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## ESTIMATION OF ANTIULCEROUS EFFECT OF MULTICOMPONENT PLANT EXTRACT IN EXPERIMENT

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#### ABSTRACT

The aim of the work was to estimate the antiulcerous effect of a new multicomponent plant extract derived from the following species of medicinal plant materials: leaves of *Plantago major* L., grass of *Gnaphalium uliginosum* L., rhizomes and roots of a *Inula helenium* L., flowers of *Matricaria chamomilla* L., roots of *Glycyrhiza glabra* L., grass of *Polygonum aviculare* L., grass of *Urtica dioica* L., fruits of *Sorbus aucuparia* L. in the ratio 3:3:3:2:2:1:1:1. Standardization of the extract was carried out in terms of flavonoids. There was used the Pauls index (PI), the index of antiulcerous effect (AE) and morphological evaluation of the gastric mucosa. In experiments on *Wistar* line rats with butadiene-induced injury of the stomach mucosa the marked antiulcerous activity of the multicomponent plant extract in a dose of 150 mg/kg has been established. The administration of the extract restricted the formation of ulcer defects which were more marked on the 14<sup>th</sup> and 21<sup>st</sup> days of observation. Such morphological features of activation of regenerative processes against the background of introduction of the given remedies as the new growth of vessels, cleanliness of the wound, inhibition of inflammatory reaction and active granulation were more expressed in rats treated with the extract and less distinct in rats treated with the preparations of comparison. The findings of the research confirm the expressed antiulcerous effect of the plant extract which is comparable to the effects of befunginum and ranitidine. The complex of biologically active substances contained in the extract promotes the acceleration of the ulcer healing due to its versatile influence on the main pathogenesis mechanisms of the given pathology. The findings of the research have shown that the tested extract has good prospects for creation of medicinal preparations for prophylaxis and treatment of gastric ulcer.

**Keywords:** multicomponent plant extract, model of the butadiene-induced ulcer, antiulcerous effect.

**Introduction.** On abundance, weight of a current, complications and mortality the stomach ulcer occupies one of the leading places among diseases of the digestive system [7, 8]. In a disease pathogenesis the main role is assigned to balance upset between factors of aggression and protection of a mucosa of a stomach and duodenum against the background of change of a neuroendocrine and immune regulation of a gastroduodenal zone [4, 9, 11].

The market of medicinal preparations with the proved antiulcerous activity exceeds 500 names, at the same time the problem of effective therapy is

far from the permission. At treatment antiulcerous tools observe development of aggravations and emergence of a recurrence in 30-80% of cases, the complicated stomach ulcer forms meet at 25-45% of patients, the resistance of gastroduodenal ulcers to pharmacotherapeutic influence meets at 15-25% of patients, the side reactions at reception of a number of medicines are observed at one third of patients. Therefore relevant is a problem of development of the effective, not having the side effect gastroprotective tools [3, 6, 10].

In this regard, the purpose of

our research was determination of antiulcerous activity of new complex plant extract.

#### Materials and research methods.

As object of researches served extract dry, received from the following types of vegetable raw materials: leaves of *Plantago major* L. (3 h), grass of *Gnaphalium uliginosum* L. (3 h), rhizomes and roots *Inula helenium* L. (3 h), flowers of *Matricaria chamomilla* L. (2 h), roots *Glycyrhiza glabra* L. (2 h), grass of *Polygonum aviculare* L. (1 h), leaves of *Urtica dioica* L. (1 h), fruits of *Sorbus aucuparia* L. (1 h). The received extract contains carotenoids,

polysaccharides, flavonoids, tanning agents, saponin, steroids, proteins, sesquiterpene lactones, mucilage's, pitches, organic acids, vitamins, macro- and minerals, essential oils and other natural connections. Standardization of extract is carried out on the sum of flavonoids.

Work is performed according to "The guide to carrying out preclinical researches of pharmaceuticals". Experiments are executed on 104 white rats males of the Wistar line with an initial weight of 180-200. Animals received from "Scientific center of biomedical technologies" and contained in conditions of the certified vivarium with the free access to a forage and water. Pharmacological researches were conducted according to "Rules of work with use of the experimental animals", "The rules adopted by the European convention on protection of the vertebrate animals used for the experimental and others scientific". Keeping of animals and design of researches are coordinated with ethical committee.

Beforehand on model of stressful ulcers experimental and therapeutic doses of the received extract with use of the antiulcerous index of Pauls which corresponded to 100-350 mg/kg are defined. Doses of 400 mg/kg had also above no advantages therefore all subsequent experiments are made with use of a dose - 150 mg/kg.

Antiulcerous activity of an aqueous solution of complex plant extract studied in an experimental and therapeutic dose 150 mg/kg and comparison medicines: in isoeffective doses befunginum (0.3 ml/kg) and ranitidine (50 mg/kg).

The antiulcerous activity of extract was studied in the conditions of model of "a butadiion ulcer". White rats in this experiment were distributed on groups: intact (8 rats); control (24 rats); experienced 1 (24 rats); experienced 2 (24 rats); experienced 3 (24 rats). The canker mucous a stomach was reproduced intraperitoneal introduction of butadiion in a dose by 100 mg/kg of 1 times a day within 3 days in a row. From the fourth day to skilled groups 1, 2, 3 entered into a stomach extract, befunginum, ranitidine in the specified doses, respectively, 1 times a day within 10 days. In monitoring to rats entered an equivalent amount of water cleaned in the similar mode. Intact animals served as additional monitoring. The euthanizing of animal skilled groups 1, 2, 3 and monitoring was carried out for the 7, 14 and 21 day since the beginning of experiences in  $\text{CO}_2$  to the camera. After opening of animals measured the area of ulcer defects in  $\text{mm}^2$  and also calculated

the index of antiulcerous effect of the studied extract and reference medicines [5, 8]. The Paul's Index (PI) was calculated on a formula:  $\text{PI} = \text{A}^* \text{V} / 100$ , where A – average quantity of ulcers on one animal; V – quantity of animals with ulcers in group. About the antiulcerous action (AA) of extract judged by the relation of PI in monitoring to PI in skilled group of animals ( $\text{AA} = \text{PI}_c / \text{PI}_e$ ); at  $\text{AA} = 2$  and more considered that extract and also reference medicines have antiulcerous effect [4,8].

Morphological researches of a stomach were conducted for the 7, 14 and 21 day since the beginning of experiences. For these purpose stomachs of rats fixed in 10% solution of neutral formalin, filled in paraffin, prepared cuts 5 microns thick and painted a hematoxylin and eosin [8]. By means of a microscope of Axistar plus (C. Zeiss) estimated a condition mucous, submucous and muscular layers, expressiveness of inflammatory reaction and activity of regenerator processes.

1. Statistical processing of the obtained data was carried out with use of the Statistic 6.0 software package (USA). The importance of differences between selections with the distribution coming to normal was estimated by means of "t" - a criterion of Student [1]. Distinctions accepted significant at  $P \leq 0.05$ .

**Results and discussion.** Influence of the received multicomponent plant extract on the course of butadiion stomach ulcer at white rats is studied.

Apparently from the data provided in table 1, introduction of extract limits the formation of ulcer defects which is most expressed for 14 and 21 days of experience. Reference medicines also reduced the sizes of cankers mucous a stomach, conceding by efficiency to extract. The expressed antiulcerous effect of extract on late lines of a course of pathological process, apparently, is caused by mucous accelerated by regeneration of an epithelium.

The index of antiulcerous effect of extract corresponds to 3.4; at introduction of befunginum – 2.5; and at ranitidine introduction it corresponds 2.0 (table 2).

Histologically in this experiment it is established that for the 7th day in monitoring the deep ulcer defect filled with necrotic masses is found (a desquamated epithelium, clots, muci-

lage's), around an ulcer of a fold mucous are thickened, infiltrate leukocytes. Edges of defect irregular, the expressed hypostasis and infiltration of mucous and submucous layers is observed by granulocytes, found lymphocytes in a small amount. Far from ulcer defect numerous erosion are visible. Against the background of introduction of extract at animals of skilled group 1 in an infiltrate of mucous and submucous lymphocytes dominate, folds mucous are hydropic, hyperemic, the extent of defect it is much less, than in monitoring, and around an ulcer found single erosion and dot hemorrhages. In skilled groups 2 and 3 also the extent of ulcer defect was less, than at rats of control group, observed also hypostasis and infiltration of mucous and submucous layers mainly leukocytes.

For the 14th day of an experiment at rats of control group the extensive and deep defect with a necrotic masses remained, the stomach wall in the field of an ulcer is thickened, infiltrate, the expressed inflammatory reaction with hypostasis of all layers was observed. In infiltrate macrophages, fibroblasts are already noticeable. At the bottom of an ulcer found new growths of vessels, hypostasis and the expressed hyperemia remained. Mucous pieces of iron were found in edges of an ulcer. In skilled group 1 processes of the fissile adhesion of defect with the granulation filling ulcer cavities are noticeable sites of an integumentary epithelium with the high content of mucilage are found. Against the background of introduction of reference medicines observed a similar trend with less expressed activity of regenerator processes.

21 day of experience in control group

Table 1

Influence of extract on ulcer defects at «butadiion» stomach ulcer at white rats

Groups of animals	Area of ulcer defects, $\text{mm}^2$		
	7 days	the 14th day	21 day
Intact	0	0	0
Control	84,2 $\pm$ 2,30	69,8 $\pm$ 2,80	51,6 $\pm$ 2,10
Experienced 1	66,1 $\pm$ 1,20*	46,8 $\pm$ 1,60*	14,2 $\pm$ 1,00*
Experienced 2	72,5 $\pm$ 1,30*	48,0 $\pm$ 1,70*	20,8 $\pm$ 0,90*
Experienced 3	70,3 $\pm$ 1,00*	51,0 $\pm$ 1,10*	25,2 $\pm$ 1,00*

Note: hereinafter \* - distinctions are significant at  $P \leq 0.05$ .

Table 2

Influence of plant extract on the course of «butadiion» stomach ulcer at white rats (21 day of experience)

Groups of animals	Quantity of rats with ulcers, %	Quantity of destructions on 1 rat	PI	AA
Intact	0	0	0	0
Control	100	12,6 $\pm$ 0,68	12,6	0
Experienced 1	50	5,5 $\pm$ 0,21*	3,7	3,4
Experienced 2	80	6,0 $\pm$ 0,24*	5,0	2,5
Experienced 3	78	7,5 $\pm$ 0,22*	6,2	2,0

of rats later observed the phenomena of partial clarification of ulcer defect from necrotic masses, hypostasis of all layers of a wall remains, a damage zone hyperemia is noticeable. In skilled group 1 processes of the fissile regeneration in a defect zone clearly are noticeable, seams practically filled defect, but the complete recovery mucous is noted that it is probably bound to a short course of introduction of extract. In groups with introduction of befunginum and ranitidine strengthening of reparative processes, and, more the first in morphological features in skilled group 2 - against the background of introduction of befunginum is also noted. The complete adhesion of ulcer defect with restitution mucous is also noted at animals.

Thus, the studied extract, befunginum, ranitidine have antiulcerous effect. The greatest activity is shown by extract in connection with existence in it of a wide range biologically of the active materials providing the fissile cell regeneration mucous and reducing the damaging action of butadionum at white rats that is bound apparently, with ability of the received extract to inhibit activity of cyclooxygenase 1 and 2 with the subsequent decrease in synthesis of prostaglandins [2, 4].

**Conclusion.** In general, data of the conducted researches confirm the expressed antiulcerous effect of the studied plant extract which is comparable to effects of befunginum and ranitidine in experiences on rats with damages mucous a stomach. Course introduction by an animal of extract and reference medicines in isoeffective doses is characterized by natural decrease in the index of Pauls and increase in the index of antiulcerous action – the main criteria of antiulcerous activity of medicinal preparations. The new growth of vessels, early clarification of a wound, restriction of inflammatory reaction, the fissile granulation which were more expressed when using extract were morphological features of activation of regenerator processes against the background of introduction of the specified tools. Rich complex biologically of the active materials which are available in extract promotes acceleration of an adhesion of ulcer defect thanks to its multilateral influence on the main pathogenesis mechanisms of a peptic ulcer. In fact, the system influence of extract counterbalancing factors of aggression and protection at its application that will be coordinated with literary data [5, 11]

is considered. The received results demonstrate antiulcerous effect of complex plant extract and are of great interest to clinical practice, reason expediency of its application as a part of the used technologies of treatment of patients with a peptic ulcer and also at the recovery stage that will increase effectiveness of the held treatment-and-prophylactic events.

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