REVIEW ARTICLE

Bovine Pericardium-Chitosan As A Biomaterial Prospect For Substitute and Accelerate Tissue Healing of Dural Defect: A Review

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ABSTRACT

In the field of neurosurgery, duraplasty is commonly performed after intradural surgery which requires excision of the dura mater and/ or in the case of head injury. Over the past century, several biomaterials have been investigated as a dural substitute for duraplasty. Bovine pericardium (xenograft) has been widely studied in clinical trials and is commercialized as a graft in cardiac surgery, as well as a dural substitute. Bovine pericardium as a dural substitute is elastic, suturable, and provides watertight capabilities. Another biomaterial that also has many biologic advantages is chitosan. Chitosan has been widely known as a good agent for wound healing, because of its hemostatic properties and its ability in reducing inflammation. The availability and benefits of both materials make it an interesting subject for further research. This review aims to provides knowledge about the potential bovine pericardium in combination with chitosan as a dural substitute, which not only provides watertight properties, but also accelerates tissue healing in dural defects.

Keywords: Bovine Pericardium, Xenograft, Chitosan, Dural Substitute, Tissue Healing

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INTRODUCTION

The outermost of the meninges membrane called dura mater, is firmly attaches to the internal tabula calvaria and protects the brain (1). As a protective membrane, the dura mater also has mechanical functions in order to protect the calvary from brain pulsations. Dura mater is able to transmit pressure from one bone to another and reduces pressure waves in the cerebrospinal fluid (2). The dura mater can be damaged if a head injury occurs or due to excision during intracranial surgeries, resulting in dural defects (3)

Dural defects are a complication that can lead to leakage of the protective fluid surrounding the brain and spinal cord, resulting in death (4). The protective fluid is called cerebrospinal fluid (CSF). Primary dura mater repair using sutures is the ideal method of avoiding CSF leakage, but if a large portion of the dura mater is excised or extensive damage is found in the dura mater, then this action is not amenable (3,5). Based on these problems, an adequate biomaterial as a dural substitute is needed to close the dura mater defect.

Currently, many materials have been used as dural substitutes for patching dural defects. The materials that are commonly used can be grouped into 4; autograft, allograft, xenograft, and synthetics. Unfortunately, each material has its own advantages and disadvantages. Current researches are heading towards the development of biomaterials that can provide solution to the challenges in clinical practice, including dura mater defects. Bovine pericardium is a biomaterial that have

a similar mechanical properties with human dura mater (6). Another biomaterial, chitosan has been widely known as a good agent for wound healing, because of its hemostatic properties and its ability to reduce inflammation (7). The properties of both materials make them a promising solution to the challenges in finding the ideal dural substitute.

CLINICAL IMPORTANCE AND HISTORY OF DURAPLASTY

Dural defects due to trauma or left uncovered after surgery, can cause serious complications, such as infection of the brain and its surrounding structures, cerebrospinal fistulas and liquorrhea, or brain's lining scars that may lead to traumatic epilepsy (8).

By the end of the XIX century, many surgeons noticed that traumatic epilepsy caused by the scars formed upon a traumatic brain injury was refractory to surgical treatment if the dural defect still exists (9). Various methods of interposing artificial materials for dural defect were proposed (10). At first, they tried to use tiny sheets of inert metals: gold, silver, platinum. Other people were using non-metallic grafts: gutta-percha or celluloid plates. These early attempts were disappointing as the body rejected the foreign materials in various ways, e.g., by destructing a plate with infiltrated connective tissue and by the attachment of the brain and the tissues around the material (11,12).

There were attempts to use biological materials as transplants. Freeman (1908) and Saar (1911) reported the experiments on dogs and rabbits where a dural defect was closed with an egg film. The results revealed the formation of a connective tissue capsule, histologically similar to the dura mater tissue, which prevented the development of adhesions provided that the underlying membranes and the cortex remained intact. However, if the underlying membranes were damaged the risk of adhesions significantly increased. Also, this foreign material was rejected by the body, which often resulted in an infection (13,14).

In the late 1970s, studies showed that grafting a fascia led to gross cicatricial fusion between the brain and the overlying tissues in experimental animals; such developments may increase the risk of epilepsy. Additional disadvantage of using autograft is the increased surgery time required to harvest material for transplantation. Resorption of the transplants is associated with a response by the surrounding tissues, leading to tissue adhesion and scar formation to the brain (15).

Subsequent developments in the repair of dural defects were based on discoveries in chemistry and physics, and the rapid growth of the chemical industry in the 1960s to 1980s. Innovations in the techniques of preserving cadaveric biological tissues allowed scientists to harvest larger amounts of materials for potential plastic surgery and store them for a longer time. Various treatments for this purpose (with formalin, lyophilization, freezing) were proposed. Among them, lyophilization was most often used to preserve cadaveric dura mater (14,16). It was found that grafts processed by lyophilization preserved not only their morphological structure but also their intact DNA and RNA, which was crucial for cell division and transplant engraftment. Lyophilized tissue is low-toxic, gradually degradable after transplantation, and gets replaced by the connective tissue of the recipient, which is very similar to the dura mater tissue (17). At present, these implants are not practical due to several reasons: a relatively strong immune response by the recipient; legal problems with the removal of cadaverous dura mater; the possibility of pathogen transmission (HIV, hepatitis, syphilis, prion infections) is not ruled out; the unusual shape and the small size of the defect make it technically difficult to provide proper closure using the transplant (18).

Unsatisfactory results of the described methods led to the development of fundamentally new materials — xenografts (19). Those are produced from type I animal collagen and treated in certain way that the material does not cause an immunological reaction in the recipient (20). The most commonly used transplants are made from pericardial tissue, Achilles tendon, and fetal skin that is derived from bovine, also small intestine tissue derived from porcine, or horse collagen (21).

MATERIALS OF DURAPLASTY

Duraplasty is a dural repair procedure to close the defect, so there is no CSF leak. The procedure can be performed using sutures or artificial dural substitutes along with other supporting materials (22). The duraplasty procedure requires a biomaterial or graft to cover the dura mater defect. Graft biomaterials for dural defects repair from biological sources are autograft, allograft, and xenograft. Autograft can be taken from the temporalis fascia and fascia lata. Allograft is taken from cadaveric dura mater (23), it is available in some countries in the form of commercial products (24). Xenografts derived from a species tissue that is different from the recipient, such as collagen fleece derived from porcine collagen, bovine pericardium, and bovine dermis collagen (23,24). Dural substitute from a synthetic source, include polytetrafluoroethylene and vicryl mesh (24).

The use of autograft to cover dural defects is quite difficult because the amount of material available is often insufficient to cover large defects and the properties of the material are still more inferior than allograft and xenograft material (24,25). Also, the use of autograft is associated with patient morbidity because graft harvesting requires another incision to be made

(26). Therefore, ready-to-use biomaterial/ graft is urgently needed and more clinically applied in dural defect closure.

However, until now it is still difficult to find the perfect dura mater replacement material because each material has its shortcomings. The ideal dural graft material has the criteria of non-toxic, does not cause a harmful immune reaction, free from infectious microorganisms, ready to use, not adherent to the brain and surrounding tissues, and also capable of triggering native dural reconstruction (5,25).

Allograft materials

The innovation of ready-to-use biomaterial derived from allograft material by taking cadaveric human dura mater has been around since the 1960s. The allograft is superior in reducing patient morbidity (such as postoperative pain and surgical time), readily available, and easier to use than autograft (27). However, the legality and possibility of viral pathogen transmission lessen the use of this material (14,16). Among the possibility of transmission infectious disease from a cadaver, viruses and prions are the most difficult to investigate from donors (27). A study reported 114 cases of Creutzfeldt-Jakob disease (CJD) worldwide that were fatal due to the use of cadaveric human dura mater and 139 cases of CJD due to growth hormone contamination (28). Compared to autograft, this graft is associated with more potential for infection and rejection. Therefore, allograft as dural substitution is no longer in demand (29).

Synthetic materials

Synthetic materials are an alternative to dural substitutes, which are inert, can be produced indefinitely and the characteristics of the materials are modifiable. Expanded polytetrafluoroethylene (E-PTFE) is often used clinically because it is relatively safe among other synthetic materials (24). This material has the following advantages: (i) inert (30,31); (ii) decreased infection risk (30); (iii) there is no possibility of disease transmission (30,32); (iv) provides no adhesion with the host tissue (33,34); and (v) unable to cause malignancy in the long-term period (31). As a dural substitute, E-PTFE has been used successfully for the sellar dural repair (15), and defect repair (16,19). However, based on several reports E-PTFE is associated with CSF leak (15) through the suture line (35), and severe infection, thus requiring removal and re-surgery immediately (35,36).

Xenograft materials

The xenograft is derived from collagen-based materials of living things, which are preferred because of its abundant sources (5,24). Collagen is the largest constituent in body tissue and contributes to the elastic properties of the biomaterial which allows it to conform to the host tissue (37). Xenografts have the following advantages: (i) in the short term unable to cause local and systemic complications (38,39); (ii) capable as a scaffold for the formation of endogenous neo dura mater (38,40–44); (iii) easy to use (40,42–46); and (iv) mechanically able to prevent drainage of the CSF outside (40,43,47). Xenograft which is commonly used for medical purposes and has been widely commercialized is the bovine pericardium. Pericardium tissue derived from bovine is superior due to higher collagen type I content, compared to porcine and equine (37). As biomaterials, several studies on the application of bovine pericardium have been published such as in the surgical procedure of angioplasty, prosthetic heart valve surgery, atrial septal defect closure, and also duraplasty (24,48–51).

The summary of the materials stated above are listed in table I below

Table I. Types of dural substitute materials and their characteristics

Material	Description	Advantages	Disadvantages
Autologous	Taken from the patient's skull periosteum, fascia lata, cap aponeurosis, or temporal muscle fascia before or during the operation to repair the dural defect	Effectively avoid the risk of immune rejection and potential transmission of pathogenic microorganisms and reduce medical costs	Limited size and shape, so it is not suitable for repairing large dural defects
Allogenic	Freeze-dried human cadaveric dura mater	Gradually being left out due to its diadvantages	High risk of complications; progressive dementia, convulsions, and other clinical symptoms, namely a specific form of Creutzfeldt–Jakob disease (CJD), dura mater graft-associated CJD (dCJD)
Xenogenic	Derived from porcine, bovine, equine, and other animal tissues, most commonly used dural substitute	Maintains the structure of fibrous scaffolds in the extracellular matrix of animal tissues Crisscrossing collagen fibers provides a favorable microenvironment for reconstruction of the dura mater	Some scholars believe that the use of animal-derived materials such as bovine or porcine tissue might increase the risk of disease transmission
Synthetic	Expanded polytetrafluoroethylene and polyurethane dural substitute (nonabsorbable) PGA, copolymer of L-lactic acid and ε-Caprolactone, or copolymer of lactide and polydioxanone (absorbable)	Adjustable degradation rate and watertightness through changing the ratio of lactic acid to glycolic acid (PLGA)	Absorbable; difficult to degrade, may lead to foreign body reactions Compared with collagen- based dural substitutes, lack the biological functions of inducing cell migration and promoting the secretion of related cytokines.

BOVINE PERICARDIUM COMBINED WITH CHITOSAN AS DURAL SUBSTITUTE

We conducted a review of the literature without publication time limits using the keywords (bovine pericardium OR chitosan) AND (dura mater OR biomaterial); PubMed and Medline reported 1137 findings. Additionally, we conducted a manual search and discovered 6 articles. We excluded articles written in non-English languages, articles with irrelevant topics or improper variables, and duplicates. Then, for this analysis, we reviewed 25 papers fulfill our eligibility criteria for describing the bovine pericardium and chitosan as dural substitute. Figure 1 depicts the flowchart of the literature findings.

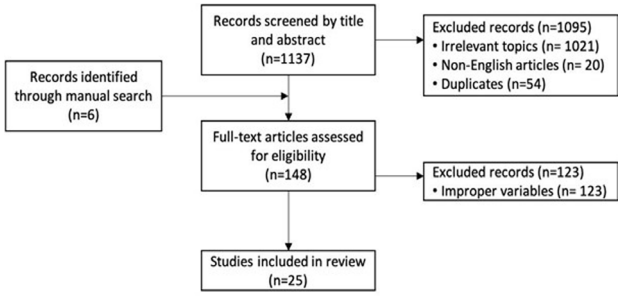

Figure 1. Flowchart of the literature review

BOVINE PERICARDIUM IN DURAPLASTY

The bovine pericardium is an abundant material that is easily available in various places, composed of a collagen network that is considered suitable for use in duraplasty. In this section, we will discuss the resume of studies about this material.

The application of dural substitute using bovine pericardium: Clinical studies

A clinical study of lyophilization bovine pericardium was conducted and reported in Malaysia in 1998-1999. A dural substitute was performed on 22 patients with various diagnoses. This study reported that 16 patients showed good outcomes, 1 patient showed moderate, and 2 patients showed poor outcomes. A total of 1 patient experienced astrocytoma relapse 1 year after surgery, and 2 patients died was not related to the surgery, but from intracranial hypertension. Four of the six patients with good outcomes in this study were trauma patients, while majority of the patients were tumor patients (50).

A good outcome related to the application of dural substitute from bovine pericardium was also found in 35 patients, in America. The diagnoses in the study included meningiomas, Chiari malformations, trigeminal neuralgia, metastatic tumors, astrocytomas, subarachnoid hemorrhage, pineal tumors, subdural hematomas, and artery venous malformations. Only one patient reported having a bad outcome in the study, namely patients who had grade 4 subarachnoid hemorrhage. Other reported complication is CSF leakage, but in exploratory surgery, the leak did not originate from the graft (6).

Another clinical study was conducted on 102 tumor patients (as majority of the cases) by comparing the lyophilization allograft and bovine pericardium. The results concluded that bovine pericardium lyophilization was superior to allograft based on its physical quality, which is watertight, easy to cut, and suturable. This study also conducted a cohort of pericardial histopathological features and found that pericardial bovine was incorporated excellently in the host membrane. However, in this study more postoperative complications were found in the pericardium bovine, namely 1 case of infection, 1 case of bleeding, 1 complication of sepsis, 2 cerebral edema, and 1 case of thrombosis (17).

Despite the good physical quality and postoperative results, clinical cases are reporting allergic reactions due to the use of bovine graft as a dural substitute. Foy et al reported postoperative complications namely CSF leak 3 weeks after the dural repair, followed by signs of allergy (52). A skin antigen and radioallergosorbent test (RAST) strongly suggesting a beef allergy. The patient recovered well after the removal of the bovine graft. This study concludes that allergic reaction is possible due to the use of bovine graft without chemically cross-linked collagen protein treatments. The decellularization process may also allow the bovine antigen to be left behind and causes the recipient to increase the immune response to the graft (52).

As commonly known, antigenicity and immune reactions to bovine tissue graft are one of the biggest barriers in clinical practice. The antibody response usually occurs due to the galactose-α-1,3-galactose (gal) component present in the xenograft (53). Therefore, bovine pericardium for clinical purposes is chemically fixed using glutaraldehyde to reduce antigenicity and increase sterility (54).

In-vitro studies of bovine pericardium use as dural substitute

In designing biomaterials as a dural substitute, mechanical properties are one of the criteria that must be considered. The mechanical properties of biomaterial can be assessed from examining its tensile strength and elongation. The bovine pericardium tensile strength value is not quite different from human dura mater in general (4,70-12,76 MPa) (55), which ranges from 6-18,96 MPa (56,57), while the artificial dura mater standard range between 4-20 MPa. If the tensile strength value is lower than the standard, the dural substitute is unable to withstand intracranial pressure (58). Also, the elongation value of bovine pericardium is not vastly different from the standard, ranging between 20,67-39,5% (56,57). While the elongation of human dura

mater ranges from 7-20% (4,59), and the artificial dura mater may preferably 30-150% (58). If the elongation value of the dural substitute is lower than the standard, it will be difficult to suture under pressure. Meanwhile if it is higher than the standard, then a dural substitute will be too stretched, resulting in CSF leakage when there is an increase of intracranial pressure and handling difficulties may arise (58). Therefore, dural substitute from bovine pericardium is claimed to have ideal physical qualities.

From the assessment of cellular and humoral immune reactions both in-vivo and in-vitro, bovine pericardium has a good biocompatibility with the host tissue (17,60). However, the bovine pericardium is still reported to be relatively toxic, based on several studies. The bovine pericardium has a relatively higher inflammatory reaction, although it is still within safe limits compared to synthetic dural substitute (60,61). Another in-vitro study reported that bovine pericardium-treated glutaraldehyde inhibited the growth of cell colonies and was causing cell death due to residual glutaraldehyde (62). Therefore, the addition of natural polymer is needed to increase its biocompatibility.

CHITOSAN IN DURAPLASTY

Chitosan is a natural polymer derived from partial deacetylation of chitin. Chitin, the second most abundant polysaccharide after cellulose, is found in the exoskeleton of crustaceans, insects, and fungi. Chitosan has many interesting biological properties, and it is often applied specifically both in medicinal and medical application. Chitosan has several advantages, i.e. its compatibility with the wound area, good resistance to the inflammatory process, and its ability to induce cell regeneration without fibrosis (7,63). Chitosan is also known to have several interesting properties that will be discussed below.

Antimicrobial

Chitosan's antimicrobial properties has been studied both in vivo and in vitro. Chitosan have antimicrobial effects on several types of organisms such as bacteria, algae, and fungi in various forms (solutions, films, and composites). Chitosan is able to kill bacteria and inhibits bacterial growth; in a recent study chitosan is found to have better bacteriostatic properties than bactericidal (64).

Haemostatic and wound healing

Chitosan is reported to increase the hemostatic effect on wound healing by triggering erythrocyte aggregation through interactions between platelet activation with sialic acid residues on erythrocyte membranes that have negative ions. In addition to trigger platelet aggregation, chitosan also increases the release of PDGF and TGFβ 1 from platelets which will accelerate the process of haemostasis and wound healing (65). Chitosan in contact with biological component does not have a high antigenic effect, has good biocompatibility, and resistant to the inflammatory process (66). The content of positive ions in the chitosan functional group (NH^{3+}) also accelerates blood clotting due to the ionic bonding with the negative ions charge on platelets and red blood cells (7,66).

Anti-inflammatory

The anti-inflammatory effect of chitosan has long been studied in various studies, both in vitro and in vivo (67,68). Chitosan has been shown to successfully reduce the inflammatory effect and the number of inflammatory cytokines released at the onset of inflammation. Chitosan is also able to reduce the release rate of lipopolysaccharide-induced nitric oxide in the inflammatory response due to cell damage, and even to prevent sepsis induced by lipopolysaccharide. Chitosan administration shows that not only it can prevent organ dysfunction, it is also able to increase survival rate in sepsis cases with increased lipopolysaccharide (68).

Chitosan as dural substitute

Chitosan has been widely researched for biomaterial applications especially a dural substitute, both as a main and supporting material. As a dural substitute, chitosan can maintain cell viability, also prevents the formation of adhesions and infection caused by microbes both early and late post-surgery (69). Unlike the classic duraplasty material that uses fascia, chitosan can ensure subdural spatial density even without suturing and it is more effective in closing dural defects. Chitosan in the form of a membrane is quite elastic and can be simulated with various complex surfaces (69). An experimental evaluation reported that bilayer scaffold from chitosan as a dural substitute provides watertight, suturable, and supports fibroblast infiltration, thereby accelerates tissue healing of the dural defect (63).

As a supportive material of dural substitute, chitosan has also been tested several times as a coating for several scaffold materials (70,71). Its advantages are non-toxic, nonimmunogenic, antimicrobial, mucoadhesive, and hemocompatible, thus it is safe and can be used as dural substitute coating or directly as a replacement dura mater (65,68). The concentration range of chitosan used as a coating material in artificial grafts applied to humans is also quite wide, given the excellent chitosan biocompatibility properties. Some tests showed that chitosan with concentration of 0.1% to 1% is safe and effective to be used as a graft coating material in human, both as a dural substitute or other grafts (70,72).

CONCLUSION

The combination of these two ingredients is still unprecedented. However, with the literature study described above, the application of combination of bovine pericardium-chitosan as a substitute for dural substitute is very promising. Bovine pericardium as dural substitute still facing drawbacks such as inflammatory

and allergic reactions, which tend to be more frequent if compared to other ingredients, but theoretically can be overcome by the anti-inflammatory and antimicrobial properties of chitosan. The hemostatic properties of chitosan can also help accelerate wound healing in the dural defect and are expected to produce better outcomes with the use of this material. Trials of dural substitute synthesis using bovine pericardium in combination with chitosan will enrich the choice of dural substitutes. It is hoped that this material can be an ideal "approach" in the process of searching for an ideal dural substitute that is still ongoing today.

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