# EFFECT OF MULTICOMPONENT CEREAL MIXTURES ON GLUCOSE LEVEL IN BLOOD OF EXPERIMENTAL ANIMALS

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**Abstract:** Recipes of multicomponent mixtures of cereals with proteins of high biological value were developed. In experiments, 35 adult male Wistar rats were used. Prior to the experiment, all animals were fed with powdered milk, grain or grain waste, germinated oats, and comprehensive multivitamin preparations, in addition to the standard balanced diet. Against this background, blood was collected from the animals for biochemical studies (control group, n = 20). Blood collection from tail vein was performed under general anesthesia, according to the recommendations of the Federation of European Laboratory Animal Science Working Group. Animals were fed with viscous-texture porridge made from ternary mixtures (rice, peas, and buckwheat; rice, barley, and maize) and the five-component cereals (rice, barley, maize, buckwheat, and peas) for 30 days. The control group received a standard vivarium diet. Postprandial glycemic curves in all groups were compared with the response to administration of glucose in the amount corresponding to the diet carbohydrates content. Postprandial glycemia was significantly lower in all groups of animals receiving the experimental diets than in the group of animals who received aqueous solution of glucose directly in the stomach by gavage at the rate of 0.03 g/g total weight (glucose tolerance test, GTT). Baudouin hyperglycemic factor was 1.52 for the control group, and in the range of 1.07–1.10, for the experimental groups. The glycemic index was 76.2 and 53.6–55.9, respectively. The results evidence that the products prepared from multicomponent mixtures of cereals belong to the products with low glycemic index.

Keywords: multicomponent mixtures of cereals, postprandial glycemia, glycemic index

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# **INTRODUCTION**

Excess influx of carbohydrates with food, especially against the background of obesity, impaired glucose tolerance, or metabolic syndrome, leads to progression of the phenomena [1]. However, not only the amount of carbohydrates, but also their qualitative composition influences the rate of absorption and, finally, glucose level in blood [2]. Simple food carbohydrates are known to be rapidly absorbed from the gastrointestinal tract increasing glucose concentration in blood [3]. After sharp increase in secretion and synthesis of insulin, glucose is eliminated by liver and muscle tissue and transformed to glycogen; then, glucose concentration in blood decreases and hunger develops [4]. Complex polysaccharides, resistant starch, and food fibers slow down glucose absorption; their absence or partial lack from the diet leads to small volume of food and consequently need for new meal, i. e. overnutrition [6, 7]. The term glycemic index was introduced as a function of the rate of carbohydrate absorption; this provided for the possibility for patients diagnosed with diabetes mellitus or other metabolic disorders, as well as healthy population, to correct diet, choosing products that do not induce high glycemia levels.

Under normal conditions, glucose is the major energy substrate for most tissues in the human and animal organisms. Its concentration in blood is an integral index, which is determined by the rate of glycogen formation from non-carbohydrate precursors, influx of carbohydrates with food, absorption in intestines, utilization by tissues, and excretion. Carbohydrate homeostasis may be referred to one of the perfect and most complexly regulated ones, controlled by both nervous and humoral effects. As a rule, concentration of glucose in blood of laboratory rats varies from 4.5 to 6.4 mmol/L and remains within this range even upon prolonged starvation. An important role in the maintenance of the constant glucose level in blood belongs to metabolic pathways through which glycogen, mainly deposited in liver and muscles, is synthesized and broken down. Hydrolysis of glucose-6phosphate, generated in liver from glycogen, is well known to serve a constant source of glucose. Metabolic pathways of its utilization in organism are described in details in a number of works [8, 9]. Glucose oxidation in a cascade of anaerobic glycolysis reactions is practically the only source of energy for such tissues as nervous tissue, renal medulla, seminal glands, and erythrocytes [8, 9], while other tissues possess the ability to use both glucose and fatty acids, ketone bodies, and other products of oxidative metabolism as energy substrates.

Cereal porridges may be considered as a complex of polysaccharides (or "slow carbohydrates"), proteins, monosaccharides, food fibers, and relatively small amount of fat. In the process of hydrolysis in the gastrointestinal tract they are cleaved to accessible forms of metabolic substrates that are transported to tissues and organs by blood. The faster the product is cleaved to simple carbohydrates, the higher is its glycemic index. Glucose with a glycemic index of 100 is considered an etalon. Due to the abundance of diabetes type I and II and a number of other metabolic disorders, functional products decreasing the burden of pancreas are required. Usually dietary actions include complete or partial disallowance of food with high glycemic index. We assume that use of cereal mixtures components of which are rich with amylose (legumes) or viscous food fibers (peeled and pearl barley, oatmeal—sources of  $\beta$ -glucans) may considerably widen the assortment of dishes and thus improve the quality of life of diabetes and metabolic syndrome patients.

The aim of the work was to develop multicomponent cereal mixtures that would have low or intermediate glycemic index.

### MATERIALS AND METHODS

Thirty five adult male Wistar rats weighing 200-300 g that were obtained from the vivarium of the Siberian Branch of the Russian Academy of Sciences and maintained in the vivarium of the Novosibirsk State Medical University according to the rules established by the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes were used in the experiment. Prior to the experiment, all animals were fed with powdered milk, grain or grain waste, germinated oats, and comprehensive multivitamin preparations, in addition to the standard balanced diet (pelleted feed PK120-3, according to the administrative order no. 179 of the Ministry of Health of USSR, 10.10.1983). Against this background, blood was collected from the animals for biochemical studies (control group, n = 20). Blood collection from tail vein was performed under general anesthesia, according to the recommendations of the Federation of European Laboratory Animal Science Working Group [10].

In function of the diet, the animals were divided into three groups, 10 animals each, and were ranged according to body weight to provide for the similarity of the parameter between the groups. Independently of the diet (Table 1), animals were kept in individual cages with free access to food and water.

After 15 and 30 days of feeding with one of the experimental diets (nos. 1, 2, or 3), blood was again collected from the animals for biochemical studies.

Postprandial glycemia was studied once, after 30 days of feeding with one of the diets. In four groups of animals, 5 male Wistar rats each, glucose concentration in blood upon 13 h starvation was measured and they gained access to forage (30 g per animal) or vivarium

standard diet feed in case of the control group. Postprandial glycemia was determined after 30, 60, and 120 min. In another group of animals on a standard vivarium diet, glucose aqueous solution was introduced directly to stomach through a gavage at the rate of 0.03 g/g weight (glucose tolerance test, GTT); glycemia level was measured at the same time points as in other experimental groups.

Glucose concentration in blood was determined by a glucose oxidase method using an EKSAN-Gm automated analyzer. The method is based on specific oxidation of glucose under the effect of glucose oxidase exhibiting high substrate specificity toward glucose.

To prepare porridges, three-component cereal mixtures were used for diet 1 (rice + peas + buckwheat) and diet 2 (rice + barley + maize) and a five-component mixture, for diet 3 (rice + barley + maize + buckwheat + peas).

Biological value of the cereal mixture proteins was calculated according to the technique proposed by Kovalev et al. [11], which allows for calculation of rational ratio of products at various combinations and determination of their biological value. The method allows for explanation of the four types of curves (according to Bressani's classification). It is based on the fact that not all essential amino acids are utilized at quantities adequate to the amino acid that has the lowest score. The remaining fraction determines the nonutilized protein ( $\Delta P$ ). The choice of optimal variant of combinations may be performed using the plot based on the linear dependence of non-utilized protein fraction ( $\Delta P$ ) from the protein ratio or products in the food mixture.

Parameter	Standard	Diet 1	Diet 2	Diet 3
Proteins, g	4.0	2.1	1.4	2.4
Fats, g	3.4	0.3	0.2	0.4
Carbohydrates, g	9.4	8.3	9.6	8.7
Energy content,	82.1	44.2	45.3	47.9
kcal				
Mass, g	50	50	50	50

Table 1. Composition and energy content per one rat

The results were processed using the Statistics 6.0 software package.

#### **RESULTS AND DISCUSSION**

In contrast to the standard vivarium diet, experimental animals received viscous porridge prepared from the relevant cereal mixtures without additives. Observations on animals revealed an interesting specific feature of group 2, which was the increased aggressiveness of the animals in the group. When taken out of the cage, the rats adopted defensive position and attempted to bite the researcher. Nevertheless, body weight of the animals of group 2 increased by day 15 of the study ( $P \le 0.02$ ). Similar results were obtained in groups 1 and 3 (Table 2).

Despite the considerable difference in qualitative composition of the diets and their energy value, all animals gained weight by day 30 of the experiment, which is explained by free access to the food. It should be noted that the percent of weight gained in group 2, receiving less protein and fat, was the lowest. Probably,

this explains behavioral reactions of the animals distinguished by aggressiveness (Table 2).

		Body weight, g		
Diet	Days of study			Weight gain, %
	Control	15	30	
	n = 30	n = 10	n = 10	
Standard		$278.5 \pm 3.1*$	$306.0 \pm 4.5*$	5.3/15.7
1	$264.4 \pm 5.5$	$270.9 \pm 3.3$	$288.4 \pm 8.4*$	2.5/9.1
2		$279.8 \pm 5.6^{*}$	$298.6 \pm 5.4*$	5.8/12.9
3		$280.5 \pm 3.9*$	$300.7 \pm 3.3*$	6.1/13.8

**Table 2.** Dynamics of body weight in experimental animals  $(M \pm m)$ 

Notes: \*statistically significant difference if compared to control group,  $P \le 0.02$ , LSD test.

After starving overnight, on day 15 and 30 of the experiment, glycemia level was determined; it was found to be significantly higher in animals on diets 1-3 (Fig. 1).



Fig. 1. Effect of the diets on glycemia in rats. \*Significant differences if compared to the control,  $P \le 0.05$ , Wilcoxon test.

We consider that the ratio between the accessible and non-accessible polysaccharides in the multicomponent porridges promoted the maintenance of a higher level of glycemia in fasted animals, which is in agreement with the literature data [7].

Calculations demonstrated that carbohydrate content in the diets was 0.03 g/g weight; therefore, it seemed interesting to study the glycemic curve progression after feeding.

After eating 30 g porridge and feed, the ascending segment of the glycemic curve in the control group evidenced rapid absorption of the glucose formed (Fig. 2).



Fig. 2. Postprandial glycemia and glucose tolerance test in experimental animals (M  $\pm$  m). \* significant differences if compared to 0 min, P < 0.01, Wilcoxon test.

The observed fact could be considered as unfavorable, since beta cells of pancreas not only secrete insulin into blood, but also synthesize it in large amounts in response to rapid influx of glucose. The load on the insulin apparatus of the pancreas increases. However, postprandial glycemia corresponded to physiologically normal values in rats [12]. The level of glycemia maintained in the course of the whole observation period and after 2 h returned to the initial values evidencing prolonged digestion of feed and gradual absorption of carbohydrates.

In animals fed with porridges, blood glucose levels practically did not change during the observation period. Glycemia gain was low if compared to the control group, and by 60 min glucose concentration returned to the initial values. Based on this, we conclude that porridges made of multicomponent cereal mixtures are characterized by low glycemic index.

It seemed important to compare postprandial glycemic curves in all groups with the response of a separate group of animals to administration of glucose at the same amount as in the relevant diet. Since rats in this group received standard vivarium diet, the glycemia value in the fasted state was the same as in the control group (Fig. 2). After administration of glucose, its blood concentration reached peak values by 30 min and exceeded considerably the values in animals of groups 1–3 (P  $\leq$  0.01). The rate of glucose absorption, judging from the ascending segment of the glycemic curve, was the highest if compared to all other groups. The peak concentration was registered by 30 min, which is in agreement with the results of other authors [13, 14]. By 60 min, glucose content in blood decreased considerably, but exceeded the values in all other groups (P  $\leq$  0.01). After two hours, glucose content returned to the initial values. One of the parameters describing the state of carbohydrate exchange is the Baudouin coefficient, or hyperglycemic coefficient, which is the ratio of glucose content after 30 or 60 min (the higher value is chosen) to its level under fasted condition (normally, below 1.7).

The obtained results evidence that carbohydrate exchange and values of the Baudouin coefficient in all groups, with the exception of those receiving glucose solution, were within the physiological ranges [14, 15].

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Diet	Hyperglycemic coefficient
Standard vivarium diet (control)	$1.52 \pm 0.03$
Diet 1	$1.10 \pm 0.06$
Diet 2	$1.10 \pm 0.07$
Diet 3	$1.07 \pm 0.03$
GTT	$1.98 \pm 0.11$

The calculations allow for a conclusion that all porridges from cereal mixtures may be referred to products with low glycemic index (Table 4). The conclusion is in agreement with a number of diet recommendations, according to which values of  $\geq$ 70 characterize products with high glycemic index, 55–69,

intermediate one, and  $\leq$ 55, low glycemic index [16].

**Table 4.** Glycemic indexes of the experimental animal diets  $(M \pm m)$ 

Diet	Glycemic index
Standard vivarium diet (control)	$76.2 \pm 4.35$
Diet 1	$55.6 \pm 3.1$
Diet 2	$55.9 \pm 3.4$
Diet 3	$53.6 \pm 1.5$

Therefore, based on the studies, precooked food from cereal mixtures 1, 2, and 3 may be recommended for arrangement of functional nutrition that provides for consumption of products with low glycemic index.

# REFERENCES

1. Lobykina, E.N., Koltun, V.Z., and Khvostova, O.I., Glycemic index of products and its application in dietary therapy of obesity, *Voprosy pitaniya (Nutrition Issues)*, 2007, vol. 76, no. 1, pp. 14–22.

2. Jenkins, D.J.A., Wolewer, T.M.S., and Jenkins, A.L., The Glycaemic Response to Carbohydrate Foods, *Lancet*, 1984, vol. 2, pp. 388–391.

3. Vloshchinskii, P.E., and Kolpakov, A.R., Structure of nutrition and glucose tolerance in the northerns, *Tekhnika i tekhnologiya pishchevykh proizvodstv (Methods and Technology of Food Production)*, 2011, pp. 17–21.

4. Jenkins, D.J.A., Lente Carbohydrate: A Newer Approach to the Dietary Management of Diabetes, *Diabetes Care*, 1982, no. 5, pp. 634–641.

5. Jenkins, D.J.A., Wolever, T.M.S., Jenkins, A.L., et al., The Glycaemic Index of Foods Tested in Diabetic Patients: A New Basis for Carbohydrate Exchange Favouring the Use of Legumes, *Diabetologia*, 1983, no. 24, pp. 257–264.

6. Jenkins, D.J.A., and Jenkins, A.L., The Glycemic Index, Fiber, and the Dietary Treatment of Hypertriglyceridemia and Diabetes, *J. Am. Coll. Nutr.*, 1987, vol. 6, no. 1, pp. 11–17.

7. Jenkins, A.L., Kacinik, V., Lyon, M., and Wolever, T.M.S., Effect of Adding the Novel Fiber, PGX, to Commonly Consumed Foods on Glycemic Response, Glycemic Index and GRIP: A Simple and Effective Strategy for Reducing Post Prandial Blood Glucose Levels – A Randomized, Controlled Trial, *Nutrition J.*, 2010, vol. 9, p. 58.

8. Kendysh, I.N., *Regulyatsyya uglevodnogo obmena* (Regulation of Carbohydrate Exchange), Moscow: Meditsyna, 1985.

9. Newsholme, E., and Start, C., Regulation in Metabolism, New York and London: John Wiley and Sons, 1973.

10. Kopaladze, R.E., Regulation of animal experiments: Ethics, laws, and alternatives, *Uspekhi physiol. nauk* (*Progress in Physiology Sciences*), 1998, vol. 29, no. 4, pp. 74–92.

11. Kovalev, N.I., Kartseva, N.N., and Krasnova, N.S., Ways to increased biological values of food products and cooled meals, in *Puti snizheniya poter' pishchevykh produktov pri khranenii i sovershenstvoveniya tekhnologii producktov obshchestvennogo pitaniya* (Methods to Decrease Losses of Food Products upon Storage and Improvement of Public Food Service Product Technology), Leningrad, 1982, pp. 160–173.

12. Nasanova, O.N., Effect of water extracts of common nettle, common burdock, dandelion, and common goat's rue on hyperglycemia and hyperlipidemia upon diabetes mellitus type 2, *Bulleten' sibirskoi meditsyny (Bulletin of the Siberian Medicine)*, 2011, no. 3, pp. 87–90.

13. Agarkov, D.Yu., et al., Hypoglycemic properties of the extract of *Gymnema sylvestre*, *Vestnik VolGMU (Bulletin of the Volgograd State Medical University)*, 2007, no. 1 (21), pp. 79–82.

14. Volchegorskiim I.A., Rassokhina, L.M., and Miroshnichenko, I.Yu., Insulin-potentiating effects of antioxidants in experimental diabetes mellitus, *Problemy endokrinologii (Problems of Endocrinilogy)*, 2010, no. 2, pp. 27–35.

15. Selyatitskaya, V.G., Palçhikova, N.A., and Kuznetsova, N.V., Activity of adrenocortical system in rats with high and low resistance to diabetogenic effect of alloxan, *Fundamental'nye issledovaniya (Fundamental Studies)*, 2011, no. 3, pp. 142–147.

16. American Diabetes Association. Nutrition recommendations and interventions for diabetes: a position statement of the American Diabetes Association, *Diabetes Care*, 2008, no. 31, suppl. 1, pp. 61–78.

