Human Germ Layer Detection Kit (HGL PCR)

Catalog Number: 0833

Product Description

The ability of human embryonic and induced pluripotent stem cells to differentiate into all three germ layers has enormous potential for basic human developmental research and regenerative medicine. ScienCell has created a convenient multiplex PCR kit for the routine detection of human pluripotent stem cell differentiation. Multiplex PCR allows two or more genes to be amplified in a single PCR reaction by using multiple primer pairs in a single reaction mixture, allowing for considerable savings in labor, cost and precious DNA samples. All required PCR reagents are supplied in this kit. Simply add DNA template and perform the PCR reaction. Tube 1 ready-mix reaction contains *NEUROD1*, *SMA* and *aFP* primers that allows for the detection of ectoderm, mesoderm and endoderm, respectively [1].

Kit Components

Cat. No.	# of vials	Name	Quantity	Storage
0833a	1	Tube 1: differentiation ready-mix	850µL	-20°C
0833b	1	nuclease-free H_2O	1mL	-20°C

Materials to be Supplied by the User

thin wall PCR tubes
DNA template
thermal cycler
agarose gel
ethidium bromide
electrophoresis system
gel imager

Quality Control

cDNAs from differentiated human pluripotent stem cells were used as template DNA. Each PCR product was sequenced to ensure specificity.

Product Use

HGL PCR kit is for research use only. It is not approved for human or animal use, or for application in *in vitro* diagnostic procedures.

Storage

Store in -20ºC upon receipt. Avoid repeated freeze thaw cycles by making five aliquots at 170µL each.

Shipping

Dry ice.

References

[1] Adewumi, O. et al. (2007) Characterization of human embryonic stem cell lines by the International Stem Cell Initiative. *Nat Biotechnol*. 25:803-816.

Procedures

1. Mix the following components in a thin-wall PCR tube:

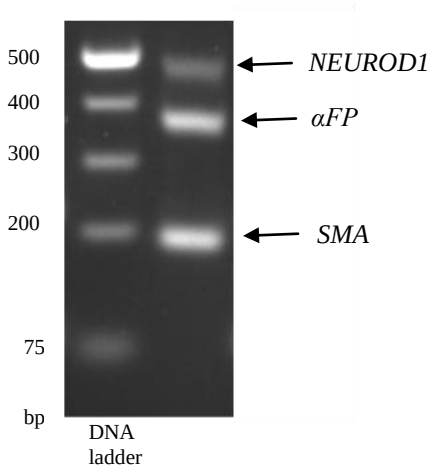
Component	Amount
Tube 1 ready mix	17µL
DNA template*	3µg
H_2O	up to 30 µL

*Optimal cDNA concentration should be ~3µg/µL

2. Perform PCR using the following conditions:

Step 1: 95 ºC	3min
Step 2: 95 ºC	1min
Step 3: 55 ºC	30sec
Step 4: 72 ºC	1min
Step 5: repeat step steps 2-4 for 40 times	
Step 6: 72 ºC	2min

3. Visualize PCR products on a 3% agarose gel containing ethidium bromide.



Expected product sizes:

Gene	Expected Size
NEUROD1	473 bps
αFP	366 bps
SMA	186 bps