Certificate of analysis

SFC112-03-01

Signature: Katja Gassner Date: 09.11.2015

Supervisor signature: Linda Lako

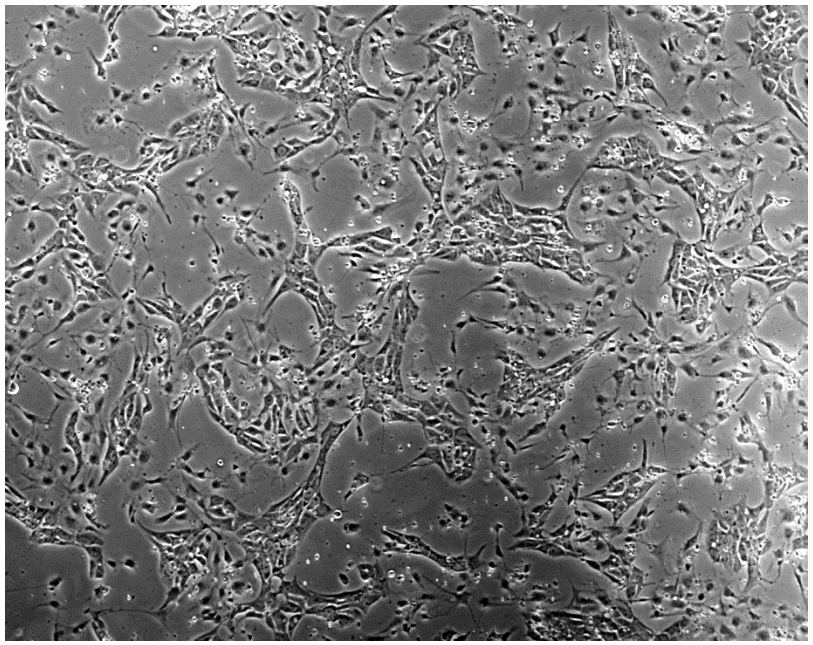
Date: 18.12 2015

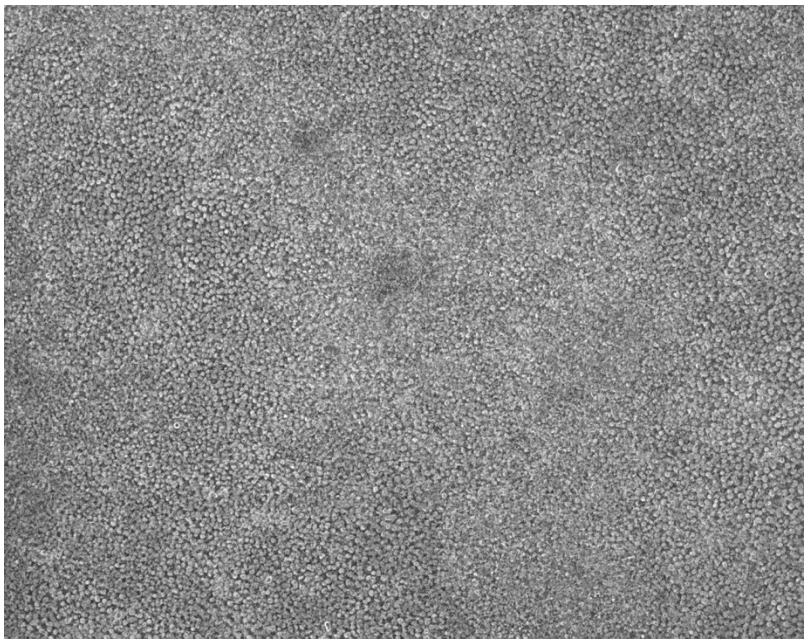
Source of fibroblasts and reprogramming information

- SF112 from UOXF
- Reprogrammed at UNEW
- Reprogrammed on 14/07/2015 at passage 6
- Cytotune 2

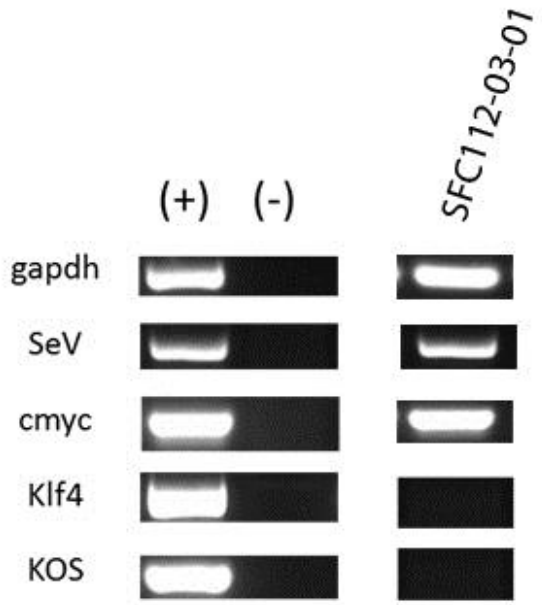
Viability post-thaw and Morphology according to SOP19 passage 13

- Cell count immediately post-thaw 1.8 x 10^6
- Viability immediately post-thaw 93%
- Photo at 24h and 5 days post-thaw:





Sendai clearance: according to WP3 SOP15 undetectable at passage 13

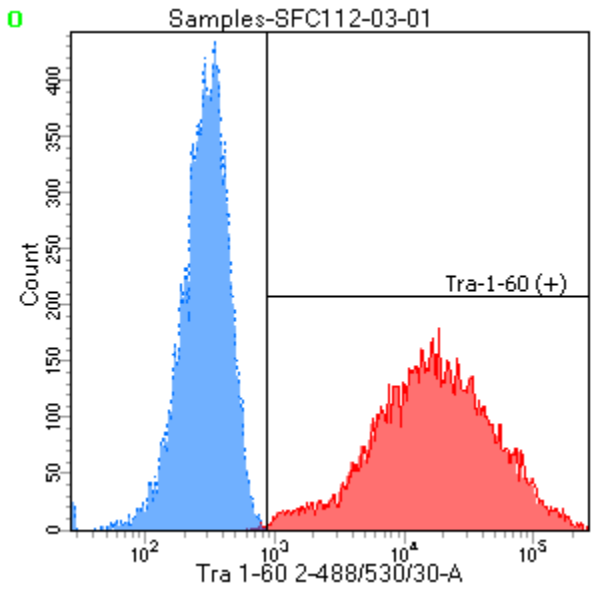


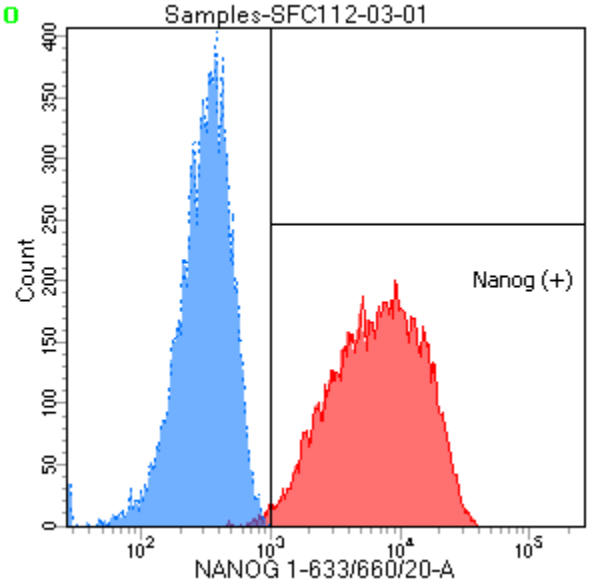
Comment: Cell line was frozen at passage 13, after thawing and 1:4/6 passaging for 3 more passages cells are still SEV-positive

Mycoplasma test:
According to MycoAlert Lonza LT07-318
Undetectable at passage 13

Owner	KG					
Date	02/11/2015					
Cell name	SFC112-03-01					
A	0.0259					
B	0.013					
B/A	0.501930502					
> 1.2		Mycoplasma Contaminated			Positive Control	Negative Control
0.9-1.2		Status Unknown - Retest within 24 hours			0.0397	0.069
0-0.9		Mycoplasma Free			0.6616	0.0055
					16.66498741	0.079710145

Flow cytometric analysis according to WP3 SOP 20 and 21 passage 13





Tube: SFC112-03-01

Population	#Events	%Parent	%Total
All Events	20,000	####	100.0
Cells	11,250	56.2	56.2
Tra-1-60 (+)	11,195	99.5	56.0
Nanog (+)	11,085	98.5	55.4
Samples/112/All Events	20,000	####	100.0
Samples/112/P1	11,074	55.4	55.4

SNP analysis
according to WP3 SOP Preparation of DNA and RNA samples for Illumina arrays

- Passage 13
- Identity to parent fibroblasts confirmed
- Karyotype abnormalities: none detected
- For details and raw data see StemDB

Comments

- Clone prone to differentiation > ReLESR Passaging after thawing recommended
  - Add 1ml ReLESR, incubate for 1 min at 37˚C
  - Remove ReLESR, incubate for 4 min at RT
  - Add medium **dropwise** to collect undifferentiated cells