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# Assessment of the rate of human aging by clinical biochemical tests

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**Abstract**. The aim of the study was the development of the accessible method for assessing the rate of human aging by laboratory biochemical parameters. There were examined 408 practically healthy people in the age from 20 to 80 years. There were determined 6 anthropometric and 14 laboratory biochemical parameters, characterizing carbohydrate and fat metabolism, liver and kidney functions. The use of stepwise multiple regression made it possible to select the most informative indicators and obtain an equation linking the age of the examined people with a number of anthropometric and metabolic indicators. The average absolute error in calculating age was 4.2 years. The method for assessing the rate of aging, developed by us, is highly accurate and can be used to assess the risk of developing of the age-dependent pathology.

**Key worlds:** biological age; metabolic biomarkers of aging

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The determination of the biological age (BA) is a technology (tool) for the assessing of the degree of development of age - dependent changes, the rate of aging and the diagnosis of accelerated aging [1-4]. BA can either coincide with chronological age (CA), and in that case the aging process is assessed as normal, physiological, or significantly exceed CA [5]. In the latter case, the aging is assessed as accelerated. It is known, that in the process of aging such changes in organs and systems occur that make a contribution to the development of a number of diseases, primarily cardiovascular diseases. Rapidly aging people have a high risk of developing of the age-associated pathology [6-9]. They need a lifestyle and nutrition correction, refusal of the bad habits and periodic medical supervision at the health center. This will slow down the rate of aging and reduce the risk of developing age- dependent pathology.

The calculation of BA in most of the works is based on the measurement of quantitative indicators that are called biological markers of aging. These indicators should correlate with age, be available for measurement in a clinical setting, and be dependent not much on the actual pathological conditions. A variety of indicators can act as biological markers: anatomical, functional, biochemical, immunological, genetic, and epigenetic [9-11].

The generally accepted mathematical method for assessing BA is the calculation of the equation of the multiple regression linking CA and a number of quantitative indicators [12-13]. For the determination of the coefficients of this equation, the survey data of a large number of practically healthy people of different ages are used. To determine the BA of the subject, a number of studies are carried out for him and his age is calculated in the accordance with the obtained formula. If the estimated age significantly exceeds CA, a conclusion is made about the accelerated aging of this person.

**Materials and methods**

The study included 408 practically healthy people aged from 20 to 80 years, who were examined at the Department of Clinical Physiology and Pathology of Internal Organs of the State Institution «D.F. Chebotarev Institute of Gerontology of the National Academy of Medical Sciences of Ukraine». People with the pathologies of the cardiovascular, respiratory, endocrine and central nervous systems, chronic liver and kidney diseases, and pathologies of the hematopoietic system were excluded from the analysis.

There were determined 6 anthropometric and 14 laboratory biochemical parameters characterizing carbohydrate and fat metabolism, liver and kidney functions.

Anthropometric measurements included determination of body weight (in kg), height (in cm), body mass index (BMI, in kg / m2), waist circumference (WC, in cm), hip circumference (HC, in cm), correlation WC/HC.

Body weight was measured with the help of stationary balance with an accuracy of 0.1 kg among the persons wearing light clothes without shoes. Height was measured with an accuracy of 0.5 cm among the persons without shoes. Body mass index was calculated as the ratio of body weight (in kilograms) to height (in meters squared).

Waist circumference was measured with a measuring tape among the persons in a standing position without clothes, at the waist after exhalation - in the middle between the back part of the lateral costal arch and the crest of the pelvic bone (WHO standard). The hips circumference was measured at the level of the large trochanter of the femur. Also the index of the ratio of the circumference of the waist to the circumference of the hips (CW/CH) was calculated. The value of CW/ CH for men> 1.0 and women> 0.85 indicates an abdominal type of obesity.

Biochemical parameters of carbohydrate metabolism included the determination of blood plasma glucose level by the glucose oxidase method. For the identifying of the latent disorders of carbohydrate metabolism, a standard oral glucose tolerance test was performed in accordance with the WHO methodology (WHO Consultation, 1999) and the American Association of studying of Diabetes (American Diabetes Association / ADA, 1997) [15].

For the assessment of lipid metabolism, the levels of total cholesterol TC, high density lipoprotein cholesterol (HDL cholesterol), low density lipoprotein cholesterol (LDL cholesterol), very low density lipoprotein cholesterol (VLDL cholesterol) and triglycerides (TG) in serum were determined. The levels of TC and TG were investigated by the enzymatic-colorimetric method using BIO SYSTEMS reagents, HDL cholesterol - by the method of precipitation with phosphoric-tungstic acid. The calculation of the indicators of LDL cholesterol and VLDL cholesterol was carried out in accordance with the accepted mathematical formulas.

All biochemical parameters, including indicators of liver function (alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase) and kidney (creatinine, urea) were determined on an automatic biochemical analyzer "BM Autolab PM 4000/3" manufactured by "Boehringer Mannheim" in the laboratory of the clinic of the Institute of Gerontology.

Statistical processing of the obtained data was carried out with the help of Excel 2007 and Statistica 7 programs. Standard statistical procedures, including variation and regression analyses, were used.

**Results and discussion**

The preliminary stage in calculating of the formula of biological age was the analysis of the dependence of the studied parameters on the age of the examined people. The table shows the correlation of the coefficients of the studied indicators with the age (R).

Table

**Correlations with Age**

| **Biological markers** | **R** |
|---|---|
| *Anthropometric indicators* | |
| Weight, | **0.12** |
| Growth | **- 0.30** |
| BMI, kg/m$^2$ | **0.33** |
| Waist, sm | **0.34** |
| Hips, sm | **0.24** |
| Waist/Hips | **0.40** |
| *Laboratory indicators* | |
| Blood glucose, mmol/l | **0.30** |
| Glucose 2 hours (GTT), mmol/l | **0.36** |
| Cholesterol, blood chemistry, mmol/l | **0.18** |
| Triglycerides, mmol/l | **0.24** |
| HDL cholesterol, mmol/l | - 0.06 |
| LDL cholesterol, mmol/l | **0.11** |
| VLDL cholesterol, mmol/l | **0.25** |
| Atherogenic index | **0.14** |
| Urea, mmol/l | **0.24** |
| Creatinine, mmol/l | **0.11** |
| Uric acid, mmol/l | **0.20** |
| ALT, u/l | **- 0.11** |
| AST, u/l | - 0.06 |
| Alkaline phosphatase, u/l | **0.41** |

**Note:** Marked correlations are significant at $p < 0.05$

The use of stepwise multiple regression made it possible to select the most informative indicators and obtain an equation linking the age of the examined people with a number of anthropometric and metabolic indicators ($r = 0.62$; $p < 0.0001$).

$$Y = 0.55\ X1 + 39.2\ X2 + 3.37\ X3 + 4.44\ X4 + 1.40\ X5 - 0.27\ X6 + 0.13\ X7 - 30.0$$

Y – Predicted age, years
X1 – BMI, kg/m$^2$;
X2 – Waist/Hips;
X3 – Glucose 2 hours (GTT), mmol/l;
X4 – VLDL cholesterol, mmol/l;
X5 – Urea, mmol/l;
X6 – ALT, u/l;
X7 – Alkaline phosphatase, u/l.

The systematic error in calculating of the age, associated with the peculiarities of constructing of the multiple regression equation is calculated using the regression equation: prognosticated age - chronological age [16]. For our data, this error is calculated by the formula:

$$\text{Age calculation error} = 38.88 - 0.646\ CA.$$

In turn, BA is calculated as the difference between the predicted age (Y) and the error in its calculation.

$$BA = Y - \text{Age calculation error}.$$

The figure shows a graph of the correlation between BA after error correction and CA. It can be seen that the dispersion of points around the regression line is small and the multiple correlation coefficient is high ($r = 0.902$; $p < 0.00001$).

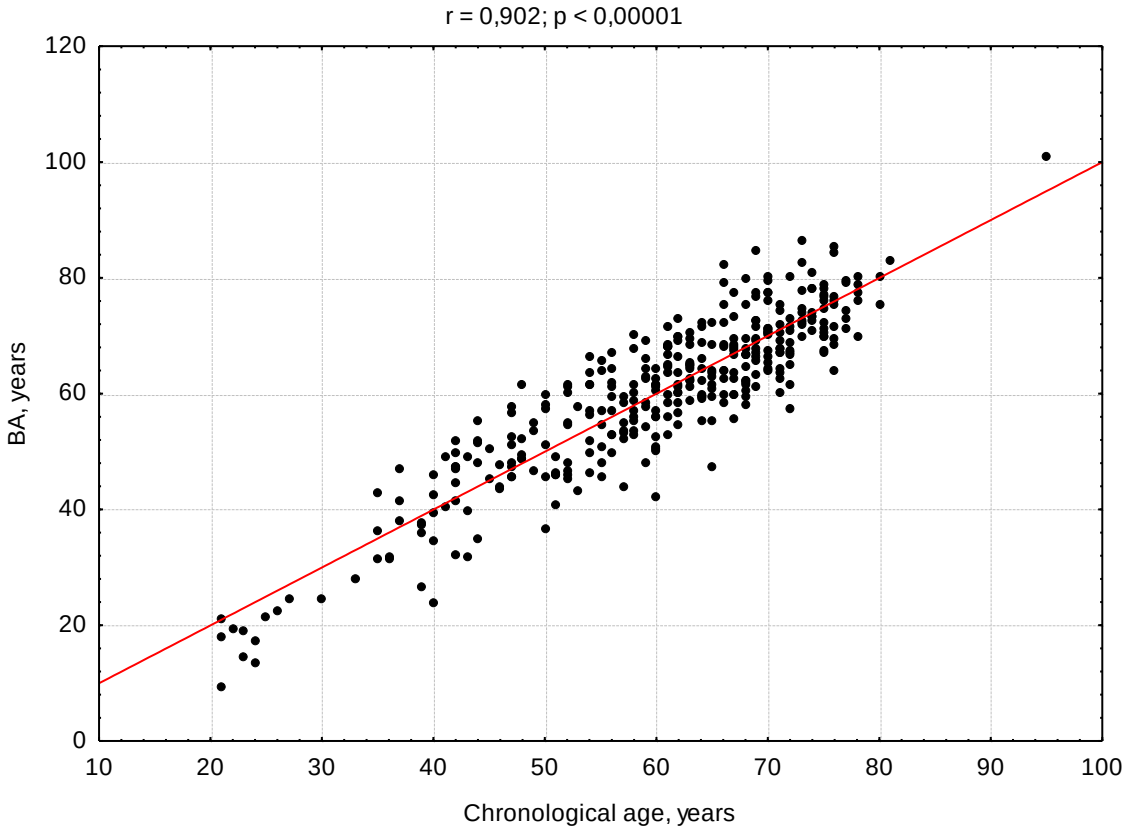


**Figure.** Correlation between biological and chronological age of people.

The average absolute value of the error of BA calculation, in this case, is 4.2 years. If we consider, as it is used to believe, people with accelerated aging, whose BA exceeds CA by 10 years, then the proportion of such people among the surveyed people is 9.1%.

Thus, the method for assessing the rate of aging developed by us, has a high accuracy and can be used to assess the risk of developing age-dependent pathology, primarily, cardiovascular and diabetes mellitus of the second type. Simple anthropometric measurements and laboratory tests used in clinical laboratories are sufficient for the use of this method. The implementation of the proposed method will allow not only to identify people with the risk of developing pathology, but also to assess the effectiveness of treatment, prophylactic and rehabilitation measures.

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