

## CULTIVATION OF MARINE INVERTEBRATE CELLS WITH CEL CULTURE® CO<sub>2</sub> INCUBATORS

### Abstract

Marine invertebrate cell culture is a very promising tool in cell research and its applications but there are no standardized protocols that cover all cell types. While there is no definitive medium, vessel, or temperature for marine invertebrate cell culture, CelCulture® CO<sub>2</sub> Incubator with Cooling System is suitable to be the standard CO<sub>2</sub> incubator for this type of culture due to its wide temperature range and competent contamination control methods.

### Introduction

In recent years, marine invertebrate cell culture served as a tool in generating models for the study of animal development, cases of malignant transformation, cell activation, and other cellular processes. Marine invertebrates, such as sponges and echinoderms, are known to produce a large variety of natural bioactive metabolites that could be processed for drug development<sup>1</sup>. Aquatic invertebrates are also interesting subjects in the study of pluripotent stem cells as they possess huge amounts of these stem cells in adulthood<sup>2</sup>. Consequently, the research on marine invertebrate cell culture has progressed to the development of immortal cell lines, to be used for a variety of application, including cancer research and regenerative medicine<sup>3</sup>. While there is a difficulty in the establishment of *in vitro* cultures for marine invertebrates, the fact still holds that this type of cell culture is a promising tool in cell biology, drug discovery, and biomedicine.

Esco recognized the significance of marine invertebrate cell culture and manufactured the CelCulture® CO<sub>2</sub> Incubators with Cooling System (Fig.1). This paper will outline the benefits of using CelCulture® for marine invertebrate cell cultivation, taking a look first on the growth requirements of marine invertebrate cells.



Figure 1. Esco CelCulture® CO<sub>2</sub> Incubators with Cooling System

<sup>1</sup> Ilan et al. 1996

<sup>2</sup> Rinkevich et al. 2009

<sup>3</sup> Rinkevich 2011

### Cultivating Marine Invertebrate Cells

While specific requirements for the culture of marine invertebrate cells vary per species, some general requirements can be noted. As in the culture of other organisms, marine invertebrate cell culture requires a suitable growth medium. There is no established culture medium yet for marine invertebrates but recent studies include the use of Leibovitz L-15 medium for the marine sponge *Latrunculia magnifica* and the zebra mussel *Dreissena polymorpha*<sup>1,4</sup> ; while epithelial cells of the shrimp *Penaeus monodon* grew best in modified Grace's medium<sup>5</sup>. Thus, optimization of a chosen medium must be done for a specific marine invertebrate cell line.

As in vertebrate cell culture, marine invertebrate cells could be kept at a variety of vessels, such as petri dishes, 24-well plates, and tissue culture flasks. Air-tight flasks are more beneficial to use as these vessels help reduce contamination<sup>4</sup>.

Invertebrate cells must be incubated at temperatures lower than that required for vertebrate cells. Marine invertebrate cells are kept at 15°C-23°C, depending on the species<sup>1,4</sup>.

### Cell Culture with CelCulture®: Guaranteed Protection and Stability

Due to the lower temperature requirement of marine environment cells, there is a need to keep them in an incubator with a lower ambient temperature and wider temperature range. This is exactly what the CelCulture® CO<sub>2</sub> Incubator with Cooling System offers. This incubator has a temperature range of 8°C below ambient to 60°C for marine invertebrate cell culture.

Considering that marine invertebrate cell culture often involves primary cell culture, the CelCulture® with Cooling System is built with maximum contamination control methods to achieve near-zero contamination. The 90°C moist heat decontamination cycle eliminates fungi, bacterial spores, and vegetative cells. The cycle completes in less than 15 hrs.

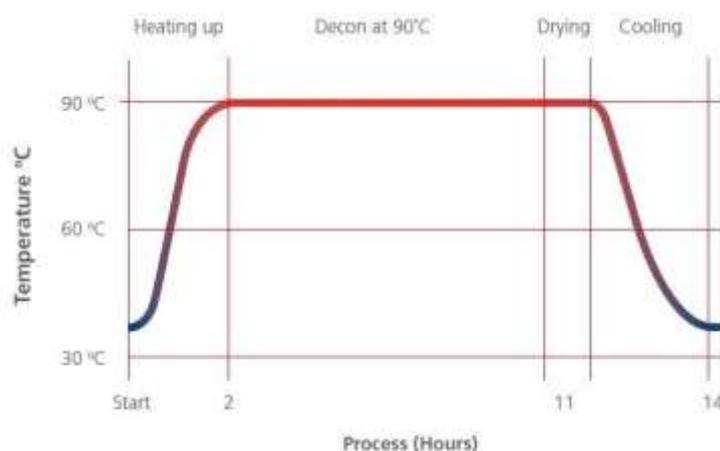


Figure 2. The moist heat decontamination cycle completes within 15 hrs. to eliminate microorganisms.

<sup>4</sup> Quinn et al. 2009

<sup>5</sup> Fraser and Hall 1999

The exterior of the incubator is coated with Isocide™ electrogalvanized antimicrobial coating that eliminates 99.99% of surface bacteria. Air is filtered thru an ULPA filter with 99.999% efficiency to ensure an ISO Class 5 Cleanliness inside the chamber. Stability and precision of environmental parameters are maintained by smart sensors for CO<sub>2</sub>, O<sub>2</sub>, and RH.

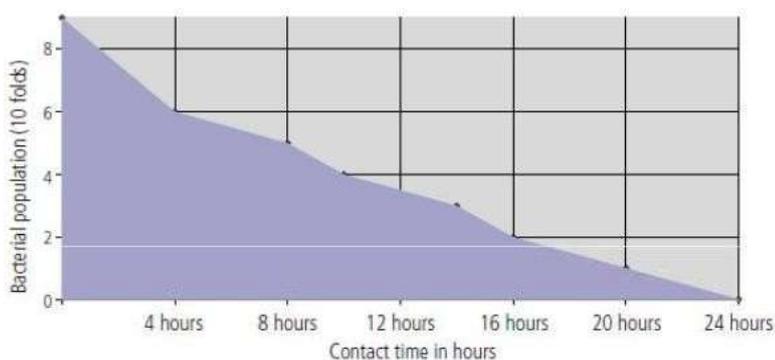


Figure 3. Isocide™ eliminates 99.9% of bacteria after a 24-exposure period.

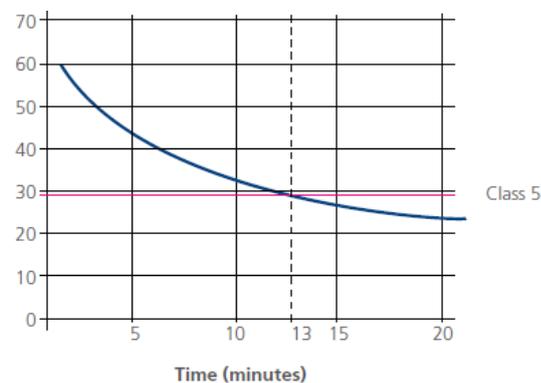


Figure 4. ULPA filtration system renders an ISO Class 5 Chamber.

These outstanding features prove that CelCulture® CO<sub>2</sub> Incubator with Cooling System is a suited for marine invertebrate cell culture.

### Conclusion

Despite having no standard cell culture protocols, culture of marine invertebrate cells is a very promising tool in cell research and its applications. CelCulture® CO<sub>2</sub> Incubator with Cooling System is appropriate for this type of culture because of its wide temperature range and complete contamination control methods.

## Conclusion

Despite having no standard cell culture protocols, culture of marine invertebrate cells is a very promising tool in cell research and its applications. CelCulture® CO<sub>2</sub> Incubator with Cooling System is appropriate for this type of culture because of its wide temperature range and complete contamination control methods.

## References

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