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*The activity of lysosomal enzymes of rabbit's muscle rhabdomyoma
during induced experimental diabetes*

The result of investigations carried out at the Department and Institute of Human Anatomy is that the activity of lysosomal enzymes of rabbit's cardiac muscle during induced experimental diabetes changes (1). Therefore, we decided to investigate the activity of this enzymes of rabbit's muscle rhabdomyoma in the process of the disease.

MATERIAL AND METHOD

The investigation was carried out on the material and method gived in the previous paper (2). We collected segments of *muscle latissimus dorsi*. The analysis of variants of values obtained in the individual investigations groups was made by means of ANOVA statistical test rang Kruskala-Wallis for variables, for which really significant differences were shown in variance analysis. The comparison of individual groups with control group was made by U Manna-Whitneya test. We agreed that if $p < 0.05$, it is a significant difference between investigation variables, but if $p < 0.01$ this difference is highly substantial statistically.

RESULTS

In table 1 the results of the investigation on the activity of free lysosomal enzymes of rabbit's muscle rhabdomyoma during induced experimental diabetes were shown, while table 2 shows that of connected lysosomal enzymes. From table 1 it appears that activity of free acid phosphatase decreases at first. In animals with 6-weeks' diabetes was lower by 23.7% in comparison with control group, which is not significant statistically. In the next part of the investigation there was an increase of free acid phosphatase whose activity for rabbits being ill the longest was increased by 32.4% in comparison with control group, which is significant statistically. From table 2 it appears that the activity of connected enzymes in the investigation was characterized by alternate increase or decrease, and it acquired the smallest value for rabbits with 6-months' diabetes, which the substantial statistically. The activity of free β -galactosidase after first decrease, in the following period of disease was higher than in control group, with the highest value for rabbits with 6-weeks' diabetes, which was not significant statistically. The investigation observed a considerable decrease of the activity of connecting β -galactosidase, which for animals with 6-months' diabetes was decreased by 80.3% in comparison with control group. The decrease of activity for rabbits with 3-weeks' diabetes was highly significant

Table 1. The activity of free lysosomal enzymes of rabbits muscle rhabdomyoma during induced experimental diabetes (mmol/mg proteine/1 hour of incubation)

Fraction of enzymes	Control group	3-weeks' diabetes group	6-weeks' diabetes group	3-months' diabetes group	6-months' diabetes group
Acid phosphatase	0.4698 ±0.1503	0.4467 ±0.1619	0.3799 ±0.0782	0.4855 ±0.1118	0.6955 ±0.2323
β-galactosidase	0.0895 ±0.1719	0.0858 ±0.0638	0.1640 ±0.1567	0.1512 ±0.1723	0.1013 ±0.0530
NAGL	0.2426 ±0.0985	0.2558 ±0.1488	0.2824 ±0.1865	0.2449 ±0.1006	0.2141 ±0.1056
Lipase	0.2520 ±0.0793	0.4058 ±0.1023	0.3601 ±0.0831	0.3456 ±0.0917	0.3904 ±0.1737
Connected sulphatase	0.003285 ±0.002729	0.001613 ±0.000833	0.001309 ±0.000480	0.001796 ±0.000742	0.1259 ±0.000433
Katepsin B	0	0	0	0	0
Katepsin D	96.5448 ±24.2993	75.0784 ±17.6613	74.9978 ±224029	79.5343 ±15.4499	98.6697 ±29.5739
Katepsin L	0	0	0	0	0

Table 2. The activity of connected lysosomal enzymes of rabbits muscle rhabdomyoma during induced experimental diabetes (mmol/mg proteine/1 hour of incubation)

Fraction of enzymes	Control group	3-weeks' diabetes group	6-weeks' diabetes group	3-months' diabetes group	6-months' diabetes group
Acid phosphatase	0.1099 ±0.0919	0.1100 ±0.0744	0.0873 ±0.0610	0.1122 ±0.0667	0.0638 ±0.0326
β-galactosidase	0.1668 ±0.1322	0.0458 ±0.0476	0.0465 ±0.0239	0.0577 ±0.0374	0.0328 ±0.0237
NAGL	0.1610 ±0.1026	0.1103 ±0.1065	0.0743 ±0.0390	0.1105 ±0.0420	0.0748 ±0.0213
Lipase	0.1477 ±0.1124	0.2994 ±0.0827	0.2485 ±0.0888	0.3183 ±0.1696	0.2077 ±0.0633
Free sulphatase	0.001213 ±0.000943	0.000719 ±0.000530	0.001622 ±0.001622	0.000964 ±0.000784	0.002551 ±0.001982
Katepsin B	0	0	0	0	0
Katepsin D	0	0	0	0	0
Katepsin L	0	0	0	0	0

statistically, whereas for the remaining animals it was substantial. The activity of N-acetylo-β-glucosaminidase (NAGL) increases up to 3rd month of disease, acquiring the highest value for the rabbits with 6-weeks' diabetes, which was not substantial statistically. For animals with 6-months' diabetes the decrease of activity of enzymes by 12.8% was observed in comparison with control group, which was not substantial statistically. The activity of connected NAGL decreased in investigation acquiring the smallest value for rabbits with 6-weeks' diabetes, which is highly significant statistically. The activity of free lipase increased in the course of diseases reaching the lowest value for animals with 3-weeks' diabetes, which is highly significant statistically. Likewise, the activity of connected lipase was increased, reaching the highest value for rabbits with 3-months' diabetes, which is the value highly significant statistically. The activity of free sulphatase alternately increased or decreased in the course of disease. The highest value was for animals with 6-months' diabetes, which is the value significant statistically. The activity of connected sulphatase decreased in the course of investigation, acquiring the smallest value for rabbits with 6-months' diabetes, which is not significant statistically. Among the investigated katepsins B, D and L, only free katepsin D demonstrated the

activity which decreased during 3-months' disease, to be higher for animals with 6-months' diabetes by 4.7% in comparison with control group, which is not significant statistically.

From our experiment it appears that the activity of free and connected lysosomal enzymes of rabbit's muscle rhabdomyoma during experimentally induced diabetes was smaller than the cardiac muscle. Dynamics of its changes in the course of disease in the rhabdomyoma muscle was different than in the cardiac muscle (1).

REFERENCES

1. J a m r o ż A.: Histological ,histochemical, biochemical cardiac muscle structure during induced experimental diabetes. Library AM, 1995.
2. W ó j t o w i c z Z. et al.: The activity of lysosomal enzymes of rabbits gingiva during induced experimental diabetes. Annales UMCS, Sectio D, 59, 2004.

SUMMARY

The investigation was carried out on New Zealand rabbits. Diabetes was provoked by means of an i.v. injection of alloxan. We then removed the segment of muscle latissimus dorsi. The muscles were homogenized in saccharose solution. The activity of lisosomal enzymes was determined by means of aid substrate. Among free enzymes, the highest was katepsin D, distinctly smaller acid phosphatase, NAGL and lipase, the smallest β -galactosidase and sulphatase, while activity katepsin B and L was zero. Among the connected enzymes β -galactosidase, NAGL and lipase were of the highest activity acid phosphatase smaller and sulphatase minimal. The activity of all connected katepsins was zero.

Aktywność enzymów lizosomalnych mięśnia szkieletowego u królika w przebiegu cukrzycy doświadczalnej

Badanie przeprowadzono na królikach rasy nowozelandzkiej białej. Cukrzycę wywołano przez dożylną podanie alloxanu. Do badań pobierano wycinek mięśnia najszerzego grzbietu, który homogenizowano w roztworze sacharozy. Aktywność enzymów lizosomalnych oznaczono przy pomocy odpowiednich substratów. Spośród enzymów wolnych największą aktywność miała katepsyna D, wyraźnie mniejszą fosfataza kwaśna, NAGL i lipaza, najmniejszą zaś β -galaktozydaza i sulfataza, natomiast aktywność katepsyny B i L była zerowa. Z enzymów związanych największą aktywność miały β -galaktozydaza, NAGL i lipaza, mniejszą fosfataza kwaśna, a minimalną sulfataza. Aktywność wszystkich związanych katepsyn była zerowa.