**Spectroscopical measurements of freezing-induced acidity jump of pharmaceutical reliable buffers**

Susrisweta Behera1, Lukáš Veselý 1, Radim Štůsek 1, Thomas Loerting 2, Dominik Heger

1 Masaryk University, Kamenice 5, 62500 Brno, Czech Republic

2 Department: Institute of Physical Chemistry, University of Innsbruck, Tyrol, Austria

 Freezing biological substances is a crucial stabilization step that extends the viability and promotes manufacturing flexibility. However, the stability of the products in frozen solutions is associated with several problems including potential destabilization by the acidity changes [1]. Until now, there have been no systematic studies related to the acidity variations in buffers due to freezing, thus creating a loophole in the freeze-drying process [2].

The work explores a method that measures the ability of frozen buffer solutions to protonate the sulfonephthalein indicators which is used as a spectroscopical acid base probe [3]. The indicators are present in the freeze concentrate solution and give precise knowledge about the change in proton activity. Most commonly used pharmaceutical buffers like carboxylic acid, amino acid, amine buffer, Good’s buffer [4] and phosphate buffer were frozen and the acidity change was investigated for various concentrations, temperatures, and rates of cooling. Furthermore, differential scanning calorimetry and optical cryomicroscopy studies were used to find the crystallisation of the buffer components leading to acidity change. At last, the results obtained were compared with the results of the most commonly used low-temperature pH electrode method [5].

Keywords: Biological buffers, cryopreservation, freeze-induced acidity change, freezing

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