

Will the Amniotic Membrane and Its Stem Cells Preserve their Capacity after Cryopreservation?

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Editorial

There has been a great attention to the capacities of the amniotic membrane and its stem cells in the recent years. This membrane has been shown to have several unique capacities such as antitumor effects [1,2] as well as angiogenesis and vascular formation capacities [3]. However it is important to notice that in order to consider the amniotic membrane as a feasible option that is practical for use in clinical settings, it is essential to make sure that this tissue can be preserved and transferred to different patients as needed and when needed, while keeping its capacities after the necessary modifications. The present study aims to summarize in brief what has been done to assess the likelihood that the amniotic membrane can be preserved keeping its viability and specific properties even after freeze and thaw procedure is performed on the membrane.

The amniotic membrane is the inner most layer of the human placenta, secretes various substances and has unique characteristics such as immunoregulatory, antiangiogenic, and pro-apoptotic activity, as well as antimicrobial properties, antifibroblastic activity, cell migration, and cell growth-promoting activity. The amniotic membrane (AM) acts as a biologic barrier that supports the fetus by preparing an anatomically, physiologically and immunologically privileged space [4,5].

The AM is composed of a thick basement membrane with a single layer of epithelial stem cells (ESCs) and an avascular stroma containing mesenchymal stem cells (MSCs). Studies have shown that the amniotic membrane owes its unique characteristics to special stem cells that it harbors on its both sides.

One of the many properties that the AM has shown to have is angiogenesis capacities in the presence of MSCs. Studies have proven a pro-angiogenesis effect for these stem cells [3,6] while ESCs of the AM are shown to have anti-angiogenesis and pro-apoptosis properties [1,2]. Pro-angiogenesis effects of AM can help in controlling vascular diseases [3] while anti-angiogenesis and pro-apoptosis effects of this membrane and its stem cells can be applicable in cancer therapy [1,2,5].

All these capacities make the AM an appealing candidate for application in clinical therapies; however, an important concern in the application of the amnion in clinic is the availability of the tissue and the procedures which make it ready to apply and the vitality and effectiveness of the tissue after certain modifications [7,8]. It is true that fresh AM could easily be obtained from elective Caesarean deliveries, and is a ready-to-use tissue which has no need for processing. But if the AM is to be transferred between hospitals to be used for different patients and there is time gap between tissue retrieval and clinical application, there is a chance that the viability of

the stem cells in the fresh tissue will continue to decrease. Thus, in order to have a reliable and ready-to-use source of the stem cells of the AM, it is essential to establish a proper preservation protocol. [9,10] Cryopreservation is a well established method to preserve tissues such as the amnion [11,12], in which the tissue is stored at -80 degrees centigrade or lower in a cryoprotectant (e.g. Me2SO or glycerol) containing solution [7].

There are controversies on the effect of the cryopreservation on the AM. While some recent studies showed cryopreservation could negatively alter histological and cellular components of the AM [9,13], in several other studies we used both fresh and preserved amniotic membrane and proved to have the same angiogenesis properties even in the thawed AM [6]. Same pattern was noticeable when using cryopreserved AM and cryopreserved Amniotic Epithelial Stem cells which were detached from the AM, cultured separately and then cryopreserved. The ESCs after being thawed and co-cultured with the cancer cell lines, had the same pro-apoptotic effect on HeLa cells as well as on MDA-MB-231 breast cancer cells [2]. In another study by Niknejad and his team, the effect of cryopreservation on the angiogenesis and anti-angiogenesis properties of different sides of the AM has been closely compared between fresh and cryopreserved AM tissue. The amount of factors that contribute to angiogenesis such as IL-8 (interleukin-8) and TIMP-2 (Tissue Inhibitor of Matrix Metalloproteinase-2) were evaluated using ELISA assay. The study showed preserved properties for AM even after cryopreservation. It is shown however that in the cryopreserved AM, certain pro-angiogenic factors such as IL-8 and anti-angiogenic factors such as TIMP-2 were significantly reduced compared to fresh AMs. But the overall effects of cryopreserved AM on vessel formation and elongation was reported to be the same as its fresh counterpart [14]. These results suggest that many factors contribute in creating properties of the AM, and even though certain elements can be affected by cryopreservation, the AM and its stem cells will still keep their main properties [15].

To summarize, recent studies suggest that the AM can be cryopreserved without losing its properties and specifications related to its stem cells. These promising results can act as a basis to confirm cryopreservation as a proper and reliable method of storing AM and its stem cells and can also make the Amniotic Membrane a great candidate for application in clinical settings as it can be easily stored, preserved for a longer duration and transferred to patients whenever needed.

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