MAINTAINING HUMAN CELL LINES IN AN ESCO CELCULTURE® CO_2 INCUBATOR FOR 3D CELL CULTURE CONDITIONS

Abstract

For years, 3D cell culture has been providing scientists good models of systems seen *in vivo*. This technology was utilized by a recent study to evaluate the absorption of the drug in the intestines. A 3D model of the intestinal architecture was constructed and seeded with intestinal myofibroblast (CCD18-Co cells), epithelial enterocytes (Caco-2 cells), and mucus-producing cells (HT29-MTX cells). These cells were cultured in an Esco CelCulture® CO_2 incubator prior to seeding. The CelCulture® CO_2 incubator was able to provide the optimum growth conditions for these cell lines.

Introduction

3D cell culture has provided a whole new method on replicating the *in vivo* cellular environment. Cells grown in a 3D format have exhibited significant similarities in morphology and behaviour with those *in vivo*.[1] Various fields have benefited from the 3D cell culture technique, one of which is drug discovery.

Early stages of drug discovery involve the evaluation of the absorption, distribution, metabolism, and excretion (ADME) of drug candidates. For instance, absorption evaluation is traditionally done thru *in vitro* intestinal models. However, such models are based on cell cultures on petri dishes or flasks, and therefore do not provide a good look into the actual intestinal architecture. 3D culture models competently address this concern, as such models demonstrate the morphology and behaviour of living cells through matrix dimensionality.[2]

Such feature of 3D culture was utilized by a recent study entitled, "*Dissecting stromal-epithelial interactions in a 3D* in vitro *cellularized intestinal model for permeability studies"* by Pereira et al. (2015) to evaluate the absorption of drugs in the intestine.[2] To mimic the *in vivo* architecture of the intestine, the study used three different cell lines. The intestinal myofibroblast (CCD18-Co cell line) was used to represent the myofibroblasts in the lamina propria. Epithelial enterocytes (Caco-2 cells) and mucus-producing cells (HT29-MTX cells) were then seeded on top of the CCD18-Co cells to demonstrate the epithelial layer.[2] These cells require specific conditions for growth, which were all satisfied by the Esco CelCulture® CO_2 incubator.

CCD18-Co cell line

The CCD18-Co cell line is an adherent human colon myofibroblast. This cell line must be maintained in ATCC-formulated Eagle's Minimum Essential Medium (Catalog No. 30-2003). Fetal bovine serum is added to the medium for a complete growth medium to a final concentration of 10%. Tumor necrosis factor alpha (TNF-α) is added to enhance the medium. The cell line must be incubated at 37°C.[3]

[1] Justice et al. 2009
[2] Pereira et al. 2015
[3] ATCC.

Caco-2 cell line

Caco-2 is an adherent human colon epithelial cell line. These cells must be maintained in ATCC-formulated Eagle's Minimum Essential Medium (Catalog No. 30-2003). To complete the medium, fetal bovine serum must be added, to a final concentration of 20%. Cells must be cultured with 95% relative humidity, 5% CO_2, and 37°C.[3]

HT29-MTX cell line

The HT29-MTX cell line is an adherent human epithelial cell line that is mucus-producing. Cells must be maintained in Dulbecco's Modified Eagle Medium (DMEM) with 2 mM glutamine, 1% nonessential amino acids, and 10% fetal bovine serum. This cell line must be incubated at 37°C with 7.5-10% CO_2.[4]

CelCulture® CO_2 Incubator

Prior to seeding into the 3D intestinal model, the abovementioned cell lines were incubated in Esco CelCulture® CO_2 incubator. The CelCulture® was able to provide optimum growth conditions for the cell lines: 37°C, 5% CO_2, in a water saturated atmosphere.[1]

CelCulture® boasts of its precise parameter control and robust contamination control methods. Through the microcontroller PID, direct heat and air jacket of the incubator is continuously maintained, ensuring optimum temperature conditions for cell lines. Moreover, with the top-of-the-line IR sensor, the CelCulture® precisely monitors CO_2. Relative humidity is maintained through a humidity pan. Air is constantly circulated inside the chamber via the forced convection method.

Aside from providing optimal environmental conditions for your samples, the CelCulture® CO_2 incubator maintains a sterile chamber through its ULPA filtration system which has a 99.999% efficiency – more efficient than conventional HEPA filters. As for decontamination, the CelCulture® CO_2 incubator is equipped with a 90°C moist heat decontamination method that completes overnight. The chamber is dry after the cycle, thus there is no need for wipe down. Contamination control is further enhanced in the CelCulture® through the Isocide™ antimicrobial electro-galvanized powder coating that eliminates 99.9% of surface bacteria within 24 hours of exposure.

Fig. 1. The CelCulture® CO_2 Incubator for maintaining cell lines for 3D culture models.

[4] Sigma Aldrich.

Conclusion

3D cell culture is another emerging field of research, as it has various applications such as drug discovery. For this new technique, the Esco CelCulture® CO_2 incubator is a reliable partner in maintaining cell lines prior to seeding to 3D culture models. With its precise parameter control and robust contamination control methods, the CelCulture® CO_2 incubator is still the best choice for cell culture applications.

References

CCD18-Co and Caco-2 cell line description and culture conditions. Retrieved from https://www.atcc.org/: June 28 2017.

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