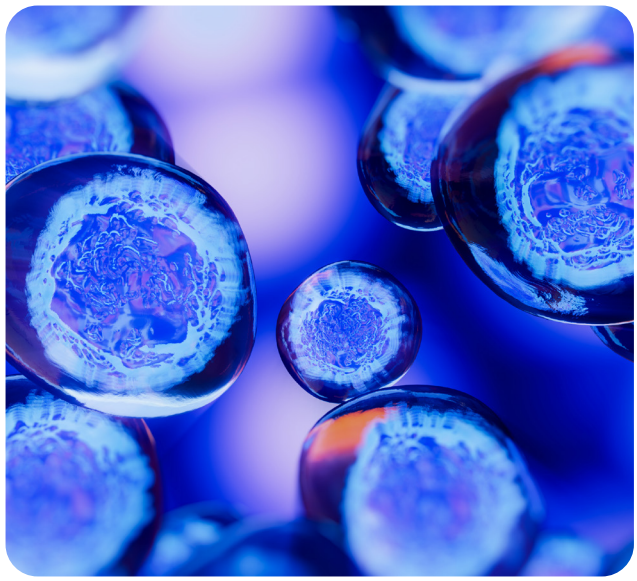
The 5 most common mistakes to avoid when culturing IVISbrite tumor cells

To ensure your success in working with our cell lines, please read our IVISbrite™ Tumor Cell Culture Guidelines, located on the tumor cell lines product webpages, as our recommendations may be different from other standard culturing practices.

Though it is important to follow all of the recommendations in the IVISbrite Tumor Cell Culture Guidelines, we have noted some of the most common pitfalls below, and our recommended culture technique.



1. Centrifuging the cells

Centrifuging cell suspensions may be standard practice to some, but we advise against centrifuging most of the IVISbrite cell lines*, especially upon thaw.

Many of the cell lines are fragile and will recover best if diluted into complete medium without antibiotics and plated gently into a flask. The addition of the vial contents into a larger volume of medium has proven sufficient to dilute out the DMSO to a relatively harmless concentration. Centrifuging the cells upon thaw may be more harmful than beneficial. After 24-48 hours, the media in the flask can be changed for fresh media.

2. Using the wrong antibiotics

The only antibiotic recommended for use in non-GFP expressing cells is Puromycin at specific concentrations (see IVISbrite Tumor Cell Culture Guidelines, Table 1). Puromycin drives the expression of the luciferase gene and should be added to the complete medium at the first passage and then removed prior to cell banking.

DO NOT include Puromycin in the medium used in the thawing or freezing stages of cell culture. The use of other antibiotics such as Penicillin/Streptomycin may be common practice, but none of the IVISbrite™ cell lines were established using Pen/Strep. We do not recommend Pen/Strep usage as it has the ability to mask low-level contamination and the potential to adversely affect gene expression and cell metabolism. (Do not use any antibiotics with GFP-expressing cell lines BW128090 and BW133416 as they do not have any antibiotic selection resistance and will not survive).

3. Passaging the cells too soon

Following thaw, cells must be given enough time to recover and begin to divide. Do not re-plate or passage cells before they have reached 80% confluency. The cells will not grow well if they are disturbed too soon or plated too sparsely. Media can be replaced within the first 24-48 hours and again every 3-4 days during the incubation period until the cells reach 80% confluency at which time the cells should be passaged.

4. Using the wrong medium or adding additional components to the medium

The IVISbrite Tumor Cell Culture Guidelines specifies the correct media and media composition (Table 1), along with the suggested manufacturer (Table 2), for each cell line. If using media from a different manufacturer, make sure to verify that the media formulation is an exact match to that of the suggested manufacturer's media. Do not use a different media formulation and do not add any additional components to the media.

5. Using both vials before contacting us for assistance

As a courtesy, we provide customers with 2 vials of cells. The second vial should be kept in liquid nitrogen gas phase as a backup. If there is an issue with the first vial, DO NOT THAW the second vial; please contact the Revvity Technical Support Team at global.techsupport@revvity.com

* The only exception is IVISbrite LNCaP Red F-luc (BW125055) which requires special culture techniques. Please refer to the LNCaP Tumor Cell Culture Guidelines available on our website.

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