

Ultrastructural analysis reveals differences in the secretory activity among four regions of amniotic membrane

Francesca Passaretta^{1,2}, Lucia Centurione^{1,2}, Maria Antonietta Centurione^{2,3}, Mariangela Basile^{1,2}, Nicoletta Zini^{4,5}, Roberta Di Pietro^{1,2}

¹ Department of Medicine and Ageing Sciences, G. d'Annunzio University of Chieti-Pescara, Italy

² StemTeCh Group, Chieti, Italy

³ Institute of Molecular Genetics, National Research Council of Italy (CNR), Section of Chieti, Italy

⁴ CNR - National Research Council of Italy, IGM, Bologna, Italy

⁵ SC Laboratory of Musculoskeletal Cell Biology, Rizzoli Orthopaedic Institute, Bologna, Italy

Human Amniotic Epithelial Cells (hAEC) from term placenta are a promising source of stem cells for regenerative medicine. In a previous study we observed histological heterogeneity, together with different expression of pluripotency markers and content in lipid granules among four regions of amniotic membrane (AM). To better investigate cell heterogeneity among different cell populations, we performed an ultrastructural study with Transmission Electron Microscopy. Term placentae from healthy women were collected after caesarean section and AM samples were freshly isolated from four regions: R1 (close to the umbilical cord); R2 (intermediate); R3 (peripheral to the placental disc); R4 (reflected amnion). Ultrastructural analysis revealed an epithelium of variable thickness, cellular shape, amount and type of vesicles in the four regions. The epithelium showed columnar hAEC with increased height in R1 and R3 and a multi-layered organization in R3, whereas it was a monolayer in the other regions. The highest amount of granules and vesicles was observed in R3, although R4 showed granules with a different density. Furthermore, in R1, R3 and R4 we noticed several vesicles of 100-150 nm in diameter, probably exosome-like structures, suggesting a consistent secretory activity. All along its length the epithelium was rich in microvilli both on the side facing the amniotic fluid and in lateral contacts (narrow desmosomal junctions) between cells. This *in situ* investigation shows for the first time differences in secretory activity and granules appearance along the AM as a proof of its heterogeneity. This could be relevant in clinical applications as the choice of the area could improve the effectiveness of AM/hAEC transplantation.

Keywords

Term placenta, amniotic membrane, ultrastructural analysis, secretory activity