitigating disease transmission risks:

Overview of safety enhancements implemented in SygnaCel™ CBM processing

Introduction

Allograft bone grafts are an important tool for surgeons treating patients undergoing procedures that require bone grafting, such as spinal fusion, orthopedic trauma, or revision surgery[1]. Often, patient's own autograft bone is in short supply and may lack quality. When these challenges arise, surgeons typically augment available autograft tissue with bone grafts, such as allografts, to enhance their likelihood of successful clinical outcomes. Allograft is derived from donated bone tissue following a donor's death and is regulated by FDA under Title 21 Code of Federal Regulations (21 CFR) Part 1271.

In spine procedures utilizing bone graft, the term "allograft" typically encompasses three primary product technology categories: mineralized bone chips, demineralized bone matrix (DBM), and cellular bone matrix (CBM). Unlike the other allograft categories, only CBM products contain live cells. The cells found in CBMs reside on fragments of mineralized bone which are often formulated in combination with demineralized bone matrix.[2] To preserve the live cells in a CBM product, the tissue recovery, processing, and final storage of the tissue in a cryoprotectant fluid, must happen in a very short period time. For SygnaCel™ CBM, all donor recovery, processing, and packaging are completed within 96 hours of a donor's time of death. SygnaCel™ CBM is stored in an optimized biocompatible cryoprotectant containing glycerol in lactated Ringer's solution. The product is stored at -70C to -80C and can be rapidly thawed and made available for use within 3-6 minutes should a surgeon require the product in surgery.[3]

While CBMs have been reported to aid in fusion,[2,4] all allografts, including CBMs, carry a risk of infectious disease transmission from the donor to the recipient. Unlike highly processed or sterilized acellular allografts, CBMs contains viable cells and therefore management of microbial risk is of elevated importance. In recent years, two *Mycobacterium tuberculosis* outbreak events have been linked to bone matrix products containing live cells. Some patients affected by these outbreaks have died. Therefore, it is imperative that CBMs are processed meticulously with enhanced safety measures to significantly reduce disease transmission risks.

Tissue recovery organizations (TROs) follow the best practices in tissue recovery and processing set by American Association of Tissue Banks (AATB) and FDA to mitigate this potential risk. TROs carefully screen all potential donors for risk factors and test donor tissues for infectious diseases. A qualified medical director reviews the screening results to assess donor eligibility for conversion into a CBM, aiming to minimize disease transmission. Only donor tissues meeting the stringent criteria set by AATB and FDA are processed into CBMs.

SygnaCel™ CBM safety enhancement process

To minimize the risk of disease transmission from donor bone tissue, Medtronic has implemented several measures. These include partnering exclusively with licensed tissue recovery organizations (TROs) who strictly adhere to our rigorous donor standards, which meet or exceed AATB and FDA screening and testing requirements. In addition, Medtronic has also developed and implemented the proprietary Xpel™ processes that mitigates microbiological contamination from donor tissues during processing of the SygnaCel™ CBM product. Finally, as part of the quality assurance program at Medtronic, each lot of SygnaCel™ CBM undergoes a series of sterility tests prior to release for customer usage.

The aforementioned processes comprise a three-stage approach aimed at minimizing disease transmission risks associated with SygnaCel™ CBM:

1. **Pre-Processing** – Strict screening of donor and testing of tissues obtained during donor tissue recovery to determine eligibility.

2. **In-Processing** – Tissue washing, comprehensive bioburden assessment, and proprietary Xpel™ process that includes broad-spectrum antibiotic/antimycotic treatment during tissue processing.

3. **Post-Processing** – Lot release testing, inclusive of finished product USP<71> sterility testing in conjunction with extended sterility tests, a validated laboratory-developed *Mycobacterium tuberculosis* Nucleic acid Amplification Test (Mtb NAT), and culture testing optimized for *Mycobacterium tuberculosis* detection.

Figure 1 shows our three-stage process surpassing the safety criteria established by both AATB and FDA, demonstrating our unwavering commitment to enhancing product safety.

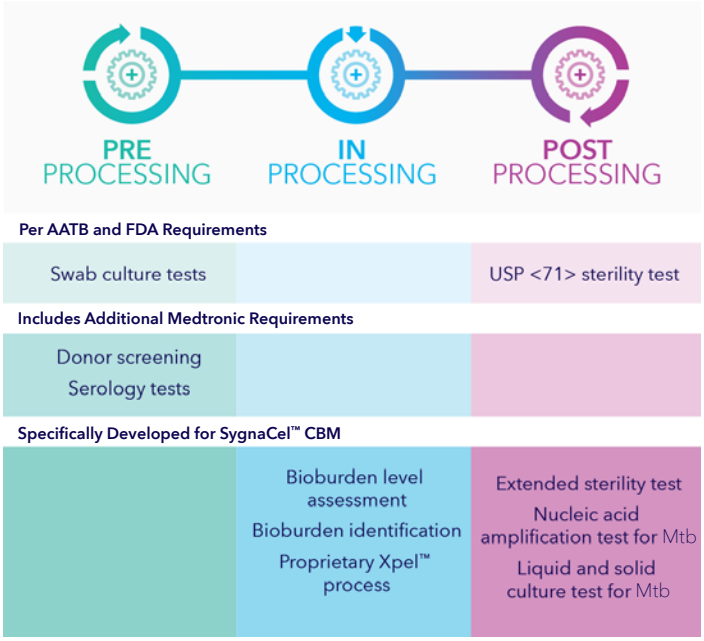


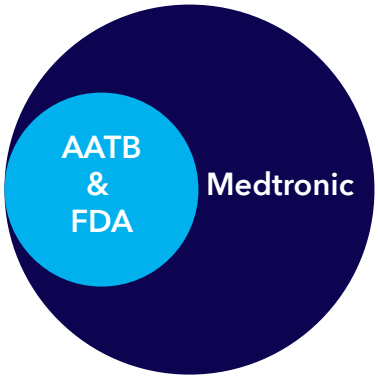
Figure 1. The Medtronic three-stage approach aimed at minimizing disease transmission risks associated with SygnaCel™ CBM.

All Pre-Processing steps are carried out by TROs recovering the donor tissues. The medical director at these TROs screens donors based on Medtronic specifications to determine donor eligibility. The bioburden level assessment and identification during In-Processing stage is conducted at a Medtronic contracted lab, while the proprietary Xpel™ processing is performed in-house. During Post-Processing stage, Medtronic contracts with specialized labs to conduct *Mycobacterium tuberculosis* testing by multiple methods. All In-Processing and Post-Processing results conducted by Medtronic contracted external labs are reviewed by Medtronic Quality and must meet predefined standards to be approved for lot release.

Pre-Processing

The Pre-Processing stage includes comprehensive donor screening, along with swab culture tests to detect microbial presence, and serology tests to detect infectious agents. Donor screening is the first line of defense against disease transmission. TROs and their associated tissue banks recovering donors for Medtronic conduct comprehensive interviews with the donor's family to identify exclusionary risk factors which may prohibit tissue donation. During tissue recovery, swab cultures are taken to assess bioburden and identify exclusionary microorganisms (e.g. *Clostridium sp.* and *Streptococcus pyogenes*) that may be present on recovered donor tissue. Serological testing with donor blood samples is conducted to detect infectious diseases. Depending on circumstances, an autopsy may also be conducted to determine the underlying cause of death. For SygnaCel™ CBM, Medtronic has developed additional donor screening and serology criteria to further mitigate risks. These criteria established by Medtronic include a strict age limit for donors and additional exclusion factors that exceed AATB requirements for tissues containing live cells. A tissue bank Medical Director at our tissue suppliers reviews these comprehensive data. Only donors meeting all Medtronic criteria are deemed eligible for SygnaCel™ CBM.

Figure 2 summarizes Medtronic donor criteria for SygnaCel™ CBM compared to FDA and AATB donor criteria for a viable cellular product.



SygnaCel™ donors
90+ screening/testing factors EXCEEDING FDA and AATB requirements

Figure 2. Graphic depicting Medtronic donor requirements for SygnaCel™ CBM exceeding FDA and AATB requirements.[5]

In-Processing

While the Pre-Processing steps are comprehensive, they do have limitations. Therefore, Medtronic employs additional In-Process techniques to safeguard against infectious agents that may have evaded detection during the initial Pre-Processing stage.

SygnaCel™ CBM is exclusively processed at a Medtronic owned and operated tissue bank with all necessary licenses and registrations for the processing of tissue products containing live cells. SygnaCel™ CBM contains live cells on fragments of mineralized cortical cancellous bone in combination with demineralized cortical bone matrix fibers. The cortical fibers undergo our proprietary D-MIN™ demineralization process, which involves acid and alcohol washes which are effective at removing or neutralizing various bacteria, fungi, and viruses.[6] However, the mineralized cortical cancellous bone chips cannot be exposed to acid or alcohol as it could kill the desirable bone forming cells associated with this tissue.

To inactivate microorganism contamination from the mineralized cortical cancellous bone component of SygnaCel™ CBM, Medtronic treats these tissues with its

proprietary Xpel™ process which incorporates extended tissue conditioning in a broad-spectrum triple antibiotic/antimycotic cocktail. This innovative technique was meticulously crafted by Medtronic engineers and microbiologists to neutralize bacteria and yeast while preserving viability of the desirable bone forming cells. The Xpel™ process has been rigorously tested to assess its performance. Drawing from over 30 years of tissue processing experience at our Medtronic tissue processing facility, and insights from AATB, a panel of representative bacteria and yeast found in donated tissues were subjected to the Xpel™ process. A summary of the kill efficiencies for these microorganisms ranges from 3 to greater than 7 log reduction, as detailed in Table 1.

Table 1. Xpel™ kill efficiencies for representative microorganisms[7]

	Microbe Type	Avg. Log Reduction	Avg. % Reduction
Staphylococcus aureus	Gram+ cocci	> 7	99.999973
Enterococcus faecalis	Gram+ cocci	5	99.99957
Pseudomonas aeruginosa	Gram- rod	> 6	99.999939
Corynebacterium xerosis	Gram+ rod	> 7	99.999971
Bacillus atrophaeus	Gram+ rod (spore-former)	3	99.86
Clostridium sporogenes	Anaerobe (spore-former)	4	99.9943
Candida albicans	Yeast	7	99.999988
Aspergillus brasiliensis	Mold	0	N/A

To ensure the Xpel™ process is operating within allowable limits, a sampling of tissues entering the process are destructively cultured to assess their bioburden levels at a Medtronic-contracted independent lab using methods optimized for bioburden detection in cadaveric cortical cancellous bone. This qualifying step measures and identifies the initial bioburden, verifying that the Medtronic Xpel™ process will be effective in neutralizing the bioburden. Additionally, the wash fluids prior to the Xpel™ process undergo culturing by Medtronic labs to identify any organisms present.
If non-compliant organisms, such as *Aspergillus brasiliensis* (mold) is detected at this stage, then the entire affected tissue batch is promptly removed from production.

Post-Processing

As the final defensive barrier against disease transmission, every lot of SygnaCel™ CBM undergoes comprehensive testing to assess product sterility. Given its limited chemical processing, this testing is critical in upholding the overall quality of the SygnaCel™ CBM product. Only product batches which pass these sterility tests, and all other lot-release criteria defined for SygnaCel™ CBM product, receive approval for release from the Medtronic tissue processing facility. This rigorous process ensures that every product delivered to our customers adheres to the highest quality standards set by Medtronic. The following provides a concise overview of the sterility tests conducted.

USP<71> sterility test

A sampling of each finished batch of SygnaCel™ CBM (including inner packaging) is subjected to destructive testing under both aerobic and anaerobic conditions. Testing is conducted at Medtronic labs in compliance with the U.S. Pharmacopeia method, with a 14-day incubation period to monitor for signs of microbial growth. This test is non-specific and can detect a wide variety of microorganisms. Test must result in "no growth."

Extended sterility test

A sampling of each finished batch of SygnaCel™ CBM (including inner packaging) is subjected to destructive testing under both extended duration aerobic and anaerobic conditions at Medtronic labs. Testing is based on the 14-day U.S. Pharmacopeia method, but cultures are assessed for growth out to 21 days. This test is non-specific and can detect a wide variety of microorganisms and is designed to detect slow-growing microorganisms that might fall below the detection limit observed in the standard USP<71> sterility test.
Test must result in "no growth."

Nucleic acid Amplification Test (NAT) for *Mycobacterium* tuberculosis complex†

A sampling of each finished batch of SygnaCel™ CBM is subjected to destructive testing to assess for DNA associated with *Mycobacterium tuberculosis* complex. This is a highly specific laboratory-developed assay, validated by a Medtronic-contracted independent testing lab, to detect *Mycobacterium tuberculosis* complex. Multiple product samples are meticulously prepared and analyzed from each batch, with each test conducted in triplicate to enhance precision and reliability of detection. Estimated limit of detection (LOD) by PCR is 10.9 CFU/cc.[8] Test result must be "negative".

†There are currently no FDA approved tests for *Mycobacterium tuberculosis* detection in cadaveric specimens. Negative results do not guarantee the product is free of Mtb organisms. This testing for Mtb may not detect inactive (latent) mycobacteria.

Liquid and solid culture test for *Mycobacterium* tuberculosis[1]

- A sampling of each finished batch of SygnaCel™ CBM is subjected to destructive testing by liquid culture method by a Medtronic-contracted independent laboratory with expertise and equipment to perform this specialized testing. Following extraction, samples are tested in an automated culture system optimized for *Mycobacterium tuberculosis* growth. Through measurement of oxygen consumption, *Mycobacterium tuberculosis* metabolism can be detected. Estimated limit of detection (LOD) for liquid culture method is 1.1 CFU/cc.[9] This 6-week duration test must result in "no growth."
- A sampling of each finished batch of SygnaCel™ CBM is also subjected to destructive testing by solid agar culture method. Testing is conducted by a Medtronic-contracted independent lab with expertise in this method. Following extraction, samples are inoculated onto agar plates optimized for *Mycobacterium tuberculosis* growth promotion. Estimated limit of detection (LOD) for solid culture method is 4.4 CFU/cc.[9] This 8-week duration test must result in "no growth."

Conclusion

In summary, SygnaCel™ CBM undergoes rigorous screening, processing, and testing at various stages from recovery to final product release, ensuring multiple layers of quality assurance. Our safety measures are incorporated into three key stages of processing;

- **Pre-Processing** - a stringent tissue screening and testing protocol that identifies exclusionary risk factors, followed by
- **In-Processing** - the Medtronic proprietary Xpel™ process that effectively neutralizes bioburden during processing, and culminates in
- **Post-Processing** - an extensive sterility testing program intended to improve detection of slow growing microorganisms.

While these measures mitigate the risk of disease transmission to patients, it is important to recognize that certain diseases and infectious agents may evade detection or elimination during processing. For more information regarding the potential risks of SygnaCel™ CBM, please refer to the Important Information section below. Medtronic is committed to providing quality products that comply with all applicable laws, regulations, and company policies. As AATB and FDA update guidelines and issue new recommendations, Medtronic is committed to evaluating and implementing them to minimize disease transmission. Through these combined efforts, Medtronic will continue to deliver SygnaCel™ CBM that meets customer safety expectations and complies with all relevant regulations.

SygnaCel™ CBM important information

DESCRIPTION: SygnaCel™ Cellular Bone Matrix is a formulation of human cryopreserved viable cortical cancellous bone matrix and demineralized bone fibers.

INDICATIONS FOR USE: SygnaCel™ Cellular Bone Matrix can be used as a bone graft in orthopedic or reconstructive procedures. Use SygnaCel™ Cellular Bone Matrix in combination with autologous bone and/or bone marrow aspirate.

STERILITY: SygnaCel™ Cellular Bone Matrix is labeled as "Aseptically Processed, Passes USP Sterility Tests." It was aseptically processed and tested for sterility according to the current US Pharmacopeia.

CONTRAINDICATIONS: The presence of infection at the implantation site is a contraindication for the use of this allograft.

WARNINGS:

- **SygnaCel™ CBM is a cellular bone matrix product containing viable cells.**
- **Bone matrix products containing viable cells have been linked to, or associated with, two Mycobacterium tuberculosis (Mtb) outbreak events in the United States.**
- **Some affected by these outbreaks have died.**
- **Current donor screening, processing, and testing cannot fully eliminate the risk of disease transmission from bone matrix products containing viable cells.**

POTENTIAL ADVERSE EVENTS: Donor screening methods are limited. Therefore, certain diseases and infectious agents may not be detected and/or eliminated during processing. The following complications of tissue transplantation may occur:

- Loss of function or integrity of transplanted tissue due to resorption, fragmentation, or disintegration including associated loss of continuity, or displacement, and/or fracture at treatment site.
- Non-union (or pseudarthrosis), delayed union, and/or mal-union.
- Immune rejection of transplanted grafts or infection.
- Transmission or causation of known diseases as well as diseases of unknown etiology and characteristics.
- Transmission of known infectious agents including HIV, Hepatitis B, Hepatitis C, and bacteria (e.g. syphilis, Mycobacterium tuberculosis).
- Death.

CAUTION: This allograft may contain trace amounts of processing agents. Caution should be exercised if the patient is allergic to any of these agents.

PRECAUTIONS: Despite extensive donor selection and qualification processes, transmission of disease is still possible. Bacterial or fungal infection may also occur.

DONOR SCREENING AND TESTING: The donor was screened in accordance with FDA regulations and standards established by the AATB. The donor was also screened for HIV, Hepatitis, and CJD/vCJD risk factors. Donor blood was tested for communicable disease by a laboratory registered with the FDA using FDA approved, licensed, or cleared tests. Results of tests were negative or non-reactive. Laboratory-developed Mtb NAT (nucleic acid amplification testing) and culture testing for Mtb was also performed. Note that there are currently no FDA approved tests for Mtb detection in cadaveric tissues. Testing for Mtb may not detect inactive (latent) mycobacteria.

For a complete list of indications, safety, and warnings, please visit https://manuals.Medtronic.com

References

1. Kang J, An H, Hilibrand A, Yoon ST, Kavanagh E, Boden S. Grafton and local bone have comparable outcomes to iliac crest bone in instrumented single-level lumbar fusions. *Spine* (Phila Pa 1976). 2012;37(12):1083-1091.
2. Eastlack RK, Garfin SR, Brown CR, Meyer SC. Osteocel Plus cellular allograft in anterior cervical discectomy and fusion: evaluation of clinical and radiographic outcomes from a prospective multicenter study. *Spine* (Phila Pa 1976). 2014;39(22):E1331-E1337.
3. Internal benchtop testing data.
4. Vanichkachorn J, Peppers T, Bullard D, Stanley SK, Linovitz RJ, Ryaby JT. A prospective clinical and radiographic 12-month outcome study of patients undergoing single-level anterior cervical discectomy and fusion for symptomatic cervical degenerative disc disease utilizing a novel viable allogeneic, cancellous, bone matrix (Trinity Evolution™) with a comparison to historical controls. *Eur Spine J.* 2016;25(7):2233-2238.
5. Internal comparison data on file.
6. In vitro testing data on file.
7. In vitro testing data on file.
8. In vitro PCR validation data on file.
9. In vitro testing data on file.

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