



Miltenyi Biotec

CliniMACS Prodigy® Adherent Cell Culture

Differentiation of human pluripotent stem cells into cardiomyocytes

Application

The CliniMACS Prodigy Adherent Cell Culture process enables expansion and differentiation of human pluripotent stem cells (PSCs) in a closed and automated system. In combination with the ready-to-use, xeno-free StemMACS™ CardioDiff Kit XF, human, it supports robust and scalable differentiation of human PSCs into cardiomyocytes in just 8 days. This application sheet provides an entire process overview in addition to highlighting information about the required materials. In addition, it illustrates the setup of the tubing set CliniMACS Prodigy TS 730 and the key performance data.

Specifications

Process capacity:	scalable
Number of PSCs for differentiation:	approx. 1.6×10^8 cells*
Number of final cardiomyocytes:	up to 2.9×10^8 cells
Total process time:	8 days
Total hands-on time:	approx. 10 h

* The optimal initial seeding density for differentiation is cell line dependent. This is the cell number we used for our cell line in a CellSTACK 1 Chamber.

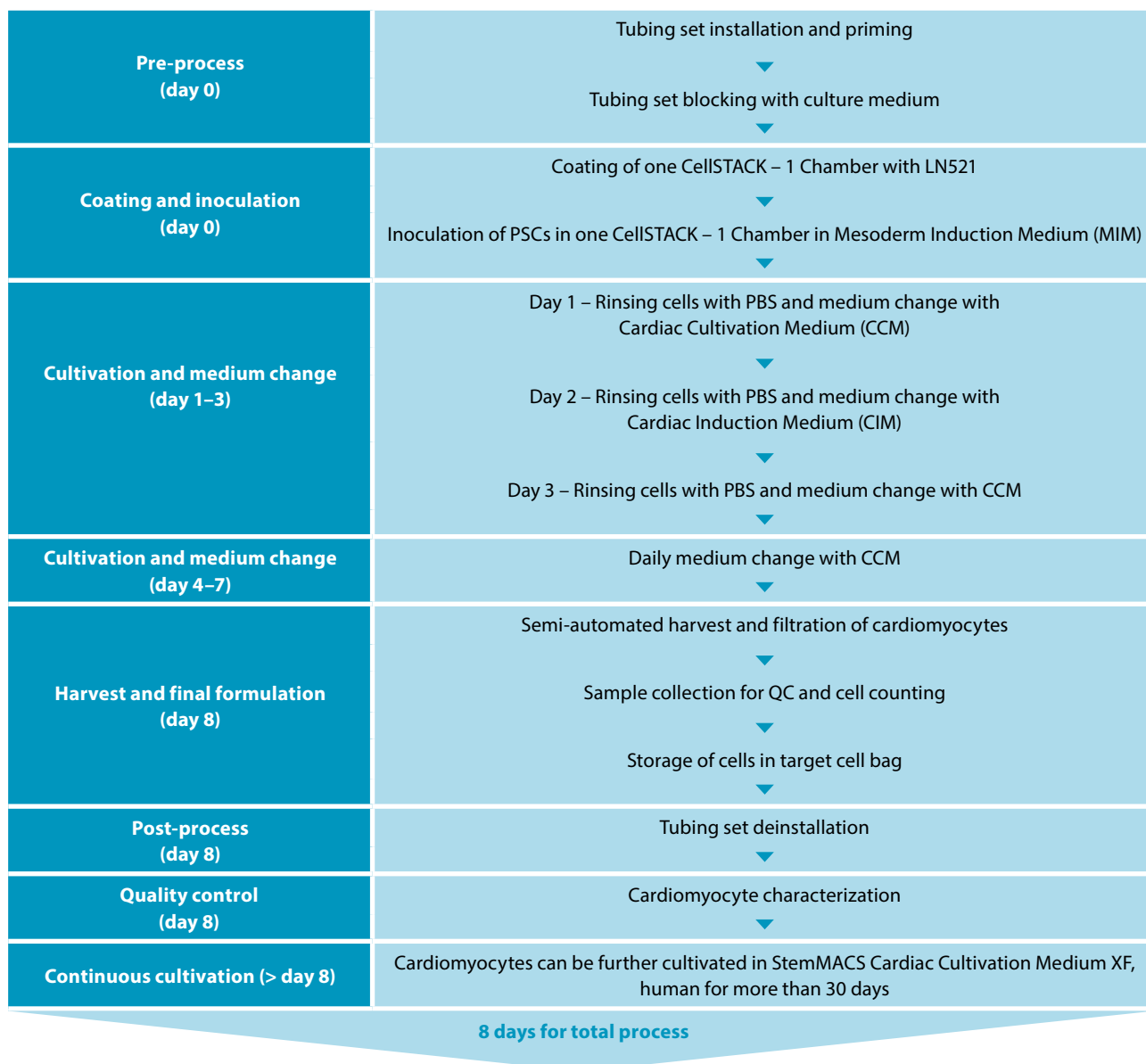
Products

Consumables	Amount required
CliniMACS Prodigy Instrument	1 piece
CliniMACS Prodigy TS 730	1 set
StemMACS CardioDiff Kit XF, human	4 kits
PBS	3 L
1 m Tube Extension	1 piece
MPC male Adapter 1/4" ID	1 piece
In-Line Filter Adapter (200 µM)	1 piece

Additional materials	Amount required
Corning® 33 mm Polyethylene Filling Cap with a Female MPC Polycarbonate with a 1/4 (6.4 mm) ID Coupling and a Male MPC Polycarbonate End Cap	1 piece
Corning CellSTACK® – 1 Chamber	1 piece
Corning 1000 mL Easy Grip Polystyrene Storage Bottles with Dip Tube, with 0.2 µm MLL/FLL Filter*	2 pieces
Flexboy Bag 500 mL, Inlet: Luer Lock male + cap, Outlet: Luer Lock female + cap, Sartorius	5 pieces
Gibco™ TrypLE™ Select Enzyme (10x), no phenol red, Thermo Fisher	100 mL
Gibco Soybean Trypsin Inhibitor, powder, Thermo Fisher	60 mL (5 mg/mL)
Biolaminin 521 LN (LN521), 500 µg, BioLamina	2 vials

* Used as alternative vessels for StemMACS CardioDiff Kit XF, human

Process overview for cardiomyocyte differentiation



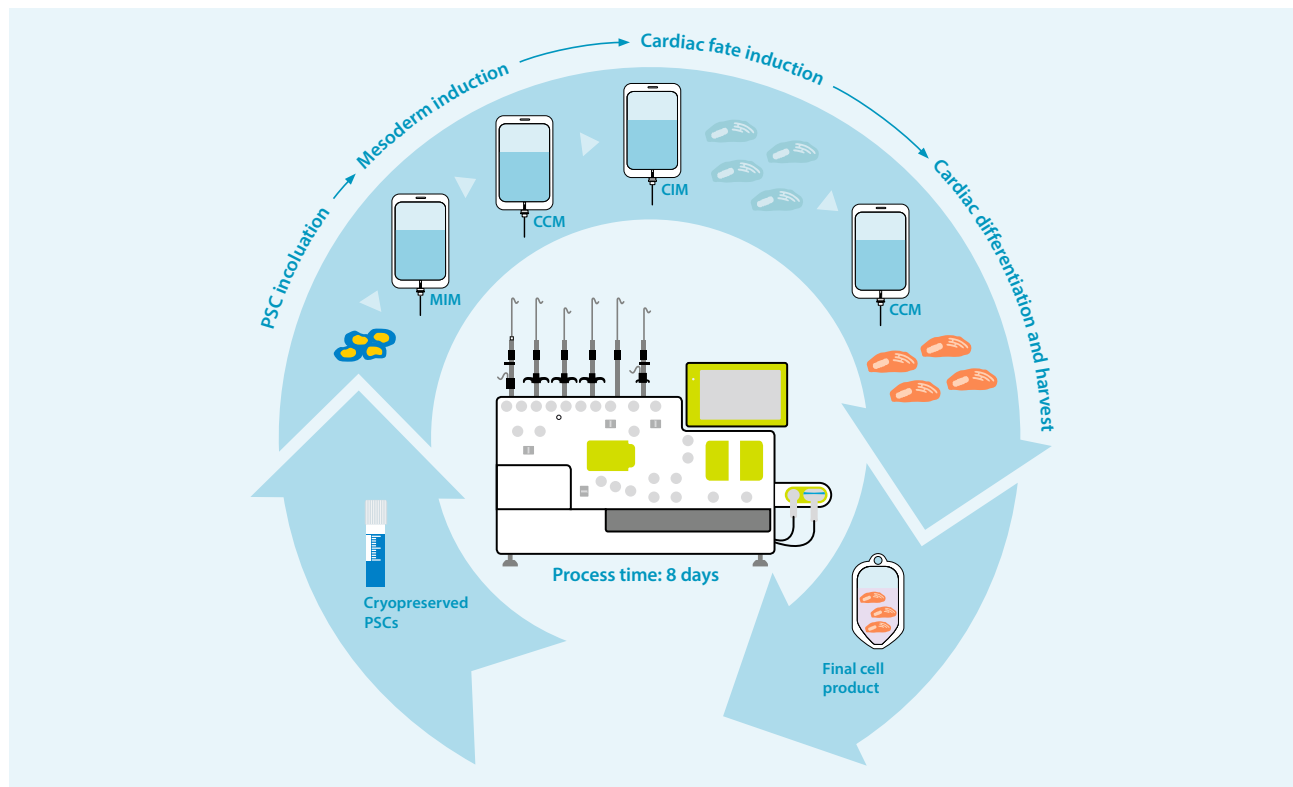
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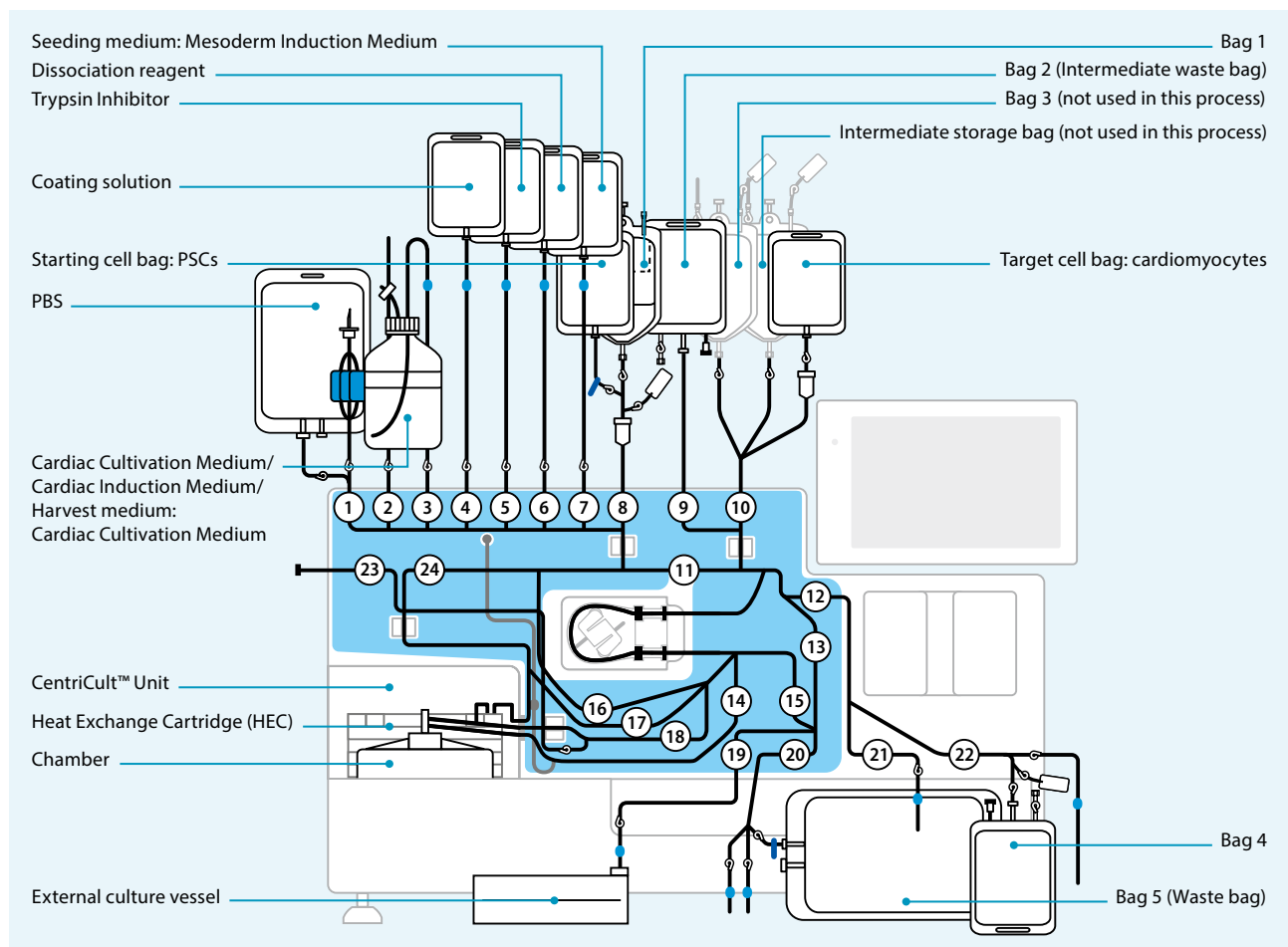
Take a look at our protocol for further information on immunophenotyping using flow cytometry

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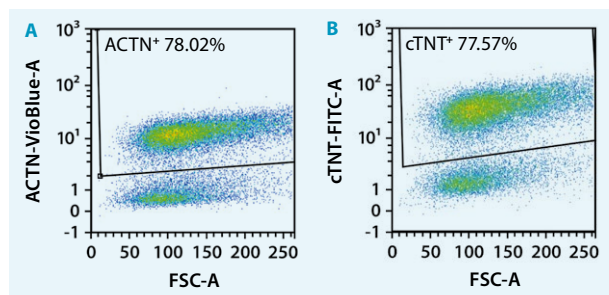
Principle of the cardiomyocyte differentiation process using the CliniMACS Prodigy



CliniMACS Prodigy TS 730 setup for cardiomyocyte differentiation



Performance data



Approximately 1.6×10^8 human PSCs were used for differentiation into cardiomyocytes using the CliniMACS Prodigy Adherent Cell Culture process with the StemMACS CardioDiff Kit XF, human. The differentiation took place in a CellSTACK – 1 Chamber from day 0 to day 8 with varying medium. (day 0 – Mesoderm Induction Medium, day 1– Cardiac Cultivation Medium, day 2– Cardiac Induction Medium, days 3–8 Cardiac Cultivation Medium). After 8 days of differentiation, the expression levels of cardiomyocyte markers α -Actinin (ACTN) (A) and Cardiac Troponin T (cTNT) (B) in the resulting cells were analyzed by flow cytometry, revealing a differentiation efficiency of approx. 78%. Furthermore, up to 2.9×10^8 ACTN⁺ cardiomyocytes could be harvested from a CellSTACK – 1 Chamber and larger external culture vessels can be employed for a scalable cardiomyocyte differentiation processes.



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