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UNIFICATION OF SOME BIOCHEMICAL METHODS OF RESEARCH IN THE PRE- AND POST-FLIGHT PERIODS

R. A. Tigranyan

Translation of "Unifikatsiya nekotorykh biokhimicheskikh metodov issledovaniy v pred- i poslepoletnom periode", Institute of Medical and Biological Problems, Ministry of Health USSR, Moscow (Report presented at the 11th Joint Soviet-American Working Group on Space Biology and Medicine, Moscow, October 1980), 1980, pp. 1-7

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16. Abstract

The report compares and discusses the unification of the biochemical methods of determination of various parameters and factors during pre- and post-flight periods, as used by American and Soviet teams dealing with space space medicine. Specifically, the discussion centers on the exchange of information on the study of the blood and urine content of space travelers before and after space flight. A series of electrolytic, enzymatic, and hormonal factors are discussed and compared.

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UNIFICATION OF SOME BIOCHEMICAL METHODS OF RESEARCH IN THE PRE- AND POST-FLIGHT PERIODS

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A resolution was adopted at the Third Congress of the Joint Soviet-American Working Group on Space Biology and Medicine (Moscow, February 26-March 3, 1973) on the exchange of information for purposes of the development of unified procedures of pre- and post-flight biochemical studies of the blood and urine of cosmonauts. /1*

During the past period of time, recommendations have been discussed and adopted on the conduct of biochemical studies of the blood and urine. According to these recommendations, the biochemical studies are carried out for the evaluation of the state of health and changes in metabolism of the body of cosmonauts, and for the obtaining of information on the effect of factors of space flight on the body. The conditions and procedure for carrying out the studies are as follows.

1. As a rule, all blood analyses are done in the morning after urination and before eating; samples of the peripheral blood are taking under sterile conditions with a single puncture of the vein.

2. Before taking of blood, it is not permissible to undertake heavy physical exercises.

3. The blood samples should be taken no earlier than 12 hours after an orthostatic test, or a test with the effect of ODNT[1].

4. The urine is collected in the course of a day and its volume is measured, after which a certain amount of urine is utilized for analysis. Fractional collection of the urine, and subsequent analysis

[1]ODNT-- expansion unknown

*Numbers in the margin indicate pagination in the foreign text.

of each portion, is carried out during certain periods of the investigation.

5. It is necessary to record the conditions under which the cosmonauts are found, and to establish their activity, as well as any symptoms of diseases, use of medicinal substances, and any effect of toxic products. /2

6. It is desirable that the results of the analysis of each urine fraction be compared with the work or load tests which the cosmonauts are undergoing at this time.

7. Nutrition during the biochemical investigation should be of full value and rational, and should completely cover energy expenditures.

Concurrence was achieved on the list of recorded parameters, which included the following biochemical indicators, determined in the blood and urine:

1. Sodium
2. Potassium
3. Calcium
4. Magnesium
5. Chlorides
6. Inorganic phosphor us
7. Glucose
8. Urea
9. General protein
10. Electrophoresis of protein
11. Creatinine
12. Cholesterol
13. Ur^ic acid
14. General and direct bilirubin
15. Alkaline phosphatase
16. Creatinphosphokinase
17. Glutamateoxalate-transaminase /3

18. Glutamatepyruvate-transaminase
19. Lactatedehydrogenase and its isoenzymes

It was agreed that the pre-flight biochemical investigations should be carried out within the limits of 30 days prior to the beginning of the flight; the post-flight investigations are carried out as soon as possible after touchdown (within 24 hours).

Subsequently, the two sides exchanged materials on procedures of biochemical determinations.

Carried out first w - the exchange of materials on procedures for determining the content of electrolytes (sodium, potassium, calcium, magnesium, chlorides and inorganic phosphorous) in the blood and urine. These materials also included the setting forth of procedures for the taking of blood samples and the daily collection of urine.

The procedure for taking blood samples was as follows. The blood was taken from a well-pronounced ulnar vein in a lying down position, utilizing a 20 or 30 milliliter syringe with a needle of sufficiently wide opening; the syringe was removed and the blood went directly into a clean dry test tube from the needle. The test tube with the whole blood remained at room temperature for a period of 15-30 minutes, and the clot was carefully enclosed with a stick and centrifuged to separate the serum and cellular elements, after which the serum was transferred to a clean test tube and covered. If rapid analysis was possible, then, in this case, the samples were stored at a temperature of +4° C, while if storage of the samples was required, they were placed in dry ice and subsequently stored at a temperature of -50-60° C. Right before analysis, all of the frozen samples had to be thawed, carefully mixed, and then /4
centrifuged to separate the fibrin, and the serum supernatant had to be transferred to a clean test tube for subsequent analysis. In the case of obtaining of blood plasma samples, the test tubes in which the blood was collected contained the appropriate anticoagulant.

The procedure of daily urine collection consists of the following. Right before urine collection, the bladder is emptied, and this portion of the urine is discarded. In the period of the subsequent 24-hour collection, all of the urine portions are collected, including that portion of the urine which is contained in the bladder by the end of the collection period. All of the urine portions are stored at a temperature of +4-8° C throughout the entire collection period, and are not preserved. After the completion of the daily collection, all of the collected urine portions must be well mixed and carefully measured, which is quite significant for the conduct of subsequent calculations. The collected samples from each daily urine portion are placed in containers and transported to the laboratory. Storage of the urine samples is accomplished at a temperature of -20° C and below. Right before analysis, all of the frozen samples are thawed at room temperature, mixed well and centrifuged to separate the precipitates; the supernatant is analyzed.

The examination of the materials according to the procedures for determining the content of electrolytes in the blood and urine showed that both sides make use of practically identical methods, utilizing the method of plasma emission spectrophotometry for the determination of sodium and potassium, the method of atomic-absorption spectrophotometry for the determination of magnesium and calcium, the colorimetric automatic method for the determination of phosphorous, and the method of amperometric titration for the determination of chlorides. All of this makes it possible to think that the procedures for determining the content of electrolytes (sodium, potassium, calcium, magnesium, chlorides and inorganic phosphorous) in the blood and urine may be considered unified. /5

Subsequently carried out was the exchange of materials on the procedures of determination of a number of biochemical indicators (glucose, urea, ureic acid, general protein, protein electrophoresis, creatinine, cholesterol, general and direct bilirubin) in the blood and urine. The examination of these materials also showed that both sides make use of practically identical methods, utilizing automatic analysis on the "Tekhnikon" analyzer for the determination of the

content of glucose, urea, ureic acid, creatinine, cholesterol, and general and direct bilirubin, and the method of separation on cellulose-acetate strips with subsequent densitometry for the determination of protein electrophoresis. For the determination of the content of general protein in the blood serum, the Soviet side utilizes the automatic method on the "Tekhnikon" analyzer; this method is based on the biuret reaction. The American side utilizes a special Bauch and Lomb protein meter for the determination of the content of general protein in the blood plasma. These methods give results which coincide well. Comparison of the procedures of biochemical determination enumerated above makes it possible to conclude that the methods for determination of the content of glucose, urea, ureic acid, general protein and protein fractions, creatinine, cholesterol and general and direct bilirubin in the blood and urine may be considered unified.

Also carried out was the exchange of materials on the procedures for determination of the activity of a number of enzymes (alkaline phosphatase, creatinphosphokinase, glutamateoxalate-transaminase, glutamatepyruvate-transaminase, lactatedehydrogenase and its isoenzymes) in the blood serum. Analysis of these materials showed that both sides make use of identical methods, utilizing the method /6 of automatic analysis on the "Tekhnikon" analyzer for the determination of the activity of alkaline phosphatase, the method of enzyme spectrophotometry at 340 nm for the determination of the activity of creatinphosphokinase, glutamateoxalate-transaminase, glutamate-pyruvate-transaminase and lactate dehydrogenase, and the method of electrophoresis on cellulose-acetate strips with subsequent densitometry for the determination of isoenzymes of lactatedehydrogenase, i.e., the procedures for the determination of the activity of the indicated enzymes in the blood serum may also be considered unified.

The placement of man under conditions of space flight is accompanied by functional changes in a number of organs and systems. As is common knowledge, the neuroendocrinal system plays a leading role in the formation of the complex of adaptational-homeostatic

reactions to the effect of space flight, and subsequent readaptation to conditions of the earth's gravitation. In this connection, the study of the content of a number of hormonal and biologically-active compounds in the blood and urine in cosmonauts is quite important and urgent for a deep understanding of the changes taking place in the body.

It is proposed to include the following in the list of recorded biochemical parameters, and to carry out exchange of materials on the following procedures of determination of the content of hormones and bioactive compounds in the blood and urine by July 1, 1981:

1. Adrenocorticotropic hormone (ACTH)
2. Somatotropic hormone (STH)
3. Parathyroid hormone (PTH)
4. Thyrotropic hormone (TTH)
5. Follicle stimulating hormone (FSH) /7
6. Luteinizing hormone (IH)
7. Hydrocortisone
8. Insulin
9. Glucagon
10. Prolactin
11. Testosterone
12. Aldosterone
13. Angiotensin I
14. Thyroxin (T_4)
15. Triiodothyronine (T_3)
16. Prostaglandine
17. Cyclic adenosine monophosphate
18. Cyclic guanosinemonophosphoric acid

These materials should include:
-description of the principle
-detailed description of the procedure
-norm boundaries
-accuracy of method
-literature