BIOLOGICAL ACTIVITY OF CHLOROPHYLLIN SODIUM

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Chlorophyllin sodium (a water-soluble preparation of natural chlorophyll) stimulates growth and multiplication of yeast cells, and when injected into rats stimulates oxygen absorption by spleen, kidney, and liver tissues and exhibits an anti-inflammatory action.

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The study of the action of chlorophyll on the body, begun by Burgi [16], has shown that it possesses hemopoietic and tonic properties, speeds up granulation and epithelization, and has a powerful deodorizing effect [2, 4, 11, 13, 16, 19, etc.].

Experimental investigation of chlorophyllin sodium, a Soviet water-soluble preparation of chlorophyll [10], has revealed its hemopoietic action in posthemorrhagic anemia, benzene leukopenia, and radiation sickness [7], and also in hemolytic anemia [1]. Investigations have also been made of the stimulant action of chlorophyll and its derivatives on metabolism [15-17, 21-23, etc].

In the present investigation some of the biological properties of chlorophyllin sodium were studied: the effect of the preparation on growth and multiplication of yeast cells, on the intensity of oxidative processes, and its anti-inflammatory action.

EXPERIMENTAL METHOD AND RESULTS

Effect of Chlorophyllin Sodium on Growth and Multiplication of Yeast Cells. Experiments were carried out on a pure culture of the yeast Saccharomyces cerevisiae, races B-C, using a nephelometric method [14].

Chlorophyllin sodium was tested in 12 dilutions, from 1:50,000 to 1:10,000,000 (each dilution in 10-12 parallel tests). The investigation showed that the preparation stimulates growth and multiplication of yeast cells, and its action begins to be manifested in a dilution of 1:350,000, increasing in the higher concentrations. The biological activity of the preparation in a dilution of 1:100,000 was 0.958 extinction unit (control 0.0017 unit; $P < 0.001$).

Effect of Chlorophyllin Sodium on Oxidative Processes. The intensity of oxidative processes was determined in a Warburg's apparatus [8].

When injected subcutaneously into rats in doses of 0.05 and 0.5 mg/kg body weight, chlorophyllin sodium stimulates absorption of oxygen by tissues of the liver, kidney, and spleen, the greatest increase being observed in the spleen tissue (by 31.3% after 1 h and by 32.2% after 2 h in a dose of 0.05 mg/kg body weight, and by 43.5 and 27.2% respectively in a dose of 0. 5 mg/kg body weight compared with control animals). This can evidently be attributed to the participation of the spleen in hemopoiesis.

In experiments in vitro, chlorophyllin sodium in a concentration of 0.1 mg/ml caused no significant change in tissue respiration, but in a concentration of 1 mg/ml it produced a statistically significant decrease in oxygen absorption by the tissue, in agreement with published data [18].

Anti-inflammatory Action of Chlorophyllin Sodium. The basis for the study of the anti-inflammatory effect of chlorophyllin sodium was its action in promoting healing of ulcers and wounds described in the literature [2, 4-6, 11, 13, 20].

The following experimental models of aseptic inflammation were used in this part of the investigation: formalin arthritis in rats [12], turpentine inflammation in mice [3], and inflammation of the skin in rabbits caused by application of xylene [9].

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TABLE 1. Effect of Chlorophyllin Sodium on Development of Formalin Arthritis in Albino Rats, M ± m

	No. of animals	Volume of limb (in percent of initial) after			
		2 h	6 h	24 h	48 h
Control	24	155,4±1,78	171,5±2,36	179,2±2,14	158,0±2,22
Cholorophyllin sodium	24	143,8±1,38	154,6±1,87	164,2±2,57	152,5±2,74
R		<0,01	<0,001	<0,01	<0,01
ACTH	15	129,6±2,05	144,5±2,50	156,5±1,87	143,3±1,31
R		<0,01	<0,01	<0,001	<0,001

TABLE 2. Effect of Chlorophyllin Sodium on Barrier Properties of a 24-h Focus of Turpentine Inflammation in Mice

	No. of mice	Survived	Died
Control	30	27	3
Chlorophyllin sodium	30	19	11
Cortisone	30	5	25

Chlorophyllin sodium was injected subcutaneously in a dose of 0.5 mg/kg body weight 2 h before injection of formalin, and again 30 min and 3 and 24 h after injection. The action of the preparation was compared with that of ACTH (2 units per rat subcutaneously) injected at the same time (Table 1).

As Table 1 shows, chlorophyllin sodium reduced the volume of formalin edema of the limb in rats, especially in the first 24 h after injection of formalin, but this effect was less marked than that of ACTH.

In other experiments chlorophyllin sodium was injected subcutaneously into mice in a dose of 0.05 mg/kg body weight 2 h before injection and 5 h after injection of turpentine. Cortisone was used for comparison (1 mg per mouse; Table 2).

Table 2 shows that chlorophyllin sodium weakened the protective function of the inflammatory focus in relation to a lethal dose of strychnine (0.003 ml of a 1% solution) injected into the focus, although its action was much weaker than that of cortisone.

Experiments on 10 rabbits showed that 1 h after injection of chlorophyllin sodium the time of coloration of the papule was increased on the average by 1.72 min ($P < 0.05$) compared with the control (after injection of cortisone this difference was 3.41 min). Consequently, chlorophyllin sodium diminishes vascular permeability.

The experiments thus showed that chlorophyllin sodium stimulates growth and multiplication of yeast cells, and if injected subcutaneously into rats it increases the oxygen absorption of spleen, kidney, and liver tissues (this effect is not found in vitro). The preparation possesses an anti-inflammatory action.

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