Cell Culture Automation in Biopharma

Cell Line Optimization and Monoclonal Antibody Production

Dr. Christoph Krüll
Product Manager Automated Solutions
Beckman Coulter Europe

BECKMAN COULTER

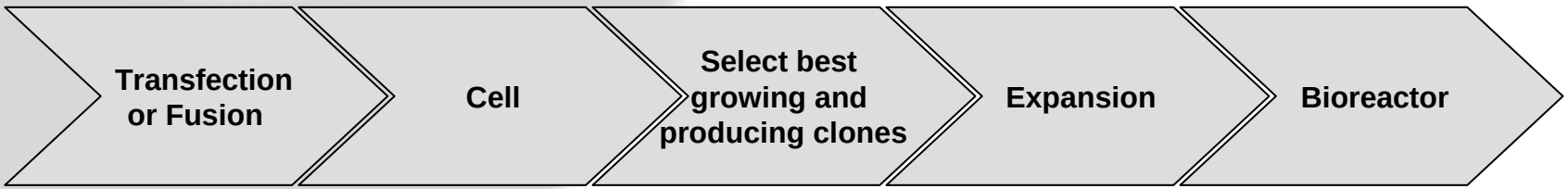
Agenda

- Cell Line Optimization and Expansion
- Monoclonal Antibody Production
- Software Control

BECKMAN COULTER

Biopharmaceuticals

Basic Process



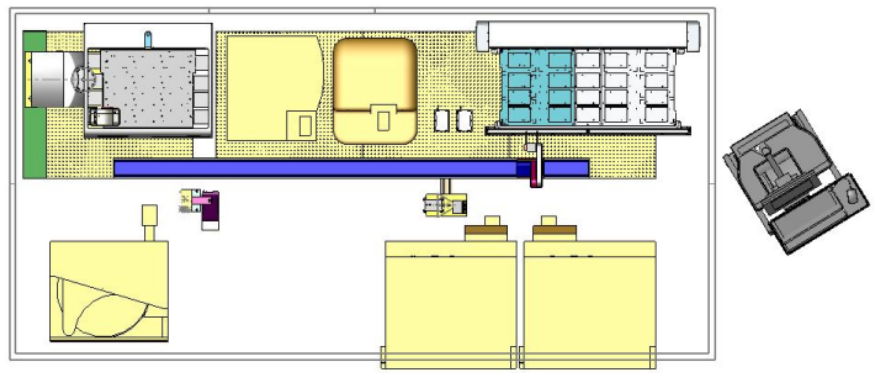
Monoclonal Antibodies

BECKMAN COULTER

Cell Line Optimization and Expansion

BECKMAN COULTER

Cell Line Optimization and Expansion System



- Requirements :
  - On-line incubation
  - Cell Line passage
  - Media exchange
  - Cell imaging for confluency and monoclonality
  - Specific protein analysis (titre)
  - Hitpicking and Expansion (96 / 24 / 6 / 1 well plates and Autoflasks)
  - Cell counting/viability assay
  - Multiple experiments occur simultaneously
    - 2 months per batch
    - 5 batches in parallel

BECKMAN COULTER

Cell Line Optimization and Expansion: Decision Making on the Fly!

- **96 well to 24 well Expansion**

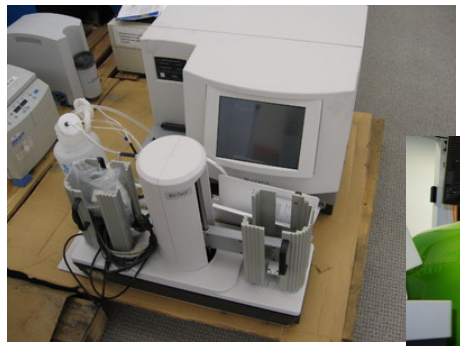
  - combined M384 and Clone Select data used to generate pick list.

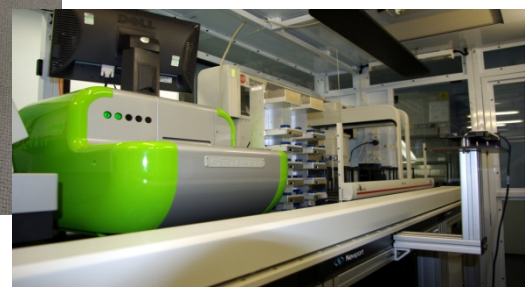
- **24 well to 6 well Expansion**

  - CloneSelect imager data generates pick list

- **Amplification in 96 well plates**

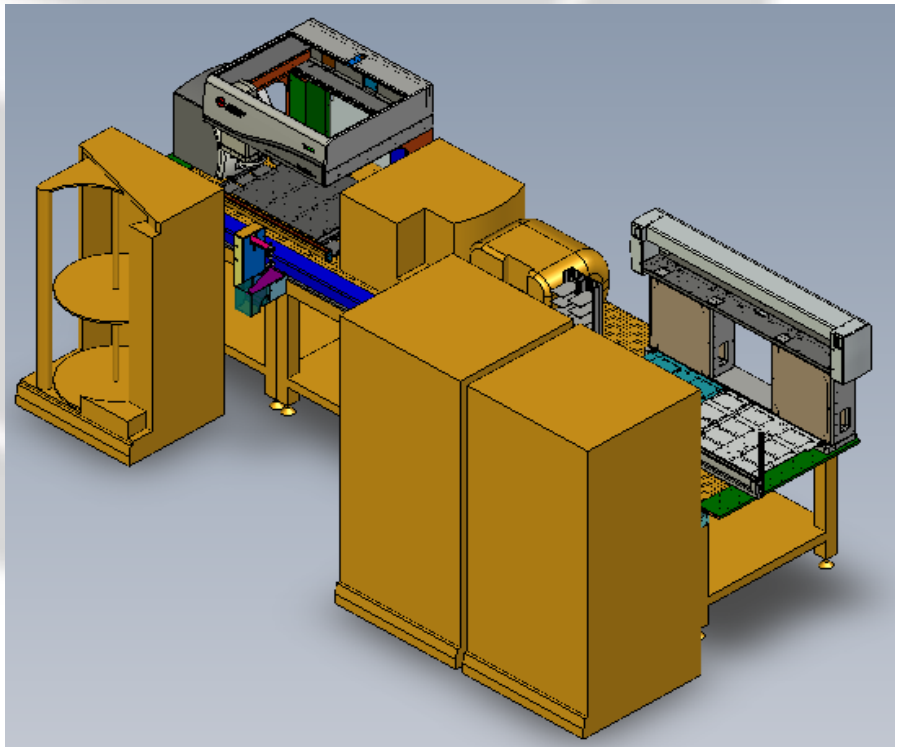
  - Vi-CELL® XR data used to plate cells back into 96 well plates at correct seeding density





BECKMAN COULTER

Cell Line Optimization and Expansion System

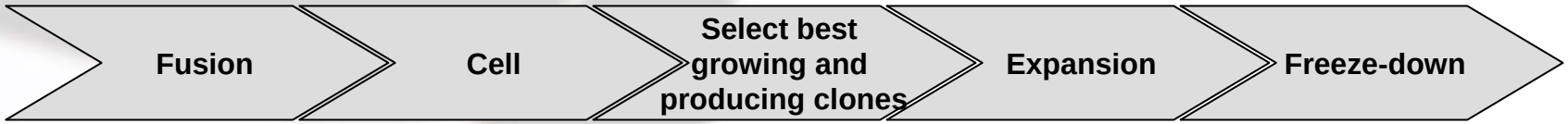


BECKMAN COULTER

Monoclonal Antibody Production

BECKMAN COULTER

Monoclonal antibody production – The Automation Mission



- Mission : Automate all non-animal tasks within the MAP
  - Confirmatory ELISA
  - Fusion
  - Clonal Expansion
  - Media Collection (Mab) and cell freeze-down
- Solution: Partial automation with separate workstations
  1. ELISA workstation
  2. Fusion workstation
  3. Expansion / Freezing

BECKMAN COULTER

Fusion Process

Spleen Harvest and Splenocyte Preparation

⇩

Viable Cell Count (Splenocytes and SP-2)

⇩

Mix Splenocytes and SP-2

⇩

Wash cell suspension

⇩

Fusion with PEG at 37ºC

⇩

Plate fused cells at limiting dilution or in Semi-Solid Medium (HAT selection)

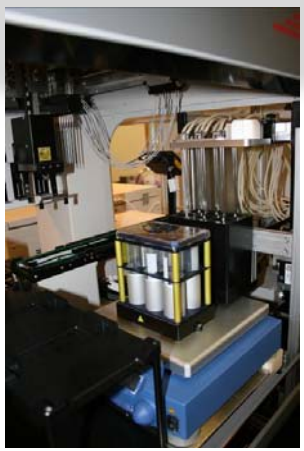
BECKMAN COULTER

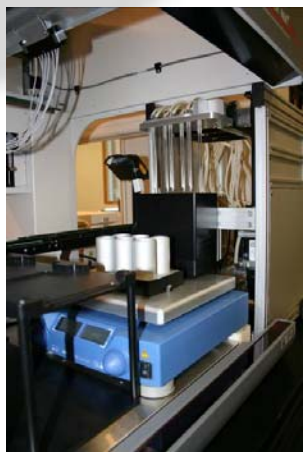
Mab Production: Fusion

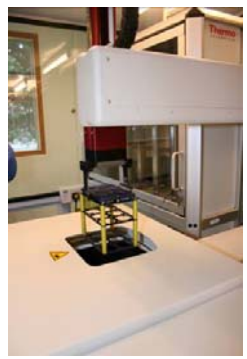
- Biomek® NX^{P} Span-8 with Gripper
- Vi-CELL XR Cell Viability Analyzer to measure viable cell count
  - splenocytes and SP-2 cells are mixed in a predefined ratio
- IKA shaker (KS130)
  - thermal exchange unit for warming tubes
- Six Position Falcon Tube Rack
  - open tube but has a micro-titre plate lid
- Bulk Dispense / Wash System
- Automated Centrifuge
- Cytomat 6001 and Cytomat Linear 8 Hotel (for tube racks)
- Sterile enclosure (Bigneat)

BECKMAN COULTER

Mab Production: Fusion





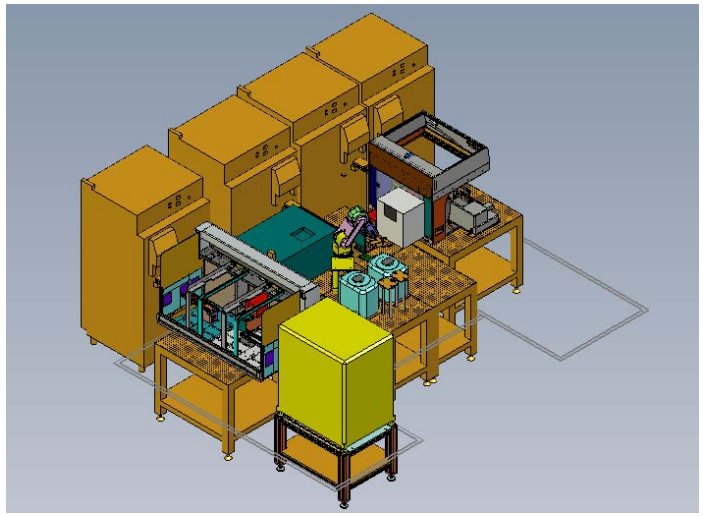


BECKMAN
COULTER

Mab Production: Expansion / Freezing

- Expansion from 96 well through 24 / 48 well plates to 6 well plates / Autoflasks
- Hitpicking based upon ELISA data
- Media exchange
- Harvesting of cells from 6 well plates / Autoflasks
- Freezedown of cells and collection of Ab-rich media

- Biomek NX^{P} Span-8 with Gripper
  - expand cells through 96, 24 and 6 well plates
  - media sampling for confirmatory ELISA; hitpicking
- Motoman HP3JC (waist-based)
  - Sigma 6K15 Centrifuge
  - FluidX Xsd-96 screwcap decapper
  - Grant Asymptote EF600 controlled rate freezers (x2)
- Biomek FX^{P} Dual Hybrid
  - Span-8 bulk transfer system
  - Simultaneous transfers using four probes across two pieces of labware
- Cytomat 24 Incubators



BECKMAN COULTER

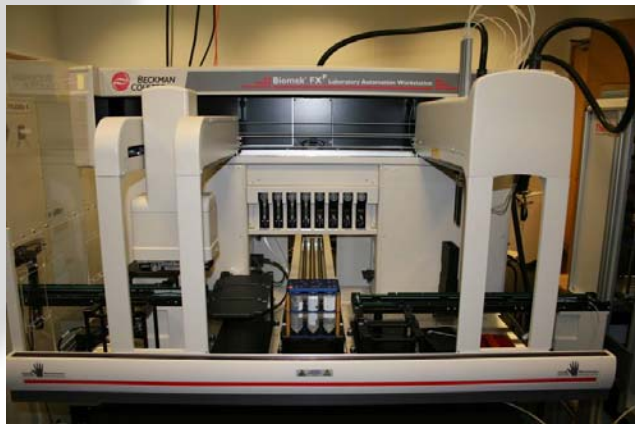
Mab Production: ZONE 1 - Expansion

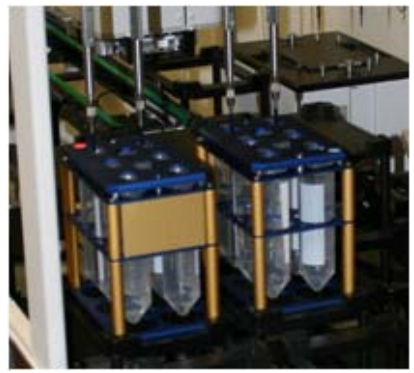
- Biomek NX^{P} Span-8 with Gripper
  - expand cells through 96, 24 and 6 well plates
  - media sampling for confirmatory ELISA; hitpicking
- Integrated Multidrop Combi
  - (custom Fillit software) for large volume media additions (24 and 6 well plates)
- Cytomat 24C incubators (x2)

BECKMAN COULTER

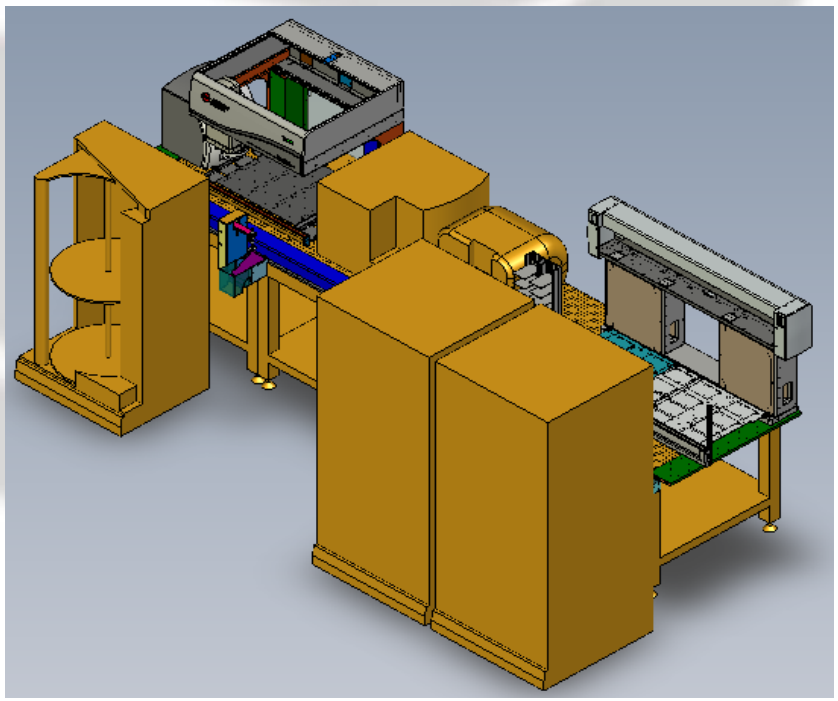
Mab Production:
ZONE 2 – Freezing & Media Harvest

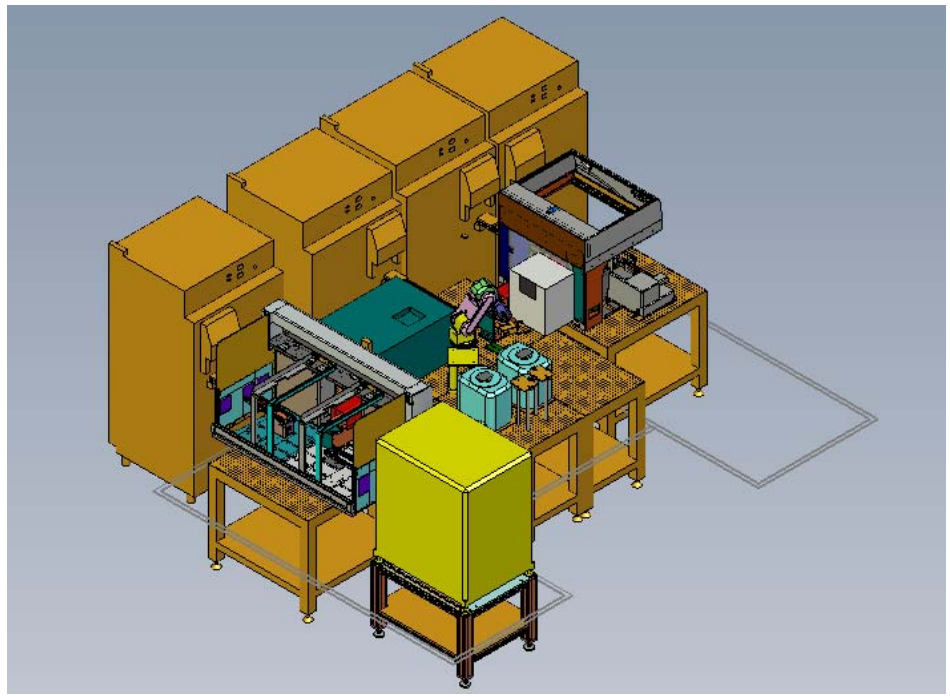
- Biomek FX^P Dual Hybrid
  - Span-8 bulk transfer system
  - Transfers from 6 well plates to 50 ml Falcon tubes
  - Cell transfer to Nunc Cryovials
- Motoman HP3JC (waist-based)
  - Sigma 6K15 Centrifuge
  - FluidX Xsd-96 screwcap decapper
  - controlled rate freezers (x2)
- Cytomat 24 Incubators





Software Control

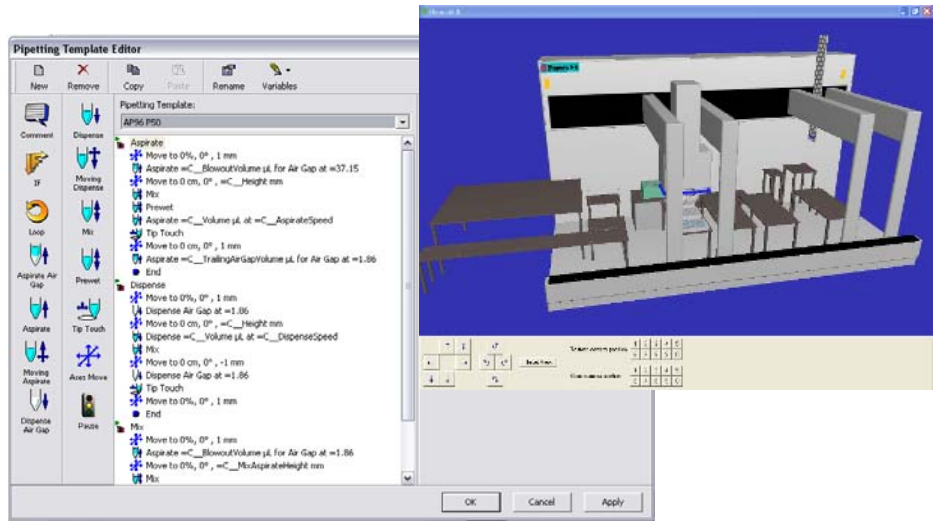




BECKMAN COULTER

Software Control

- Biomek Software
  - Common to Biomek FX^P and Biomek NX^P
  - Pipetting Template Editor controls pipetting operations (e.g. harvesting semi-adherent cells such as Hybridomas)



- SAMI® Workstation EX Scheduling Software
  - Optimising Scheduler interleaves complex processes
  - Draw the flow of plates through the process.
  - Configure liquid transfers (e.g. hitpicking), incubations (e.g. trypsinisation), device actions (e.g. multidrop media additions)
  - dynamic reschedule feature for data-driven liquid transfers (CloneSelect, Vi-CELL® XR data)

BECKMAN COULTER

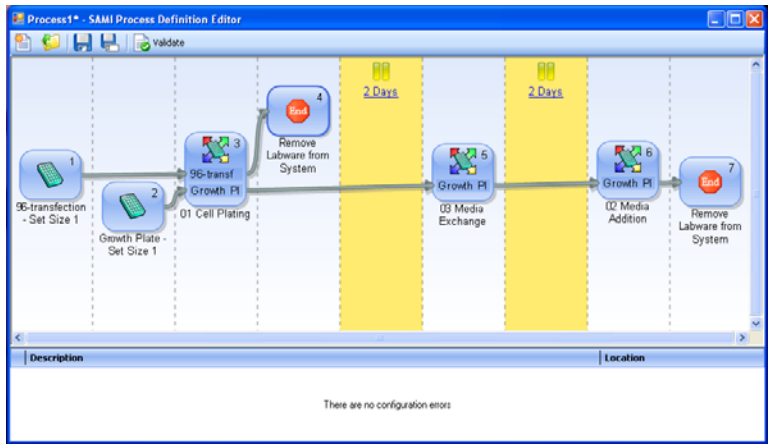
Process Management? - The Problem

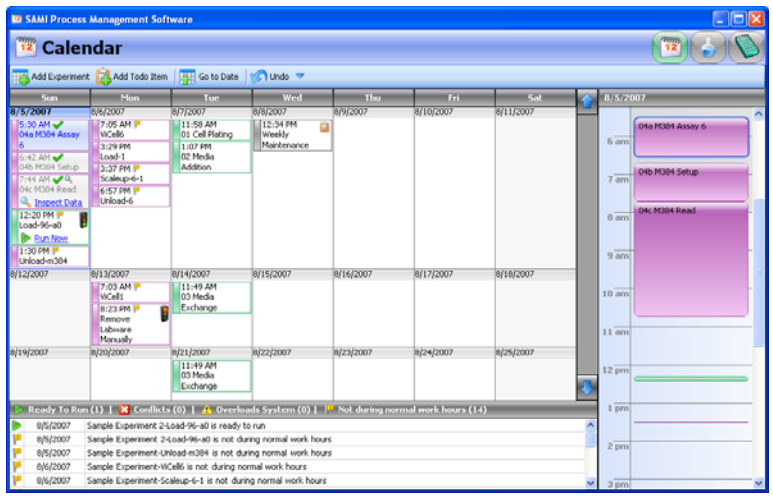
- How do I connect multiple SAMI EX methods?
- What happens to the data for these plates?
- How do I keep track of 4 batches X 6 methods per batch?
- Where are all my plates!?

BECKMAN COULTER

SAMI® Process Management Software

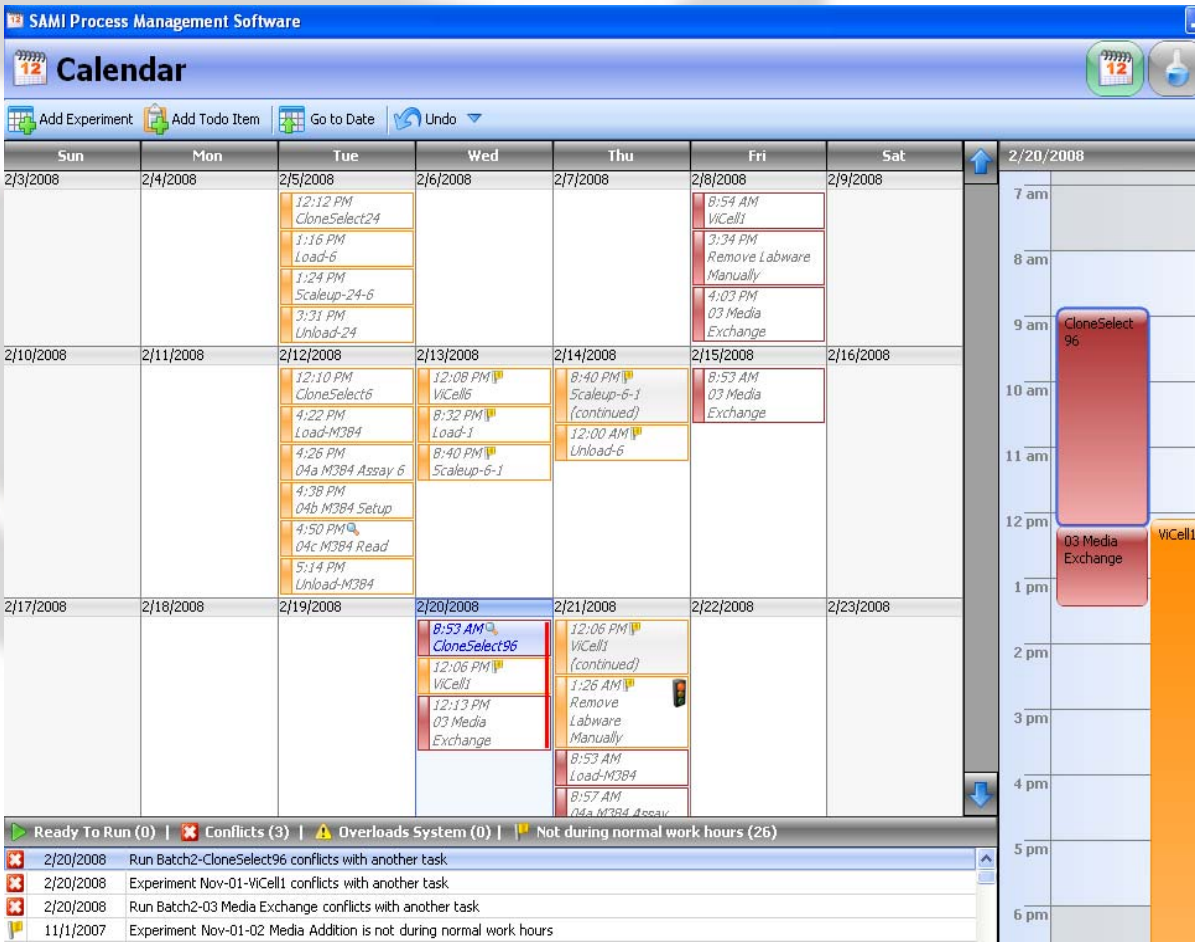
- Process Editor allows to define processes that take weeks to months to complete
- Process Management allows multiple processes to move forward independently
- Calendar view for process tasks with conflict warning / resolving options
- Integrated Database allows data flow from process to process (Data Acquisition & Reporting Tool)
- SAMI PM will track plate location and data throughout the process





BECKMAN COULTER

The Process Management Tool



- Generates conflict warnings:
  - Conflict with another task
  - Task not in normal working hours or weekends
  - System capacity (labware)
- Easy conflict resolution
  - Shift single tasks
  - Shift experiment
- Unattended start functionality for tasks occuring outside normal working hours

BECKMAN COULTER

Summary

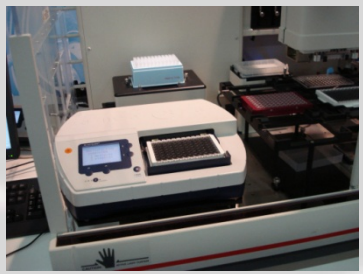
- Novel hardware solutions for automating complex cell culture automation
- First bespoke system for cell fusion from splenocyte preparation to Hybridoma
- First system for combined expansion and controlled rate freezing of cell lines
- SAMI Process Management is a unique software package allowing different cell culture processes and experiments to move forward independently
- DART enables data flow from process to process

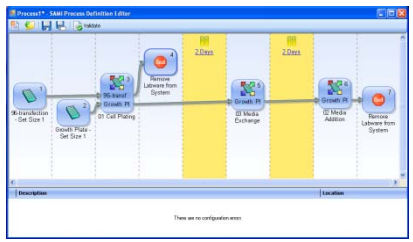
BECKMAN COULTER

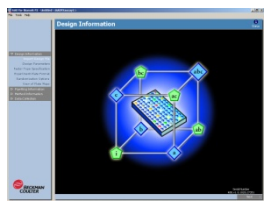
Beckman Coulter:
Your Partner in Cellular Analysis

Construct Cell Lines → **Biological Screening** → **Production**

- Transfection or Fusion
- Cell Line Optimization
- Cell Line Expansion
- Cell Production
- Production Scale Up
- Production QC







BECKMAN COULTER