Physiol. Res. 50: 433-437, 2001

Correlation Between the Blood Surface Tension and the Activity of Some Enzymes

A. KRATOCHVÍL, E. HRNČÍŘ

Clinic of Occupational Diseases, Third Faculty of Medicine, Charles University, Prague, Czech Republic

Received May 22, 2000
Accepted January 3, 2001

Summary

The aim of this study was to find the principal parameters of surface tension of the blood and verify its possible correlation with some of the commonly assessed laboratory indicators that are affected by diseases with the expected possible changes in the surface tension of the blood. The surface tension of the blood was assessed in 150 patients. At the same time, the basic biochemical and hematological parameters were determined in these patients. The results have shown a close correlation between the surface blood tension and the activity of plasma gamma-glutamyltransferase. We assume that this relationship is based on the changes in concentration of bile acids in the blood during liver or bile duct afflictions.

Key words
Surface tension • Blood • Assessment • Correlation

Introduction

The assessment of surface tension of body fluids in medicine has so far been associated mainly with the determination of surface tension of the tracheal aspirate in early diagnostics of the respiratory distress syndrome. Surface tension correlates with the amount of surfactant present in the tracheal aspirate (Friedrich *et al.* 1996). Reports on assessing the surface tension of the blood have started to appear in large numbers only recently. The first results have already indicated that the level of surface tension of the blood marks changes occurring in various disease conditions such as rheumatoid arthritis, proliferative glomerulonephritis, some tumors of the central nervous system or the urogenital tract (Kratochvíl and Hrnčíř 1999). Higher average values of surface tension of the blood serum in women have been reported to reflect the differences of some enzymes, metabolites, hormones etc. in the serum. In the same way, these can also explain the higher values of surface tension of the blood in the elderly. However, both above mentioned relationships were not present when non-coagulated blood was used as the substrate (Siniachenko *et al.* 1998, Hrnčíř and Rosina 1997).

In the available literature, however, no data have been described concerning the level of surface tension of the blood and its interindividual variability from a more representative population sample. A pilot study conducted in 1996 did not reveal any statistically significant correlation between basic biochemical and hematological indicators and surface tension of the blood (red cell sedimentation rate, blood hemoglobin levels, number of erythrocytes and leukocytes, total serum proteins, total serum cholesterol, total serum

Fax+4202 24920590
E-mail physres@biomed.cas.cz http://www.biomed.cas.cz/physiolres

triacylglycerols, creatinine blood levels, ALT and AST activity). As mentioned above, no significant dependence on the age and sex was found (Hrnčíř and Rosina 1997).

The goal of the present study was to verify the relation between the surface tension of the blood and some laboratory indicators in diseases, where one can assume a change of surface blood tension and to verify this by improving the methodology of measuring the surface tension of the blood and expanding the original group.

Surface tension, defined as the energy required to increase the surface area of a liquid by a unit of energy depends upon the fact that molecules on the surface of the liquid are not completely surrounded by other molecules of the liquid. This causes an imbalance of force. The active surface agent also known as surfactant, or detergent, is then a soluble compound that reduces the surface tension of liquids, or reduces the interfacial tension between two liquids or between a liquid and a solid (Havránek 1967).

Surface tension is an easily and rapidly measurable physical quantity. It can be assessed effectively by means of diverse procedures, particularly by the following methods:

1. Tearing off method, which enables to measure the surface tension of a liquid by determining the force F needed for tearing off a wire or a ring that is soaked in a given liquid (Wilhemy′s method using a platinum wire or plate or Du Nouy′s method using a platinum or irridium ring).
2. Drop-weight method (see below in Methods)
3. Method of capillary elevation, which is based on the degree of liquid elevation in a capillary depending upon the surface tension of this liquid.
4. Computer-assisted methods. These are based on surface tension evaluation using Laplace´s law. Tensiometer measures the pressure inside an air bubble formed in a liquid by a capillary. During oscillations of its average value, tensiometer readings depend on the surface tension of a liquid (Havránek 1967, Enhorning 1977, Cole-Parmer Instrument Co. 1999).

Material and Methods

For measuring the surface tension of blood, we chose the drop-weight method. This makes it possible to assess surface tension effectively with a relatively simple equipment. The principle of this method is based on the dependence of surface tension levels on the weight of a drop of the measured liquid sheared off from a capillary. It is possible to determine the relevant level of surface tension by comparing the weight of the drop with a standard, which in our case is distilled water ($\sigma = 72.45 \times 10^{-3}$ N.m^{-1} at 22 ^{o}C) (Havránek 1967). Surface tension is a unit dependent on the temperature. There is a difference in distilled water of approximately 2.3 % over a temperature gradient of 10 ^{o}C. Therefore, the surface tension of distilled water was corrected according to the actual temperature.

Within the framework of this study, the surface tension of 150 samples of citrate blood was assessed (72 men and 78 women aged 20 to 65 years). They belonged to a group of patients who were examined for a preventive check-up. At the same time, their basic biochemical and hematological parameters were assessed (levels of urea, creatinine, uric acid, bilirubin, cholesterol, triacylglycerol, glucose, total proteins, AST, ALT, GMT activity in the blood, red cell sedimentation rate, blood count and differential count of leukocytes, chemical examination of urine and microscopic examination of the urinary sediment). The above mentioned parameters were determined in order to distinguish the groups of “healthy patients” and to verify the possible correlation of some of these parameters (see below) with the surface tension of the blood.

A correlation of the surface blood tension values with total serum cholesterol, triacylglycerol levels, creatinine, red cell sedimentation rate, number of leukocytes or total serum proteins was not found (see above). The possible dependence between the surface tension of blood and levels of urea, uric acid and glucose cannot be established and there is no report concerning these relationships in the available literature. Surface tension was measured three times for each sample of citrate blood and the arithmetical average of measured levels was considered to be the definite level. For weight assessment, we used the Electronic Balances Ohaus CT 10 with readability 0.002 g. The accuracy correctness of this method was verified by measuring the surface tension of liquids the surface tension of which is known.

After assessing how the presence of an anticoagulant agent affects the surface tension of blood in the final sample, basic statistical parameters of the surface tension of blood were determined in samples obtained from all subjects and in samples obtained from subjects with no detectable laboratory abnormalities. In the next stage, we raised the question concerning the possible existence of correlation between the surface

tension of the blood and the commonly observed laboratory levels. We mainly investigated those parameters for which it was theoretically possible to assume that their changes could be connected with a shift of the surface tension of blood levels. We hypothesized that the surface tension of the blood might correlate with those laboratory abnormalities that are usually accompanied by changes in bile acid blood levels. To confirm this hypothesis, we were looking for a correlation between the surface tension of blood and the following parameters: level of total bilirubin in the blood, activity of alanine aminotransferase (ALT), aspartate aminotransferase (AST) and gamma-glutamyltransferase (GMT, GGT). The correlation between the surface blood tension and the aforementioned parameters was assessed in case that the upper limit of their chart levels was exceeded (for the level of total bilirubin in blood above 20μmol/l, for the activity of alanine aminotransferase above 0.55 μkat/l, aspartat aminotransferase above 0.5 μkat/l and gamma-glutamyltransferase above 0.6 μkat/l).

The data were statistically processed by the method of selective Spearman′s coefficient of serial correlation and the relevant coefficient of determination. Values of $\alpha<0.01$ were considered to be statistically significant (Strnad 1968, Kubánková and Hendl 1986).

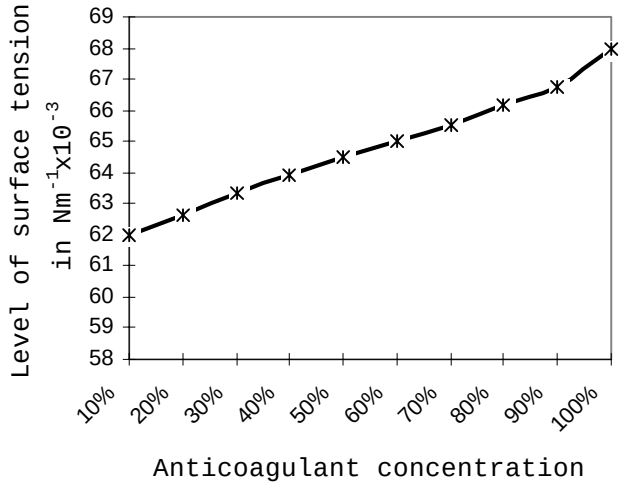

Fig. 1. *The linear dependence of the surface tension of blood level on the concentration of anticoagulant agent – sodium citrate.*

Results

After evaluating basic biochemical and hematological parameters, it was found that all the aforementioned parameters were normal in 41 patients (19 men and 22 women). Some differences were detected in case of the remaining 109 patients (53 men and 56 women). The following exceptions of laboratory reference values were found (the upper limit of their chart levels was exceeded): red cell sedimentation rate in 33 cases, total serum cholesterol levels in 24 cases, the level of glucose in 9 cases, the level of ALT in 15 cases, the level of AST in 13 cases, the level of GMT in 13 cases, the level of total serum proteins in 8 cases, the level of uric acid in 6 cases, urine and urinary sediment abnormalities in 6 cases, the number of leukocytes in 6 cases, triacylglycerol levels in 4 cases and levels of urea and creatinine in 3 cases.

When evaluating the surface tension of blood, it is necessary to consider the fact that the substrate is always blood with anticoagulant agent. In our case, it was 3.8 % sodium citrate, forming a standard 25 % in each of the measured samples. The chart below illustrates the surface tension dependence of full blood and anticoagulant agent mixture in different substitutions. (Fig. 1) The dependence is linear. Each increase of anticoagulant substitution by 10 % in the final sample, surface tension increases by $0.6\text{x}10^{-3}$ N.m^{-1}.

Below illustrated values of the surface tension of non-coagulating blood are presented approximated for full blood (free of an anticoagulant agent). The arithmetical average of the surface tension of blood level, obtained by means of data processing of all observed subjects ($n = 150$), is $\sigma = 64.33\text{x}10^{-3}$ Nm^{-1} by 22 ^{0}C, standard difference $\sigma = 1.35$ by dispersion $s^2 = 1.82$ and by the variation coefficient $V = 2.1$. The arithmetical

average of the surface tension of the blood, obtained by means of data processing of all subjects with no detected laboratory abnormalities (n = 41) is $\sigma = 64.60 \times 10^{-3}$ Nm^{-1} by 22 ^{0}C, standard difference $\sigma = 1.69$ by dispersion $s^2 = 1.30$ and the variation coefficient V = 2.01. The histogram below demonstrates the distribution of blood surface tension in healthy population and in population with some abnormalities in the given parameters. (Fig. 2)

Furthermore, correlation between the surface tension of blood and the activity ALT, AST, GMT in blood or the level of total bilirubin in blood was assessed using the selective Spearman's coefficient of serial correlation and the relevant coefficient of determination (Table 1). The above mentioned results show that a statistical correlation only exist between the surface tension of blood and the activity of gamma-glutamyltransferase.

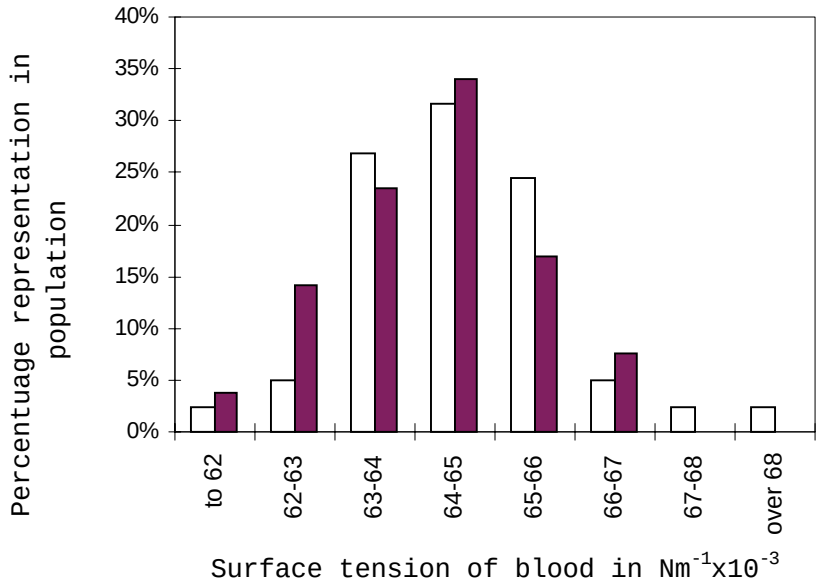

Fig. 2. *Histogram showing the frequency of distribution of the surface tension of blood levels: blood samples of patients with no detectable laboratory abnormalities in basic biochemical and hematological parameters (n = 41, 19 men and 22 women, open columns), blood samples of patients with some abnormalities in determined parameters (n = 109, 53 men and 56 women, full columns).*

Table 1. Statistical evaluation of correlated data (samples taken from group n=53)

Correlated levels	**Number of samples (n)**	**Spearman coefficient (ρ_s)**	**Statistical significance (p)**	**Coefficient of determination (r^2)**
σ/ALT	15	– 0.279	> 0.05	0.078
σ/AST	13	0	> 0.05	0
σ/GMT	13	– 0.835	< 0.01	0.697
σ/bilirubin	12	– 0.490	> 0.05	0.240

Discussion

The surface tension of blood is not a currently assessed parameter. Although first reports on changes in the surface tension of the blood have already appeared in literature concerning the development of some pathological conditions, there is still a need for an extended report providing data about the surface tension of the blood and its distribution in the population that would serve as a material for further research (Kazakov *et al.* 1998, Kratochvíl and Hrnčíř 1999). To our knowledge, there was only one such description, but the determinations were only carried out in a small group of patients (n=29) and with no information about the temperature. With the use of the drop method, the average level of the blood surface tension was 63.8×10^{-3} Nm^{-1}, which is slightly lower than that found by us (Hjelde *et al.* 1994). As mentioned above, for the interpretation and comparison of data it is important to respect the modification of data by the anticoagulant

agent. It is also important to evaluate the results in relation to the native sample.

The fact that blood exhibits lower surface tension values than water is caused by the presence of substances with active surface (detergent) effects (Hrnčíř and Rosina 1997). To clarify the correlation of different quantities others than blood GMT activity with the surface tension of the blood, it is necessary to process a larger number of samples. The correlation we detected between the vaues of blood surface tension and the activity of plasma gamma-glutamyltransferase thus reflects changes in a substitution of surface active substances in the blood. In our opinion, this might be caused by a greater concentration of bile acids in the blood accompanied by an increased activity of gammaglutamyltransferase during liver or bile duct afflictions, which is the subject of a future research.

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Reprint requests

Doc. MUDr. Evžen Hrnčíř, CSc., Clinic of Occupational Diseases, Third Faculty of Medicine, Charles University, Šrobárova 50, 100 34 Prague 10, Czech Republic.