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Original Contribution

PHAGOCYTIC ACTIVITY OF CHICKENS FROM VARIOUS ALKALINE PHOSPHATASE GENOTYPES HATCHED FROM GAMMA-IRRADIATED EGGS

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SUMMARY

The phagocytic activity of White Rhode Island chickens from various alkaline phosphatase genotypes, hatched from eggs irradiated with different doses of gamma rays prior to the incubation, was studied. In pullets, a tendency towards higher phagocytic numbers, compared to cockerels, was observed. The phagocytic number of birds from the AkpSS alkaline phosphatase genotype was stimulated by irradiation at 0.15 Gy. This dose resulted in higher phagocytic numbers and phagocytosis percentages in birds with AkpFF genotype.

Key words: Phagocytic activity; phagocyte number, phagocyte percentage, chickens, gamma rays, alkaline phosphatase genotype.

INTRODUCTION

Phylogenetically, phagocytosis is the most ancient and the earliest form of natural cellular immunity during the ontogenesis of animals. According to some authors (1, 7, 15, 16), the phagocytosis in vertebrates and men is a function of polymorphonuclear granulocytes as well as of cells from the mononuclear-phagocytic system that reflect the status of systemic resistance and adaptation

Under the influence of radiation (gamma and x-rays), the immunological and, especially the phagocytic, activity is changed depending on the dose and the time of exposure (6). Numerous authors (3, 4, 5, 8, 9, 10, 11, 14) have studied the effects of morphological and functional changes in hematopoietic organs and the blood, DNA structures, the factors of both specific and non-specific immunity.

The cells of chicken embryos possess a significantly lower radiosensitivity compared to those of mammals (21). According to Zakaria (22), the gamma irradiation at doses ranging between 0.05 and 0.6 Gy prior to the incubation had no effect on embryonic development, whereas the dose of 2.1 Gy was detrimental. Meada et al. (19) showed that gamma irradiation at 8 Gy impeded the development of chicken embryos. Kozlov and Mageldadze (18) reported the stimulating effect of very low gamma irradiation doses upon chicken embryo development.

Todorov (12, 13, 14) showed that the phagocytic mechanisms of defense were impaired following a total irradiation at sublethal doses of gamma radiation. This phenomenon was most pronounced after irradiation at lethal doses (1000 rad) due to the impaired membrane permeability and the injuring effect of radiation on enzymes. A dose of 3 rad was found to stabilize membrane structures of liver mitochondria in 12-, 13-, 15-, 20- and 21-day old embryos as well as in 1-, 2-, 3- and 7-day old chickens.

The present study aimed to study the phagocytic activity of chickens from various alkaline phosphatase genotypes hatched from eggs, irradiated at different gamma ray doses.

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MATERIALS AND METHODS

Seventy-two pullets and 72 cockerels from the White Rhode Island breed, aged 3 months, were studied. The chickens hatched from eggs, irradiated at various gamma rays doses were grouped into the following: group I – non-irradiated, group II – irradiated at 0.15 Gy, group III – irradiated at 0.3 Gy and group IV – irradiated at 0.6 Gy. The eggs were irradiated immediately prior to the incubation using a ^{60}Co gamma source "Rokus" with a density of 0.526 rad/s, irradiation area of 40 × 40 cm and a distance to eggs of 150 cm. The exposure time of group II was 30 s, of group III – 60 s and of group IV – 120 s.

The alkaline phosphatase genotype was determined at the age of 6 weeks by electrophoresis of blood (aseptically sampled from the wing vein) in a horizontal starch gel according to the method of Gahne (17).

The phagocytic reaction was determined by the method of Valchanov (2). A 24 h microbial suspension of *Staph. aureus* strain 209 with a density of 1-2 mlr/ml (the third tube according to the MacFerland scale) was used. The phagocytic activity of chickens hatched from eggs irradiated at various gamma rays doses was determined via two parameters – phagocytosis percentage (% phagocytosis) and phagocytic number. The latter was the number of bacteria, phagocytized by one leukocyte. The phagocytosis percentage was determined as a ratio of phagocytized leukocytes and the total leukocyte counts, multiplied by 100.

Table 1. Gender-related differences in phagocytosis in White Rhode Island chickens depending on the alkaline phosphatase genotype and the gamma irradiation dose (%)

Groups	Gender	Alkaline phosphatase genotype					
		AkpFF		AkpFS		AkpSS	
		n	x ± Sx	n	x ± Sx	n	x ± Sx
Controls (non-irradiated)	♀	6	88,0 ± 1,9	6	86,0 ± 2,3	6	88,0 ± 0,0
	♂	6	86,0 ± 2,3	6	85,0 ± 1,2	6	86,0 ± 1,3
	♀ + ♂	12	87,0 ± 1,3	12	85,5 ± 1,1	12	[a]87,0 ± 0,7
Irradiated at 0.15 Gy	♀	6	88,0 ± 1,9	6	86,0 ± 1,3	6	85,0 ± 22,0
	♂	6	88,0 ± 1,9	6	84,0 ± 1,9	6	80,0 ± 1,9
	♀ + ♂	12	[b]88,0 ± 1,1	12	85,0 ± 1,1	12	82,5 ± 1,6
Irradiated at 0.3 Gy	♀	6	85,0 ± 1,2	6	86,0 ± 1,3	6	85,0 ± 11,0
	♂	6	84,0 ± 1,9	6	87,0 ± 1,2	6	82,0 ± 2,9
	♀ + ♂	12	84,5 ± 0,9	12	86,5 ± 0,8	12	83,5 ± 1,5
Irradiated at 0.6 Gy	♀	6	84,0 ± 1,9	6	83,0 ± 1,2	6	85,0 ± 11,0
	♂	6	83,0 ± 1,2	6	86,0 ± 1,3	6	86,0 ± 2,9
	♀ + ♂	12	83,5 ± 0,9	12	84,5 ± 0,9	12	85,5 ± 1,4

[a] $p<0.05$ *vs* chickens from the AkpSS genotype, irradiated at 0.15 Gy
[b] $p<0.05$ *vs* chickens from the AkpFF genotype, irradiated at 0.6 Gy

RESULTS AND DISCUSSION

Phagocytosis percentage

Table 1 presents the phagocytosis percentages in the various avian genotypes according to irradiation doses. In non-irradiated pullets and cockerels from homozygous genotypes (AkpFF and AkpFS), phagocytosis percentages were equal whereas in the heterozygous one (AkpFS) – lower ($p<0.05$). The dose of 0.15 Gy stimulated phagocytosis percentages in cockerels while the dose of 0.3 Gy and especially 0.6 Gy decreased the values of this parameter in both genders ($p<0.05$). The dose of 0.6 Gy decreased phagocytosis percentages in pullets from the heterozygous genotype AkpFS although not significantly. The lower doses had no effect. In AkpSS chickens, the dose of 0.15 Gy decreased phagocytosis percentages in both genders ($p<0.05$). The irradiation at 0.3 Gy (better manifested in cockerels) and

0.6 Gy also lowered phagocytosis percentages. The differences for the higher dose were significant ($p<0.05$). It must be emphasized that the irradiation at 0.15 Gy was found to be stimulating in AkpFF chickens ($p<0.05$) and inhibiting in AkpSS chickens.

Phagocytic numbers

The data on **Table 2** showed that the phagocytic numbers in pullets were higher compared to those in cockerels in all three genotypes. A tendency towards increase in phagocytic numbers in pullets from the homozygous genotypes AkpFF and AkpSS was present. The irradiation at 0.15 Gy was stimulating for all genotypes (with the exception of AkpFS pullets). This trend was most apparent in cockerels ($p<0.01$) and pullets from the AkpSS genotype.

The dose of 0.3 Gy was feebly stimulating in birds carrying the AkpF allele and in AkpS birds, was found to be stimulating only in cockerels.

The irradiation at 0.6 Gy decreased insignificantly the phagocytic numbers in AkpFF and AkpFS pullets and stimulated them in cockerels. In AkpSS pullets and cockerels, this tendency was the opposite.

The results presented on both tables showed that the percentage of decrease of phagocytized macrophages resulted in increased activity of these with phagocytic activity that is probably some kind of a compensatory mechanism of gamma rays influence, especially in AkpSS and AkpFS chickens.

Table 2. Gender-related differences in phagocytic numbers in White Rhode Island chickens depending on the alkaline phosphatase genotype and the gamma irradiation dose

Groups	Gender	Alkaline phosphatase genotype					
		AkpFF		AkpFS		AkpSS	
		n	x ± Sx	n	x ± Sx	n	X ± Sx
Controls (non-irradiated)	♀	6	1,73 ± 0,126	6	1,68 ± 0,033	6	1,70 ± 0,067
	♂	6	1,64 ± 0,027	6	1,65 ± 0,076	6	^{bb}1,54 ± 0,072
	♀ + ♂	12	1,68 ± 0,058	12	1,66 ± 0,036	12	^{aa}1,62 ± 0,053
Irradiated at 0.15 Gy	♀	6	1,99 ± 0,064	6	1,68 ± 0,105	6	1,87 ± 0,035
	♂	6	1,79 ± 0,100	6	1,85 ± 0,195	6	2,39 ± 0,239
	♀ + ♂	12	1,89 ± 0,055	12	1,76 ± 0,100	12	2,13 ± 0,147
Irradiated at 0.3 Gy	♀	6	1,77 ± 0,074	6	1,71 ± 0,035	6	1,68 ± 0,082
	♂	6	1,65 ± 0,092	6	1,65 ± 0,048	6	^{b}1,71 ± 0,091
	♀ + ♂	12	1,71 ± 0,056	12	1,68 ± 0,028	12	^{aa}1,69 ± 0,053
Irradiated at 0.3 Gy	♀	6	1,67 ± 0,076	6	1,56 ± 0,019	6	1,75 ± 0,022
	♂	6	1,72 ± 0,065	6	1,74 ± 0,055	6	^{bb}1,62 ± 0,085
	♀ + ♂	12	1,69 ± 0,044	12	1,65 ± 0,440	12	^{aa}1,68 ± 0,046

^{aa} $p<0.01$ vs chickens from the AkpSS genotype, irradiated at 0.15 Gy
^{bb} $p<0.01$ vs cockerels from the AkpSS genotype, irradiated at 0.15 Gy
^{b} $p<0.05$ vs cockerels from the AkpSS genotype, irradiated at 0.6 Gy

The low doses of ionized radiation stimulate mononuclear phagocytic system and particularly, chicken macrophages (20).

The low doses of gamma and x-rays (0.15 and 0.3 Gy), applied prior to egg incubation, stimulate the growth and development of pullets and cockerels from the Worren hybrid (9). In our previous studies (11) we found that the dose of 0.15 Gy stimulated the phagocytic activity in broiler chickens. Gender-related differences in phagocytic activity were observed by Yotova (10) too. In Worren hybrids, the irradiation at 0.15 Gy stimulated the phagocytic index of homozygous (AkpFF and AkpSS) chicken genotypes. The phagocytic numbers of chickens from the AkpSS genotype were increased after exposure to the three radiation doses, but the effect of stimulation was dependent on the genotype, the gender and the

breed of chickens.

CONCLUSIONS

1. A tendency towards higher phagocytic numbers in pullets compared to cockerels was observed.

2. The phagocytic numbers in birds with AkpSS alkaline phosphatase genotype were stimulated by irradiation at 0.15 Gy, whereas in AkpFF birds the same dose resulted in increased phagocytic numbers and phagocytosis percentages.

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