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SCIENTIFIC ACHIEVEMENTS OF MODERN SOCIETY



**ABSTRACTS OF VIII INTERNATIONAL
SCIENTIFIC AND PRACTICAL CONFERENCE
APRIL 1-3, 2020**

**LIVERPOOL
2020**

SCIENTIFIC ACHIEVEMENTS OF MODERN SOCIETY

Abstracts of VIII International Scientific and Practical Conference
Liverpool, United Kingdom
1-3 April 2020

**Liverpool, United Kingdom
2020**

UDC 001.1
BBK 83

The 8th International scientific and practical conference “Scientific achievements of modern society” (April 1-3, 2020) Cognum Publishing House, Liverpool, United Kingdom. 2020. 912 p.

ISBN 978-92-9472-193-8

The recommended citation for this publication is:

Ivanov I. Analysis of the phaunistic composition of Ukraine // Scientific achievements of modern society. Abstracts of the 8th International scientific and practical conference. Cognum Publishing House. Liverpool, United Kingdom. 2020. Pp. 21-27. URL: <http://sci-conf.com.ua>.

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TABLE OF CONTENTS

1.	<i>Bugaieva L., Beznosyk Yu.</i> AN INTELLIGENT SYSTEM FOR CHOOSING A METHOD OF GAS PURIFICATION.	15
2.	<i>Dolgiy G., Maslak H., Tsokur N., Chernenko G.</i> FIBRONECTIN ON THE SURFACE AND INSIDE THE LEUKOCYTES IN LIVER FIBROSIS.	24
3.	<i>Dmytriieva O. I.</i> INNOVATIVE STRATEGY OF DEVELOPING TRANSPORT INFRASTRUCTURE OF UKRAINE.	27
4.	<i>Husiev A., Prakhovnik N.</i> CONFORMAL BEHAVIOR AND SAFE BEHAVIOR IN THE WORKING PROCESS.	33
5.	<i>Honcharenko T., Mihaylenko V.</i> VERIFICATION OF BIM-MODELS FOR LIFECYCLE OF CONSTRUCTION SITE.	36
6.	<i>Karasova O.</i> THE ORGANIZATION OF STUDENTS' INDEPENDENT SCIENTIFIC RESEARCH WORK AT UNIVERSITIES.	42
7.	<i>Kaplunenko A. A., Kaplunenko A. N., Kavko A. A., Orlyuk M. I., Pigulevskiy P. I.</i> ON THE QUESTION OF CONNECTION BETWEEN SICKNESS RATES OF LARGE CATTLE LEUCOSIS AND NATURAL MAGNETIC FIELD OF THE EARTH.	46
8.	<i>Kats Ju.</i> LINGUISTIC MODELS OF FAREWELL DISCOURSE.	51
9.	<i>Loiuk O., Hritchenko T.</i> THINKING AS THE BASIS FOR THE DISCOVERY OF NEW KNOWLEDGE.	60
10.	<i>Lytvynenko O., Maliutina O.</i> CREATION OF METHODOLOGICAL SUPPORT FOR INDIVIDUAL WORK OF ENGLISH STUDENTS IN LEARNING UKRAINIAN.	64
11.	<i>Lykhochvor V. V., Andrushko M. O., Andrushko O. M.</i> PEA (PISUM SATIVUM) YIELD OF OTAMAN VARIETY, DEPENDING ON THE SOWING RATE.	70
12.	<i>Mayboroda I., Nahlov O.</i> CIRCADIAN FEATURES OF THE INFLUENCE OF WHITE AND RED ILLUMINATION ON THE BEHAVIORAL RESPONSES OF RATS.	75
13.	<i>Makovska T., Tkachenko N., Sevastyanova O., Ruda M.</i> PERFUME FRAGRANCES: TYPES AND OPTION POSSIBILITIES TO SELECT.	80

FIBRONECTIN ON THE SURFACE AND INSIDE THE LEUKOCYTES IN LIVER FIBROSIS

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Annotation. The aim of the study was to determine the level of fibronectin in blood leucocytes in patients with liver fibrosis. The level of fibronectin on the surface and inside in the blood monocytes, lymphocytes and granulocytes was determined by flow cytometry. The level of lymphocytes fibronectin in patients with liver fibrosis decreased compared to its normal value, both inside and on the cell surface, by 45.3% ($p < 0.05$) and 16.2% ($p < 0.05$), respectively. The decreasing of granulocytes fibronectin on surface and inside of cells was by 25.0% ($p < 0.05$) and 36.5% ($p < 0.05$), respectively.

Key words: liver fibrosis, fibronectin, exponents, lymphocytes, granulocytes.

Liver fibrosis (LF) is a key link in the development of the pathological process in the liver tissue, and the degree of fibrosis is highly sensitive nonspecific marker for pathological changes in the liver under the influence of various causative factors [1, c. 350]. Stellate cells, myofibroblasts and bone marrow cells converge in a complex interaction with hepatocytes and immune cells to cause scarring in response to liver

damage, which is provided by various extracellular matrix proteins, including fibronectin [2, c. 223]. As fibronectin molecule has near eight different domains for connection with collagen, proteoglycan, hyaluronic acid, heparin it can perform an integrative role in the organization of intercellular matter, promote cell adhesion [3, c. 399]. The diagnostic value of fibronectin and its role in the development of fibrosis has not yet been elucidated and contradictory. It has now been established that leukocytes are no longer merely “observers” in the pathogenesis of liver fibrosis, but are directly involved in this process [4, c. 1139]. The aim of the study was to determine the level of fibronectin on the surface and inside in blood lymphocytes, monocytes and granulocytes of patients with LF (n = 15) aged 28-46 years. The control group consisted of healthy volunteers (n = 15) aged 25 to 52 years. The level of fibronectin on the surface and inside of blood cells was determined by flow cytometry using monoclonal antibodies to matrix fibronectin and the corresponding secondary antibodies to mouse immunoglobulins conjugated to fluorescence isothiocyanate. Statistical processing was carried out using software packages «R» and «EasyROC 1.3.1». It was obtained, that the fibronectin level in lymphocytes of patients with LF decreased compared to its normal value, both inside and on the cell surface, by 45.3% ($p < 0.05$) and 16.2% ($p < 0.05$), respectively. This declining trend was also observed when investigating fibronectin level on the blood granulocytes surface and inside them: by 25.0% ($p < 0.05$) and 36.5% ($p < 0.05$), respectively. The level of exposure of fibronectin on the surface and inside the monocyte in a group of fibrosis and a healthy donors was not different. To assess the effectiveness of using of the level fibronectin expression in lymphocytes and granulocytes as diagnostic markers the traditional ROC-analysis method was used [5, c. 763]. Diagnostic efficiency of the test fibronectin exposure in lymphocytes is such: Se = 0.923, Sp = 0.769. AUC = 0.899 ($p = 2.01 \cdot 10^{-11}$), indicating a very good diagnostic informativeness of the method. The test of the fibrosis diagnosis according to level of fibronectin on the surface of lymphocytes is: Se = 0.923; Sp = 0.692. AUC = 0.869 ($P = 6.57 \cdot 10^{-8}$), the diagnostic informativeness of method is high. The diagnosis opportunities of the fibronectin level test inside the granulocytes is: Se = 1.0;

Sp = 0.846. AUC = 0.970 (P = 6.46e-65) – the excellent diagnostic informativeness. On the surface of granulocytes this indicators were: Se = 1.0, Sp = 0.923. AUC = 0.964 (P = 8.44e-36) – also excellent diagnostic informativeness. The results obtained, of course, require further research with CD differentiation of granulocytes and lymphocytes, which may lead to the development of additional criteria for the diagnosis of fibrosis. This can lead to decrease in the need for liver biopsy, a painful and risky procedure, which remains the main type of diagnosis.

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