

## STUDYING THE THE EFFECT OF GAMMA ( $\gamma$ )–RADIATION ON AMYLOLYTIC ACTIVENESS IN THE BLOOD AND HOMOGENATE OF RATS AT DIFFERENT PHYSIOLOGICAL AGE STAGES

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### ABSTRACT

*The aim of the research lies on studying the the effect of gamma ( $\gamma$ )–radiation on amylolytic activeness in the blood and homogenate of young (2 months old), mature (6 months old) and old (36 months old) rats at different physiological age stages. In the experiments, the effect of  $\gamma$ –radiation (4 Gr) was carried out on the rats by means of radiation device "Luch" (Latvia) based on the <sup>60</sup>Co radioactive isotope. In the experiments, it was found that amylolytic activity in blood of young rats was decreased by ~20% in comparison with the control group within 20 days by the effect of  $\gamma$ –radiation, and its restoration in blood and liver homogenate until the value of control group on the 60–day. It was identified that as the result of  $\gamma$ –radiation the effectiveness of amylolytic activity in the blood of mature rats decreased by 58±0.3% in comparison with the control group on the 20–day, it approximated to the control group value on the 60–day, and amylolytic activity in liver homogenate was lower than that of the control group even on the 60–day after  $\gamma$ –radiation. It was found out that amylolytic activity in the blood of old rats reduced by ~20% in comparison with the control group on the 20–day after  $\gamma$ –radiation, it approximated to the control group on the 30–day, amylolytic activity in liver homogenate decreased in compariron with the control group after  $\gamma$ –radiation and it did not regenerate even on the 60–day. The results obtained can be explained by the significant difference in experimental animals' different digestive system enzyme secretion/incretion mechanisms at different age stage, as well as significant difference of their compensatory/adaptive mechanisms of responses to the effect of  $\gamma$ –radiation.*

**Keywords:** amylolytic activity, blood, liver homogenate, gamma( $\gamma$ )–radiation.

### INTRODUCTION

The functional activity of the organism is directly related to the state of the digestive system, including enzymes of the system, with homeostasis [Rothman et al., 2002; Korotko, 2005a; Korotko, 2005b; Korotko, 2016]. Under the influence of various internal/external factors, including stress factors, specific reactions as the type of adaptation/compensation in the body are induced. Numerous researchers have discovered that various stress factors have a significant impact on the homeostasis of these enzymes in biological organisms [Smirnov, 1990; Mirzarakhimova and Kadyrov, 2018]. Changes in the activity of digestive system enzymes in biological organism in the case of thermal shock [Romanov, 2004; Bozhenkova, 2004], psycho–emotional stress [Izatulin et al., 2005], hypokinesia [Smirnov, 1990; Kamskova, 2000; Stelnikova, 2008; Mirzarakhimova and Kadyrov, 2018] were analyzed in the researches.

Also, radiotherapy is widely used in the treatment of cancer in clinical practice. However, it was known that radiation rays have a negative effect on human organisms by the generation

of free radicals in the form of biological membrane structure/function disorder, modulation of physiologically important macromolecules (proteins, nucleic acids) [Gundarova et al., 2008; Erofeeva, 2008]. Therefore, it has an actual importance from the view-point of studying the mechanism of  $\gamma$ -radiation, improving the methods of radiotherapy utilization in practice. [Giardi et al., 2013; Mansour et al., 2014].

Based on the information given above, the purpose of this study is to investigate the effect of gamma ( $\gamma$ )-radiation on amylolytic activity in blood and liver homogenate of rats in their different physiological age stages.

## **MATERIAL AND METHODS**

### **Experimental animals and method of $\gamma$ -radiation**

White outbred rats in the weight range of ~90–270 gr. were used in conditions provided with standard nutrients and water supply during the experiments. In the experiments, the groups of white outbred rats of different experimental physiological age (body weight ~90–110 gr., 2 months old), mature (body weight ~170–200 gr., 6 months old) and old (body weight ~250–270 gr.; 36 months old) were used.

During the study, the standard method was used for the study of radiation fluctuations in the rats [Li, 2007].

In the field of oncologic medicine, a series of radiation therapeutic devices («GUT-Co-20», «GUT-Co-400», «RUM-3», «RUM-7», «Agat-C») were created on the direction of oncotherapy of  $\gamma$ -radiation and the telegram-therapeutic device "Luch-1" (Latvia) of distant electromechanical control unit was developed from 1971 to 1973 and started to be used in practice since 1975 [Kamneva and Jukova, 2007]. It should be noted that it is recorded that the "Luch" ( $^{60}\text{Co}$ )  $\gamma$ -radiation device is a standard device for use in scientific researches in this area [Ulyanenko et al., 2010].

The effect of  $\gamma$ -radiation on the experimental rats was performed by the help of radiation device "Luch" (Latvia) based on the  $^{60}\text{Co}$  radioactive isotope. The  $\gamma$ -radiation surface area was 20×20 cm and the focal length was 75 cm in the experiments. The experimental intensiveness of experimental animals is equal to 0.86–0.85 Gr/min. and the dose value is 4 gray (Gr).

### **Method of determining amylolytic activity**

In the experiments,  $\alpha$ -amylase activity in blood and liver homogenate is identified by the method developed by Smith-Roy and modified by Ugolev A.M. [Ugolev, 1969; Ugolev, 1985; Ershova and Volkova, 2008]<sup>i</sup>. The amylolytic activity was determined by photorocymetric method based on the amount of starch dissolved over the given time [Fisinin et al., 2017].

The preparation of rat liver homogenate was carried out by using the standard method [Ohkawa et al., 1979; Tkachenko, 2011].

### **Data Analysis**

The obtained results were statistically processed by means of special software packages – Excel 2003 (Microsoft Office, USA) and OriginPro c. 8.5 SR1 (EULA, Northampton, MA

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<sup>i</sup> Standard 20264.4–89. Ferment preparations. Methods of identifying amylolytic activity // Moscow. – Standards publishing, 1989.

01060–4401, USA). The results are given in the form of  $M \pm m$  results of the  $n$ -repeat experiments and  $M$  is the average arithmetic value and  $m$  is the default error value. The level of statistical reliability between the values of the control and experimental group was also calculated based on the Student  $t$ -criterion and in  $p < 0.05$  values it was evaluated as statistically reliable [Dospheov, 1985; Lakin, 1990].

## RESULT AND DISCUSSION

Thus, the effect of  $\gamma$ -radiation on amylolytic activity in blood and liver homogenate of different age rats was analyzed. The experiments showed that amylolytic activity in the blood of young rats almost unchanged by the effect of  $\gamma$ -radiation on the 1-day, it decreased by  $9 \pm 3\%$  in the liver homogenate in comparison with the control group (Table 1).

Also, it was found out that amylolytic activity in the blood of young rats decreased by  $\sim 20\%$  after the 20-day by the effect of  $\gamma$ -radiation. After the 60-day of  $\gamma$ -radiation, the ratio of amylolytic activity in the blood and liver homogenate of young rats was estimated to be close to the value of the control group (Table 1).

**Table 1. The effect of  $\gamma$ -radiation on the amylase activity in the blood and liver homogenate of young rats ( $M \pm m$ )**

Durability of hypokinesia (days)	Blood content (mg/min./ml)		Liver homogenate (mg/min./g)	
	Control group	Experimental group	Control group	Experimental group
1	$101 \pm 0,7$	$101 \pm 1,0(0,1)$	$2380 \pm 20$	$2157 \pm 79(0,1)$
	100	$100 \pm 0,9(0,1)$	100	$91 \pm 3(0,05)$
3	$97 \pm 1,5$	$86 \pm 1,2(0,001)$	$2470 \pm 41$	$1838 \pm 70(0,001)$
	100	$89 \pm 1,0(0,001)$	100	$75 \pm 2,1(0,001)$
10	$96 \pm 1,1$	$83 \pm 3,3(0,001)$	$2740 \pm 11$	$1760 \pm 22(0,01)$
	100	$86 \pm 3,7(0,001)$	100	$64 \pm 0,8(0,01)$
20	$101 \pm 1,5$	$81 \pm 3,0(0,001)$	$2860 \pm 15$	$1630 \pm 60(0,01)$
	100	$80 \pm 2,3(0,001)$	100	$57 \pm 2,6(0,001)$
30	$102 \pm 2,0$	$91 \pm 3,0(0,05)$	$2323 \pm 17$	$1710 \pm 26(0,01)$
	100	$89 \pm 2,6(0,01)$	100	$91 \pm 1,2(0,01)$
60	$101 \pm 2,0$	$98 \pm 3,2(0,1)$	$2407 \pm 12$	$2350 \pm 27(0,1)$
	100	$97 \pm 2,7(0,1)$	100	$98 \pm 1,2(0,1)$

Note: Amylase activity in the numerator (absolute value); its percentage in denominator; the statistical reliability level ( $p$ ) of the experimental group compared to the control group in brackets (in all cases  $p < 0,05$ ;  $n=3-4$ ).

It was found out that there was a significant decrease in amyloitic activity in blood and liver homogenate of mature rats  $\gamma$ -radiation on the 1-day. In particular, it was identified that the value of amylolytic activity in the blood was  $58 \pm 0.3\%$  in comparison with the control group on the 20-day, and it came closer to the control group value on the 60-day. The experiments showed that amylolytic activity in the liver homogenate was found to be lower than the control group even on the 60 days after  $\gamma$ -radiation (Table 2).

**Table 2. The effect of  $\gamma$ -radiation on the amylase activity in the blood and liver homogenate of mature rats ( $M\pm m$ )**

Durability of hypokines (days)	Blood content (mg/min./ml)		Liver homogenate (mg/min./g)	
	Control group	Experimental group	Control group	Experimental group
1	$780 \pm 6,2$	$460 \pm 3(0,001)$	$1401,8 \pm 14$	$1300 \pm 12(0,001)$
	100	$59 \pm 1,0(0,001)$	100	$93 \pm 0,6(0,001)$
3	$780 \pm 6,4$	$490 \pm 14(0,001)$	$1500,0 \pm 6$	$1210 \pm 18(0,001)$
	100	$63 \pm 1,4(0,001)$	100	$81 \pm 0,8(0,001)$
10	$790 \pm 7,0$	$456 \pm 3,6(0,001)$	$1519,8 \pm 7$	$1240 \pm 12(0,001)$
	100	$58 \pm 0,8(0,001)$	100	$82 \pm 1,1(0,001)$
20	$760 \pm 6,0$	$445 \pm 7(0,001)$	$1526,0 \pm 8$	$1260 \pm 9(0,001)$
	100	$58 \pm 0,8(0,001)$	100	$83 \pm 1,6(0,001)$
30	$750 \pm 4,0$	$620 \pm 6(0,001)$	$1604,0 \pm 10$	$1400 \pm 6(0,001)$
	100	$83 \pm 0,7(0,001)$	100	$87 \pm 1,0(0,001)$
60	$740 \pm 6,0$	$720 \pm 10(0,1)$	$1670,0 \pm 11$	$1620 \pm 7(0,05)$
	100	$97 \pm 1,0(0,05)$	100	$97 \pm 0,9(0,05)$

Note: Amylase activity in the numerator (absolute value); its percentage in denominator; the statistical reliability level ( $p$ ) of the experimental group compared to the control group in brackets (in all cases  $p < 0,05$ ;  $n=3-4$ ).

It was identified that amylase activity decreased after  $\gamma$ -radiation, including reduction of amylolytic activity in blood by 20% in comparison with the control group on the 20-day, it approximated to the value of the control group on the 30-day. In addition, it was found out that amylolytic activity in liver homogenate of old rats decreased in comparison with the control group after  $\gamma$ -radiation and it was regenerated even on the 60-day (Table 3).

**Table 3. The effect of  $\gamma$ -radiation on the amylase activity in the blood and liver homogenate of old rats ( $M\pm m$ )**

Durability of hypokines (days)	Blood content (mg/min./ml)		Liver homogenate (mg/min./g)	
	Control group	Experimental group	Control group	Control group
1	$800 \pm 19$	$680 \pm 7,0(0,001)$	$3630 \pm 45$	$3200 \pm 40(0,001)$
	100	$85 \pm 0,9(0,001)$	100	$88 \pm 1,1(0,001)$
3	$815 \pm 17$	$690 \pm 10(0,01)$	$3670 \pm 63$	$3300 \pm 19(0,05)$
	100	$85 \pm 1,1(0,001)$	100	$90 \pm 1,6(0,001)$
10	$836 \pm 18$	$650 \pm 9,0(0,001)$	$3665 \pm 60$	$3320 \pm 60(0,001)$
	100	$78 \pm 1,1(0,001)$	100	$91 \pm 1,7(0,001)$
20	$717 \pm 16$	$580 \pm 9,0(0,001)$	$3710 \pm 56$	$3314 \pm 27(0,001)$
	100	$81 \pm 1,2(0,001)$	100	$89 \pm 0,8(0,001)$
30	$720 \pm 15$	$620 \pm 8,0(0,001)$	$3620 \pm 60$	$3440 \pm 25(0,1)$
	100	$86 \pm 1,1(0,001)$	100	$95 \pm 0,7(0,1)$
60	$710 \pm 18$	$686 \pm 6,0(0,05)$	$3567 \pm 60$	$3460 \pm 40(0,1)$
	100	$97 \pm 0,8(0,01)$	100	$97 \pm 1,1(0,1)$

Note: Amylase activity in the numerator (absolute value); its percentage in denominator; the statistical reliability level ( $p$ ) of the experimental group compared to the control group in brackets (in all cases  $p < 0,05$ ;  $n=4-6$ ).

Based on the obtained results, it is possible to conclude that the functional activity of old rats' salivary and pancreatic gland, which provides amylolytic activity in the blood by the incretion mechanism, can not be restored even on the 60-day  $\gamma$ -radiation.

Also, the research carried out has established that synthesis of amylase enzyme and intensity of incretion in the saliva of rats by the effect of  $\gamma$ -radiation (1–6 Gr) reduce significantly [Dumaeva et al., 2017].

## **CONCLUSION**

Thus, the experiments revealed that amylolytic activity in the blood of young rats decreased by 20% on the 20-day under the effect of  $\gamma$ -radiation in comparison with the control group and it was regenerated in the blood and liver homogenate of young rats after the 60-day. The experiments let us identify that amylolytic activity in the blood of mature rats was  $58 \pm 0.3\%$  in comparison with that control group on the 20-day by the effect of  $\gamma$ -radiation, it approximated to the control group value on the 60-day, and amylolytic activity in the liver was lower than the control group on the 60-day after  $\gamma$ -radiation. The research revealed that after the  $\gamma$ -radiation, amylolytic activity in the blood of old rats reduced to 20% in comparison with the control group on the 20-day, there was an approximation to the control group value on the 30-day, and that after the  $\gamma$ -radiation amylolytic activity in liver homogenate was lower than the control group, it did not regenerate even on the 60-day. The results obtained can be explained by the significant difference in experimental animals' different digestive system enzyme secretion / incretion mechanisms at different age stage, as well as significant difference of their compensatory / adaptive mechanisms of responses to the effect of  $\gamma$ -radiation.

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