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Culture of Human Mesenchymal Stem Cells Culture in Xeno-Free Serum-Free Culture Conditions on Laminin-521

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Abstract: Introduction: Maintenance of stem cell properties during culture necessitates the recreation of the natural cell niche. Studies reported the promising outcome of mesenchymal stem cells (MSC) properties maintenance after using extracellular matrix such as CELLstart™, which is the recommended coating material for stem cells cultured in serum-free and xeno-free conditions. Laminin-521 is known as a crucial adhesion protein, which is found in natural stem cell niche, and plays an important role in facilitating the maintenance of self-renewal, pluripotency, standard morphology, and karyotype of human pluripotent stem cells (PSCs). The aim of this study is to investigate the effects of Laminin-521 on human umbilical cordderived mesenchymal stem cells (UC-MSC) characteristics as a step toward clinical application. Methods: Human MSC were isolated from the umbilical cord via the explant method. Umbilical cord-derived-MSC were cultured in serum-free and xeno-free conditions in the presence of Laminin-521 for six passages. Cultured cells were evaluated by morphology and expansion index for each passage. Phenotypic characterization of UC-MSCs cultured on Laminin-521 was evaluated by assessment of cell surface markers. Results: Umbilical cord derived-MSCs formed small colonies and expanded as a homogeneous monolayer when cultured on Laminin-521. Umbilical cord derived-MSCs reached confluence after 4 days in culture. No statistically significant difference was detected in all passages when comparing the expansion index of UC-MSCs cultured on LN-521 and CELLstart™. Phenotypic characterization of UC-MSCs cultured on LN-521 using flow cytometry revealed positive expression of CD73, CD90, CD105 and negative expression of CD34, CD45, CD19, CD14 and HLA-DR.Conclusion: Laminin-521 is comparable to CELLstart™ in supporting UC-MSCs expansion and maintaining their characteristics during culture in xeno-free and serumfree culture conditions.

Keywords: mesenchymal stem cells, culture, laminin-521, xeno-free serum-free

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