Hemorheological and Actoprotective Activity of *Serratula coronata* Extract in Exhausting Physical Exercise in Rats

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Studies on rat model of exhausting physical exercise showed that *Serratula coronata* extract administered in a dose of 150 mg/kg for 14 days exhibited hemorheological activity, which manifested in reduced blood viscosity and erythrocyte aggregation and in increased erythrocyte deformability. Actoprotective effect of the test extract was observed only in trained animals.

Key Words: *Serratula coronata extract, blood rheology, actoprotective activity*

Tissue oxygenation and microcirculation are the main processes limiting working capacity under conditions of intensive physical exercise [1]. Hemorheological properties of the blood is an important factor determining the intensity of blood supply at the level of microcirculation and oxygen-transporting function of the blood [6,8]. In this context, the search and development of drugs improving hemorheological parameters and increasing working capacity is an urgent problem. Ecdysteroids-containing plant preparations (*e.g.* extract from *Serratula coronata L.* in 40% ethanol) are promising in this respect. We previously demonstrated hemorheological activity of *Serratula coronata L.* extract (SCE) on *in vitro* models [5].

Here we evaluated hemorheological and actoprotective activities of SCE on the model of exhausting physical exercise (EPE) and the influence of previous training on the magnitudes of hemorheological and actoprotective effects of EPE.

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MATERIALS AND METHODS

Experiments were carried out on 40 Wistar rats weighing 180-250 g. The animals were divided into 5 groups: intact, untrained controls (control I), untrained experimental animals (experiment I), trained controls (control II), and trained experimental animals (experiment II). The experimental animals received SCE in a dose of 150 mg/kg through a gastric tube daily for 14 days; control animals received an equivalent volume of distilled water.

Control II and experiment II animals were subjected to treadmill training (30 min daily, 22.5 m/min treadmill rate) starting from day 7 of SCE administration. On day 14 of the experiment, all animals (except intact) were presented EPE (exhausting criterion was refusal from running despite electrical stimulation); the blood was taken from the common carotid artery under light ether anesthesia immediately after EPE. The animals were euthanized by ether overdose.

Blood samples were stabilized with 3.8% sodium citrate (9:1). Blood and plasma viscosity was measured on an AKR-2 rotation hemoviscosimeter. Reversible erythrocyte aggregation was studied by

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TABLE 1. Effect of Course Treatment with SCE on Blood Viscosity at Different Shear Rates during EPE in Untrained and Trained Rats (*M*±*m*)

Group	Blood viscosity (mPa×sec) at shear rates, sec^{-1}						
	3	5	7	10	50	100	300
Intact	7.3±0.1	6.7±0.2	6.3±0.1	6.0±0.1	4.4±0.1	4.2±0.1	3.9±0.1
Control I	8.3±0.2*	7.3±0.2*	6.7±0.1*	6.2±0.1*	4.6±0.1*	4.4±0.1*	4.1±0.1*
Experiment I	7.7±0.4	7.4±0.3	6.7±0.3*	6.3±0.2*	4.5±0.1	4.2±0.1+	3.9±0.1+
Control II	8.0±0.3	7.4±0.3	7.0±0.3	6.8±0.3	4.8±0.1	4.5±0.1	4.1±0.1
Experiment II	7.3±0.2	6.7±0.2+	6.4±0.1+	6.1±0.2+	4.7±0.1	4.3±0.1	4.1±0.1

Note. Here and in Tables 2-4: $p<0.05$ compared to *intact animals, +corresponding control.

the method of syllectometry on an MKMF-1 modified microcalorymeter connected to an N306 plotter [3]. Fibrinogen concentration in blood plasma was measured photometrically by the method of Klauss using a Cormay KG-4 hemocoagulometer. Hematocrit was measured by centrifugation in glass capillaries on a PC-6 centrifuge at 2000 rpm for 15 min. Erythrocyte deformability was evaluated by the method of ektacytometry [7]; the index of erythrocyte deformability was calculated as the ratio (L-H)/(L+H), where L and H are the maximum and minimum diameters of the first diffraction maximum.

The data were processed statistically using Student's *t* test, and nonparametric Mann—Whitney *U* test.

RESULTS

Hemorheological parameters in untrained rats (control I) subjected to EPE considerably differed from those in intact animals. For instance, blood viscosity at shear rates 3-300 sec^{-1} in untrained rats increased by 14% compared to intact animals (Table 1), erythrocyte aggregation increased by 15%, while erythrocyte deformability decreased by 22-47% at shear rates 360-890 sec^{-1} (Table 2). Changes in plasma viscosity, fibrinogen concentration, and hematocrit did not attain statistical significance from the intact group (Table 3). The mean duration of running in untrained rats subjected to EPE was 166±48 min (Table 4). This load led to pronounced hypoglycemia: blood glucose level was 2.8 mmol/liter, which was 1.5-fold lower than in intact animals. The concentration of lactate did not change, while the concentration of pyruvate significantly increased by 2 times compared to intact animals (Table 5).

Thus, single EPE induces increased blood viscosity syndrome (IBVS) in experimental animals primarily due to shifts in blood rheology parameters (decreased deformability and increased aggregation of erythrocytes).

Course treatment with SCE decreased blood viscosity at high shear rates 100-300 sec^{-1} in untrained animals subjected to single EPE (experiment I) by 5% compared to control I (Table 1). SCE treatment improved erythrocyte deformability at shear rates of 360 and 890 sec^{-1} by 13 and 15%, respectively, compared to the corresponding parameters in the control group (control I; Table 2) and decreased erythrocyte aggregation by 25% compared to the corresponding parameters in control animals. In rats treated with SCE we observed a moderate, but significant decrease in plasma fibrinogen concentration by 9% compared to rats of the control group (Table 3). The capacity to decrease fibrino-

TABLE 2. Effect of Course Treatment with SCE on Erythrocyte Deformability during EPE in Untrained and Trained Rats (*M*±*m*)

Group	Index of erythrocyte deformability, arb. units			
	90 sec^{-1}	180 sec^{-1}	360 sec^{-1}	890 sec^{-1}
Intact	0.146±0.010	0.218±0.012	0.321±0.009	0.432±0.027
Control I	0.110±0.004*	0.174±0.012*	0.242±0.017*	0.348±0.016*
Experiment I	0.104±0.012*	0.180±0.022	0.275±0.054+	0.400±0.063+
Control II	0.104±0.007	0.173±0.012	0.268±0.012	0.367±0.024
Experiment II	0.139±0.002	0.208±0.003	0.320±0.011+	0.427±0.012+

TABLE 3. Effect of Course Treatment with SCE on Plasma Viscosity, Fibrinogen Concentration, Hematocrit, Erythrocyte Aggregation Half-Time during EPE in Untrained and Trained Rats (*M*±*m*)

Group	Plasma viscosity, mPa×sec	Fibrinogen concentration, g/liter	Hematocrit, %	Erythrocyte aggregation half-time, sec
Intact	1.3±0.1	2.5±0.1	43±2	34±2
Control I	1.4±0.1	2.1±0.6	40±4	29±1*
Experiment I	1.4±0.1	2.0±0.5*	41±2	39±2+
Control II	1.4±0.1	2.6±0.1	42±1	29±6
Experiment II	1.3±0.1+	2.3±0.1	45±1+	30±2*

gen concentration is typical of ecdysteroid-containing extracts [4].

The mean duration of treadmill running during single EPE in untrained rats receiving SCE for 14 days was 165±65 min (Table 4). Course treatment with SCE in animals also prevented the development of hypoglycemia during single EPE. The concentration of pyruvate in the blood increased by 28% (similarly to control animals). Blood lactate content in these rats increased 3-fold (Table 5). This probably determines short running duration in untrained rats (Table 4).

Thus, course treatment with SCE reduces the degree of IBVS caused by single EPE in untrained animals. SCE reduces the severity of hemorheological disturbances due to its effect on erythrocyte functions (aggregation and deformability) and due to decreased plasma fibrinogen content. At the same time, SCE exhibited no actoprotective effects in untrained animals under conditions of single EPE.

In trained animals (control II), hemorheological parameters during single EPE were similar to those in intact animals. However, in trained animals EPE did not cause hemorheological disturbances, in contrast to untrained rats, in whom IBVS developed after EPE. Moreover, some hemorheological parameter improved under these conditions. For instance, erythrocyte aggregation in trained rats decreased by 45% compared to intact animals. Other hemorheological parameters in these animals did not differ from those in intact rats (Tables 1-3). The mean duration of running in trained rats subjected to EPE was 210±20 min (Table 4). During EPE, the concentration of glucose in animals of control II group decreased by 39%, the content of pyruvate increased 3-fold, and the concentration of lactate increased 8.5-fold compared to the corresponding parameters in intact animals (Table 5).

Thus, single EPE did not lead to the development of IBVS in trained rats; on the contrary, some

TABLE 4. Effect of Course Treatment with SCE on Body Weight and Running Duration during EPE in Untrained and Trained Rats (*M*±*m*)

Group	Initial body weight, g	Body weight before blood sampling, g	Body weight gain, g	Running duration, min
Intact	217±29	243±45	42±12	—
Control I	223±19	253±36	47±9	166±48
Experiment I	213±26	248±37	57±3	165±65
Control II	235±15	310±10	75±5*	210±20
Experiment II	240±10	314±14	74±7*	300±13+

TABLE 5. Effect of Course Treatment with SCE on Biochemical Parameters during EPE in Untrained and Trained Rats (*M*±*m*)

Group	Glucose, mmol/liter	Lactate, mmol/liter	Pyruvate, μmol/liter	Lactate/pyruvate
Intact	6.6±0.1	1.05±0.34	32±7	0.04±0.02
Control I	2.4±0.5*	1.07±0.38	65±18*	0.02±0.01
Experiment I	5.2±0.9*	3.07±1.58*+	55±11*	0.05±0.02
Control II	4.3±0.9*	3.4±2.8	49.3±17.0*	0.07±0.03
Experiment II	7.2±0.2+	1.0±0.1+	33.2±3.5*	0.03±0.01

hemorheological parameters even improved (*e.g.* erythrocyte aggregation decreased), which one more time confirms the positive effect of moderate physical work on rheological properties of the blood. Decreased blood viscosity and improved hemorheological parameters were observed in athletes after long-term exercises [2]. Accumulation of lactate and pyruvate attested to predominant utilization of glucose (glycogen) during physical exercise in animals trained in the specified regimen (running, 30 min daily for 5 days).

In trained animals receiving course treatment with SCE (experiment II), blood viscosity at shear rates 5-10 sec^{-1} decreased by 10% during single EPE (Table 1). In trained animals receiving SCE erythrocyte deformability at shear rates of 360 and 890 sec^{-1} increased by 19 and 16%, respectively, compared to the corresponding parameters in control II group (Table 2). Other hemorheological parameters in experiment II group did not differ from the corresponding parameters in control animals (Table 3). In trained animals receiving SCE, running duration during EPE increased by 30% (Table 4), the content of pyruvate decreased by 32%, and the content of lactate decreased by 70% compared to the corresponding parameters in control II group. Other hemorheological parameters in experiment II group did not differ from the corresponding parameters in control animals (Table 5).

Thus, course treatment with SCE in a dose of 150 mg/kg for 14 days in trained rats led to normalization of the hemorheological status during EPE and optimization of carbohydrate metabolism, which was seen from the utilization of lactate and pyruvate during physical work.

Our experiments showed that ecdysteroid-containing preparation SCE improved hemorheological status during EPE in both trained and untrained animals. However, actoprotective activity of SCE under conditions of EPE was observed only in trained animals.

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