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Anti-Aging Effect of *Cynara cardunculus* L. var. *Cynara scolymus* L. Extract in D-Galactose-Induced Skin Aging Model in Rats

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Abstract

The aim of the study was to evaluate the efficacy of 2% *Cynara scolymus* L. extracts in reverse disturbances of collagen metabolism and inflammation in D-galactose induced skin aging model in rats. D-galactose induced aging reproduced in 48 animals of main group, and 12 rats in control group. All animals in main group were randomized for 4 groups: I. aging + saline, II - IV. different manufacturers 2% artichoke extracts (with content of chlorogenic acid < 2.5%) in a dose of intradermal injection 0.13 mg twice at weeks during 4 weeks. Influence of artichoke extracts restored skin relative weight and led to an increase of solubility in neutral salt and acid, and decreased pepsin solubility collagen fraction, restored the hexosamine/collagen (hydroxyproline) ratio and decreased the activity of nuclear transcription factor (NF-kB). Local prolonged treatment with artichoke extracts improved collagen metabolism and attenuated the progression of inflammation in D-galactose induced skin aging model.

Keywords

Skin, Aging, Collagen, Pure Water *Cynara cardunculus* L. var. *Cynara scolymus* L. Extract, Nuclear Transcription Factor Kappa B

1. Introduction

Intrinsic or innate aging, an unpreventable process, affects the skin in the same pattern as it affects all internal organs. Age-relating skin changes and its prevention as a dream of human being strongly associated with loss of elasticity, epi-

dermal atrophy, wrinkling of the skin and cause by hormonal (estrogens and androgens) rearrangement inversed turnover collagen type I/III and collagen degradation in dermis, dryness, worsening of the texture and appearance of skin [1]. Skin aging is inevitably associated with a disturbance in collagen metabolism (adult skin is composed of type I (80% - 85%) and type III collagen 10% - 15%) due to the decreased activity of fibroblasts and their collagen synthesis, as well as elastin [2]. Thus, the search of new medicinal technologies modulating metabolic processes in the skin and improving skin resistance to oxygen demand and oxidative stress development (the oxidative stress is one of the major mechanisms for skin aging and dermatological conditions [3]) and preventing the collagen isoforms imbalance, is an urgent problem in dermatology. A significant amount of evidence has been pointed to the beneficial effect of natural plants extracts rich in phenolic and flavonoid compounds, on cellular nucleus and collagen metabolism disturbances, reducing the amount of free radicals in dermal fibroblasts [4]. Such substances are present in extracts of artichoke *Cynara scolymus* L. var. *Cynara cardunculus* L., Asteraceae). The artichoke is emerging like a promising medicinal plant and ancient crop in Italy, Spain, France, Egypt and now in Georgia (Italian cultivars var., *Cynara cardunculus* L., Grosso Romanesco) with favorable climatic condition. Extracts of artichoke on the tissue level have been found to decrease the production of reactive oxygen species, the oxidation of low-density lipoproteins and lipid peroxidation, protein oxidation and increase the activity of glutathione peroxidase (for review see [5] [6] [7] [8]). Among health-promoting properties, there were identified and characterized antioxidant, hepatoprotective, choleretic, hypoglycemic, hypocholesterolemic, hypotriglyceridaemic, cardiovascular, antifibrotic, anti-inflammatory and antimicrobial, anti-dyspepsia and prebiotic effects [8]-[17]. Hepatoprotective effect of artichoke leaf extract has a cholesterol reducing property, decrease oxidation of low density lipoproteins throughout in part by facilitating bile production assisted fat digestion together with toxin removal [18] [19]. The lipolytic action of *Cynara scolymus* L. occurs by enhancing the synthesis of the NAD-$NADH_2$ and NADP-$NADPH_2$ coenzymes involved in reducer anabolic processes that are mainly used in fatty acid synthesis in the adipose tissue and the liver. Appropriate formulation of luteolin [17], sylimarin [20] and cynaropicrin [21], as components of artichoke extracts, promising to candidate to target local and systemic treatments of inflammatory skin diseases [20] [22], however, little known about direct ability of water extracts of artichoke to improvement in skin conditions and skin inflammation in aging. The aim of the present study is to assess the mechanism of therapeutic effects of lyophilized water extracts of drying leaves of *Cynara cardunculus* L. on markers of collagen metabolism and inflammation in skin in experimental animals with D-galactose induced skin aging.

2. Materials and Methods

2.1. Plant Materials and Extraction

The experiment was carried out during the 2016-17 years, in a representative

area for *Cynara cardunculus* L. (Grosso Romanesco) var. *scolymus* L., femaly Aesreraceae) globe artichoke cultivation in Mtskhetis region (Rosenthal, Georgia, latitude 41˚56'02"N and longitude 44˚34'36"E), average minimum temperature −1˚C and maximum 35˚C. Preparation for comparison: pure Artichoke extract, 2%, ampoule 5 ml, from "Veluderm International S.L.", Spain and 2% Artichoke extract, manufacturer "Group Companies Martinex", Russia. The fresh leaves of the artichoke were collected at harvest maturity from the June to the middle of October. The leaves were separated washed, cleaned, and drying in according with Eur Ph monograph 01/2008:1866 corrected 6.0. Extraction of dried leaves artichoke, separation and identification of Artichoke (*Cynara cardunculus* L.) performed in according with Eur Ph monograph 01/2009:2389 (content of chlorogenic acid < 2.5%). Crude aqueous extracts of dried leaves (100 g) were prepared by infusion with distilled water (plant:solvent ratio of 5:1) at 96˚C to the homogenized leaves for 120 min, and extraction for four hours using bi-distillated water as a solvent. Prepared extracts filtered through a metallic mesh to remove any kind of solid particle, cooled at room temperature and centrifuged at 5000 - 6000 rpm (revolutions per minute) for 15 min. The obtained primary extract was filtered throughout closed sterile filtration systems with 0.45 μ and 0.2 μ. After sterile filtration extracts concentrated by lyophilization with a FTS Systems Lyostar II LYOACC3P1, USA lyophilizer (initial temperature of −30˚C, the time of lyophilisation 24 h, additional drying at 32˚C for 6 h), previous freezing at −55˚C in a Ultra-Low-Temperature Freezer VT 407 (Vestfrost Solutions, Danish). The resulting yields were 13.7 g for dry leaf water extracts. The toxicity of artichoke extract is very low (LD50 for mice with intraperitonially application is more than 2.0 g/kg body and in rats is more than 1 g/kg body weight. No microscopic lesions registries in skin of Wister rats after application of artichoke extract twice daily, for 14 days, at concentration up to 10% and moderately irritating when applied to intact and abraded rabbit skin. Pilot patch test on 7 volunteers (occlusive patch 48 hours) documented absence of irritant reactions at 15 min and 24 hours after patch removal and indicated on very good skin compatibility.

2.2. Determination of the Total Phenolics, Flavonoids and Antioxidant Capacity (AOC)

Total phenols (TPC) was determined spectrophotometrically by measured the absorbance at 765 ± 2 nm following the Folin-Ciocalteu reagent assay using gallic acid for the preparation of calibration curve (12.5, 25, 50, 62.5, 100 and 125 mg/L). Dilutions of lyophilized water extract of artichoke leaves ranging from 4.25 to 0.02 mg/ml were used. The total phenolic content expressed as mg gallic acid/100 mg of extract. Total flavonoid content of was determined by the aluminium chloride method using quercetin as a standard [23] and results were expressed as mg of quercetin equivalents measured by absorption at 420 ± 2 nm/100 mg of extracts [24]. Total antiradical activities was determinate by evaluation of 1,1-diphenyl-2-picrylhydrazyl (DPPH) methods. 1 ml extract of arti-

choke was mixed with in 4 ml of 0.004% solution of DPPH in 80% methanol (a radical at the concentration of (working solution 24 mg/L (60.8 μmol/L) DDPH in 80% methanol, an absorbance of 0.8 at 517 ± 2 nm) was prepared for calibrated curve preparation. The sample was incubated for about 60 min in dark at 22˚C and then absorption at 517 ± 2 nm was measured. AOC was calculated as IC50 (quantity of extract neutralizing 50% of DDPH amount).

2.3. Animal Study Design

Experiments carried out in 72 female Wistar rats weighing 180 - 200 g, 170 - 180 days old. Animals received humane care in compliance with "Guide for the Care and Use of Laboratory animals" (National Institutes of Health publication 86-23, Revised 1996) and was performed with approval of the local Interinsitutional (International Scientific Centre of Introduction of New Biomedical Technology, Department of Medical Pharmacology and Pharmacotherapy, Tbilisi State Medical University, Tbilisi) Animal Care and Use Committee. All animals were secured under specific pathogen free conditions according to the Federation of European Laboratory Animal Science Associations guidelines in humidity- and temperature-controlled environment, with a daylit environment for at least 1 week before the experiments. Animals were fed commercial laboratory rat's food pellet and allowed drink tap water *ad libitum* before the experiments. After 7 days of adaptation, all animals randomized into two groups: control I and main. Animals in main group after randomization received injection with D-galactose (reducing sugar, is a naturally occurring substance in the body, 150 mg/kg/day, i.p. [25] [26]), while in control I group received saline injection (1 mg/kg/day, i.p.), for 8 weeks. At 21 days after injection with D-galactose the 3 cm round tattoo area was prefabricated on each side of rats previously disinfected hip under sterile condition and general anesthesia with pentobarbital (40 mg/kg). All animals in main group (40 animals) were secondly randomized into 4 groups in dependence to treatment (twice in week of intradermal injection under general anesthesia) for 5 weeks: control II group animals treated with microinjection of saline, main I group treated with 0.12 ml of 2% lyophilized powder of Artichoke extracts salivated in water for injection, Georgia (equivalent of average intradermal dose for patients 10 mg), n = 12; main II – 0.12 ml of pure Artichoke extract, 2% (Spain), n = 12 and main III – 0.12 ml of 2% artichoke extracts (manufacturer "Group Companies Martinex", Russia), n = 12. After the experiments, all the rats were euthanized by pentobarbital (60 mg/kg intraperitonially).

2.4. Body Weight and Skin Oedema Evaluation

The mean body weight of all the animals was taken from the start of the study, 8 weeks and on the end of experiments. Appearance of skin, fur and eyes were observed during these days. Skin oedema was measured as decreases in skin (1 cm^2) and calculated by dividing the wet-to-dry weight ratios after tissue drying to constant weight at 80˚C.

2.5. Total Collagen (Hydroxyproline) and Hexosamine Contents and Its Fraction

Immediately, skin a weighted amount of skin (3 g) was shaved, cleaned from the underlying connective, muscular and adipose tissues was frozen in nitrogen and minced. Lipids were removed with chloroform:methanol (2:1) mixture to determine the amount of hydroxyproline and hexasamine [27]. Total content of collagen was measured assuming a hydroxyproline content of 0.91 mmol·1^{-1} (mg collagen)$^{-1}$ or by multiplying the hydroxyproline content by a factor 7.46. Solubility pattern of collagen was determined as described by [28].

2.6. Preparation of Nuclear Extracts and Determination of NF-kB

Skin sample frozen in nitrogen, minced in 10 volumes of solution 1, containing 10 mmol/L KCl, 10 mmol/L HEPES-buffer, 0.1 mmol/L ethylene glycol tetraacetic acid (EGTA) and 0.1 mmol/L ethylenediaminetetraacetic acid (EDTA), 0.1 mmol/L phenylmethylsulfonyl fluoride (PMSF) nd 0.1 mmol/L dithiothreitol (DTT), pH 7.8. IGEPAL (octylphenoxypolyethoxyethanol, Sigma Aldrich) was added to a final concentration 0.7%. Skin homogenate were centrifugated twice at 7000 g for 30 s after resuspended in 1 ml of new portion solution 1. Then pellet was resuspended in 0.3 ml cold solution 2 containing: 0.4 mol/L NaCl, 20 mmol/L HEPES-buffer, 1.0 mmol/L EGTA and 1.0 mmol/L EDTA, 1.0 mmol/L PMSF and 1.0 mmol/L DTT, pH 7.8, left on ice for 20 min and the pellet of centrifugated at 10,000 g for 30 min was used in the study. The activity of NF-kB(p60) was registrated as described early [29]. The Bradford method (Coomassie brilliant blue G-250 staining) was used to detect protein content.

2.7. Statistical Analysis

All variances in the measurement data expressed as mean ± standard deviation of mean (SD), and statistical significance assessed using Student t-test for normally distributed variables and $p < 0.05$ is considered significant. All statistical calculations were performed using the Statistical Sciences (SPSS, version 23.1).

3. Results

3.1. Properties of the Studying Extracts of Artichoke (*Cynara cardunculus* (Grosso Romanesco), Subsp *Cynara scolumus* L.)

The studying water dry extracts of artichoke (*Cynara cardunculus*, cultivated in Georgia, Mtskhetis region), 2% Pure Artichoke extract from Spain and Russia related to water artichoke extracts with content of chloroagenic acid < 2.5% (Assessment report on *Cynara scolymus* L., folium EMA/HMPC/150209/2009) and about 10% of total phenolic acids. No significance difference in the contents of total phenolic, flavonoids compounds and total antioxidant activities in the extract made in our laboratories and VD Pure Artichoke extract, 2% (Spain) eliminated. While values of all parameters of Artichoke extract, 2% (Russia) are smaller (Table 1).

Table 1. Comparison of phenolic, flavonoids content and total antioxidant activity of different 2% extracts of artichoke.

Properties	Liophylized extract artichoke, Georgia	Pure Artichoke extract, 2% (Spain)	Artichoke extract, 2% (Russia)
Chlorogenic acid, mg/100 mg extract	1.97 ± 0.18	1.86 ± 0.14	1.25 ± 0.14**#
Total phenolic compounds, %*	9.6 ± 0.6	8.8 ± 0.8	7.7 ± 1.1*
Flavonoids, %	1.45 ± 0.09	1.51 ± 0.10	1.23 ± 0.10*#
Total antioxidant activity, 50% inhibitory of DDPH	14.3 ± 1.8	11.4 ± 1.2	8.4 ± 0.8*#

Note: values are mean of 5 samples of 5 times production of extracts and in each samples determination performed in triplicate in case of "Lyophilized extract artichoke, Georgia" and values of 4 bath of commercial Artichoke extract (Spain) and 5 bath of Artichoke extract (Russia); *- total phenols in absolute value in "Lyophilized extract artichoke, Georgia" is 0.29 mg GAE/100 mg extract; *- compared with "Lyophilized extract artichoke, Georgia", # - with Artichoke extract (Spain), significance of between the extracts, significance of difference of comparison: one symbol – $p < 0.05$, two – $p < 0.01$, absence of symbol indicated that differences is not significance ($p > 0.05$).

3.2. Beneficially Therapeutic Effect of Different Artichoke Extracts on Maintenance Level and Balanced Soluble/Insoluble Collagens in D-Galactose-Induced Skin Aging of Rats

Animals with D-galactose induced skin aging during 12 weeks demonstrated evident symptoms of aging, including slow movement, a lag in response, listlessness, and withered and lackluster fur, decreased behavioral activity, some hypodynamia, neurological impairment, established hair color changes and an unique skin appearance, with wrinkling's and furrows indicative of skin aging appearing at the age (Table 1). Prior to euthanized, no morbidity/mortality and clinically relief differences in food intake and water consumption in subgroups of main group were not observed. The mean body weight of animals in treated group when compare with mean body weight of animals in control III groups showed no significant alterations in the weight gain, but was decreased in all groups in comparison to mean body weight in first day and to control II animals the same age as in main group (Table 2).

However, the relative weight of skin markedly decreases in D-galactose model of aging. Water content in the skin is presumed to be a critical determinant in cutaneous aging. Treatment with all preparation of artichoke extracts restores the water dysbalanced in the aging skin in the same manner. Treatment with lyophilized extract artichoke, Georgia and pure Artichoke extract, 2% (Spain) and in less extent in case of 2% artichoke extracts manufacturers by "Group Companies Martinex", Russia leads to increase the content of hydroxyproline in aging skin which compared to aging groups without treatment (Table 2). In the samples of skin from animals of control group III occurs decreased fractions of solubility in neutral salt and acid collagen while the collagen fraction solubility with pepsin increased about two 2-fold. Local prolonged treatment with artichoke extract preparation improved solubility of skin collagen in neutral salt and acid as compared with D-galactose treated group, meaning that the content of

Table 2. Body weight, relative weight of skin, hydroxyproline, hexosamine and content and NF-kB activity in skin in control and experimental animals.

Groups/Parameters	Control I, n = 12	Control II, n = 12	Main group, = 48 D-galactoside induced aging group			
			Control III	I	II	III
Body weight, g 180 days at the beginning of the studying	186 ± 19	298 ± 23^{x}	257 ± 18	277 ± 17	270 ± 21	259 ± 21
Relative weight, mg dry/100 mg wet weight	31.4 ± 1.4	32.1 ± 1.2	24.4 ± 1.5**	28.9 ± 1.4#	28.0 ± 1.6#	27.1 ± 1.5#
Hexosamine mg/g wet tissue	16.4 ± 1.4	18.2 ± 1.2	23.7 ± 2.3*	17.8 ± 4.1#	19.3 ± 4.3#	21.8 ± 1.2*
Hydroxyprolinemg/g wet tissue	15.0 ± 0.8	17.2 ± 0.9^{x}	13.7 ± 0.8**	15.9 ± 0.9#	15.4 ± 1.1#	14.8 ± 1.2*
Total collagen, mg/g wet tissue	112 ±10	129 ± 10	102 ± 6*	119 ± 8#	115 ± 8#	111 ± 9
NaCl-Soluble collagen, mg/g wet tissue	7.1 ± 1.1	7.7 ± 1.0	4.4 ± 0.8**	6.5 ± 0.5##	6.35 ± 0.63##	5.97 ± 0.78#
Acid-Soluble collagen, mg/g wet tissue	0.12 ± 0.02	0.13 ± 0.03	0.07 ± 0.01**	0.10 ± 0.01*#	0.10 ± 0.01*#	0.09 ± 0.01*
Pepsin-Soluble collagen, mg/g wet tissue	0.85 ± 0.08	0.88 ± 0.08	1.8 ± 0.2***	0.95 ± 0.10###	1.09 ± 0.09###	1.47 ± 0.15#
Insoluble collagen, mg/g wet tissue	105 ± 9	121 ± 9	97 ± 9*	113 ± 6#	109 ± 6	105 ± 7
NF-kB(p65) activity, o.u. at 450 nm	0.07 ± 0.02	0.09 ± 0.01	0.21 ± 0.03	0.10 ± 0.02	0.10 ± 0.03	0.14 ± 0.02

Note: control II practically healthy animals of control I group (170 - 180 days old) at 230 - 240 days old as in main groups. *- compared with control II, # - with control III, §- main I group, Δ - main II group, x – between control I and control II; significance of between the extracts, significance of difference of comparison: one symbol – $p < 0.05$, two – $p < 0.01$, absence of symbol indicated that differences is not significance ($p > 0.05$).

newly synthesized collagen had been increased and its level is known to decrease with age in skin [30]. Early it was shown that soluble collagen extractable by 0.5 M NaCl showed rise after birth, reach maximum at the age of 8 weeks and then decline with animal aged. While level of insoluble collagen in skin rose continuously over the 12 month and revealed a strong correlation with tensile strength. Alternatively, the increase of hexosamines up to 23.7 ± 2.3 (vs. 18.2 ± 1.2 mg/g wet tissue in the control) in D-galactose induced aging skin indicated of presence of some signs of inflammation. Decrease the absolute levels of hexasamines and ratio between collagen (hydroxyprolyne): hexosamine under treatment with artichokes extracts could indicate to elimination of on acute fibrosis processes.

3.3. Changes of Inflammatory Markers in D-Galactose-Induced Skin Aging and Efficacy of Artichokes Extracts Action

Several studies point to a role for NF-κB in modulating epidermal thickness and apoptotic susceptibility of fibroblast [31] and keratinocytes [32] [33]. In D-galactose subacute model, skin aging activity of NF-kB (p65) increases by 133% (Table 2). Benefit for positive action of artichokes extracts inflammation confirmed by decreasing the activity of NF-kB (p65) by 50% up to level characterized to skin rats at the same age.

4. Discussion

The present study designed to investigate the potential protective effect of low-dose Georgian lyophilized extracts of artichoke against the skin markers of

elasticity caused by inversed turnover of collagen type ratio I/III dysbalance in D-galactose induced skin aging model in rats. Skin aging is a complicated multi-targets dysbalancing progression in the epidermal and dermal layers resulting from many intimal intrinsic and extrinsic factors. The major topic of research of pathogenesis and molecular mechanism of skin aging focused on the main dermal constituent collagen (about 60% - 80% of skin-mass [33]) and ability to maintenance collagen regulatory system, to prevent a loss of the skin's ability to resist stretching and degradation in aging. Artichoke extracts, a preparation which represents a natural source of phenolic compounds with dicaffeoylquinic acid along with their precursor chlorogenic acid (5-caffeoylquinic acid) as the predominant molecules has been traditionally used as a hepatoprotective, diuretic, detoxifying, lipid lowering agents with antioxidant effects and ability to reduce postprandial blood glucose (for review see [5] [6] [7] [8]). Pretreatment of *Cynara scolymus* extract in N-nitrodiethylamine and carbon tetrachloride-induced hepatocarcinogenesis in rats limited not only oxidative stress production, but as much important, decrease level of proinflammatory cytokines TNF-alpha and nuclear transcription factor-kappa B (NF-kB) [20] [22]. Canonical NF-κB pathways signaling has a crucial role in the maintenance of skin homeostasis [34]. Chronic, low-grade inflammation is also recognized as a major characteristic of the aging process [35]. This phenomenon is called "inflammaging". Given that the mechanism of skin "inflammaging" is far from being thoroughly understood, little progress is made to develop targeted treatments. Suppression activity of NF-kB included in the intrinsic mechanism of inhibition of tumor cell proliferation and apoptosis in N-Nitrosodiethyleamine induced hepatic injury [20] and blocking NF-kB activity could reverse effects of advanced glycation end-products on cell proliferation and migration, but not adhesion [36]. Previous studies also suggest that the NF-kB pathway plays an essential role in psoriasis progression [37] [38], where there is marked elevation of active phosphorylated NF-kB p65 in psoriases, as an inflammatory skin disease in which activated immune cells and the pro-inflammatory cytokine TNF are well-known mediators of pathogenesis [17]. Luteolin could be inhibiting inflammatory mediator production via blockade of NF-kB activation. However, it is also possible that luteolin acts on signaling molecules upstream of NF-kB activation, such as the mammalian target of rapamycin (mTOR) that governs cell size, growth, and metabolism [39]. Which of following or other molecular mechanism is responsible for undergoes collagen metabolism changes under D-galactose induced subacute skin aging and is it the same with normal skin aging processes is subject to a further research.

5. Conclusion

In conclusion, the results of the current study demonstrated the anti-aging effect of black seed oil in a mouse model of aging induced by D-galactose. Obtained data indicate that the concomitant use of 2% artichoke extract could be the only

way to exert anti-aging action in this model of skin aging in experimental animals, way to prevent imbalance in collagen metabolism and de-remodeling function of aging skin. Influence of artichoke extracts restored skin relative weight and led to an increase of solubility in neutral salt and acid and decreased pepsin solubility collagen fraction, restored the hexosamine/collagen (hydroxyproline) ratio and decreased the activity of nuclear transcription factor (NF-kB). Local prolonged treatment with artichoke extracts improved collagen metabolism and attenuated the progression of inflammation in D-galactose induced skin aging model.

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Conflicts of Interest

The authors declare that they have no conflict of interests regarding the publication of this paper. The authors alone are responsible for the content and writing of this article. No competing financial interest or financial conflict with the subject matter or materials discussed in the manuscript exists.

References

[1] Gragnani, A., MacCornic, S.M., Chominski, V., Ribeiro de Noronha, S.M., Correa de Naronha, S.A.A. and Ferreira, L.M. (2014) Review of Major Theories of Skin Aging. *Advances in Aging Research*, **3**, 265-284. https://doi.org/10.4236/aar.2014.34036

[2] Ganceviecene, R., Liakou, A.I., Theodoridis, A., Mafrantonaki, E. and Zouboulis, C.C. (2012) Skin Anti-Aging Strategies. *Dermato-Endocrinology*, **4**, 308-319. https://doi.org/10.4161/derm.22804

[3] E Silva, S.A.M., Leonardi, G.R. and Michniak-Kohn, B. (2017) An Overview about Oxidation in Clinical Practice of Skin Aging. *Anais Brasileiros de Dermatologia*, **92**, 367-374. https://doi.org/10.1590/abd1806-4841.20175481

[4] Mukherjee, P.K., Maity, N., Nema, N.K. and Sarkar, B.K. (2011) Bioactive Compounds from Natural Resources against Skin Aging. *Phytomedicine*, **19**, 64-73. https://doi.org/10.1016/j.phymed.2011.10.003

[5] Ben Salem, M., Affes, H., Ksouda, K., Dhouibi, R., Sahnoun, Z., Hammami, S. and Zeghal, K.M. (2015) Pharmacological Studies of Artichoke Leaf Extract and Their Health Benefits. *Plant Foods for Human Nutrition*, **70**, 441-453. https://doi.org/10.1007/s11130-015-0503-8

[6] Priechna, L. and Karklina, D. (2014) Natural Antioxidant Changes in Fresh and Dried Spices and Vegetables. *International Journal of Nutrition and Food Engineering*, **8**, 492-498.

[7] Sahebkar, A., Pirro, M., Banach, M., Mikhailidis, D.P., Atkin, S.A. and Cicero, F.G.

(2017) Lipid-Lowering Activity of Artichoke Extracts: A Systemic Review and Meta-Analysis. *Critical Reviews in Food Science and Nutrition*. https://doi.org/10.1080/10408398.2017.1332572

[8] El-Deen, S.A.S. (2018) Artichoke, the Ancient Hepatic Friend and Promising Multifunctional Therapeutic Agent. *Journal of Pharmacology and Therapeutic Research*, **2**, 4-5.

[9] Englisch, W., Beckers, C., Unkauf, M., Ruepp, M. and Zinserling, V. (2000) Efficacy of Artichoke Dry Extract in Patients with Hyperlipoproteinemia. *Arzneimittelforschung*, **50**, 260-265.

[10] Holtmann, G., Adam, B., Haag, S., Collet, W., Grünewald, E. and Windeck, T. (2003) Efficacy of Artichoke Leaf Extract in the Treatment of Patients with Functional Dyspepsia: A Six-Week Placebo-Controlled, Double-Blind, Multicentre Trial. *Alimentary Pharmacology & Therapeutics*, **18**, 1099-1105. https://doi.org/10.1046/j.1365-2036.2003.01767.x

[11] Pittler, M.H., White, A.R., Stevinson, C. and Ernst, E. (2003) Effectiveness of Artichoke Extract in Preventing Alcohol-Induced Hangovers: A Randomized Controlled Trial. *CMAJ*, **169**, 1269-1273.

[12] Speroni, E., Cervellati, R., Govoni, P., Guizzardi, S., Renzulli, C. and Guerra, M.C. (2003) Efficacy of Different *Cynara scolymus* Preparations on Liver Complaints. *Journal of Ethnopharmacology*, **86**, 203-211. https://doi.org/10.1016/S0378-8741(03)00076-X

[13] Zhu, X., Zhang, H. and Lo, R. (2004) Phenolic Compounds from the Leaf Extract of Artichoke (*Cynara scolymus* L.) and Their Antimicrobial Activities. *Journal of Agricultural and Food Chemistry*, **52**, 7272-7278. https://doi.org/10.1021/jf0490192

[14] Joy, J.F. and Haber, S.L. (2007) Clinical Uses of Artichoke Leaf Extract. *American Journal of Health-System Pharmacy*, **64**, 1904-1909. https://doi.org/10.2146/ajhp070013

[15] Huber, R., Müller, M., Naumann, J., Schenk, T. and Lüdtke, R. (2009) Artichoke Leave Extract for Chronic Hepatitis C—A Pilot Study. *Phytomedicine*, **16**, 801-804. https://doi.org/10.1016/j.phymed.2009.03.007

[16] Wider, B., Petter, M.H., Thompson-Coon, J. and Ernst, E. (2009) Artichoke Leaf Extract for Treating Hypercholesterolaemia. *Cochrane Database of Systematic Reviews*, **7**, CD003335. https://doi.org/10.1002/14651858.CD003335.pub2

[17] Weng, Z., Patel, A.B., Vasiadi, M., Therianou, A. and Theoharides, T.C. (2014) Luteolin Inhibits Human Keratinocyte Activation and Decreases NF-kB Induction That Is Increased in Psoriatic Skin. *PLoS ONE*, **9**, e90739. https://doi.org/10.1371/journal.pone.0090739

[18] Küskü-Kiraz, Z., Mehmetçik, G., Dogru-Abbasoglu, S. and Uysal, M. (2010) Artichoke Leaf Extract Reduces Oxidative Stress and Lipoprotein Dyshomeostasis in Rats Fed on High Cholesterol Diet. *Phytotherapy Research*, **24**, 565-570.

[19] Heidarian, E., Jafari-Dehkordi, E. and Seidkhani-Nahal, A. (2011) Lipid-Lowering Effect of Artichoke on Liver Phosphatidate Phosphohydrolase and Plasma Lipids in Hyperlipidemic Rats. *Journal of Medicinal Plants Research*, **5**, 4918-4924.

[20] Afifi, N., Ramadan, A., Yassin, N.Z., Fayed, H. and Abdel-Rahman, R.H. (2015) Molecular Mechanisms Underlying Hepathoprotective Effect of Artichoke Extract: Modulates TNF-Induced Activation of Nuclear Transcription Factor (NF-kB) and Oxidative Burst Inhibition. *World Journal of Pharmacy and Pharmacology*, **4**, 1546-1562.

[21] Tanaka, Y.T., Tanaka, K., Kojima, H., Hamada, T., Masutani, T., Tsuboi, M. and Akao, Y. (2013) Cynaropircin from *Cynara scolymus* L. Suppresses Photoaging of Skin by Inhibiting the Transcriptional Activity of Nuclear Factor Kappa-B. *Bioorganic & Medicinal Chemistry Letters*, **23**, 518-523. https://doi.org/10.1016/j.bmcl.2012.11.034

[22] Yasukawa, K., Matsubara, H. and Sano, Y. (2010) Inhibitory Effect of the Flowers of Artichoke (*Cynara cardunculus*) on TPA-Induced Inflammation and Tumor Promotion in Two-Stage Carcinogenesis in Mouse Skin. *Journal of Natural Medicines*, **64**, 388-391. https://doi.org/10.1007/s11418-010-0403-z

[23] Georgievs, S.S., Boyadzhieva, S.S. and Angelov, G. (2016) Identification of Extraction of Bioactive Substances from Artichoke Wastes. *Bulgarian Chemical Communication*, **48**, 451-455.

[24] Zhishen, J., Mengcheng, T. and Jianming, W. (1999) The Determination of Flavonoid Contents in Mulberry and Their Scavenging Effects on Superoxide Radicals. *Food Chemistry*, **64**, 555-559. https://doi.org/10.1016/S0308-8146(98)00102-2

[25] Liu, S., Chen, Z., Cai, X., Sun, Y., Zhao, C., Liu, F. and Liu, D. (2014) Effects of Dimethylaminoethanol and Compound Amino Acid on D-Galactose Induced Skin Aging Model of Rat. *The Scientific World Journal*, **2014**, Article ID: 507351.

[26] Liu, C., Hu, J., Mao, Z., Kang, H., Liu, H., Fu, W., Lv, Y. and Zhou, F. (2017) Acute Kidney Injury and Inflammatory Response of Sepsis Following Cecal Ligation and Puncture in d-Galactose-Induced Aging Rats. *Clinical Interventions in Aging*, **12**, 593-602. https://doi.org/10.2147/CIA.S132277

[27] Zadorozhnyi, A.V., Popkov, V.L., Galenko-Yaroshevskii, Antelava, N.A., Chikobava, E.A., Sukoyan, G.V. and Meladze, V.N. (2003) Effect of Richlokaine Alone and in Combination with Energostim on the Severity of Endotoxemia and Survival of the Skin under Conditions of Reduced Blood Flow. *Bulletin of Experimental Biology and Medicine*, **136**, 200-203. https://doi.org/10.1023/B:BEBM.0000008976.49593.9c

[28] Thirunavukkarasu, V., Nadhini, A.T. and Anuradha, C.V. (2004) Fructose Diet-Induced Skin Abnormalities Are Prevented by Lipoic Acid. *Experimental Diabetes Research*, **5**, 237-244. https://doi.org/10.1080/154386090506148

[29] Sukoyan, G.V., Golovach, S.V., Dolidze, N.M., Kezeli, T.D., Gongadze, N.V., Chipashvili, M. and Okujava, M.V. (2016) Hypertrophic and Inflammatory Markers in Isoproterenol-Induced Cardiac Hypertrophy and Its Pharmacological Correction. *Journal of Cardiovascular Pharmacology*, **6**, 5.

[30] Heikkinen, E. and Kulonen, E. (1968) Effect of Age on the Maturation of Rat-Skin Collagen. *Biochimica et Biophysica Acta*, **160**, 464-466. https://doi.org/10.1016/0005-2795(68)90218-3

[31] Reelfs, O., Tyrrell, R. and Pourzand, C. (2004) Ultraviolet a Radiation-Induced Immediate Iron Release Is a Key Modulator of the Activation of NF-kB in Human Skin Fibroblast. *The Society for Investigative Dermatology*, **122**, 1440-1447. https://doi.org/10.1111/j.0022-202X.2004.22620.x

[32] Saliou, C., Kitazawa, M., McLaughlin, L., Yang, J., Lodge, J., Tetsuka, K., Iwasaki, K., Cillard, J., Okamoto, T. and Packer, L. (1999) Antioxidants Modulate Acute Solar Ultraviolet Radiation-Induced NF-kappa-B Activation in a Human Keratinocyte Cell Line. *Free Radical Biology & Medicine*, **26**, 174-183. https://doi.org/10.1016/S0891-5849(98)00212-3

[33] Koyama, K. and Goto, E. (1986) Effect of Physical Activity on Collagen Content in Mice. *Fitness Japan*, **35**, 177-181. https://doi.org/10.7600/jspfsm1949.35.177

[34] Grinberg-Bleyer, Y., Dainichi, T., Oh, H., Heise, N., Klein, U., Schmid, R.M., Hayden, M.S. and Ghosh, S. (2015) Cutting Edge: NF-κB p65 and c-Rel Control Epidermal Development and Immune Homeostasis in the Skin. *The Journal of Immunology*, **194**, 2472-2476. https://doi.org/10.4049/jimmunol.1402608

[35] Odorisio, T. (2016) Epigenic Control of Skin Re-Epitalization: The NF-kB/JMJD3 Connection. *Journal of Investigative Dermatology*, **136**, 738-740. https://doi.org/10.1016/j.jid.2016.01.010

[36] Tian, M., Lu, S., Niu, Y., Xie, T., Dong, J., Cao, X., Song, F., Jin, S. and Qing, C. (2012) Effects of Advanced Glycation End-Products (AGEs) on Skin Keratinocytes by Nuclear Factor-Kappa B (NF-kB) Activation. *African Journal of Biotechnology*, **11**, 1132-1142.

[37] Liu, Z.G. (2005) Molecular Mechanism of TNF Signaling and Beyond. *Cell Research*, **15**, 24-27. https://doi.org/10.1038/sj.cr.7290259

[38] Goldminz, A.M., Au, S.C., Kim, N., Gottlieb, A.B. and Lizzul, P.F. (2013) NF-kappaB: An Essential Transcription Factor in Psoriasis. *Journal of Dermatological Science*, **69**, 89-94. https://doi.org/10.1016/j.jdermsci.2012.11.002

[39] Laplante, M. and Sabatini, D.M. (2012) mTOR Signaling in Growth Control and Disease. *Cell*, **149**, 274-293. https://doi.org/10.1016/j.cell.2012.03.017