

Use of trypsin dry substance and trypsin/EDTA solutions from Biochrom AG

Information from Biochrom AG, December 20, 2010

An animal tissue cell group is composed of a complex matrix of proteins (such as collagen), glycoproteins, lipids, glycolipids, and mucopolysaccharides. This matrix has to be carefully digested with enzymes, such as collagenase or trypsin, without the cell surface or the inter-cellular structure being damaged irreversibly, in order to isolate single cells or produce primary cultures.

The trypsin generally used in cell culture or a trypsin/EDTA solution attacks the matrix slowly, damaging cellular as well as intercellular mucoproteins. In addition, it may cause irreversible damages to released cells. Serum may be used to inhibit trypsin. Trypsin/EDTA solutions are used in different concentrations to passage monolayer cultures.

The following includes data on the activity of trypsin dry substance, as well as notes on the different trypsin and trypsin/EDTA solutions. Before being released, trypsin dry substance is tested for mycoplasma and parvoviruses.

1 Specifications of trypsin dry substance

- storage temperature: +2 - +8 °C
- solubility: 2 % (w/v) in water
- weight loss on drying ≤ 5 %; pH value 3.0-4.0.
- specific activity range: 275 USP U/mg (+/-25)

Trypsin as dry substance (powder) normally features an activity of 1250 USP U/mg^{1,2} (cat. no.: L 2103-20G, unit 20 g). Its activity is five times higher than that of the trypsin solution offered by Biochrom AG, which is used as initial solution for further dilution processes. The activity of this initial solution corresponds to approx. 1:250 (1250:5 = 250) of the normal activity featured by the dry substance.

2 Production of solutions from the trypsin dry substance offered by Biochrom AG

Example: producing 100 ml of a 1 % trypsin solution (1:250 U/mg) from the trypsin dry substance offered by Biochrom AG (cat. no.: L 2103-20G).

1. Take 200 mg of dry substance.
2. Dissolve in 100 ml PBS without Ca²⁺, Mg²⁺ (cat. no.: L 1825).
3. Sterile filter the dissolved substance using a 0.22 µm sterile filter.

Biochrom AG offers 0.25 % as well as 2.5 % trypsin solutions from trypsin dry substance (cat. no.: L 2103-20G).

¹ USP: The United States Pharmacopeia (USP) uses the term *USP unit* instead of *international unit* (IU).

² One USP unit equates the enzyme amount that provokes the hydrolysis of BAEE (benzoyl arginine ethyl ester), causing the light absorption to increase at 253nm by 0.003 absorbance units per minute at 25 °C.

tab. 1: trypsin solutions from Biochrom AG

solution	cat. no.	unit
Trypsin (1:250), 0.25 % in PBS without Ca ²⁺ , Mg ²⁺ storage temperature: -20 °C	L 2123	100 ml
Trypsin (1:250), 2.5 % in PBS without Ca ²⁺ /Mg ²⁺ storage temperature: -20 °C	L 2133	100 ml

3 Production of trypsin/EDTA solutions from the trypsin dry substance offered by Biochrom AG

Biochrom AG's trypsin dry substance (cat. no.: L 2103-20G) is used to produce trypsin/EDTA solutions in different dilutions, trypsin dry substance and EDTA previously being diluted in PBS.

tab. 2: Trypsin/EDTA solutions from Biochrom AG

solution	cat. no.	unit
Trypsin (1:250)/EDTA (0.05 %/0.02 %) in PBS without Ca ²⁺ /Mg ²⁺ storage temperature: -20 °C	L 2143	100 ml
Trypsin (1:250)/EDTA (0.25 %/0.02 %) in PBS without Ca ²⁺ /Mg ²⁺ storage temperature: -20 °C	L 2163	100 ml
(10x) Trypsin (1:250)/EDTA (0.5 %/0.2 %) in (10x) PBS without Ca ²⁺ /Mg ²⁺ storage temperature: -20 °C	L 2153	100 ml

4 More information on all Biochrom AG enzymes

- enzymes at a glance:
<http://www.biochrom.de/en/products/enzymes/>
- collagenase:
<http://www.biochrom.de/en/products/enzymes/collagenase/>
- comparison of collagenase and trypsin features:
http://www.biochrom.de/fileadmin/user_upload/service/produktinformation/englisch/100430_h_intergrundinformation_collagenase_tabelle_en.pdf