ANALYSIS OF LYOPHILIZED BLOOD PRODUCT PROTEINS USED AS GAS MASK ABSORBENTS THROUGH THE PROCESS OF ELECTROPHORESIS

Veselin Ivanov*, Svetla Dyankova, Rosica Ruseva***

** Trakiya University, Medical Faculty, Stara Zagora, Bulgaria*
*** Institute of Cryobiology and Food Technology, Sofia, Bulgaria*
Correspondence to: Veselin Ivanov, Department of Chemistry and Biochemistry, Medical faculty, Trakia University - Stara Zagora, Bulgaria,
veskoasenov@abv.bg

ABSTRACT

Lyophilized blood products can have many different applications. Their properties, however, change in the course of time. After the process of lyophilisation certain amounts of water remain in them which can sometimes lead to hydrolysis. The purpose of the present study is to analyze the proteins of lyophilized sheep blood plasma and lyophilized sheep erythrocyte mass which were produced in 2002. The aim of the study is to analyze the product changes which have occurred due to time exposure. These products have been tested as gas mask absorbents through electrophoresis in poliacrilamid gel. The results are presented in table 1. The protein compound of the lyophilized blood plasma, stored for 9 years at room temperature, is very well preserved.

Key words: *gas mask, chemical warfare agents, chemical disaster*

INTRODUCTION

Gas masks were first used during the First World War to protect soldiers against poisonous gases. Apart from that they have been used in dealing with the aftermath of industrial accidents, fires and natural disasters. The importance of gas masks has recently increased with the occurrence of terrorist attacks. Problems with the use of masks are discussed widely in the training of medical students in several disciplines (1). It has been shown that lyophilized blood products have high sorption capacity for toxic gases and toxic compound vapours. (2-5). It can be thus concluded that they can be used as absorbents for gas masks. The purpose of this study is to find out if it is possible for lyophilized blood products to be stored for a long time while preserving their properties.

MATERIALS AND METHODS

Materials: Defibrinized sheep blood (BulBio Institute of Communicable and Parasitic Diseases, Sofia); lyophilized sheep blood plasma and erythrocyte mass, produced on 01.06.2002 (BulBio Institute of Communicable and Parasitic Diseases, Sofia,), lyophilized bovine albumin (Fluka). When opening the samples the buffer Tris-glycine, pH 8,3, which contains 10% glycine; 1% mercaptoethanol; 1 % SDS and 0,01% bromophenol blue was used. Protein marker LMW-SDS Calibration Kit, produced by Amersham Bioscences (molecule mass от 97 kD до 14 kD).

Method: Electrophoresis in poliacrilamid gel (by Laemmli, U.K.,1970) (9).

Blood samples are put in test tubes and centrifuged for 10 minutes at 5000 rpm and temperature of + 4°C. 100 μl of the supernatant are solved in 2 ml of buffer until reaching concentration of 1,5 mg/ml. The samples are then thermally denaturized and stored in a refrigerator until electrophoresis occurs.

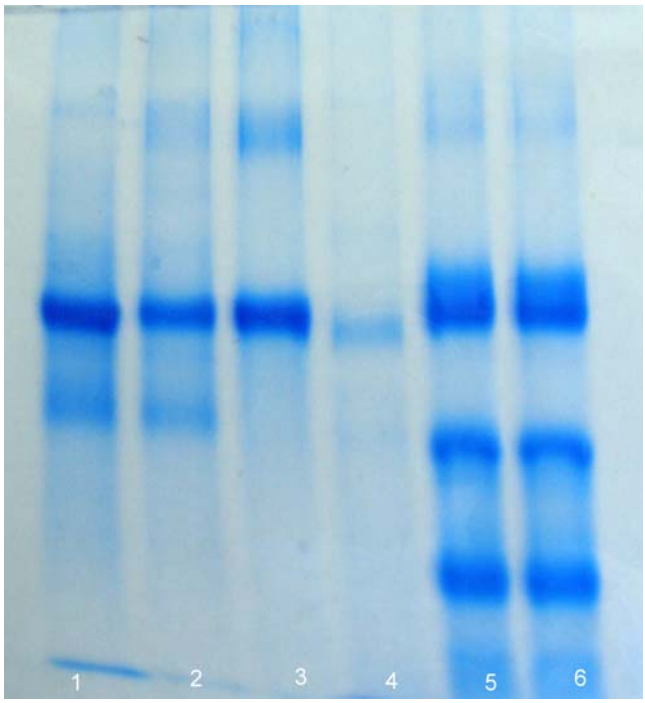
Electrophoresis in poliacrilamid gel by Laemmli (1970) and O'Farrell's (1975) method was applied.

Electrophoresis was performed under the following conditions: concentration of the cumulative and separation gel 6 % and 10 % respectively; electricity power сила – 25mA; duration – 4h. Electrode buffer solution - 0.025 M Tris, 0.192 M Glycine and 0.1% SDS; pH 8.3. 20 μl of

each sample and 10 μl of protein marker were used. Protein fractions were colored with Commassie Brilliant Blue R-250.

RESULTS AND DISCUSSION

The results are presented in picture 1



Picture 1. SDS-PAGE of blood products sample: 1. defibrinized sheep blood; 2. lyophilised sheep plasma (stored for 9 years); 3. lyophilized bovine albumin; 4. lyophilized sheep erythrocyte mass (stored for 9 years); 5 and 6 – protein markers

It is clearly seen from figure 1 that the electrophoretic profile of sample 2 is identical with that of sample 1, i.e. lyophilized plasma has the same protein fractions. The molecule masses of fraction proteins were calculated by comparing their electrophoretic mobility with the electrophoretic mobility of protein markers. The most intensive stripe refers to serum albumin (sample 3) – MW 66 kDa. The molecule mass of the other clearly distinguishable protein fraction was calculated to be 52 kDa. Under electrophoresis, a light, not clearly distinguishable stripe appeared in sample 4, corresponding to traces of serum albumin.

CONCLUSIONS:

According to existing literature, due to low humidity (1-6%) (6) in lyophilized blood products, stored in air-tight, gas and vapour-proof packaging, these products can be preserved at room temperature for 2 to 5 years, without changing their properties (8). After being electrophorized, the results confirm that the protein compound of the lyophilised blood plasma, stored for 9 years at room temperature, is very well preserved. In comparison the shelf life of active carbon sorbent is 5 years (7).

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