The amniotic membrane is a layer that directly covers the amniotic cavity derived from trophoblast cells with various types of pluripotent cells so that it has clinical potential. This membrane is essential for intrauterine fetal growth and development. The amniotic membrane has been widely studied to have various functions, one of which is in the process of wound healing, both acute and chronic. One type of acute wound that begins to use the amniotic membrane as a dressing material is a cesarean section wound. In cesarean section, there is an acute wound due to the incision of the abdominal wall and uterus. The process of wound healing, especially in the epithelial part, is the same as other acute wound healing, which consists of the process of migration, proliferation and cross-linking between cells in the epithelium. In general, the amniotic membrane has clinical potential, especially in wound healing. This relates to the various substrates and the molecules contained therein. In some clinical studies also found a positive effect on wound healing. However, specifically discussing its clinical effectiveness in caesarean section wounds is still limited. Therefore, in this literature, we will discuss the use of the amniotic membranes in wound healing for cesarean sections.

Keywords: Amniotic membrane, cesarean section, wound healing.
the amniotic membrane has the function of accelerating epithelialization, its use as a dressing material for surgical wounds in general and cesarean section wounds in particular has the potential to accelerate healing. Several studies have confirmed the clinical effectiveness of using the amniotic membrane [3], [4].

Especially in cesarean section, several studies have shown the effectiveness of the use of the amniotic membrane. The use of these substances has a positive effect quantitatively based on redness, edema, ecchymosis, discharge, approximation (REEDA) on cesarean section surgical wounds. The quantity of pain is also reduced by using the membranes as a dressing. Likewise with healing time, incidence of infection, allergies, and the maintenance cost are better compared to ordinary dressing [5].

In general, the amniotic membrane has clinical potential, especially in wound healing. This relates to the various substrates and the molecules contained therein. In some clinical studies also found a positive effect on wound healing. However, specifically discussing its clinical effectiveness in cesarean section wounds is still limited. Therefore, in this literature, we will discuss the use of the amniotic membranes in wound healing for cesarean sections.

II. AMNIOTIC MEMBRANE

The amniotic membrane is a layer that directly covers the amniotic cavity. These membranes are derived from trophoblast cells which contain many types of pluripotent cells so that they have clinical potential. This membrane consists of three layers, namely the epithelium, the basement membrane and the outermost layer which is in direct contact with the chorion. Each layer has a different structure and function [6].

The amniotic membrane has a translucent structure without nerve, muscle or lymphatic tissue. This tissue serves as a source of nutrients, oxygen, for the fetus through the process of diffusion. Energy is mainly obtained anaerobically because oxygen supply is limited. Glucose transporter proteins (GLUT) 1 and 3 are located on the apical surface of amniotic membrane epithelial cells. The thickness of the amniotic membrane ranges between 0.02 and 0.5 mm. The amniotic membrane is made up of three layers: an epithelial layer, a thick foundation membrane layer, and avascular mesenchymal tissue. The innermost layer, just adjacent to the amniotic fluid, is made up of a single layer of cuboidal epithelial cells that are firmly attached to the basement membrane. These epithelial cells adhere directly to the basement membrane's acellular layer, which is composed of collagen types I, II, and V. Microvilli on the apical surfaces of amniotic epithelial cells may have intra and transcellular secreatory and transport functions [1], [7].

While the outermost layer consists of mesenchymal cells such as fibroblasts which may originate from embryonic mesoderm. This layer contains a lot of collagen, which helps the amniotic membrane to be stronger. This layer is also known as the spongy zone because it includes a lot of proteoglycans and glycoproteins that give histological preparations a spongy look. This layer lies next to the chorion, which has a nearly acellular structure and a non-fibrillar meshwork made of type III collagen [8]. However, the composition and structural properties of the amniotic membrane change throughout the gestation period.

The membranes are a rich source of progenitor, fetal, multipotent, and even pluripotent stem cells in general. The amniotic membrane's epithelial lining contains epithelial stem cells that help with reepithelialization and wound healing. The presence of collagen IV, V, and VII, fibronectin, proteoglycans, glycosaminoglycans, laminin, and fibrinectins in the amniotic membrane aids in membrane strengthening, mechanical support, and cell migration in the wound region. Because of the low expression of HLA A, B, C, and DR and the greater suppression of neovascularization, the chance of a host versus graft reaction happening in membrane therapy is likewise low. The presence of matrix metalloproteinases and their inhibitors, as well as many growth factors, aids in the maintenance of appropriate growth equilibrium [9].

The contents of the amniotic membrane have various functions. Collagen acts as a resistance factor against various types of proteolytic factors. Elastin functions to maintain membrane elasticity, as well as transport fluids and solutes and produce bioactive peptides, growth factors and cytokines. This membrane also contains various types of growth factors that facilitate epithelial cell migration, adhesion to bulk membranes, epithelial differentiation, prevent cell apoptosis, and promote wound healing. Progenitor cells, endothelin-1 and parathyroid hormone-related proteins were also found. In addition, brain natreuric peptide and corticotrophine releasing hormone were also obtained which function to increase cell proliferation and calcium metabolism [10].

Fig. 1 shows wound healing with the amniotic membrane with polyurethane (PU) giving better results compared to controls using only PU or PU foil. In the picture, it can be seen that the scar formed is smaller in treatment with the amniotic membrane [7].

III. WOUND HEALING IN CESAREAN SECTION

Wound healing in the skin occurs through a process of migration and proliferation. In acute wounds, there is an inflammatory response of neutrophils, monocytes, and macrophages. Then there is activation of keratinocytes caused by the expression of several cytokines and growth factors. The activation is characterized by changes in the

Fig. 1. Wound condition treated using membranes + PU (a-c), only using PU (d-f), and PU foil (g-i) on day -1, 12 and 75.
cytoskeletal network and receptors on the cell surface that are important in the reepithelialization process such as keratin (K) 6 and K16 [11].

Migration of keratinocytes to close the skin defect must first result in loosening of the adhesions between the keratinocytes, as well as the keratinocytes with the basal lamina. This is accomplished by releasing links between cells via desmosomes or hemidesmosomes. Keratinocytes can migrate from the wound edge to the center in this condition, as illustrated in Fig. 2. KGF (keratinocyte growth factor), PDGF-bb (platelet-derived growth factor bb), VEGF (vascular endothelial growth factor), GM-CSF (granulocyte-macrophage colony-stimulating factor), TGF (transforming growth factor)-beta, and IL-8 (interleukin) all play a role in this process [12].

![Image](https://via.placeholder.com/150)

Fig. 2. Keratinocyte migration.

Cell proliferation is heavily influenced by growth factors, cell differentiation, and cell attachment. Only the basal cells have the potential to multiply between the layers of the skin. Several growth factors, including HB-EGF, EGF, TGF-alfa, and KGF, play a part in this proliferation. Keratinocyte cells communicate with other keratinocytes, fibroblasts, endothelium, immune cells, and several other cell types via autocrine and paracrine pathways during proliferation and migration [13].

IV. PREPARATION AND STORAGE OF AMNIOTIC MEMBRANE

Human amniotic membranes are created from a placenta retrieved after a planned cesarean operation following an uneventful pregnancy. The membranes were rejected based on signs of infection in babies, labor before 34 weeks of gestation, and rupture of membranes more than 12 hours before birth. The donors provided written permission for the donation and use of the membranes. One placenta can yield four to five pieces of membrane tissue with a diameter of 5 cm [14].

The placenta was cleansed with physiological saline and placed in the collection vessel with the antimicrobial solution until preparation within 2 hours following cesarean delivery at the time of collection. The membrane structure is completely submerged in a sterile packing container. In a container, the amniotic membrane is combined with antibiotics and antifungals. The membranes are then sliced into various sizes, and membranous membrane tissue pieces are produced. For cryopreservation of the amniotic membranes, a cryoprotective agent was applied and the membranes were preserved at -80 °C in separate containers. Two vials of aerobic and anaerobic organisms were inoculated with placental rinse fluid (8-10 mL) for bacteriological testing [15].

In addition, the placenta is processed for pathological examination. Test tubes containing maternal blood were obtained on the day of the cesarean section at the moment of amniotic membrane removal for the following serological tests: HIV-1 and -2, Ag p24, HCV, HTLV; syphilis: VDRL-TPHA; and HBV: HBs antigen-HBc antibodies. After extensive serological testing, the membranes were finally validated by re-testing the donor lady after 120 days. The membranes can be carried to the hospital and kept on dry ice for up to 24 hours before use, or in normal saline at room temperature for up to 2 hours after thawing [15].

Unused membranes can be stored at temperatures above and below 0 degrees Celsius in a variety of medium compositions. Cell viability in the amnion decreased during storage as expected; in fact, the effect was more pronounced when the amnion was stored frozen; however, the higher viability of the amnion obtained by storage above 0 degrees C with the medium was associated with a short storage time limitation of only 28 days [16].

![Image](https://via.placeholder.com/150)

Fig. 3. Preparation of amniotic membrane.

V. UTILIZATION OF AMNIOTIC MEMBRANE FOR SURGICAL WOUND HEALING

Amniotic membranes have been used in many types of surgery. Several types of surgery have used this preparation, such as reconstruction of the mouth, tongue, nasal mucosa, larynx, tympanic membrane, vestibular, bladder, urethra, vagina, and tendons. Peritoneal substitution in pelvic reconstructive surgery, prevention of adhesions in abdominal and pelvic surgery and use in omphalocle reconstruction. In addition, in the field of ophthalmology, these membranes are also often used in the reconstruction of ocular organs [6].

Animal studies showed that there was no significant difference between the group that was given amniotic membranes and the group that was given polyurethane (PU). Bleeding, inflammation, and infection were also no difference between the two groups. However, quantitatively, it was found that 87.5% of wounds treated with amniotic membranes resembled normal skin on day 60, whereas in the
PU group only 50% resembled normal skin. On the seventh day, the skin epithelium treated with amniotic membranes was thicker than the PU group (p<0.001), but after 60 days became thinner (p<0.0001). Clinically, the effectiveness of the use of the amniotic membrane is shown in Figure 2. below. Group A was treated with membranes, group B was treated with PU while group C was treated with PU + praffin similar to skin grafts. The figure shows lower exudation in the membrane-treated group (p<0.001), groups A and B requiring less wound care than group C, the lowest pruritus rate (p<0.05) and more comfort (p<0.05) compared to the other two groups [7].

![Fig. 4. Amniotic membrane.](image)

![Fig. 5. Comparison of the effectiveness of using A (amniotic membrane), PU (B), and PU + praffin (C).](image)

![Fig. 6. Relative risk of amniotic membrane usage as surgical wound healing.](image)

The epithelium of the wound treated with the amniotic membrane was thinner after 60 days of observation, indicating a possible reduced risk of cicatrization. This reduction in cicatrization risk is related to the anti-inflammatory activity of the amniotic membrane and inhibition of fibrosis [17]. This is supported by the skin color obtained in the group treated with the membranes that resembled normal skin more [7]. The effectiveness of the amniotic membrane in wound healing was also demonstrated by a meta-analysis on the treatment of diabetic feet. In this meta-analysis of 311 patients, the relative risk of wound healing with the membranes was 2.75 times (95% CI 2.06-3.67, p<0.001) [8].

The use of the amniotic membrane in wound healing is still a challenge. The pluripotent cell subpopulation is not homogeneous. Differences in culture media used in several studies have the potential to cause the growth of different cell populations so that there is a risk of bias [18].

Because the amniotic membrane provides a natural scaffold, or the supporting matrix on which cells and tissues develop, it is regarded as an important component of tissue healing with a variety of clinical uses. Furthermore, as previously noted, the amniotic membrane contains various biological features that are necessary for tissue regeneration, including as anti-inflammatory, antibacterial, anti-fibrotic, anti-scarring, and low immunogenicity. As a result, the amniotic membrane may be regarded as a bio-therapeutic product composed of a single layer of epithelial cells positioned in the basement membrane and a nonvascular collagenous stroma. These three components provide the membrane with therapeutic features such as antiadhesive, bacteriostatic, wound protection, pain relief, and epithelializing actions. Another study found that the duration between tissue creation and strengthening ranged from 14 to 60 days, with an average of 33.314 days. This research included 14 foot ulcers with healing rates ranging from 0.064 to 2.22 with an average of 0.8960.646 cm2/day with a 100% reduction in lesion size [19].

VI. MECHANISM OF AMNIOTIC MEMBRANE IN INCREASING THE REEPITHELIZATION OF SURGICAL WOUND SKIN

The amniotic membrane is the deepest layer of the placenta and is readily separated from the chorion. The method by which the membranes stimulate reepithelialization remains unknown. This membrane may promote keratinocyte migration from the wound tip and encourage differentiation, allowing the epithelium to remain intact [6]. Other investigations have discovered a stimulatory impact on wound epithelialization assisted by growth factors and progenitor cells produced by the amniotic membrane [14]. Furthermore, this membrane protects the integrity of the basement membrane and stromal matrix, which promotes healing potential and is critical in initiating re-epithelialization.

When the membranes are applied to keratinocytes, they phosphorylate ERK1/2, JNK1/2, and Sn p58. A similar reaction was observed in another investigation employing amniotic membrane media. It also stimulates the expression of c-JUN, an AP1 family member involved in keratinocyte migration and wound healing. JUN N-terminal kinase (JNK) 1 and two other kinases are phosphorylated by membranes. This JNK1 is a c-JUN positive regulator that helps with phosphorylation and stability. The expression of c-JUN was very robust in membrane-treated wounds, especially in the basal epithelium adjacent to the wound edge and in the epithelialized tongue. This shows that c-JUN expression is vital in the wound epithelialization process [6].
These membranes exhibit unique structural, biochemical, and immunological properties. This membrane has a re-epithelializing function and is immunogenic. The amniotic membrane is also anti-inflammatory, antifibrotic, antibacterial, analgesic, and non-tumorigenic. These extensive effects are connected to their capacity to manufacture and release biologically active molecules such as cytokines and signaling factors such as tumor necrosis factor- (TNF-), transforming growth factor (TGF)-, TGF, basic fibroblast growth factor (TGF), b FGF), epidermal growth factor (EGF), keratinocyte growth factor (KGF), hepatic growth factor (HGF), interleukin4 (IL4), IL6, IL8, natural metalloprotease inhibitor, defenidin, and prostaglandins are all examples of growth factors [20].

The amniotic membrane is also a site of proliferation and differentiation. The adoption of such membranes has various benefits over skin transplantation. Its usage does not result in rejection because of its low immunogenicity and lack of uncontrolled expansion [6]. As previously stated, all of these impacts are tied to its ability to release and manufacture biologically active chemicals.

The amniotic membrane may swiftly adhere to the wound region and manage the balance between angiogenesis and mesenchymal stem cell regulation, as well as the manufacture of MMP and its inhibitors such as TIMP. Due to the presence of amniotic membrane epithelial cells, this procedure helps the third stage of wound healing by promoting re-epithelialization of the wound region. Furthermore, it is considered that the amniotic membrane plays a key function in suppressing protease production, PMN filtration, and growth factor release from donor fibroblast cells [4].

Anti-inflammatory properties of amniotic membranes have also been discovered. The amniotic membrane traps inflammatory cells and drives them to apoptosis via pro-apoptotic agents; its epithelial cells produce anti-inflammatory factors; pro-inflammatory cytokines such as interleukin 1 alpha and 1 beta are suppressed; MMP inhibitors are produced; migration inhibition factor (MIF) is expressed; anti-inflammatory cytokines such as IL-1 receptor antagonists are expressed; and anti-inflammatory factors such as PGE2, TGF This anti-inflammatory action is critical in the wound healing process [15].

Surgical wound healing is determined by the degree of inflammation and the anti-infective properties of the amniotic membrane have been reported. The anti-infective is thought to be due to the synthesis of anti-inflammatory and decreased expression of TGF beta and pro-inflammatory cytokines. Cells in the amniotic membrane synthesize innate immune system peptides such as defending beta, elastase inhibitors, elfn, lactoferrin and IL-1 RA. All of these factors may have an antimicrobial effect. In Loffelbein's study getting wound care with amniotic membrane therapy showed almost no infection [7]. One model describes the pathway of the amniotic membrane in wound healing as shown in Fig. 7.

VII. THE BENEFIT OF AMNIOTIC MEMBRANE IN SURGICAL WOUND HEALING AFTER CESAREAN SECTION

The use of amniotic membranes in cesarean section has not been widely studied. A randomized clinical trial study with 45 interventions with amniotic membrane dressings and 45 controls showed that wound healing scores consisting of redness, edema, ecchymosis, discharge, approximation (REEDA) at 24 hours after surgery (p=0.003) were better in the intervention group, although on day 8 there was no difference [9].

The use of amniotic membranes also reduces discomfort during cesarean section surgery. The amount of discomfort was not substantially different between the amniotic membrane dressing group and the control group after 4 and 12 hours postoperatively, according to Mohseni's study. It was, however, significant 24 hours (p=0.026), 36 hours (p=0.026), and 48 hours (p=0.004) after surgery. The membrane dressing group required fewer analgesics than the control group (p=0.041) [10].

Another trial with 72 patients found that the group that used the amniotic membrane dressing healed faster. Healing time was 2.92 1.02 days in the amniotic membrane dressing group and 5.51 3.81 days in the control group (p = 0.000). Local infection was considerably lower on day 3 (p=0.000) in the group with amniotic membrane dressing, and allergic responses were similarly reduced on day 3 (p=0.023) and day 5 (p=0.003). The cost of maintenance every day is likewise reduced [11].

The amniotic membrane is an autologous form that comes from the patient's body and is free. This membrane plays a role in reducing infection in the surgical wound area and reducing pain and accelerating healing. Meta-analysis and systematic review showed that the membranes improved the surgical wound outcome with a relative risk (RR) of 2.057-3.665 (p<0.001) within 6 weeks. The acceleration of wound healing is thought to be most closely related to angiogenesis. The amniotic membrane contains a lot of pro-angiogenic substances so that it accelerates wound healing. The mechanism for accelerating surgical wound healing using amniotic membrane dressings is as shown in Figure 7 above [20].

Another study discovered that the amniotic membranes can also help with wound healing following a cesarean surgery. This research was a randomized, double-blind clinical experiment. Women who had cesarean section took part in this study. Patients were split into two groups at random (N =
45 in each). Cesarean wounds were dressed using amniotic membranes in one group, whereas a simple dressing was used in the other. The Redness, Edema, Ecchymosis, Discharge, Approximation (REEDA) scale was used to measure wound healing 24 hours and 8 days following the cesarean surgery. According to the findings, the amniotic membrane group had a lower mean REEDA score, which implies significantly different wound healing across the groups 24 hours after cesarean surgery. However, there was no significant difference between the groups on day 8 following cesarean section (P = 0.078). The outcomes of this study suggest that using amniotic membrane dressings can help with wound healing during the early phases after cesarean section [9].

VIII. CONCLUSION

The amniotic membrane is a layer that covers the amniotic cavity. The tissue contains many types of pluripotent cells so that it has clinical potential. In addition, this tissue also contains or can secrete various factors that play a role in growth that can accelerate the epithelialization process. Clinically several studies have shown its effectiveness in healing surgical wounds. In addition, the amniotic membrane also has the potential to reduce abnormal healing such as the formation of scar tissue. Specifically for cesarean section wounds, data on its effectiveness are still limited. However, several studies have shown that the amniotic membrane has a positive effect on healing, the incidence of infection, allergies and the cost of wound care.

CONFLICT OF INTEREST

Authors declare that they do not have any conflict of interest.

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