

Hep-G2/2.2.15 Human Hepatoblastoma Cell Line

Cancer Cell Line

Cat. # SCC249

Pack size: $\geq 1 \times 10^6$

viable cells/vial

Store in liquid nitrogen

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Data Sheet

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Background

Hepatocellular carcinoma is the primary malignancy of the liver and a leading cause of cancer-related death in the world. The etiology of hepatocellular carcinoma is closely linked to infection with hepatitis B virus.¹ Because of the causative role of HBV in liver cancers, cellular models of HBV-infected hepatocellular carcinomas have been developed for understanding the progression and fundamental biology of liver cancers.

The Hep-G2/2.2.15 human hepatoblastoma cell line, a subclone of HepG2 human hepatoblastoma that stably expresses the hepatitis B virus, is one of the most widely used models for HBV-associated liver disease. Hep-G2/2.2.15 cells possess lower proliferation and invasion activity than parental HepG2 cells and have low tumorigenicity in nude mice.² Like parental HepG2 cells, Hep-G2/2.2.15 cells express the microtubule-associated marker doublecortin (DCX). The Hep-G2/2.2.15 cell line is a suitable model for research into many aspects of HBV-associated liver cancer.

Source

Hep-G2/2.2.15 was produced from Hep-G2 cells stably transformed with the hepatitis-B virus (HBV) genome. Hep-G2/2.2.15 cells are resistant to G418 and contain two head-to-tail dimers of the HBV type D genome.³ The parental HepG2 cell line was isolated from hepatoblastoma tissue of a 15-year-old male patient.⁴

Short tandem repeat (STR) Profile

D3S1358: 15, 16	D16S539: 12, 13
TH01: 9	CSF1PO: 10, 11
D21S11: 29, 31	Penta D: 9, 13
D18S51: 13, 14	vWA: 17
Penta E: 15, 20	D8S1179: 15, 16
D5S818: 11, 12	TPOX: 8, 9
D13S317: 9, 13	FGA: 22, 25
D7S820: 10	Amelogenin: X, Y

Cancer cell lines are inherently genetically unstable. Genetic instability may arise in the form of loss of heterozygosity of alleles at one or more genetic sites with increased passages.

Storage & Handling

Hep-G2/2.2.15 human hepatoblastoma cell line should be stored in liquid nitrogen. The cells can be cultured for at least 10 passages after initial thawing without significantly affecting the cell marker expression and functionality

Quality Control Testing

- Each vial contains $\geq 1 \times 10^6$ viable cells.
- Cells tested positive for Hepatitis B virus (HBV) and negative for all other infectious diseases in the Human Essential CLEAR panel by Charles River Animal Diagnostic Services.
- Cells are verified to be of human origin and negative for inter-species contamination from rat, mouse, Chinese hamster, Golden Syrian hamster, and non-human primate (NHP) as assessed by a Contamination CLEAR panel by Charles River Animal Diagnostic Services.
- Cells are negative for mycoplasma contamination.
- Each lot of cells is genotyped by STR analysis to verify the unique identity of the cell line.

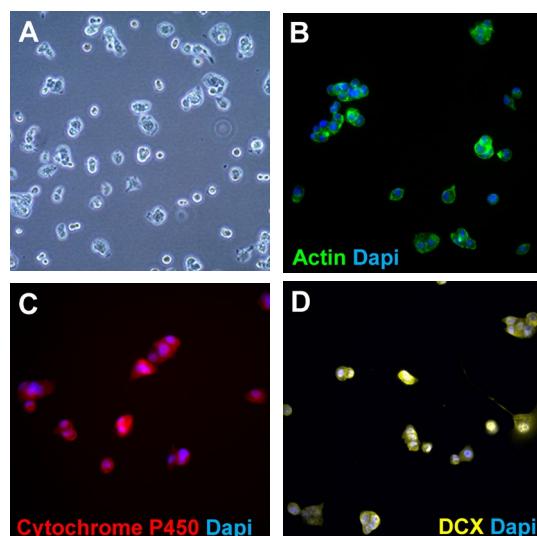


Figure 1. Hep-G2/2.2.15 cells one (A) day after thawing in a T75 flask. Cells express actin (B, Phalloidin-FITC; Sigma P5282), Cytochrome P450 (C, Sigma SAB4500606) and Doublecortin, DCX (D, Sigma MABN707).

References

1. *World J Gastroenterol.* 2007; 13(32): 4295-42305.
2. *World J Gastroenterol.* 2011; 17(9): 1152-1159.
3. *Proc Natl Acad Sci USA.* 1987; 84(4): 1005-1009.
4. *Nature.* 1979; 282(5739): 615-616.

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Protocols

Thawing Cells

1. Do not thaw the cells until the recommended medium is on hand. Cells can grow on normal tissue cultureware surfaces without any additional coating.

Hep-G2/2.2.15 Expansion Medium: Cells are thawed and expanded in DMEM-High Glucose (Sigma D5796) supplemented with 10% FBS (Millipore ES-009-B).

2. Remove the vial of frozen Hep-G2/2.2.15 cells from liquid nitrogen and incubate in a 37°C water bath. Closely monitor until the cells are completely thawed. Maximum cell viability is dependent on the rapid and complete thawing of frozen cells.

IMPORTANT: Do not vortex the cells.

3. As soon as the cells are completely thawed, disinfect the outside of the vial with 70% ethanol. Proceed immediately to the next step.

4. In a laminar flow hood, use a 1 or 2 mL pipette to transfer the cells to a sterile 15 mL conical tube. Be careful not to introduce any bubbles during the transfer process.

5. Using a 10 mL pipette, slowly add dropwise 9 mL of Hep-G2/2.2.15 Expansion Medium (Step 1 above) to the 15 mL conical tube.

IMPORTANT: Do not add the entire volume of media all at once to the cells. This may result in decreased cell viability due to osmotic shock.

6. Gently mix the cell suspension by slowly pipetting up and down twice. Be careful not to introduce any bubbles.

IMPORTANT: Do not vortex the cells.

7. Centrifuge the tube at 300 x g for 2-3 minutes to pellet the cells.

8. Decant as much of the supernatant as possible. Steps 5-8 are necessary to remove residual cryopreservative (DMSO).

9. Resuspend the cells in 15 mL of Hep-G2/2.2.15 Expansion Medium.

10. Transfer the cell mixture to a T75 tissue culture flask.

11. Incubate the cells at 37°C in a humidified incubator with 5% CO₂.

Subculturing Cells

1. Do not allow the cells to grow to confluence. Hep-G2/2.2.15 cells should be passaged at ~80-85% confluence.

2. Carefully remove the medium from the T75 tissue culture flask containing the Hep-G2/2.2.15 cells.

3. Rinse the flask with 10 mL 1X PBS. Aspirate after the rinse.

4. Apply 5-7 mL of Accutase and incubate in a 37°C incubator for 3-5 minutes.

5. Add 5-7 mL of Hep-G2/2.2.15 Expansion Medium to the plate.

6. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells to a 15 mL conical tube.

7. Centrifuge the tube at 300 x g for 3-5 minutes to pellet the cells.

8. Discard the supernatant, then loosen the cell pellet by tapping the tip of the tube with a finger.

9. Apply 2-5 mL of Hep-G2/2.2.15 Expansion Medium to the conical tube and resuspend the cells thoroughly.

IMPORTANT: Do not vortex the cells.

10. Count the number of cells using a hemocytometer.

11. Plate the cells to the desired density. Typical split ratio is 1.6.

Cryopreservation of Cells

Hep-G2/2.2.15 human hepatoblastoma cell line may be frozen in the expansion medium plus 10% DMSO using a Nalgene slow freeze Mr. Frosty container.

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