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**SCIENTIFIC
JUSTIFICATION
OF ACUTE TOXICITY
PARAMETERS
OF SEMI-FINISHED
PROTEINS
AND
CARBOHYDRATES
WITH FOOD
SYSTEMS'
STABILIZER**

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The topicality. Today, the use of traditional stabilizers, such as flour, starch, gelatin, is ineffective. To improve the properties of semi-finished protein-carbohydrate (NBV) was chosen stabilizer of food systems “Astri Gel”, consisting of 8 additives with an index of E and d-glucose. Each of the E-components, provided excessive intakes, can cause intoxication. Therefore, scientific studies aimed at determining the parameters of acute toxicity of NBV using the stabilizer “Astri Gel” are relevant. **The purpose of the article** is to identify possible manifestations of acute toxicity from the consumption of NBV with stabilizer of food systems “Astri Gel” and determine the class of its toxicity. **The research methods** on biological objects have been used (*in vivo*), experimental planning, the method of Probit-analysis of Litchfield and Wilcoxon. **Results.** The effect of food stabilizer has been investigated “Astri Gel” on the body of white linear rats. The general fluidity of a clinical picture at acute toxicity at laboratory animals after introduction of NBV with the “Astri Gel” stabilizer has been studied. Thus, no animal died during the experiment, only some of them experienced moderate poisoning due to the introduction of maximum doses of NBV (7.5 ... 9.0 g per rat). The dependence of “effect-dose” after consumption by rats of NBV with the stabilizer “Astri Gel” has been established on the basis of which parameters of acute

toxicity of the stabilizer have been calculated: indicators of lethal and effective doses, degree of its safety. Thus, according to the results of in vivo studies, the LD50 for NBV in terms of stabilizer “Astri Gel” was 17.87 g / kg (with estimated values of LD16 = 4.75 g / kg, LD84 = 67.20 g / kg, and LD99 = 395.95 g / kg), which corresponds to the fifth class of toxicity of additives and certifies its practical non-toxicity. **Conclusions and discussions.** The safety of the stabilizer for the mammalian body indicates its feasibility in the technology of food and semi-finished products, including NBV and dessert products.

Keywords: toxicity, semi-finished product, protein, carbohydrate, stabilizer, additive, “Astri Gel”.

The topicality of the problem

Formulation of the problem. Development of new combined products nowadays tends to decrease in their share of raw materials of animal origin. At the same time, increasing the share of plant components, although it allows to obtain semi-finished and finished culinary products with improved biological value and mineral composition, contributes to undesirable technological factors, such as stratification or instability of dispersed systems (especially in dessert products), reduction of shear stress, etc. To eliminate them, resort to the inclusion in the prescription mixtures of stabilizing components.

Today, the use of traditional stabilizers, such as flour, starch, gelatin, is inefficient, because they do not have all the properties necessary to create the structure of dessert products (Sefikhanova et al., 2020). To improve the properties of semi-finished products, the authors (Deynychenko et al., 2013) chose the stabilizer “Astri Gel” manufactured by “ASTRI”, Kyiv. It is a stabilization system, which includes the following components: sodium citrate (E331), citric acid (E330), potassium phosphates (E340), potassium citrate (E332), carrageenan (E407), pectin (E407), gum Arabic (E414), locust bean gum (E410), dextrose (d-glucose).

The safety of the food supplement is confirmed only if it does not cause acute and chronic toxicity, does not contain carcinogenic substances, mutagens, does not have teratogenic and gonad toxic properties (Golovko et al., 2015). Moreover, the daily amount of such substances that enters the body is crucial.

The state of solving the problem. Specialized international organization of the Joint FAO / WHO Committee of Experts on Food Additives and Contaminants (JECFA) regulates the activities of food manufacturers in the use of food additives. In Ukraine, they fall under the jurisdiction of the State Sanitary Supervision Department of the Ministry of Health.

Since the last decade, most scientists have been inclined to believe that the primary stage of hygienic regulation of the content of food additives in human foods and diets should be a preliminary toxic and hygienic evaluation (Zhang & Spallholz, 2011, Benko et al., 2012). So, nowadays all over the world researches directed on revealing of parameters of acute toxicological poisoning of living organisms not only because of consumption of food additives are carried out (Ghosh et al., 2019, Žabka & Pavela, 2018), but also to determine the extent of the some insecticides impact on pests of crops (Saba et al., 2018), cypermethrin on catfish (Akanksha, S., Kannez, Z., 2017), γ -radiation on some legumes (Veni et al., 2017), allopathic phenomenon (Dafaallah, 2020) etc. Computer software should be used to optimize the process of statistical processing of experimentally obtained data (Hermsen et al., 2011). To calculate the parameters of acute toxicity of NBV with stabilizer “Astri Gel” we used the software package Probit Analysis, recommended for this type of research (Dafaallah, 2020).

Unresolved issues. One of the main questions that arise in toxicological studies of any substance is the parameters of its acute toxicity, the potential danger. Knowledge of these parameters is necessary to establish the degree of danger of the additive, as well as for further research, which requires information on the maximum tolerated doses. The obtained information on the toxic properties of the new food additive is needed to determine the coefficient of its toxicity: the ratio of the dose corresponding to LD50 (the dose at which 50% of the studied animals die or experience non-lethal effects from the consumption of the additive) to therapeutic. The stabilizer of food systems “Astri Gel” contains 8 additives with index E, each of which, in case of excessive intake, can cause intoxication. Therefore, scientific research aimed at determining the parameters of acute toxicity of NBV using the stabilizer “Astri Gel” is relevant.

Purpose and research methods

The purpose of the article is to identify possible manifestations of acute toxicity from the consumption of semi-finished protein-carbohydrate with stabilizer of food systems “Astri Gel” and determine the class of its toxicity.

The methodological basis of the study is the process of establishing the possible toxicological effect of semi-finished protein-carbohydrate with stabilizer of food systems “Astri Gel” consumption *in vivo* with subsequent processing of test results to establish the toxicity class of the stabilizer.

Research methods are on biological objects (in vivo), experimental planning, Litchfield and Wilcoxon Probit analysis method.

Information base of research is scientific articles, patents, materials of international congresses and symposiums, scientific and practical conferences, normative and technical documentation.

The object of research is the scientific substantiation of the parameters of acute toxicity of semi-finished protein-carbohydrate products with the stabilizer of food systems “Astri Gel”.

Subject of research is a stabilizer of food systems “Astri Gel”, its toxicological indicators, semi-finished protein-hydrocarbons with stabilizer of food systems “Astri Gel”, indicators of their acute toxicity.

The scientific novelty of the obtained results is: substantiation of scientific parameters of acute toxicity of semi-finished protein-carbohydrate products with stabilizer of food systems “Astri Gel” and establishment of their influence on the body of white linear rats; method of determining the nature of the biochemical processes course in model laboratory animals after consumption of NBV with a stabilizer. Data on the safety class of “Astri Gel” food system stabilizer and ways to use it in food technology have been further developed.

Research results

The experiment was performed on 170 white linear rats with a body weight of 155 ... 175 g breeding vivarium DZ “Dnieper Medical Academy of the Ministry of Health of Ukraine”. The experimental animals were divided into 17 groups of 10 heads each. Conditions for keeping animals and providing them with food were in accordance with the normative requirements of the International Convention: at a temperature of 20 ... 22 °C and humidity 40 ... 60% (Zharmukhamedova et al., 2011).

Pre-starved animals (for 4 ... 6 hours) were orally administered NBV with Astri Gel stabilizer using a metal probe with oil at the end. NBV with stabilizer “Astri Gel” was administered once a day with the maximum permissible for oral administration volume – 1.0... 9.0 g per animal, or 6.25... 56.25 g / kg.

The animals have been observed for two weeks with the study of the clinical picture of acute experimental poisoning: continuously for the first day after the introduction of NBV with stabilizer “Astri Gel”; in the future – twice a day for 13 days.

The general status and behavior of animals have been recorded, the state of neuromuscular and autonomic functions, coat, eating food, water consumption. Particular attention was paid to the development of toxicities signs, assessed their severity, recovery time. The general clinical picture of the in vivo experiment is shown in table 1.

Tabl. 1. The general fluidity of the clinical picture of acute toxicity in rats with the introduction of PCs-fP with “Astri Gel” stabilizer

№ groups	Dose		Number of rats			Clinical picture
			total	with clinical effect		
	d / goal	g / kg		absent	was observed	
1	1,0	6,25	10	10	0	There are no deviations from physiological norms
2	1,5	9,38	10	10	0	
3	2,0	12,50	10	10	0	
4	2,5	15,63	10	9	1	One rat had a decrease in activity
5	3,0	18,75	10	9	1	
6	3,5	21,88	10	9	1	
7	4,0	25,00	10	9	1	
8	4,5	28,13	10	9	1	Two rats showed lethargy, drowsiness, depression
9	5,0	31,25	10	8	2	
10	5,5	34,38	10	8	2	
11	6,0	37,50	10	8	2	
12	6,5	40,63	10	8	2	
13	7,0	43,75	10	8	2	Three rats showed lethargy, drowsiness, depression
14	7,5	46,88	10	7	3	
15	8,0	50,00	10	7	3	
16	8,5	53,13	10	7	3	
17	9,0	56,25	10	7	3	

Source: own development

The clinical picture after the introduction of NBV with stabilizer “Astri Gel” in the stomach of white rats in toxic and effective (non-lethal) doses developed after 10 ... 12 hours. Clinical symptoms of acute poisoning of white rats by oral administration were not observed. Aggression and periods of arousal or increased motor activity in most individuals were not observed.

At the same individuals who underwent toxic effect, after the short period of excitement sharply expressed depression, drowsiness developed. It should be noted that no laboratory animals died during the experiment. Some rats only experienced

moderate poisoning due to the introduction of maximum doses of NBV with stabilizer “Astri Gel” (7.5 ... 9.0 g per rat).

Toxicity of NBV with “Astri Gel” stabilizer was determined by the following parameters: maximum tolerated dose LD0, average lethal dose LD50, as well as LD16 and LD84 to establish confidence limits of the average lethal dose LD50 and LD99 using Probit-analysis using computer software Probit Analysis v.2.0.0.6.

Data on the maximum tolerated (effective) dose of LD0, the average lethal (effective) dose of LD50, as well as LD16 and LD84 to establish confidence limits for the dose of LD50 are given in tables 2 and 3.

Tabl. 2. Initial data for determining the parameters of acute toxicity of PCs-fP using the “Probit Analysis” software (in terms of “Astri Gel”) (n=17, p>0,05)

Nº group	Number of rats	Review, r	Share of the total number of rats, Pemp (p), %	Dose “Astri Gel” (d), mg / goal
1	10	0	0,0	100,0±5,0
2	10	0	0,0	150,0±8,0
3	10	0	0,0	200,0±10,0
4	10	1	10,0	250,0±13,0
5	10	1	10,0	300,0±15,0
6	10	1	10,0	350,0±18,0
7	10	1	10,0	400,0±20,0
8	10	1	10,0	450,0±23,0
9	10	2	20,0	500,0±25,0
10	10	2	20,0	550,0±28,0
11	10	2	20,0	600,0±30,0
12	10	2	20,0	650,0±33,0
13	10	2	20,0	700,0±35,0
14	10	3	30,0	750,0±38,0
15	10	3	30,0	800,0±40,0
16	10	3	30,0	850,0±43,0
17	10	3	30,0	900,0±45,0

Source: own development

Tabl. 3. The results of determining the parameters of acute toxicity of PCs-fP (in terms of “Astri Gel”) (n=17, p>0,05)

Percentage rate, (p%),%	“Astri Gel” dose (Dose), g / kg	“Astri Gel” dose maximum (Dmax), g / kg	The dose of “Astri Gel” is minimal (Dmin), g / kg
1,0	0,81	1,76	0,05
16,0	4,75	6,74	2,88
50,0	17,87	174,92	10,43
84,0	67,20	713,01	24,06
99,0	395,95	854,88	71,55

Source: own development

The figure below shows a graph expressing the effect-dose relationship after consumption of NBV rats with “Astri Gel” stabilizer.

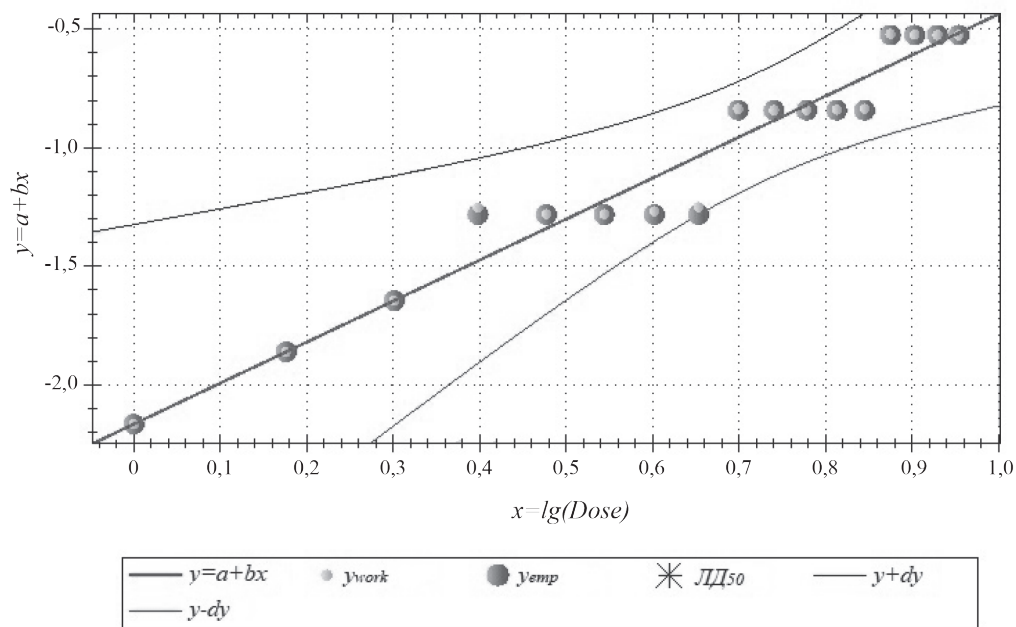


Fig. 1. Dependence of the shown effect after consumption by rats of PCs-fP with the «Astri Gel» stabilizer on quantity of the stabilizer
 Source: own development

The abscissa axis shows the logarithms of the doses of the stabilizer “Astri Gel”, and the ordinate axis is the value of the range. The straight line, which is drawn through the found points, by interpolation, allows determining the LD50 or any other dose at which the effect is observed.

Experimental sample points coincide completely with the calculated ones, which confirm the 95% reliability of the study results. The obtained results of Probit-analysis allow to establish the degree of safety of the stabilizer of food systems “Astri Gel” in accordance with the data given in table 4 (Sirohman & Zavgorodnya, 2009).

Tabl. 4. Toxicity of substances depending on the value of LD_{50}

Toxicity class	LD_{50} , mg / kg	Toxicity characteristics
1st	Less 5	Extremely toxic
2nd	5...49	Highly toxic
3rd	50...499	Moderately toxic
4th	500...4999	Low toxicity
5th	Over 5000	Practically non-toxic

Source: Syrokhman I. V. & Zavgorodnja V. M., 2009

The higher the LD50 value, the lower the acute toxicity of the food additive. According to the results of the experiment, it was found that the average value of LD50 for the stabilizer of food systems “Astri Gel” corresponds to 17.87 g / kg. Thus, according to Table 4, its toxicity class is fifth, i.e. it is practically non-toxic.

Conclusions and discussion of results

Thus, according to the results of *in vivo* studies, the fifth class of toxicity of the stabilizer of food systems “Astri Gel” was established, which testifies to its practical non-toxicity. The dependence “effect-dose” after consumption by rats of NBV with the stabilizer “Astri Gel” was established on the basis of which parameters of acute toxicity of the stabilizer were calculated: indicators of lethal and effective doses, its safety degree. The LD50 indicator for semi-finished protein-carbohydrate products in terms of the stabilizer of food systems “Astri Gel” was 17.87 g / kg (with calculated values of LD16 = 4.75 g / kg, LD84 = 67.20 g / kg, and LD99 = 395,95 g / kg), which proves its safety when consumed in moderate doses. In general, no laboratory animals died during the experiment, only some of them experienced moderate poisoning (lethargy, drowsiness, depression) due to the introduction of maximum doses of NBV (7.5 ... 9.0 g per rat). For its part, this affected the overall fluidity of the clinical picture of the experiment. In general, the safety of the stabilizer for mammals indicates its feasibility in the technology of food and semi-finished products, including NBV and dessert products. Data on the toxicology of the food system stabilizer “Astri Gel”, obtained in the course of the study presented in this work will form the basis for determining its daily allowable dose and in-depth clinical studies, such as biochemical studies of blood of laboratory animals.

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НАУКОВЕ ОБГРУНТУВАННЯ ПАРАМЕТРІВ ГОСТРОЇ ТОКСИЧНОСТІ НАПІВФАБРИКАТІВ БІЛКОВО-ВУГЛЕВОДНИХ ЗІ СТАБІЛІЗАТОРОМ ХАРЧОВИХ СИСТЕМ

Актуальність. На сьогодні використання традиційних стабілізаторів, таких як борошно, крохмаль, желатин, є малоефективним. Для покращення властивостей напівфабрикатів білково-вуглеводних (НБВ) був обраний стабілізатор харчових систем «Астрі Гель», що складається із 8 добавок з індексом Е та d-глюкози. Кожен з Е-компонентів, за умови надмірного надходження до організму, може викликати інтоксикаційні процеси. Тому наукові дослідження, спрямовані на визначення параметрів гострої токсичності НБВ із використанням стабілізатора «Астрі Гель», є актуальними. **Метою статті** є виявлення можливих проявів гострої токсичності від споживання НБВ зі стабілізатором харчових систем «Астрі Гель» та визначення класу його токсичності. При написанні статті використовувались **методи дослідження** на біологічних об'єктах (*in vivo*), планування експерименту, метод Probit-аналізу Літчфілда і Вілкоксона. **Результати.** Досліджений вплив стабілізатора харчових систем «Астрі Гель» на організм білих лінійних щурів. Вивчена загальна плінність клінічної картини при гострій токсичності у лабораторних тварин після введення НБВ зі стабілізатором «Астрі Гель». Так, у ході експерименту жодна тварина не загинула, лише деякі з них відчували помірне отруєння через введення максимальних доз НБВ (7,5...9,0 г на одного щура). Встановлена залежність «ефект-доза» після споживання щурами НБВ зі

стабілізатором «Астрі Гель», на основі чого розраховані параметри гострої токсичності стабілізатора: показники летальних і ефективних доз, ступінь його безпечності. Так, за результатами проведених досліджень *in vivo* показник ЛД₅₀ для НБВ у перерахунку на стабілізатор «Астрі Гель» склав 17,87 г/кг (при розрахункових величинах ЛД₁₆=4,75 г/кг, ЛД₈₄=67,20 г/кг, а ЛД₉₉=395,95 г/кг), що відповідає п'ятому класу токсичності добавок та засвідчує його практичну нетоксичність. **Висновки та обговорення.** Безпечність стабілізатора для організму ссавців свідчить про його можливість застосування у технологіях харчових продуктів і напівфабрикатів, зокрема НБВ та десертній продукції.

Ключові слова: токсичність, напівфабрикат, білок, вуглевод, стабілізатор, добавка, «Астрі Гель».

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НАУЧНОЕ ОБОСНОВАНИЕ ПАРАМЕТРОВ ОСТРОЙ ТОКСИЧНОСТИ ПОЛУФАБРИКАТОВ БЕЛКОВО-УГЛЕВОДНЫХ СО СТАБИЛИЗАТОРОМ ПИЩЕВЫХ СИСТЕМ

Актуальность. На сегодняшний день использование традиционных стабилизаторов, таких как мука, крахмал, желатин, малоэффективно. Для улучшения свойств полуфабрикатов белково-углеводных (ПБУ) был выбран стабилизатор пищевых систем «Астри Гель», состоящий из 8 добавок с индексом Е и d-глюкозы. Каждый из Е-компонентов, при условии

избыточного поступления в организм, может вызвать интоксикационные процессы. Поэтому научные исследования, направленные на определение параметров острой токсичности ПБУ с использованием стабилизатора «Астри Гель», актуальны. **Целями статьи** являются выявление возможных проявлений острой токсичности от употребления ПБУ со стабилизатором пищевых систем «Астри Гель» и определение класса его токсичности. При написании статьи использовались **методы исследования** на биологических объектах (*in vivo*), планирования эксперимента, метод Probit-анализа Литчфилда и Вилкоксона. **Результаты.** Исследовано влияние стабилизатора пищевых систем «Астри Гель» на организм белых линейных крыс. Изучено общее протекание клинической картины при острой токсичности у лабораторных животных после введения ПБУ со стабилизатором «Астри Гель». Так, в ходе эксперимента ни одно животное не погибло, лишь некоторые из них испытывали умеренное отравление из-за введения максимальных доз ПБУ (7,5 ... 9,0 г на одну крысу). Установлена зависимость «эффект-доза» после употребления крысами ПБУ со стабилизатором «Астри Гель», на основе чего рассчитаны параметры острой токсичности стабилизатора: показатели летальных и эффективных доз, степень его безопасности. Так, по результатам проведенных исследований *in vivo* показатель ЛД₅₀ для ПБУ в пересчете на стабилизатор «Астри Гель» составил 17,87 г/кг (при расчетных величинах ЛД₁₆=4,75 г/кг, ЛД₈₄=67,20 г/кг, а ЛД₉₉=395,95 г/кг), что соответствует пятому классу токсичности добавок и свидетельствует о его практической нетоксичности. **Выводы и обсуждение.** Безопасность стабилизатора для организма млекопитающих свидетельствует о его возможности применения в технологиях пищевых продуктов и полуфабрикатов, в частности ПБУ и десертной продукции.

Ключевые слова: токсичность, полуфабрикат, белок, углевод, стабилизатор, добавка, «Астри Гель».