10. Шутова Н. А., Кузьміна І. Ю., Морозов О. В. Гібридні технології навчання в ХНМУ: Сьогодення. *Сучасні аспекти досягнень фундаментальних та прикладних медико-біологічних напрямків медичної та фармацевтичної освіти та науки* : матер. І наук.-практ. інтернет-конф. з міжнар. учас., яка присвячена до 90-ї річниці з дня народження професора Л. Т. Киричок (Харків, 17 листопада 2022 р.). Міністерство охорони здоров'я України. Харків : ХНМУ, 2022. 472 с.

DOI https://doi.org/10.30525/978-9934-26-446-7-23

ANTIOXIDANT STATUS OF EXTRACTS FROM STEMLESS CARLINE THISTLE (*CARLINA ACAULIS* L.), MOUNTAIN ARNICA (*ARNICA MONTANA* L.) AND POT MARIGOLD (*CALENDULA OFFICINALIS* L.)

АНТИОКСИДАНТНИЙ СТАТУС ЕКСТРАКТІВ ВІДКАСНИКА БЕЗСТЕБЛОВОГО (*CARLINA ACAULIS* L.), АРНІКИ ГІРСЬКОЇ (*ARNICA MONTANA* L.) ТА КАЛЕНДУЛИ ЛІКАРСЬКОЇ (*CALENDULA OFFICINALIS* L.)

Yaremkevych O. S.
Ph.D. of Biological Sciences,
Associate Professor at the Department of Technology
of Bioactive Compounds, Pharmacy and Biotechnology

Fedoryshyn O. M.
Ph.D. of Technology,
Assistant at the Department of Technology
of Bioactive Compounds, Pharmacy and Biotechnology

Krychkovska A. M.
Ph.D. of Pharmacy Sciences,
Associate Professor at the Department of Technology
of Bioactive Compounds, Pharmacy and Biotechnology
Lviv Polytechnic National University
Lviv, Ukraine

Stress accompanies humans throughout their lives. The problem of stress and its impact on various functional systems of the body has remained relevant for modern biology and medicine for many years, especially in the present time when people are affected by the many stressful situations

related to the war in Ukraine. There are two types of stress: eustress and distress. Eustress is a normal, moderate level of stress that does not disrupt the body's homeostasis. On a physiological level, distress, which disrupts homeostasis, can cause many diseases [1, 2] and exceeds the limits of adaptation. Due to distress, a shift in redox balance occurs in the body towards the formation of free radicals and production of lipid and protein peroxides. In biological systems, the most active are free radicals and ion radicals, which have unpaired electrons in their structure and form so-called reactive oxygen species (ROS), nitrogen species (RNS), sulfur species (RSS), and chlorine species (RCS). Free radical oxidation is a universal mechanism by which the most important homeostatic physics-chemical parameters of the cell are controlled: selective permeability, viscosity, and the integrity of cell membranes [3]. It is known that oxidative stress is enhanced as a result of many chronic diseases [4–6], which leads to the activation of lipid peroxidation (LPO) processes, oxidative modification of proteins (OMP), and the destruction of nucleic acids and carbohydrates. This causes significant changes in the metabolic processes of cells and cellular membranes. Membranes damaged by LPO and OMP processes lose their electro excitable function, energy potential, and control over ion flows and mediator functions. As a result, pathological processes occur in tissues – such as inflammatory, neurodegenerative, and malignant processes – which lead to cell death. In most cases, the hydroxyl radical ($OH\cdot$) aggressively initiates this process, acting as a hydrogen atom acceptor from organic compounds, forming a free radical ($RH + OH \rightarrow R\cdot + H_2O$) [7].

Given this, avoiding various complications in the course of diseases can be achieved by timely blocking the initiating mechanisms of pathology, specifically by reducing the intensity of lipid peroxidation (LPO) and oxidative modification of proteins (OMP) in the body through the use of antioxidants.

Therefore, the search for new medicinal preparations for stress correction, as well as the comprehensive use of antioxidants and natural products as stress correctors, is of great practical importance [8, 9]. Recently, considerable attention has been paid to studying the antioxidant activity of medicinal plant extracts, which are widely used in both traditional and official medicine, as well as in cosmeceuticals.

The aim of this work is to obtain extracts from medicinal plant raw materials (MPRM) under laboratory conditions and to investigate their antioxidant properties.

In the study, plant raw materials (MPRM) were collected from natural habitats (Skole District, Lviv Region). The herbs were dried according to standard requirements for medicinal herb preparation – in a dark, dry, and well-ventilated place. The dried MPRM was ground in a mortar and sieved through a sieve with a pore diameter of 3.0 mm. The extraction was carried

out using the maceration method with a raw material-to-extractant ratio of 1:10 at a temperature of 20˚C for 7 days, with periodic stirring. An aqueous-alcohol solution containing 40% and 70% ethanol was used as the solvent. After extraction, the extract was filtered.

The antioxidant properties of the investigated extracts were determined under conditions of in vitro free radical oxidation initiation using two indicators of oxidative stress – thiobarbituric acid reactive substances (TBARS) and carbonyl groups (CG) of proteins. Both indicators of oxidative stress were determined in a single sample – the content of TBARS was determined in the supernatant, while CG was determined in the sediment using the method described by V.I. Lushchak [10].

The research data was statistically processed taking into account the arithmetic mean (M) and the standard error (SE) in the form of (M±SE), with the number of repetitions n=5. Differences between experimental data were determined using Tukey's test for one-way analysis of variance (ANOVA), where differences were considered significant at $p<0.05$ [11]. The processing results are presented in the form of diagrams.

As a result of the conducted research on POL and OMP (Fig. 1–4), it was found that compared to the control, all investigated extracts significantly (> 50%) reduced the content of TBARS and the formation of protein carbonyl groups. This demonstrates the high antioxidant properties of the obtained extracts. As can be seen from the diagrams, the most effective among the studied 40% and 70% aqueous-alcoholic extracts in terms of oxidative stress indicators were arnica and calendula.

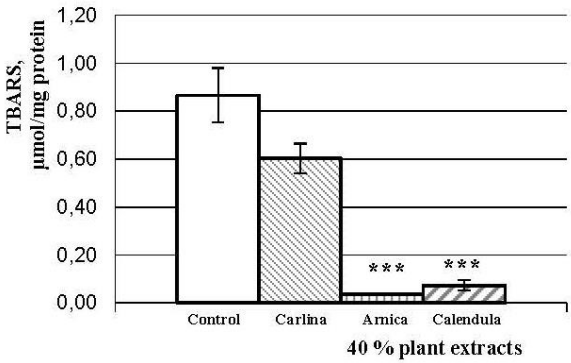

Fig. 1. The content of TBA-reactive substances (TBARS) in rat liver homogenate under the influence of 40% plant extracts (*– p≤0.005; M±m; n=5)**

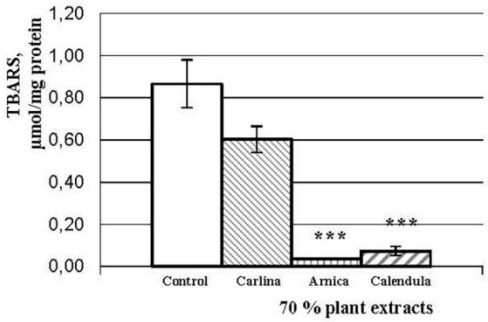

Fig. 2. The content of TBA-reactive substances (TBARS) in rat liver homogenate under the influence of 70% plant extracts (*– p≤0.001; M±m; n=5)**

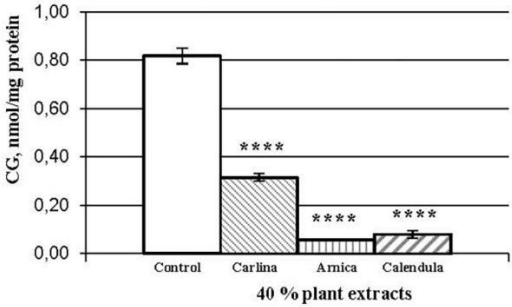

Fig. 3. The content of protein carbonyl groups in rat liver homogenate under the influence of 40% plant extracts (**– p≤0.001; M±m; n=5)**

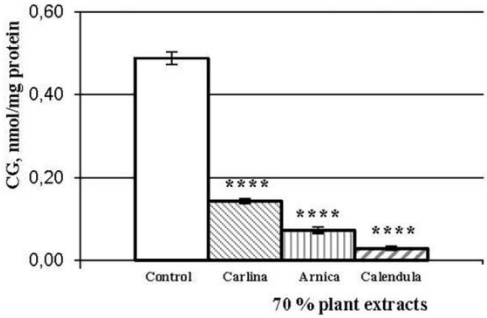

Fig. 4. The content of protein carbonyl groups in rat liver homogenate under the influence of 70% plant extracts (**– p≤0.001; M±m; n=5)**

Under the influence of these extracts at a concentration of 40%, there is a reduction in TBARS content by 95.8% for arnica and by 91.5% for calendula ($p \leq 0.005$), and protein carbonyl content by 93.1% and 90.3% respectively compared to the control ($p \leq 0.001$). Under the influence of 70% extracts, there is a reduction in TBARS content by 86.7% for arnica (*Arnica Montana* L.) and by 95.9% for calendula (*Calendula officinalis* L.) ($p \leq 0.001$), and protein carbonyl content by 85% and 93.9% respectively compared to the control ($p \leq 0.001$).

Less pronounced, yet still high antioxidant activity compared to the control, was exhibited by stemless carline thistle (*Carlina acaulis L.*) at concentrations of 40% and 70%.

Under the action of stemless carline thistle at a concentration of 40%, the TBARS content was 69.6±10.3%, and the protein carbonyl content was 38.6±5.0% ($p \leq 0.001$). The use of stemless carline thistle at a concentration of 70% contributed to a reduction in TBARS content by 57.6% and in protein carbonyl content by 70.7%, respectively ($p \leq 0.001$). Our experi mental studies confirmed the presence of antioxidants of various origins in the medicinal plant raw materials.

References:

1. Rabasa, C., Dickson, S. L. Impact of stress on metabolism and energy balance. *Current Opinion in Behavioral Sciences*. 2016. Vol. 9. P. 71–77. https://doi.org/10.1016/j.cobeha.2016.01.011

2. Gupta, R. K., Patel, A. K., Shah, N., Choudhary, K. A., Jha, U. K., Yadav, U. C., Gupta, P. K., & Pakuwal, U. Oxidative stress and antioxidants in disease and cancer: A Review. *Asian Pacific Journal of Cancer Prevention*. 2014. Vol. 15(11). P. 4405–4409. https://doi.org/10.7314/APJCP.2014.15.11.4405

3. Petersen, R. C. Free-radicals and advanced chemistries involved in cell membrane organization influence oxygen diffusion and pathology treatment. *AIMS Biophysics*. 2017. Vol. 4(2). P. 240–283. https://doi: 10.3934/biophy.2017.2.240

4. Bhatti, J.S., Sehrawat, A., Mishra, J., Sidhu, I.S., Navik, U., Khullar, N., Kumar, S., Bhatti, G.K., & Reddy, P.H. Oxidative stress in the pathophysiology of type 2 diabetes and related complications: Current therapeutics strategies and future perspectives. *Free Radical Biology and Medicine*. 2022. Vol. 184(1). P. 114–134.

5. Barrera, G. Oxidative stress and lipid peroxidation products in cancer progression and therapy. *ISRN Oncol*. 2012. P. 137289.

6. Simonian, N. A., & Coyle, J. T. Oxidative stress in neurodegenerative diseases. *Annu. Rev. Pharmacol Toxicol.* 1996. Vol. 36. P. 83−106.

7. Shapoval, H. S. Mechanisms of antioxidant protection of the body under the action of active forms of oxygen. *Ukraine biochem. Journal*. 2003. Vol. 75(2). P. 5–13.

8. Mizutani, T., & Masaki, H. Anti-photoaging capability of antioxidant extract from *Camellia japonica* leaf. *Experimental Dermatology*. 2014. Vol. 23(1). P. 23–26. https://doi:10.1111/exd.12395

9. Ahn, K. The worldwide trend of using botanical drugs and strategies for developing global drugs. *Biochemistry & Molecular Biology Reports*. 2017. Vol. 50(3). P. 111–116. https://doi: 10.5483/BMBRep.2017.50.3.221

10. Лущак, В. І., Багнюкава, Т. В., & Лужна, Л. І. Показники оксидативного стресу. 2. Перекиси ліпідів. *Український біохімічний журнал*. 2006. Вип. 78(5). С. 113–119.

11. Morgan, G. A., Leech, N. L., Gloeckner, G. W., & Barrett, K. C. *IBM SPSS for Introductory statistics. In: Use and Interpretation*, 4-th ed. ; Routledge Taylor & amp; Francis Group, New York. 2012. 243 p.