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The AlphaSTEM Test™

For optimizing therapeutic tissue stem cell production: more stem cells, less production costs

Background: Tissue stem cells play essential roles in the renewal and repair of human tissues, making them a major focus for biomedical research. Ironically, given their importance, major challenges limit the use of tissue stem cells for medical applications. Their tissue fractions are extremely low (typically as low as 1 per thousand tissue cells), they are difficult to isolate, and it is difficult to produce them in the quantities needed for cell therapy. A particularly troublesome, long-standing barrier to their therapeutic development has been the lack of a means to count them specifically. This difficulty has persisted for more than half a century because of the well known lack of specific molecular biomarkers that identify tissue stem cells, but not their more numerous progeny, early committed progenitor cells. Asymmetrex recently solved the tissue stem cell-specific counting problem with its AlphaSTEM Test™ technology. The AlphaSTEM advance opens a door to greater production of therapeutic tissue stem cells (*e.g.*, human mesenchymal stem cells, hMSCs) with greatly reduced production costs.

Applications for producers of tissue stem cell culture products: The AlphaSTEM Test™ permits, for the first time, monitoring of specific tissue stem cell number during expansion culture. As shown, in Fig. 1, the test can be used to evaluate the effects of culture supplements on tissue stem cell expansion. In addition, being a computer simulation technology, the AlphaSTEM software can be used to design culture procedures for more effective stem cell expansion. ***Optimization studies based on published hMSC expansion culture data doubled the number of hMSCs produced, while cutting production time in half and reducing production costs by more than 95%!***

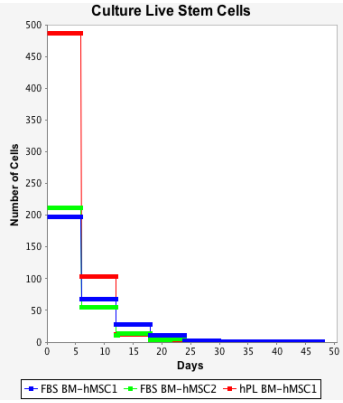

Fig. 1. An example of the use of the AlphaSTEM Test™ to detect and quantify the effect of human platelet lysate on serial cultures of human bone marrow-derived mesenchymal stem cells. Both human platelet lysate (**hPL, red**; Cook Regentec)-supplemented and fetal bovine serum (**FBS**)-supplemented hMSC cultures (from two independent donors, **hMSC1** and **hMSC2, blue and orange**) exhibit a decline in stem cell fraction with serial passage intended for stem cell expansion. The decline in tissue stem cell number with successive culture passages is predicted due to continued asymmetric self-renewal by tissue stem cells in culture (1,2). After the first six days of culture, hPL-supplemented cultures produce >75% more stem cells that FBS-supplemented cultures ($p = 0.012$). The initial stem cell fraction corresponds to approximately 1 stem cell per 1000 total cells ($p = 0.004$). Published data from Heathman *et al.*, 2016 was used for this analysis (3).

The AlphaSTEM Test™ Service: The AlphaSTEM Test™ service has been validated for stem cells from six different human tissues. These include lung, liver, hematopoietic cells for bone marrow and cord blood, and mesenchymal stromal cells from bone marrow and amniotic fluid. The service has been certified for detecting both stem cell-toxic and stem cell-activating agents. In addition to stem cell-specific fraction, the AlphaSTEM Test™ provides other stem cell-specific cell kinetics factors like generation time, self-renewal rate, and death rate. The AlphaSTEM Test™ is also the first technology that can delineate effects on tissue stem cells from effects on their lineage-committed progeny cells.

For more information contact: jtonkinson@asymmetrex.com or jsherley@asymmetrex.com

1. Rambhatla L *et al*. (2001) Cellular Senescence: Ex vivo p53-Dependent Asymmetric Cell Kinetics. *J. Biomed. Biotech.* 1, 27-36.
2. Paré J-F & Sherley JL (2006) Biological Principles for Ex Vivo Adult Stem Cell Expansion. In *Current Topics in Developmental Biology*, ed. G. Schatten, Elsevier, Inc. (San Diego), Vol. 73, pp. 141-171.
3. Heathman TRJ *et al*. (2016) Scalability and Process Transfer of Mesenchymal Stromal Cell Production from Monolayer to Microcarrier Culture Using Human Platelet Lysate. *Cytotherapy* 18, 523-535.