THE INFLUENCE OF EXTRACTS FROM PRUNUS DOMESTICA ON DISORDERS OF INTESTINAL PERISTALSIS INDUCED BY BARIUM CHLORIDE ACTION IN MICE

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THE AIM OF THE WORK. To confirm by experimental studies the laxative activity of fiber-containing extract and extract with polysaccharide complex obtained from the Prunus domestica fruits in a model of intestinal peristalsis damage induced by barium chloride in mice, to identify the most effective extract among them and determine its effective dose.

MATERIALS AND METHODS. Dry extracts from the Prunus domestica fruits (fiber-containing extract (PEF) and extract with polysaccharide complex (PEPC)) were used in the experimental studies. The investigated extracts were obtained by the original method at the Department of Chemistry of Natural Compounds of the National University of Pharmacy. The study of the laxative effect of the PEF and PEPC extracts was conducted in the model of intestinal peristalsis damage induced by barium chloride in mice. The studied effect was determined by the rate of contrast mass passage through the intestines.

RESULTS AND DISCUSSION. The introduction of PEF and PEPC at doses of 75 mg/kg, 100 mg/kg, 200 mg/kg and the reference drug natrii picosulfas at a dose of 2.3 mg/kg abolished the spasm of smooth muscles of the stomach and intestine. It was evidenced by the lengthening of contrast mass pathway through the intestines of mice to the level of intact control. Moreover, in the case of the use of PEF extract, the intensity of the laxative activity had a dose-dependent manner: with increasing dose, the intensity of the action increased. At a dose of 200 mg/kg, in contrast to the doses of 75 and 100 mg/kg, the contrast mass pathway was statistically significantly higher than in the control group. The laxative effect of 200 mg/kg PEF extract was 29% (by percentage of the contrast mass pathway to the entire length of the intestine), which exceeded the maximum activity of 100 mg/kg PEPC extract (26 %) and was at the level of the reference drug 2.3 mg/kg natrii picosulfas (27 %).

CONCLUSIONS. The 200 mg/kg PEF extract exhibited laxative activity (29 %) on the model of intestinal peristalsis damage in mice induced by barium chloride. The studied effect of PEF extract was higher than maximum activity of the 100 mg/kg PEPC extract (26 %) and was at the level of the reference drug 2.3 mg/kg natrii picosulfas (27 %).
The aim of the work – to confirm by experimental studies the laxative activity of PEF and PEPC extracts obtained from the *Prunus domestica* fruits in a model of intestinal peristalsis damage by barium chloride in mice, to identify the most effective extract among them and determine its effective dose.

Materials and Methods. Dry extracts from the *Prunus domestica* fruits (fiber-containing extract (PEF) and extract with polysaccharide complex (PEPC)) were used in the experimental studies. The investigated extracts were obtained by the original method at the Department of Chemistry of Natural Compounds of the National University of Pharmacy. The phytochemical analysis showed, that the PEF extract contains homopolysaccharides, the complex of phenolic compounds (anticyanin and hydroxyl-cinnamic acids), organic acids, proteinogenic amino acids, and the PEPC extract contains a heteropolysaccharide complex, bound amino acids, organic acids [13, 14].

Standardization of the investigated extracts was performed by the content of neutral sugars at the Department of Chemistry of Natural Compounds of the National University of Pharmacy. The presence of the homopolysaccharides in the PEF extract and the heteropolysaccharides in the PEPC extract detected by their hydrolysis, which resulted in the formation of neutral sugars.

The study of the laxative effect of the PEF and PEPC extracts was conducted in the model of intestinal peristalsis damage induced by barium chloride in mice. The studied effect was determined by the rate of contrast mass passage through the intestines.

In the experiments, sixty-three outbred mice of both sexes were used. The animals were divided into five groups. The mice of the first group were given equivalent volume of distilled water via gastric tube – control group. The second group was control pathology (CP); the animals of this group were induced constipation without further administration of the test extracts and the reference drug. The mice of this group were given equivalent volume of distilled water via gastric tube. The third experimental group – animals with constipation treated with reference drug sodium picosulfas drops ("Picolax") via gastric tube. The fourth and fifth experimental groups – animals with constipation treated with the PEF and PEPC extracts via gastric tube.

Animals were deprived of food for 24 hours before testing. The mice of CP group were given 10 ml/kg of the 0,025 % barium chloride solution via gastric tube 30 minutes after administration of the investigated extracts. The PEF extract and PEPC extract were administered at doses of 75 mg/kg, 100 mg/kg and 200 mg/kg, and the reference drug Natrii picosulfas ("Picolax", drops, 0.3 ml/kg) at a dose of 2.3 mg/kg. A contrast mass – 20 % suspension of coal in 1 % starch paste – was introduced 30 minutes after administration of test samples. Each animal was given 0.6 ml contrast mass. Twenty minutes

(IIS), cancer, etc. [1, 2]. The IBS that can manifest in the form of functional constipation remains the current problem of modern medicine. The treatment of this disease necessitates a significant increase in health care costs for patients – direct (through IBS and its associated diseases) and indirect (due to increased days of temporary disability) [3, 4].

Constipation is a polyetiological, multifactorial symptom complex of general and gastrointestinal disorders. In recent years, there has been a significant increase in the incidence of constipation in patients of all ages. They are diagnosed in 30–50 % of the adult population and 3 % of children who visit a doctor. In addition, in the presence of chronic diseases of the digestive system – in 10–25 % of patients [5].

According to official data of the Ministry of Health of Ukraine in 2017, the number of first reported cases of diseases with intestinal damage function over 1 million was registered, among them 60 % were detected in women [6].

The laxatives are used in clinical practice. An analysis of the pharmaceutical market in Ukraine showed that laxative drugs are different in origin, chemical composition and mechanism of action. The national manufacturers produce 44 % of laxatives, international – 56 %. International laxatives includes 31 items. However, only six laxatives are herbal origin, three of which are mono-constituent and three are complex [7, 8]. Thus, the market analysis showed that in pharmacies of Ukraine there is a low choice of laxatives of herbal origin. In addition, most of drugs have side effects, and some of them can cause an addiction (e.g. natrii picosulfas). Research and development of new effective and safety herbal laxative is current task for pharmacy and medicine.

The *Prunus domestica* is one of the prospective plant. It is quite widespread in Ukraine and well known for its medicinal properties and application in folk medicine. It has the variety of pharmacological effects: laxative, hepatoprotective, anti-inflammatory, antioxidant, membrane-stabilizing, etc [9, 10, 11]. In addition, the absence of mono-component medicines based on *Prunus domestica* gives evidence for pharmacological studies as a perspective phytoobject for production of effective and safe remedy for treatment gastrointestinal diseases.

Previous experimental studies of the laxative properties of four extracts obtained from the *Prunus domestica* fruits on the model of loperamide-induced constipation made it possible to identify the most effective phytoobjects with the ability to stimulate the intestinal motility and thus accelerate the intestine emptying. Such extracts are the extract containing fibers (PEF) and the extract containing polysaccharide complex (PEPC) [12].

Therefore, for further experimental studies of the laxative effect of extracts selected by primary screening, the barium chloride-induced model of intestinal peristalsis damage in mice was used.
after administration of contrast mass, all animals were sacrificed. The whole intestine was isolated (from the stomach to the anal opening) and unfolded into one straight line. The total intestine length and the length of contrast mass pathway were measured. Obtained results expressed in percent [15].

The obtained experimental data were processed by the methods of variation statistics (mean (M), its standard error (m) or minimum (min) and maximum (max) sample values) using parametric (Newman-Keyless test) methods of analysis. The differences were considered statistically significant at $p<0.05$. For statistical analysis of the obtained data, we used the standard 

Results and Discussion. Intra-gastric administration of barium chloride to mice lead to spasm of smooth muscle fibers of the stomach and intestines, resulting in delayed evacuation of gastric contents and reduced the length of contrast mass path along the intestine (Table 1). The percentage of intestine with a contrast mass from the total length of the intestine of the control pathology group was significantly reduced by 26 % compared with control, indicating a slowing of intestinal peristalsis (Table 1).

The administration of PEF and PEPC extracts at doses of 75 mg/kg, 100 mg/kg, 200 mg/kg, and the reference drug natrii picosulfas at a dose of 2.3 mg/kg eliminated smooth muscle spasm of the stomach and intestines [18, 19, 20]. It evidenced by the increase in the pathway contrast mass through the intestines in mice, to the level of control (Table 1). Moreover, with the use of the PEF extract, the intensity of the laxative effect had a dose-dependent manner: with increasing dose, the activity of the extract increased. At a dose of 200 mg/kg, the contrast mass pathway was statistically significantly higher than in the control group. Thus, the laxative activity of the PEF extract at a dose of 200 mg/kg was 29 %, at doses of 100 mg/kg and 75 mg/kg – 22 % and 16 %, respectively.

The PEPC extract in this model pathology showed the maximum laxative effect at a dose of 100 mg/kg at the level of 26 % (Table 1). The laxative effect of both the investigated extracts does not statistically differ from the reference drug natrii picosulfas. However, unlike the PEPC extract and the reference drug, the PEF extract not only restored the peristalsis of the intestine disrupted by chloride, but also stimulated it. The percentage of the contrast mass pathway through the intestinal tract from its total length statistically significant increase compared with control group (Table 1).

The mechanism of the laxative effect of extracts from the Prunus domestica fruits is probably caused by presence of polysaccharides in their chemical composition, which are not absorbed and, with adequate hydration,

| Group of animals     | Dose, mg/kg | % of contrast mass pathway from length the intestines | Activity, % |
|---------------------|-------------|------------------------------------------------------|-------------|
| Control             | -           | 67.41±2.29                                           | -           |
| Control pathology   | -           | 59.87±2.34**                                         | -           |
| Natrii picosulfas   | 2.3         | 75.76±2.36**                                         | 27          |
| PEF                 | 75          | 69.21±2.61**                                         | 16          |
|                     | 100         | 73.00±3.37**                                         | 22          |
|                     | 200         | 77.26±2.37**                                         | 29          |
| PEPC                | 75          | 68.76±2.27**                                         | 15          |
|                     | 100         | 75.73±2.13**                                         | 26          |
|                     | 200         | 71.96±2.86**                                         | 20          |

Notes:
* – $p<0.05$ versus intact control group;
** – $p<0.05$ versus control pathology group;
n – amount of animals in the group.
increase the intestinal content, facilitating the activation of peristalsis. In this way, the transit of intestinal contents is activated, facilitating emptying. In addition, due to the inability of the polysaccharides to absorb, an osmotic gradient can be created that promotes fluid retention in the intestinal lumen. That leads to softening of fecal masses and mechanical stretching of the intestinal walls, increased volume of its contents, which is generally accompanied by stimulation of peristalsis. The described possible mechanisms are consistent with published data of literature [21].

Proved intensive laxative effect of the PEF extract probably related to the presence in its chemical composition of homopolysaccharides (59 %).

Conclusions. Experimental studies have confirmed the laxative properties of the PEF and PEPC extracts from *Prunus domestica* fruits in the model of intestinal peristalsis damage induced by barium chloride in mice. The PEF extract showed laxative activity at a dose of 200 mg/kg (29 %), which was higher than the maximum activity of the PEPC extract observed at a dose of 100 mg/kg (26 %) and was at the level of the reference drug natrii picosulfas at a dose of 2.3 mg/kg (27 %). Thus, the fiber-containing extract was identified as the most active between the two extracts. The dose of 200 mg/kg was determined as conditionally effective. Proved intensive laxative effect of the PEF extract probably related to the presence in its chemical composition of homopolysaccharides (59 %).

**Conflicts of interest:** authors have no conflict of interest to declare.

**Конфлікт інтересів:** відсутній.
Цель работы. Экспериментальными исследованиями подтвердить слабительную активность экстрактов, полученных из плодов сливы домашней СЭВ и СЭПК на модели нарушения перистальтики кишечника мышей, вызванное действием бария хлорида, выявить среди них наиболее эффективную субстанцию и определить ее условноэффективную дозу.

Материалы и методы. В экспериментальных исследованиях использованы сухие экстракты из плодов сливы домашней (экстракт, содержащий волокна (СЭВ) и экстракт, содержащий полисахаридный комплекс (СЭПК)), которые были получены по оригинальной методике на кафедре химии природных соединений Национального фармацевтического университета. Изучение слабительного действия экстрактов СЭВ и СЭПК проводили на модели нарушения перистальтики кишечника, вызванное действием бария хлорида по методу наблюдения за скоростью прохождения контрастной массы по кишечнику мышей.

Результаты и обсуждение. Введение СЭВ и СЭПК в дозах 75 мг/кг, 100 мг/кг, 200 мг/кг и препарата сравнения натрия пикосульфата в дозе 2,3 мг/кг устраивало спазм гладких мышц желудка и кишечника, о чем свидетельствовало удлинение пути следования контрастной массы по кишечнику мышей до уровня интактного контроля. Причем в случае использования экстракта СЭВ выраженность слабительной активности имела четкий дозозависимый характер: с повышением дозы выражение действия усиливалось. В дозе 200 мг/кг, в отличие от доз 75 мг/кг и 100 мг/кг, путь контрастной массы статистически значимо превышал в группе интактного контроля. Экстракт СЭВ проявлял слабительное действие в дозе 200 мг/кг (29 %), которое превышало максимальную активность экстракта СЭПК в дозе 100 мг/кг (26 %) и была на уровне препарата сравнения натрия пикосульфата в дозе 2,3 мг/кг (27 %).

Выводы. Экспериментальными исследованиями на модели нарушения перистальтики кишечника, вызванное действием бария хлорида, были подтверждены слабительные свойства экстрактов из плодов сливы домашней СЭВ и СЭПК. Экстракт СЭВ проявлял слабительную активность в дозе 200 мг / кг (29 %), что превышало максимальную активность экстракта СЭПК, наблюдаемая в дозе 100 мг/кг (25 %) и была на уровне препарата сравнения натрия пикосульфата в дозе 2,3 мг/кг (27 %). Таким образом, среди двух экстрактов было обнаружено наиболее активный экстракт СЭВ и определена его условноэффективная доза, которая составляет 200 мг/кг. Подтвержденный выраженный слабительный эффект экстракта СЭВ, вероятно, связан с наличием в его химическом составе гомополисахаридов (59 %).

Ключевые слова: желудочно-кишечный тракт; кишечник; моторно-эвакуаторная функция; функциональный запор; растительные волокна; плоды сливы домашней.

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