

Case Study

CMO's Cannot Produce SP2/0 Working Cell Bank: CytoVance Identifies Problem and Successfully Produces WCB

Benefits of Case:

- A viable Working Cell Bank (WCB) that thaws with good viability and demonstrates logarithmic growth necessary for expansion
- Further Characterization of Cell Line

Background and Challenge:

- The client had purchased a Master Cell Bank had had been manufactured in 1999.
- The client is able to grow cells successfully in their R&D laboratory.
- The Client transferred vials of MCB to a well known CMO to manufacture a WCB
- The CMO failed several times to successfully manufacture a WCB.
- Poor Viability and no growth.

Strategy:

- CytoVance to perform tech transfer utilizing data from Client R&D and CMO
- CytoVance to manufacture Engineering lot using tech transfer parameters from R&D to demonstrate transfer
- Engineering Run Data:
- The MCB thawed with good viability and had adequate growth to scale-up to achieve cell numbers to lay down 350 vial WCB
- Post Freeze Testing of the engineering run showed good viability at thaw, but viability declined and growth was not achieved.
- The beginning, middle and end of bank was tested with the same results.
- Filling Process run at room temperature and cells were exposed to DMSO for 60 minutes.
- Cryogenic freeze profile utilized forced nucleation.

Development Strategy:

- Using MCB utilize Manufacturing and Process Development technicians to study DMSO hold times
- Using MCB Utilize Manufacturing and Process Development technicians to study Room temperature versus cold process temperatures
- Using MCB Utilize Manufacturing and Process Development technicians to study Cryogenic freeze profile (1° per minute, $< 1^{\circ}$ per minute)

Development Data:

- Extended DMSO hold time of 60 minutes had no detrimental effect on viability or growth
- Process temperature favored room temperature process
- Cryogenic development data indicated that $< 1^{\circ}$ per minute worked at this scale.
- Question: Can the Controlled Rate Freezer (CRF) operate with this freeze rate and for the extended freeze time?
- Mock runs indicate CRF can maintain this rate and LN2 supply is no issue.

Value for the Client:

- A GMP run was commenced with the new freeze profile and after Post Freeze Testing of the newly manufactured WCB the bank thawed with good viability and achieved the required growth rate
- The client may now continue production and get their product into clinical trials
- Client ecstatic!