Main Factors Influencing The Processes Of Growth And Development Of The Organism Of Laboratory Animals

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Abstract:
The study of the influence of extreme conditions (high air temperature, insufficient protein diet, benzene) on the growth and development of the body of laboratory animals is an urgent problem in experimental medicine. **Purpose:** to study the mechanism of the influence of adverse factors on the peri- and postnatal periods of growth and development of laboratory rats, i.e. from birth to puberty. **Methods:** Studies were carried out on white laboratory rats in the postnatal period of both sexes, weighing from 5.78 to 208.5 g. The studied biochemical and morphological development indicators (body weight) in laboratory animals were compared with those of the control group. The study of the effect of exposure to benzene, high air temperature and insufficient amount of proteins in the diet at a dose of 112.0 mg/kg was carried out in the prenatal and postnatal periods of development of the body of laboratory rats for 105 days. Results: Under the influence of benzene, high air temperature and lack of protein in the diet, the growth of laboratory animals significantly lagged behind those of the control group of laboratory animals that received a normal protein content in the diet. A sharp decrease was observed from 21 and 28 days of age (by 6.4 and 12.2 g) - during the transition from breastfeeding to artificial. Conclusions: Conducted experimental studies on the influence of high air temperature, benzene, lack of protein in the diet, revealed a steady decline in the growth and development of laboratory animals in the postnatal period.

Key words: laboratory animals, ontogeny, control group, high temperature, benzene, protein deficiency in the diet.

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Introduction. During the development of the body, the most significant changes are experienced by the anabolic phase of metabolism, which includes various forms of biological syntheses, and, to a lesser extent, its catabolic phase [1].

An indicator of the intensity of anabolic processes is the value of the true growth rate [7, 10]. In most complex organisms, the growth rate changes with age, as a result of which, in the course of development, along with growth processes, complex processes of tissue differentiation occur [3, 5].

All forms of synthesis connected with each other weaken, albeit unevenly, over the course of individual development. At the same time, growth synthesis undergoes the greatest changes, associated with an increase in body weight and the formation of specific proteins in the processes of differentiation, mainly associated with the progressive phase of development [2, 8]. To a lesser extent, functional synthesis is weakened, which ensures the replenishment of specific, mainly protein, substances necessary for various forms of life during the stable and regressive phases of synthesis [4, 6].

In this regard, we studied the effect of extreme conditions (high air temperature, insufficient protein diet, chemicals) on the growth and development of the body of laboratory animals [9, 11].

The aim of the work was to study the mechanism of the influence of unfavorable factors on the pre- and postnatal periods of growth and development of laboratory rats, i.e. from birth to puberty.

Materials and methods. Experimental studies were carried out in accordance with the European Convention for the Protection of Vertebrate Animals used for Experimental or other Scientific Purposes (ETS 123, 2006). The studies were carried out on the basis of the Research Institute SGPE MH RUz in the laboratory of biomedical research in hygiene.

The studies were carried out on white laboratory rats in the postnatal period of both sexes, weighing from 5.78 to 208.5 g. Laboratory rats - females and males, weighing 180-200 g, were quarantined in a vivarium for 2 weeks; The control group of laboratory animals were kept under the same conditions as the experimental group. A comparative assessment of growth and development indicators of body weight in laboratory animals with indicators of the control group was carried out.

In the experiment on laboratory animals, the main attention was paid to studying the effect of adverse factors on the growth and development of the body, starting from fertilization, after childbirth and during lactation of the female. Breast milk is the most vulnerable to the effects of harmful factors, especially to chemicals and insufficient intake of essential nutrients in the diet during lactation [12].

The objects of the study were outbred white rats in the pre- and postnatal periods of their growth and development. After 12 days of fertilization and childbirth, during lactation, rats were exposed to adverse factors (benzene, high temperature and protein deficiency in the diet). The experimental group of laboratory animals were divided into 6 groups: group 1 - laboratory rats were kept under the influence of benzene until the 35th day of their life, and were also observed in the postpartum period, while the lactation period of the female was carried out under conditions of normal air temperature; group 2 - exposed to high air temperature during the period of feeding before sexual development of laboratory rats; group 3 - laboratory rats were administered benzene intragastrically after feeding with mother's milk at high air temperature (33-35 °C); group 4 - with a protein-insufficient diet during the feeding period of laboratory rats; group 5 - the influence of benzene on the growth of animals receiving an insufficient amount of proteins from mother's milk and young animals in the diet was studied; Group 6 - a control group of laboratory animals that were in vivarium conditions, receiving a general diet with a sufficient content of essential nutrients (proteins, fats, carbohydrates, vitamins and minerals).

To identify the impact of adverse factors on the offspring of laboratory animals, experimental studies included the perinatal and postnatal periods of growth and development of the organism.

Experimental laboratory animals (females and males) were exposed to harmful external influences before fertilization of females and after the onset of pregnancy (20-26 days) they were separated from males. Females continued to be exposed to the above factors before giving birth and during the period of feeding the young with breast milk.

After birth, the young of laboratory rats were exposed to unfavorable factors: benzene was administered with mother's milk, kept under conditions of high temperature and protein-malnutrition.

The slaughter of laboratory animals was carried out at each stage of ages, incl. female rats after lactation, by decapitation.

Thus, laboratory animals (females and males) were affected by unfavorable factors before and after fertilization, during lactation, after feeding, during periods of their growth and development.

The study of the reaction of the body when exposed to high air temperature, an insufficient amount of proteins in the diet with combinations of hydrocarbon - benzene at a dose of 112.0 mg/kg was carried out in the prenatal and postnatal periods of development of the rat organism for 105 days.

Animals exposed to high air temperature were preliminarily (before mixing females with males) adapted to conditions of high air temperature (15 days). To do this, laboratory animals were kept in cages in the summer and left for 4 hours (from 12 to 16 pm) in an open place under a canopy, where the ambient air temperature ranged from 38-41 °C for females, and for individuals of the litter of rats - 32-33 0C. During the stay of the animals on the thermal site, constant monitoring of the air temperature was carried out.

The protein-deficient diet consisted of the following products: bread, corn, barley, oats, oatmeal, hay, grass, root crops, bone meal. At the same time, meat, fish, fish oil, milk, meat meal and peas, which contain large amounts of proteins, were excluded from the general (standard) diet of the vivarium.

The obtained data were subjected to statistical processing on a personal computer using the Microsoft Office Excel (2016) software package with built-in statistical processing functions. Calculated indicators "M" - the arithmetic mean, "m" - the error of the arithmetic mean, "t" - Student's t-test. The statistical significance of the differences between the compared indicators was taken at the level of $p<0.05$.

Results and discussion. The impact of protein deficiency in the diet and the introduction of a chemical during the feeding of laboratory rats with breast milk is reflected in its quality.

As can be seen from the table, in the initial period after the birth of rats in the control group, the growth rate - body weight in rats was 5.78 ± 0.29 g, starting from the second day, on the 7th day (35.78 g) growth increased to 3 .61 g, on the next day 28 was 1.07 g per day; and on days 50 and 90 - 1.53 and 1.81 g, respectively. By the end (105 days) of the experiment, the increase in body weight of rats increased by an average of 1.93 g per day.

Changes in the body mass index of rats in ontogenesis under the influence of physical, chemical factors

Age periods, days		Control group under conditions of air temperature 22˚C	Benzene1/50 LD_{50}	38 ˚C	Benzene+ 38 ˚C	Benzene+ Protein deficiency	Protein deficiency
Lactation period	1	5,78±0,29	4,66±0,13	4,79±0,17	4,13±0,19	4,25±0,21	4,31±0,19
	2	6,0±0,24	4,91±0,17	5,13±0,17	4,72±0,24	4,40±0,21	5,23±0,14
	3	6,39±0,24	5,37±0,27	5,63±0,22	5,11±0,24	4,70±0,26	5,64±0,19
	4	6,72±0,18	5,69±0,33	6,05±0,26	5,50±0,29	5,10±0,26	6,14±0,19
	5	7,67±0,29	6,13±0,33	6,45±0,26	6,72±0,29	5,50±0,21	6,47±0,19
	6	8,44±0,41	6,63±0,33	7,05±0,32	6,40±0,35	6,0±0,24	6,82±0,14
	7	9,61±0,41	7,0±0,19	7,50±0,32	6,60±0,24	6,44±0,27	7,18±0,19
	14	13,72±0,35	11,75±0,73	13,2±0,84	10,61±0,35	9,19±0,53	11,68±0,48
	21	23,17±0,65	19,81±1,20	20,6±1,32	16,9±0,77	15,56±0,73	16,82±0,52
	28	35,78±0,59	30,43±1,63	29,24±2,06	26,81±0,87	23,56±1,80	23,55±0,67

	35	50,94±0,65	47,63±1,23	50,2±2,22	32,96±2,33	34,57±1,53	34,82±1,48
Puberty	50	82,17±1,94	75,64±1,20	20,6±1,32	16,9±0,77	15,56±0,73	16,82±0,52
	75	130,5±3,89	113,29±4,77	97,30±3,16	77,06±4,60	67,57±2,46	65,23±2,67
	90	169,11±4,00	145,29±4,93	129,9±4,48	104,06±5,47	94,83±5,39	85,05±3,62
Young growth	105	208,5±5,01	188,43±7,86	160,0±5,27	146,63±7,73	120,36±4,77	104,0±3,53

*Note: the significance of the value compared with the indicators of the control group: * - p<0.05; - p<0.01; * - p<0.001.*

Thus, the growth of laboratory rats in the control group in the first 28 days during breastfeeding was characterized by a slow pace, and from the 21st day after birth, their growth rate increased compared to the initial period, and by day 105, a progressive phase of weight gain in rats began.

At the birth of rats from a female exposed to chronic poisoning with benzene (112 mg/kg) during breastfeeding, the average body weight was 4.66 ± 0.13 g, which corresponded to 80.6% of the body weight of animals in the control group.

On the 28th day after birth, the body weight of rats treated with benzene increased to 30.4±1.63 g, and among the animals of the control group - up to 35.8±0.59 g, i.e. the difference in the average weight of the animals of the experimental and control groups was 15%. On the 50th day of the postnatal growth period, the average body weight of the experimental group of animals reached 75.6 ± 1.85 g, and in the control animals it was 82.2 ± 1.94 g. On the 75th day, 90th and 105th days of growth, the weight the body of the experimental group of rats was 113.4±4.77 g, 145.3±4.93 g and 188.4±7.86 g, respectively, and the difference with the control group was 13.4; 14.0 and 10.3% respectively.

Consequently, when benzene was exposed to the organism of animals in the initial period (up to 28 days), when benzene entered the body of rats from breast milk, the weight decreased sharply compared to the data of the control group, and in subsequent periods, the difference in the growth of the experimental and control groups decreased and amounted to from 6.5 to 14.0%.

On the basis of the data obtained, we believe that the daily administration of benzene to animals (up to 28 days with breast milk, and after - intragastric administration) at a dose of 1/50 LD0, the weight gain of young laboratory rats decreased.

Pregnant animals that were daily for 4 hours in conditions of high air temperature gave birth to rats with a lower weight compared to the control group. At the same time, on the first birthday, the body weight of the animals was 4.79 ± 0.17 g, almost 1 gram less than in the control group; With daily exposure of animals to benzene (before and after birth) under conditions of high air temperature, the weight of rat pups on the first day was 4.13 ± 0.19 g, which is 28.5% lower than the average weight of the control group. On the 6th day of the experiment, the body weight of the animals of the experimental group (rat pups) was 2 g lower than in the control group. On the 28th day of the experiment, this difference was 9.0 g, and on the 50th and 75th days of the experiment, respectively, decreased and amounted to 28.9 and 53.5 g. On the 105th day of the experiment, the weight of the rats became 62, 0 g, and in control animals by this time the body weight increased by 30%.

Thus, the increase in average body weight in animals of the experimental group when exposed to the chemical benzene at high air temperature decreased significantly more than when exposed to the chemical at the optimum air temperature.

A particularly sharp lag in the development of rat pups was observed with insufficient intake of protein through breast milk and from the 28th day on a diet with a protein deficiency throughout the entire period of the

experiment. In the initial period of up to seven days, the body weight of rat pups (which were fed by females receiving a protein-deficient diet) decreased by 9–19%, and on days 7, 14, and 21 the lag increased and amounted to 74.7%, respectively; 85.1 and 72.5% compared to the norm. Starting from the 28th day of age, the development of rats sharply decreased, and on the 90th and 105th days of the experiment, the body weight of the animals of the experimental group lagged behind the control group by 50%.

Consequently, with a lack of protein in the diet, the growth of the experimental group of animals lagged significantly behind the growth rates of the control group, which received a normal protein content in the diet. A particularly sharp decrease was observed from 21 and 28 days of age (by 6.4 and 12.2 g) - when there is a transition from breast milk to a formula-feeding diet.

In animals of the experimental group that received benzene with an insufficient amount of proteins in the diet, during all periods of the experiment, body weight and general development decreased from 24 to 44%, but in these rats, regardless of whether they were fed breast milk and an insufficient amount of protein in the diet during all periods, the body weight index decreased.

Conclusions. Conducted experimental studies on the influence of unfavorable factors (high air temperature, lack of protein in the diet, benzene) revealed a steady decrease in the growth and development of rat pups and rats after birth. A particularly sharp decrease in the body weight of the animals of the experimental group was observed under the influence of two factors - high air temperature with benzene and a protein-insufficient diet. Of the individual factors that significantly affect the delay in the development and growth of animals, was the use of a diet with a lack of protein substances.

Conflict of interest. All authors declare that there is no potential conflict of interest requiring disclosure in this article.

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