

**Myeloid progenitor cells: Aseptic Concentration collection and cell wash. UniFuge® Single Use Centrifuge. David Richardson, Pneumatic Scale Angelus, Clearwater, 33760, USA [david.richardson@psangelus.com](mailto:david.richardson@psangelus.com)**

**Summary:** Based on the results from the first trial in June 2012 (K562 cell line), we adjusted the G force parameter. 10 liters of myeloid progenitor suspension cells were grown in a Wave bioreactor. Aseptic collection of cells with the UniFuge single use centrifuge using the AC module. The viability of the grown cells measured 95%. The cells suspension was grown for 4 days, doubling each day. The diameter size of the cells measured between 15 -20 microns. The cells reached a final cell density of  $4.9 \cdot 10^6$  cells/ml at time of harvest. To harvest the cells, the UniFuge centrifugation parameters were feed rate (LPM) and G force. The cells were layered on a PBS buffer- a manual prefill step was utilized by switching around supply lines. The suspension cells were collected aseptically. After Harvest, the cells were washed with 3 liters of buffer, and then re-suspended in freezing medium. Cells were frozen for 24 hours, thaw and viabilities measured.

**The Objectives of this trial:**

1. Collect >90% of cells at 500 ml/min & 1500 ml/min feed rate @ 700 x G force
2. Cell viability above 90 percent after freeze thaw-most critical

**Process**

**Test 1-** 5 liters of pooled cells viability are collected into a feed bag. Connections are made with a tube welder. The cells are grown to  $4.9 \cdot 10^6$  cells/ml. Feed viability is 95%. UniFuge process parameters are 0.5 L/min @ 700 G. Fill bowl with PBS buffer until seen in centrate line. Switch buffer to feed bag for 6 minutes and 40 seconds.

**Test 2-** 5 liters of pooled cells viability are collected into a feed bag. Connections are made with a tube welder. The cells are grown to  $4.9 \cdot 10^6$  cells/ml. Feed viability is 95%. UniFuge process parameters are 1.5 L/min @ 700 G. Fill bowl with PBS buffer until seen in centrate line. Switch buffer to feed bag for 3 minutes and 20 seconds.

**Results:**

The feed cell concentration and viability were taken from the starting material ( $4.9 \cdot 10^6$  C/ml @ 95%). The cell culture was transferred to a secondary container that allowed for higher flow rates through the UniFuge (0.5L/min for the first 5L volume, 1,5L/min for the second 5L volume).

**500 mL/min –4.2L processed, 1.6L recovered,  $1.1 \cdot 10^7$  @ 93%**  
**1.500 L/min –5.6L processed, 1.6L recovered,  $1.6 \cdot 10^7$  @ 93%**

**Conclusion:** For the viable collection myeloid progenitor like suspension cells, the UniFuge single use centrifuge **met all objectives:**

1. Collect 93% of cells at 700G.
2. Cells maintained > 90 viability after 24 hours thaw.

