REVIEW ARTICLE

Processing and Use of Radiation Sterilized Human Amniotic Membrane Allografts as Biological Dressings of Wounds in Children: A Review

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Abstract

Radiation sterilized human amniotic membrane allografts are used as biological dressing of wounds in rehabilitative surgery. The amniotic membrane is used in many different clinical situations such as: heat burn, chemical burns, diabetic wound/ diabetic foot ulcer, leprotic ulcer, abdominal wall reconstruction, pterigium removal site, peripheral corneal ulcer, in the management of pressure sore, etc.Non-viable lyophilized/oven-dried radiation sterilized amniotic membrane allografts could be processed for utilization as temporary biological dressing of wounds. The allografts used clinically should not be the carriers of germs or a source of infection. Human chorio-amniotic membranes collected for processing as tissue allografts to be used as biological dressings were reported to be contaminated with microorganisms such as species of Staphylococcus, Micrococcus, Bacillus and Pseudomonas.However, oven dried (40ºC) or freeze dried (-50ºC) human amniotic membranes were found to be sterilized by irradiation with the dose of 25 kiloGray (kGy) of gamma radiation.Careful screening and selection of tissue donors, proper processing and gamma radiation sterilization of human amniotic membranes minimize the risk of disease transmission to recipients through allografts.

***Keywords**: Human amniotic membrane, gamma radiation, sterilization.*

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Introduction

Davis et al[1] in 1910 and Sabella et al[2] in 1913 reported the clinical use of foetal membranes for transplantation. The application of human amniotic membranes has been increased, in time, to many areas of surgical treatments.[3] The amniotic membranes have been used in burns,[4,5] in the treatment of venous ulcers[6], as a physiological wound dressing,[7,8] for leg ulcers,[9-11] otolaryngologic, head and neck surgery,[12,13] pelvic surgery,[14,15] etc. Human amniotic membrane grafts are used preferably as temporary biological dressings because of its unique characteristics and properties. No immunogenic rejection phenomenon does occur after the application of amniotic membrane grafts on the wound.[16,17] Healing effect of the foetal membranes when used for treating different surgical and skin defects are thought to be either due to their content

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of some antibacterial, angiogenic or other biochemical factors, or to the biomechanical characteristics of the foetal membranes as such.[18]

The amniotic membraneallografts prevent local inflammatory response[19] and its antimicrobial properties reduce risk of post-operative infection.[20] The transplanted membrane may perform as a structural barrier to impede proliferation of fibrous tissue and thus decrease scarring.[21,22]

Microbial contamination of human tissue allografts

The risk of infectious disease transmission with human tissue allografts is a major concern in tissue banking practice. Microorganisms can be introduced into the grafts during tissue collection, processing and storage, but even if all these procedures are done under aseptic conditions, the possibility of bacterial and viral disease transmission of donor origin cannot be excluded.[23] In order to minimize the risk of disease transmission through allografts, several steps should be undertaken by tissue banks, including careful donor-screening, proper tissue processing and sterilization of tissue allografts.[23]

Islam et al[24] and Begum et al[25] reported bacterial isolates obtained from human amniotic membranes and one of the bacterial isolates was reported to be highly radio-resistant. Chakraborty et al (2012)[26] and Chakraborty et al (2015)[27] reported association of microorganisms with human chorio-amniotic membranes. Chakraborty et al[26] isolated bacterial contaminants from all the studied 34 post-delivery chorio-amniotic membranes collected from labor rooms of hospitals and the contaminants were identified as *Staphylococcus aureus*, *S. cohnii*, *S. epidermidis*, *S. sciuri*, *Micrococcusroseus*, *Bacillus* sp., *B. megaterium*, *Pseudomonascepacia* and *P. diminuta*.

Sterilization of tissue allografts with ionizing radiation

Radiation sterilization of medical materials on a commercial scale was applied for the first time in the middle of the 1950s with less than 10% of medical products sterilized by ionizing radiation in the 1970s, and nearly 50% at the period of report by Dziedzic-Goclawska et al[23] with an increasing tendency. The International Atomic Energy Agency (IAEA) in Vienna promotes and supports the use of ionizing radiation for sterilization purposes. IAEA has elaborated and issued many guidelines and standards applicable for radiation sterilization.[28] The Agency's promotion and support resulted in the development of national programmes on tissue banking and application of ionizing radiation for the sterilization of tissue allografts. Tissue banks were created in many countries of the Asia Pacific region under the auspices of IAEA.[23]

The killing effect of radiation may be due to direct effect or indirect effect. The direct action of radiation involves the interaction between ionizing radiation and critical biological molecules, which results in excitation, lesion and scission of polymeric structure such as DNA.[29] The indirect effect of radiation is due to biochemical changes within the organism. The interaction of ionizing radiation with water molecules within living cells leads to the production of short-lived free radicals and peroxy radicals. These radiolytic products interact with biological molecules including DNA; hence inactivating the reproduction process. This indirect effect of radiation normally occurs as an important part of the total chain of reactions of ionizing radiation.[30,31] The indirect effect is more prominent and very much influenced by environmental factors including oxygen, water content, medium, temperature and chemicals present during irradiation.[32-34] Chakraborty et al[35] reported the influence of irradiation medium and initial viable cell numbers on the gamma radiation sterilization of the bacterial isolates recovered from human amnion membranes and bones. The sterilization efficacy of ionizing radiation lies in its good penetrability inside the matter and its high killing efficiency of microorganisms.[23] Ionizing radiation is effective in the inactivation of microorganisms in the bulk of any material without associated problems of heat exchange, pressure differences or hindrances by diffusion barriers.[36] Chakraborty et al[26] studied the effect of gamma radiation on the sterility quality of tissue allograftsand found that human amniotic membrane allografts irradiated with a dose of 25 kiloGray (kGy) ofgamma radiationwere sterilized.

Transmission of hepatitis C virus (HCV) by non-sterilized cadaveric tissue allografts has been described, while allografts irradiated with a dose of 17 kGy did not evoke infections of the graft recipients.[37] There is no data on the sensitivity of hepatitis B virus (HBV) to ionizing radiation;

however, the virus is known to be resistant to heat sterilization.[23]

Most of the studies on the effectiveness of ionizing radiation to inactivate viruses were done on the inactivation of human immunodeficiency viruses (HIV), particularly in blood plasma. The dose required to eliminate all active HIV-1 from the particular tissues depends on the amount of virus originally present (initial contamination).[23] It has been postulated that the dose of irradiation needed to reduce the viral load by 1 $\log_{10}$ is 4 kGy[38] or even 5.6 kGy[39]. Fideler et al[40], using polymerase chain reaction (PCR), found that in order to inactivate HIV in fresh frozen bone-patellar ligament allografts, a dose in the range of 30-40 kGy was required. The sensitivity of HIV to ionizing radiation is reported to depend on the temperature during irradiation. The inactivation of the virus titre of 5 to 6 $\log_{10}$ was achieved at doses of 50 to 100 kGy in frozen (-80ºC) plasmas and at the dose of 25 kGy at 15ºC.[41] Dziedzic-Goclawska et al[23] reported that prions, responsible for Creutzfeldt-Jacob disease (CJD) in humans, are extremely resistant to most of the chemical and physical sterilizing/disinfecting agents including ionizing radiations.

Selection of tissue donors

Tissues could be procured from both living and cadaveric donors. Tissues from living donors mainly fall into two categories, namely amniotic membranes and bones obtained after surgical procedures. Tissues from cadaveric donors (*e.g.*, skin, bone, cartilage, tendon etc.) are obtained under sterile or non-sterile conditions.[42] The main goal of the tissue bank is to provide safe and high quality tissue grafts to the patients. Therefore, each potential donor must be evaluated through a careful review of the donor's medical and sexual history, physical examination and laboratory screening. Generally, the ideal donor is a young, healthy adult under 55 years of age.[42] Donor selection is based on a review of social and medical history, physical examination, autopsy results (if performed) and blood tests. Donors are excluded if any elements of past social, sexual and medical history indicate the risk of HIV (human immunodeficiency virus), hepatitis B and C, or jaundice of unknown etiology, infectious diseases including tuberculosis, malignant diseases, presence or suspicion of neurodegenerative symptoms including dementia, the use of pituitary derived hormones, systemic connective tissue diseases (*e.g.*, lupus erythematosus, rheumatoid arthritis), chronic steroid treatment, exposure to toxic substances and exposure to irradiation at the site of tissue donation.[23]

Serological as well as microbiological examinations are needed to fully screen donors. The minimum testing accepted universally includes VDRL (Venereal Disease Research Laboratory test)/RPR (rapid plasma reagin), hepatitis B surface antigen, hepatitis C antibodies, HIV antibody and microbiological (culture) testing of small donor tissue samples or swabs. Other tests can be done selectively when indicated such as: CMV (cytomegalovirus) test, hepatitis B core antibody, histopathological examination, antigen testing for HIV using antibody or polymerase chain reaction (PCR) and HTLV (Human T-lymphotrophic virus) I and II.[42]

Collection and preservation of tissues

Tissues are collected aseptically in airtight sterile containers and transported to the tissue processing laboratory in an insulated container that can keep the tissues at a temperature of 4ºC for at least 12 hours (h). Normally ice slabs or dry ice are placed in the container to keep the temperature low. Chorio-amniotic membranes collected after delivery should be kept completely immersed in sterile normal saline in a plastic container and stored in a refrigerator (4ºC) for not more than 24 h.[42]

Processing of radiation sterilized human amniotic membranes for use in rehabilitative surgery

The procedure of processing of radiation sterilized human amnion membrane allografts for use in rehabilitative surgery is described elsewhere.[26,43] Post-delivery human chorio-amniotic membranes from clinically suitable donors are collected aseptically in sterile normal saline and temporarily preserved in the freezer. The frozen tissues are first thawed and then the amnion is separated aseptically from the chorion using sterile surgical instruments. The amniotic membranes are washed several times with sterile normal saline using an electric orbital platform shaker to remove blood and tissue debris. The membranes are then aseptically cut into pieces, stretched on surgical gauge and mounted on surface sterilized (70% ethanol) plastic frames and dried overnight at 40ºC temperature in an electric controlled oven. The dried membranes are then cut under a laminar air flow into pieces and each piece

of amniotic membrane is then packaged in triple layer with polyethylene packages, labeled, vacuum sealed, and then irradiated with gamma radiation at the dose of 25 kiloGray (kGy). The irradiated membranes are preserved at refrigerating temperature. The sterility of the irradiated membranes is determined by sterility testing using microbiological culture media, and then the sterile membranes are supplied to hospitals/clinics for use in rehabilitative surgery.[26,43]

Effect of processing and radiation sterilization on the biomechanical properties of tissue allografts

Chakraborty et al (2007)[44] studied the effect of oven drying (40ºC), freeze drying (-50 ºC) and gamma irradiation (25kGy) on the tensile strength of human amnion membranes. When the tensile strength of the amnion membranes was measured without rehydration of dried samples, it was found to be increased at different stages of processing including oven dried (7.93% increase), freeze dried (6.38% increase), oven dried-gamma irradiated (7.57% increase) and freeze dried-gamma irradiated (6.97% increase) as compared to that of the controls. Rehydration of dried and irradiated membranes resulted in a decrease in tensile strength. After rehydration in normal saline for 5 minutes, the retention of tensile strength by oven dried-irradiated and freeze dried-irradiated amnion membranes was respectively about 70% and 61% of that of controls.

Clinical uses and effectiveness of radiation sterilized amniotic membranes

Human amniotic membrane is used in many different clinical situations such as: superficial burns,[17, 45] mixed burns,[46] leprotic ulcer,[47,48] chemical and thermal burns,[49] deep corneal ulcers,[50] in the management of pressure sore[51] etc. It is reported to reduce pain, decrease electrolyte, fluid and protein loss, and promote epithelialization.[9] It is also reported to prevent local inflammatory response and decrease corneal haze.[19] Bari et al[52] reported a prospective study of 40 patients with deep and deep dermal burns in which radiation sterilized (25 kGy) human amniotic membranes were used. Nessa et al[43] reported the use of oven dried (40ºC) and gamma radiation sterilized (25kGy) human amniotic membranes in the treatment of 168 patients with heat burn, acid burn, diabetic wound/diabetic foot ulcer, leprotic ulcer, abdominal wall reconstruction, chemical (acid) burn (corneal surface Rt), chemical (lime) burn, pterigium removal site, peripheral corneal ulcer, and orbit reconstruction in hospitals/ clinics in 2008 in Bangladesh. For utilization as temporary biological dressing of wounds, non-viable lyophilized/oven-dried radiation sterilized amniotic membrane grafts could be processed.[17]

Use of amniotic membrane allografts in the treatment of burns in children

Khan et al[17] reported treatments of children with superficial burns using radiation sterilized amniotic membranes as biological dressing. The authors mention that the amnion grafts were very easily applied to the wounds and the patients did not feel pain during repeated dressings. No side effect was also noticed after the application of grafts and all the patients healed satisfactorily without any post operative complication. Chakraborty et al[46] reported management of mixed (superficial and deep) burn in children with allogeneic amnion and autogeneic skin grafting. Shahid et al[53] reported the use of radiation sterilized amniotic membrane as local burn wound coverage and demonstrate that amniotic membrane is a suitable and effective biological dressing in reducing patient's morbidity when compared with non-biological medicated tulle (sofratulle). Ullah et al[54] reported the use of radiation sterilized amniotic membranes in the treatment ofa total of 370 patients (mean age 2.76 years) with superficial partial thickness burn. The authors[54] mention that amnion dressing suppresses bacteria in the wound as well as reduces infection. Amnions have good adherent characteristics, which reduces infection as well as reduction of oozing of plasma from the wound.[54]

Burn injuries are one of the most devastating physical and psychological injuries that a person can suffer.[55] From 1981 to 1990 it was the second most common surgical problem in paediatric age group in the department of pediatric surgery, Dhaka Shishu Hospital.[56] Ullah et al[54] reported the use of radiation sterilized amniotic membranes in the treatment of a total of 370 patients with partial thickness burn. The authors mention that amnion dressing suppresses bacteria in the wound as well as reduces infection. Amnions have good adherent characteristics, which reduces infection and also reduce oozing of plasma from the wound. It has a role on burnt pain reduction, frequency of dressing change, rate of healing, cost, and duration of hospital

stay. Ullah et al[54] conclude that amniotic membrane is one of the effective biological skin substitutes used in burn wounds, with efficacy of low bacterial counts, has advantages of reducing loss of protein, electrolytes and fluids, decreasing the risk of infection, minimizing pain, accelerated wound healing and good handling properties. It is readily available and does not present immunological problem and allergy response. It is cost effective and very helpful for developing countries.

Conclusion

The facility of preservation of oven dried and radiation sterilized human amniotic membranes in a refrigerator, makes it possible to build up a large stock of allografts for use as biological dressing of wounds. Processing and preservation of radiation sterilized tissue allografts in a tissue bank can ensure ready availability of tissue allografts for safe clinical use.

References

1. Davis JS. Skin transplantation with a review of 550 cases at the John Hopkins Hospital. *JHH Report* 1910;**15**:307 (cited from Farazdaghi M, Adler J, Farazdaghi SM. Electron microscopy of human amniotic membrane. In: Phillips GO, Nather A, Strong DM, von Versen R, editors. Advances in Tissue Banking vol. 5, World Scientific Publishing Co Pte Ltd, Singapore, 2001;P.149-71).
2. Sabella N. Use of foetal membranes in skin grafting. *Med Rec NY* 1913;**83**:478 (cited from Farazdaghi M, Adler J, Farazdaghi SM. Electron microscopy of human amniotic membrane. In: Phillips GO, Nather A, Strong DM, von Versen R, editors. Advances in Tissue Banking vol. 5, World Scientific Publishing Co Pte Ltd, Singapore, 2001;P.149-71).
3. Farazdaghi M, Adler J, Farazdaghi SM. Electron microscopy of human amniotic membrane. In: Phillips GO, Nather A, Strong DM, von Versen R, editors. Advances in Tissue Bankingvol.5, World Scientific Publishing Co Pte Ltd, Singapore, 2001;P.149-71.
4. Kirschbaum SM, Hernandez H. Use of amnion in extensive burns. Proc. Third Int. Cong. Plastic Surg. Excerpta Medica, Amsterdam 1963 (cited from Farazdaghi M, Adler J, Farazdaghi SM. Electron microscopy of human amniotic membrane. In: Phillips GO, Nather A, Strong DM, von Versen R, editors. Advances in Tissue Banking vol. 5, World Scientific Publishing Co Pte Ltd, Singapore, 2001;P.149-71).
5. Quinby WC, Jr., Hoover HC, Scheflan M, Philmon TW, Sumner AS, Conrado CB. Clinical trials of amniotic membranes in burn wound care. *Plastic Reconstructive Surg* 1982;**70**:711-16.
6. Ward DJ, Bennett JP, Burgos H, Fabre J. The healing of chronic venous leg ulcers with prepared human amnion. *Br J Plastic Surg* 1989;**42**:463-67.
7. Colocho G, Graham WP, Green AE, Matheson DW, Lynch D. Human amniotic membrane as a physiologic wound dressing. *Arch Surg* 1974;**109**:370-73.
8. Gruss JS, Jirsch DW. Human amniotic membrane a versatile dressing. *J Can Med Assoc* 1978;**118**:1237-46.
9. Trelford JD, Trelford-Sauder M. The amnion in surgery, past and present. *Am J Obstet Gynecol* 1979;**134**:833-45.
10. Faulk WP, Matthews RN, Stevens PJ, Bennett JP, Burgo H, His BL. Human amnion as an adjunct in wound healing, *Lancet* 1980;1158 (cited from Farazdaghi M, Adler J,Farazdaghi SM. Electron microscopy of human amniotic membrane. In: Phillips GO, Nather A, Strong DM, von Versen R, editors.Advances in Tissue Bankingvol. 5. World Scientific Publishing Co Pte Ltd, Singapore 2001;P.149-71).
11. Ward DJ, Bennett JP. The long term results of the use of human amnion in the treatment of leg ulcers, *Br. J. Plastic Surg*. 1984;**37**:191-93.
12. Zohar Y, Talmi YP, Finkelstein Y, Shvili Y, Sadov R, Laurian N. Use of human amniotic membrane in otolaryngologic practice *Laryngoscope* 1987;**97**:978-80.
13. Talmi YP, Finkelstein Y, Zohar Y. Use of human amniotic membrane as a biologic dressing. *Euro J Plastic Surg* 1990;**13**:160-62.
14. Rennekamff HO, Dohrman P, Fory R, Fandrich F. Valuation of amniotic membrane as adhesion prophylaxis in a novel surgical gastrochisis model. *Invest Surg* 1994;**7**:187-93.
15. Gharib M, Ure BM, Klose M. Use of amniotic grafts in the repair of gastrochisis. *Pediatr Surg* 1996;**11**:96-99.
16. Akle CA, Adinolfi M, Welsh KI, Leibowitz S, McColl I. Immunogenicity of human amniotic epithelial cells after transplantation into volunteers. *Lancet* 1981;**2**:1003-05.
17. Khan AR, Aziz MA, Ullah MS, Nessa A, Chakraborty SK, Miah MM. Radiation sterilized amnion membrane grafts in the management of superficial burn in children. *Dhaka Shishu (children) Hospital Journal* 2002;**18**;:52-54.

18. Panakova E, Koller J. Utilization of foetal membranes in the treatment of burns and other skin defects. In: Phillips GO, Strong DM, von Versen R, Nather A, editors. Advances in Tissue Banking vol. 1, World Scientific Publishing Co Pte Ltd, Singapore 1997; P. 165-81.

19. Choi YS, Kim JY, Wee WR, Lee JH. Effect of application of human amniotic membrane on rabbit corneal wound healing after excimer laser photorefractive keratectomy. *Cornea* 1998;**7**: 389-95.

20. Talmi YP, Sigler L, Inge E. Antibacterial properties of human amniotic membranes. *Placenta* 1991;**12**:285-88.

21. Shimazaki J, Shinozaki N,Tsubota K. Transplantation of amniotic membrane and limbal autograft for patients with recurrent pterigium associated with symblepharon. *Br J Ophthalmol* 1998;**82**:235-40.

22. Tseng SCG, Li DQ, Ma X. Suppression of transforming growth factor beta isoform, TGF-â receptor type II and myofibroblast differentiation in cultured human corneal and limbal fibroblasts by amniotic membrane matrix. *J Cell Physiol* 1999;**179**:325-35.

23. Dziedzic-Goclawska A, Stachowicz W. Sterilization of tissue allografts. In:Phillips GO, Strong DM, von Versen R, Nather A, editors. Advances in Tissue Banking vol. 1, World Scientific Publishing Co Pte Ltd, Singapore 1997;P.261-21.

24. Islam MS, Begum R. Microbiological and radiation sensitivity studies of bacterial isolates obtained from human amniotic membranes. In: Phillips GO, Kearney JN, Strong DM, von Versen R, editors. Advances in Tissue Banking vol. 3, World Scientific Co Pte Ltd, Singapore, 1999;P.457-62.

25. Islam MS, Begum R. A highly radio-resistant bacterial isolate from human amnion membranes, In: Phillips GO, Kearney JN, Strong DM, von Versen R, editors.Advances in Tissue Banking vol. 3, World Scientific Co Pte Ltd, Singapore, 1999;P.463-67.

26. Chakraborty SK. Microorganisms associated with donor tissues and the impact of physico-chemical treatments on the clinical applicability of tissue allografts. PhD Thesis, Department of Microbiology, Faculty of Biological Sciences, University of Dhaka, Dhaka 1000, Bangladesh, 2012.

27. Chakraborty SK, Rashid H, Gomes DJ. Status of contamination, and isolation and identification of microorganisms from some human donor chorio-amniotic membranes and femoral head bones. Internal Report, *SIU-**3***, March 2015, Chief Editor: Dr. Md. Sirajul Islam, Director, SIU, Published by Scientific Information Unit, Atomic Energy Research Establishment, Ganakbari, Savar, Dhaka.

28. Mukherjee RN. IAEA standards applicable for sterilization. In: Morrisey RF, Prokopenko YL, editors. Sterilization of Medical Products Johnson and Johnson, Polyscience Publication Inc., Morin Heights, Canada 1991;P.117-24.

29. Yusof N. Effect of radiation on microorganisms-Mechanism of radiation sterilization. In: Phillips GO, Nather A, Strong DM, von Versen R, editors. Advances in Tissue Banking vol.5, World Scientific Publishing Co Pte Ltd, Singapore, 2001;P.342-57.

30. Antoni F. The effect of ionizing radiation on some molecules of biological importance. In: Manual on Radiation Sterilization of Medical and Biological Materials. *IAEA Technical Reports Series* 1973;**149**:13-36.

31. Coggle JE, Biological effects of radiation, Wykeham Science Series, London 1973; p. 150 (cited from Yusof N. Effect of radiation on microorganisms-Mechanism of radiation sterilization. In: Phillips GO, Nather A, Strong DM, von Versen R, editors. Advances in Tissue Banking vol. 5, World Scientific Publishing Co Pte Ltd, Singapore, 2001;P.342-57).

32. Tallentire A. Aspects of microbiological control of radiation sterilization. *J Rad Ster* 1973;**1**:85-103.

33. Gardner JF, Peel MM. Introduction to Sterilization and Disinfection, Churchill Livingstone, Edinburgh, 1986 (cited from Yusof N. Effect of radiation on microorganisms-Mechanism of radiation sterilization. In: Phillips GO, Nather A, Strong DM, von Versen R, editors. Advances in Tissue Banking vol. 5, World Scientific Publishing Co Pte Ltd, Singapore, 2001;P.342-357).

34. Whitby JL. Microbiological aspects relating to the choice of radiation sterilization dose. *Rad Phys Chem* 1993;**42**:577-80.

35. Chakraborty SK, Rashid H, Deen NS, Gomes DJ. Effect of suspending medium and bacterial load on the gamma radiation sterilization of some bacterial isolates from human amnion membrane. *Bangladesh J Medical Science* 2011;**17**:57-62.

36. Christensen EA, Kristensen H, Miller A. Radiation sterilization. In: Russell AD, Hugo WB, Ayliffe GAJ, editors. Principles and Practice of Disinfection, Preservation and Sterilization 2nd edn. Blackwell Science, Oxford, 1992 (cited from Dziedzic-Goclawska A, Stachowicz W. Sterilization of tissue allografts. In: Phillips GO, Strong DM, von Versen R, Nather A, editors. Advances in Tissue Banking vol.1, World

Scientific Publishing Co Pte Ltd, Singapore 1997; P.261-321).

37. Conrad EU, Gretch D, Obermeyer K, Moogk M, Sayers M, Wilson J, Strong DM. The transmission of hepatitis C virus through tissue transplantation. *J Bone & Joint Surg* 1995;**77**:214-24.

38. Conway B, Tomford WW, Mankin H, Hirsch MS, Schooley RT. Radiosensitivity of HIV-1. Potential application to sterilization of bone allografts. *AIDS* 1991;**5**:608-09.

39. Conway B, Tomford WW. Radiosensitivity of human immunodeficiency virus type 1. *Clin Infect Dis* 1992;**14**:978-79.

40. Fideler BM, Vangsness CT, Moore T, Li Z, Rasheed S. Effects of gamma irradiation on the human immunodeficiency virus. *J Bone & Joint Surg* 1994;**76**-A:1032-35.

41. Hiemstra H, Tersmette M, Vos AH, Over J, Van Berkel MP, De Bree H. Inactivation of HIV by gamma radiation and its effect on plasma and coagulation factors. *Transfusion* 1991;**31**:32-39.

42. Multi-Media Distance Learning Package on Tissue Banking, Module 4, Phillips GO. coordinating editor, p. 4-5, 10-11, 16-17, IAEA/NUS Regional Training Centre, IAEA/NUS International Training Centre, National University of Singapore, Singapore.

43. Nessa A, Chakraborty SK, Chakma K, Hasan MZ, Miah MM. Collection, processing and gamma radiation sterilization of human amniotic membranes for use in rehabilitative surgery. Technical Report 2008 (published in 2010), AERE/ TR-14, ISSN 2074-2371, Atomic Energy Research Establishment, Ganakbari, Savar, Dhaka;P.219-22.

44. Chakraborty SK, Nessa A, Ali Y, Alam J, Haque ME, Rashid H, Gomes DJ. The effect of oven drying, freeze drying and gamma irradiation on the tensile strength of human amnion membrane. *Bangladesh J Medical Science* 2007;**13**:144-47.

45. Goebel P, Schubert W. Personal experiences with covering burns in children with amnion. *Brit Orthop Traumatol* 1990;**37**:495-98.

46. Chakraborty SK, Nessa A, Mia MM, Khan AR, Aziz MA, Ullah MS. Management of Mixed (superficial and deep) burn with allogeneic amnion and autogeneic skin grafting. *Bangladesh Journal of Medical Science* 2003;**9**:91-94.

47. Bari MM, Begum R, Chakraborty SK, Islam MS. Tissue Banking: Use of radiation sterilized amniotic membrane grafts as temporary biological dressings for the treatment of leprotic ulcers. *Acta Orthopedica Hellenica* 1995;**46**:88-93.

48. Bari MM, Begum R. Use of radiation-sterilized amniotic membrane grafts as temporary biological dressings for the treatment of leprotic ulcers. In: Phillips GO, Kearney JN, Strong DM, von Versen R, Nather A, editors. Advances in Tissue Banking vol.3, World Scientific Publishing Co Pte Ltd, Singapore, 1999;P.477-83.

49. Shimazaki J, Yang HY, Tsubota K. Amniotic membrane transplantation for ocular surface reconstruction in patients with chemical and thermal burns. *Ophthalmol* 1997;**104**:2068-76.

50. Kruse FE, Rohrschneider K, Volcker HE. Multilayer amniotic membrane transplantation for reconstruction of deep corneal ulcers. *Ophthalmol* 1999;**106**:1504-11.

51. Hoque F, Muldun S, Rahman S, Islam M. An initial study of pressure sore management with amniotic membrane dressing. In: Phillips GO, Kearney JN, Strong DM, von Versen R, Nather A, editors.Advances in Tissue Banking vol. 3. World Scientific Publishing Co Pte Ltd, Singapore, 1999;P.491-93.

52. Bari MS, Choudhury MIM, Khan AR, Nessa A. Role of human foetal membranes (amniotic membrane) in the management of burn wounds. *Annals of Burns and Fire Disaster* 2002;**15**:3-8.

53. Shahid SMA, Khan AR, Rahman MM, Lina KSN. Effectiveness of radiation sterilized amniotic membrane in reducing patient's morbidity in comparison to non biological dressing like medicated Tulle (Sofra Tulle). *IOSR Journal of Dental and Medical Sciences* 2019;**18**:44-50.

54. Ullah MS, Ferdous KMN, Mullick MMH, Alam MR, Rahman MS, Khan AR. Amnion membrane graft dressing in superficial partial thickness burn in pediatric patients. *Chattagram Maa-O-Shishu Hospital Medical College Journal* 2015;**14**: 22-25.

55. Demling RH. Burns and other thermal Injuries. In: Lawrence WW, Doherty GM. editors. Current Surgical Diagnosis and Treatment, 11th ed. New York, McGraw-Hill, 2003;P.267-69.

56. Masood AFM, Khan AR, Islam KK. Surgical problems in children-10 years survey in Dhaka Shishu (Children) Hospital. *Dhaka Shishu (Children) Hospital Journal* 1991;**7**:24-29.