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BIOLOGICAL ACTIVITY OF A THERAPEUTIC PASTE PREPARED FROM MEDICINAL AND AROMATIC PLANTS AS A BIOACTIVE FOOD SUPPLEMENT

Tulkinov Ikboljon Makhamad ugli
Andijan State University, Andijan Uzbekistan

Abstract

This article describes the bioactive therapeutic ingredients used in the formulation of the dietary supplement called “ASTOL” formulated from nettle (Urtica dioica) and sea buckthorn (Hippophae rhamnoides). This research focuses on the composition of the supplement chemically, and underlines the presence of over 60 essential macro- and microelements that are needed for balancing metabolism and maintaining physiological functions. One specific element to be evaluated in this research was the antioxidant activity of “ASTOL” that included the inhibiting of adrenaline autooxidation. Based on the testing results, the supplement has high antioxidant activity as shown in above 10% results, indicating possible active properties towards reducing oxidative stress and free radicals. Furthermore, the article explains toxicological and pharmacological findings from the evaluation as part of the work of “ASTOL” safety analysis. Acute toxicity assays were carried out, using the OECD 2002 process and administered orally in designated dosages. This study found that the LD50 value in the supplement is over 2000 mg/kg, proving the lower degree of toxicity for this material, as well as a very high tolerance to toxicity. Additional evaluations of its anti-inflammatory activity also produced encouraging results, suggesting “ASTOL” represents a valuable therapeutic candidate for inflammatory disorders.

Keywords

nettle, sea buckthorn, toxicity, biological activity, ASTOL, dietary supplement.

INTRODUCTION.

It is widely acknowledged that the need for compounds derived from natural plants has experienced a continuous upswing of late. Dietary aids formulated from these substances are distinguished from artificially created medications through their minimal harmful effects, compatibility with biological systems, and targeted physiological actions. At present, investigating naturally occurring substances exhibiting potent biological effects, alongside the creation of nutritional supplements leveraging these findings, stands as a highly pertinent field within contemporary scientific research [1, 2].

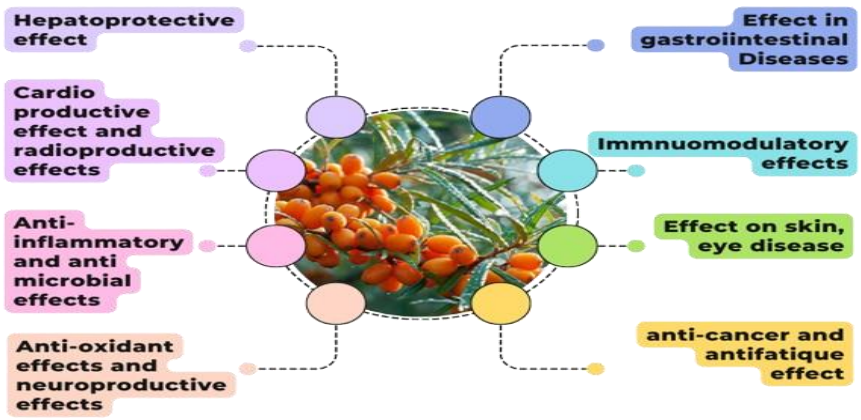

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Figure 1.

ASTOL nutritional addition consists of medicinal plant extracts composed of (*Urtica dioica*) and (*Hippophae rhamnoides*) extracts. We tested the efficacy of this supplement in vitro (at laboratory setting) on the basis of the in vitro test method for anti-adrenaline autooxidation. The results suggested that the measured antioxidant capacity of each tested formulation was over 10%, suggesting the above-mentioned antioxidant capacity to the formulation-treated formulation. Using inductively coupled plasma optical emission spectrometry (ICP-OES), the elemental content of the novel dietary aid was determined. A total of 61 different elements were found in the original botanical material as a result of investigation. The highest concentrations included phosphorus, potassium, and calcium among the macronutrients. The most commonly consumed micronutrients are iron, aluminum, manganese, and zinc than others. Concentrations of toxic elements were also found to be very low in the study. Results of scientifically proven calculations confirm that these plants have positive medicinal activity owing to high levels of the macro- and micronutrients [5]. Biobediotics including the vitamins, flavonoids, nutrients, sugars, amino acids and proteins of medicinal substances are essential for biochemical stimulation, metabolic reabsorption and for the functioning of vital organs and systems in the human body [3]. The primary objective was to systematically appraise the anti-inflammatory biological activities of dry powders extracted from *Urtica dioica* and *Hippophae rhamnoides* (ASTOL) in the context of the carrageenan-induced hind paw swelling model.

MATERIALS AND METHODS.

A powder-based extract (ASTOL) extracted from nettle and sea buckthorn was synthesized. For animal testing, rats' inflammation severity was determined by the extent of carrageenan-induced paw swelling distanced from the injection site. Rodents were placed in an acclimatization room for 10 days prior to testing in an enclosed environment. The housing had all the requirements according to the regulations (including temperature in the air): 25 ± 3°C, 12 hours of daylight, 30–70% air humidity. Standard care and unlimited water were utilized for the testing participants.

Following acclimatization, the rats were divided into four groups with 6 rats (n=6 per group). To evaluate the efficiency of superior animal extracts, laboratory rats were divided into four cohorts, with six animals in each cohort. Inflammation in the paw was stimulated by subcutaneous injection of 0.1 ml of 1% sodium carrageenan solution into the connective tissue layer of the right rear paw of each rat. The tested substances were orally administered an hour prior to the carrageenan injection. The control population received a measured volume of vehicle solution (5 ml/kg), and the experimental groups were prepared with ASTOL at doses of 50, 100 and 150 mg/kg. The degree of reduction in swelling was calculated in comparison with the control group defined as the baseline (0%). Paw diameter was measured using a digital caliper (manufacturer: Insize; model: 7140; specification 0–150 mm, 0-6 hours; origin: China). Measurements were done before injection of carrageenan (PV0) and after the injection at time points 1, 2, 3, 4, 5 and 24 h after administration (PVt). The percent of the edema that was ameliorated in the treated versus standard control group (compared to the untreated control group) was calculated as:

$$\%\text{Inhibition of edema} = \frac{(PV_t - PV_0)_{\text{control}} - (PV_t - PV_0)_{\text{treated}}}{(PV_t - PV_0)_{\text{control}}} \times 100$$

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The adopted methodology facilitated a precise assessment of how the anti-inflammatory effect of ASTOL varied with dosage, simultaneously guaranteeing replicability consistent with current benchmarks in pharmacological study.

RESULTS AND DISCUSSION.

For the determination of the direct detrimental effects of the proposed nutritional additive "ASTOL" trials in compliance with OECD 2002 guidelines on the assessment of acute hazard from ingested chemical agents, which were administered in a precisely calibrated level were performed. While comparing test and control group of the untreated baseline, no statistically significant mass decrement in the mice during the full periods of observation (1 week and 2 weeks) was seen, with a probability value greater than 0.05. The 50% mortality threshold (LD50) for "ASTOL" following a single ingestion at the level of 2000 mg/kg body weight, was established as LD50 > 2000 mg/kg, signifying the minimal hazardous nature of the product. These observations are summarized in Table 1.

Table 1.

Group	5 mL·kg^{-1} (saline)	Animals quantity per group/number of dead animals per group	Average body weight, g (M ± m)			LD_{50}, mg/kg
			Day 1	week 1	Week 2	
Sing	0.5 mL distilled water	5/0	20,0± 0,3	21,8 ± 0,3	22,6± 0,35	-
"ASTOL" dietary supplement	2000 mg/kg	5/0	19,7 ± 0,2	20,4 ± 0,25	21,8± 0,3	>2000 mg/kg

Note: *π < 0.05 compared to the control group.

The outcome of a single oral administration of ASTOL (a dietary supplement) at 2000 mg/kg to mice in accordance with OECD protocols showed chemical Class V, defined as practically non-toxic (LD_{50} values exceeding 2000 mg/kg) as the substance was identified. In order to stabilize the anti-inflammatory medicinal preparations, plant-derived sources of bioactive compounds with anti-inflammatory properties were the chosen sources. This included 50, 100, and 150 mg/kg of aqueous extracts from sea buckthorn and nettle (ASTOL). The anti-inflammatory activity shown is reported in Table 2 with extracts of botanical extracts at concentrations 50, 100 and 150 mg/kg (ASTOL), expressed as the hourly percentage of inhibition. By comparison the tabulated data shows that paw swelling reached its peak in the control group at 3 hours, yielding a 90.0 ± 8.8% increase compared to the baseline, and up to 23.4 ± 2.1% even at 24 hours. On the other hand, in the treated groups it became similar 3 hours thereafter when carrageenan entered the mix (peak edema); however, the supplement by 24 hours had virtually restored the physical size of the paw to the baseline level. Table 2. Effect of the dietary supplement ASTOL, derived from carrageenan and prepared from nettle and sea buckthorn plants, on paw edema expressed as a percentage relative to baseline (M ± m; n = 5).

Table 2.

Groups	"Inflammation inhibition, %"					
	"1 h"	"2 h"	"3 h"	"4 h"	"5 h"	"1 day"
Control	19,1 ± 2,9	38,2 ± 3,8	90,0 ± 8,8	56,8 ± 5,8	47,3 ± 4,3	23,4 ± 2,1
ASTOL 50 mg/kg	28,8 ± 3,6	51,3 ± 4,7	62,5 ± 5,9	51,3 ± 4,6	33,8 ± 3,2	15,3 ± 1,9
ASTOL 100	11,8 ± 2,7	47,2 ± 3,9	59,0±4,5	47,2 ±3,8	23,6±2,4	11,8 ± 2,2

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mg/kg						
ASTOL 150 mg/kg	11,8 ± 2,6	41,0 ±3,8	47,2± 4,1	35,4±3,4	35,4±3,6	8,9 ±1,6

Swelling of the paws involves increased vascular permeability, migration of cells into the paw area, and an accumulation of fluid where inflammation is occurring. Diameter of paw assessment is a standard technique used to determine the effectiveness of anti-inflammatory agents. A decreased diameter of the paw means lower inflammatory substances, such as IL-6 and TNF-α, and is an important indicator for successful anti-inflammatory intervention. Based on literature summarized in Table 2, we find the anti-exudative activity of all ASTOL supplements, across the dose range, varies from 34.4% to 48.0%. Except for the 50 mg/kg level, the effect was somewhat lower at 30.5% (Table 3). Anti-exudative activity of nettle and sea buckthorn plant extract (ASTOL) at different doses compared with the control (M ± m; n = 5).

Table 3.

	“Increase in paw size 3 h after induction, %”	“Anti-exudative effect, %”
Sing	90,0 ± 8,8	---
ASTOL 50 mg/kg	62,5 ± 5,9	30,5
ASTOL 100 mg/kg	59,0 ± 4,5	34,4
ASTOL 150 mg/kg	47,2 ± 4,1	48,0

CONCLUSION

Immune system modulation is one of the most important and evolving fields of contemporary biomedical research that focuses on this area at present. This ability to inhibit or regulate immune responses is very important because to treat inflammatory and allergic disease you need to be able to control the immune response in a very specific situation to suppress an immune system when necessary, meanwhile to treat immunodeficiency and infectious disease, you need to stimulate or boost immune function on the contrary. As a consequence, the quest for safe and efficacious natural sources of immunoprotective compounds has attracted remarkable scientific interest. Regarding anti-inflammatory properties, those were tested with three levels of the plant-based dietary supplement ASTOL made from nettle (Urtica dioica) and sea buckthorn (Hippophae rhamnoides). The results proved ASTOL has powerful anti-exudative activity due to its ability to reduce edema and inflammatory fluid build ups. Of the doses under study and controlled doses, the 150 mg/kg concentration exerted the superior biological effects, exceeding both those for lower and higher doses. As a result, this concentration was chosen as the dose for additional pharmacological and toxicological studies. ASTOL, with its high content of essential macro- and microelements, vitamins, flavonoids and other biologically active phytochemicals, is a strongly promising natural therapeutic agent. Its widespread bioactive constituents indicate it may be particularly relevant to the treatment of inflammation in the gastrointestinal tract. A follow-up full study is recommended to be conducted to investigate its mechanisms of action, therapeutic effects, and applications in clinic.

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