ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN – POLONIA
VOL. LX, N 2, 150 SECTIO D 2005

Department and Institute of Human Anatomy, Medical University of Lublin

ZBIGNIEW WÓJTOWICZ, FRANCISZEK BURDAN, MAŁGORZATA PLISZCZYŃSKA-STEUDEN, GRAŻYNA KIŚ, JANUSZ GOLAN, MAGDALENA BŁASZCZAK

The activity of pancreatic lysosomal enzymes of rabbit during induced experimental diabetes mellitus

According to literature, the activity of lysosomal enzymes changes during pancreatitis acute and pancreatic neoplasm (1, 2, 4). For that reason we decided to follow it during experimental diabetes mellitus.

MATERIAL AND METHODS

Material and methods like in previous study (3).

RESULTS

Table1 presents the activity of free pancreatic lysosomal enzymes of rabbit during experimental diabetes mellitus, and table 2 presents the connecting ones. According to table 1, the activity of free acid phosphatase decreased in the course of disease, primarily slightly so that it decreased by 66.1% in rabbits within 3-months' diabetes and by 62.3% in rabbits with 6-months' diabetes in comparison with the control group, which is extremely significant statistically. The activity of connecting enzyme in the course of diabetes decreased in comparison with the control group. It was the greatest in the group with 6-months' diabetes (by 54.3%), which is also extremely significant statistically.

The activity of free β-galactosidase decreased significantly in the course of disease and reached the level 83.3% lower than in the control group, which is extremely significant statistically. The activity of connecting β-galactosidase primarily decreased by 75.6% and 77.4% and then started to increase slightly in the group with 3- and 6-months' diabetes. The activity of connecting β-galactosidase during diabetes was statistically different in comparison with the control group and this difference was extremely significant.

The activity of free N-acetyl-B-D-glucosaminidase (NAGL) decreased during the experiment. A decreasing trend was the most significant in 6-weeks' diabetes group of rabbits (by 70.2%), 3-months' diabetes group of rabbits (by 72.4%) and 6-months' diabetes group (85.5%). The activity of connecting enzyme also decreased in the course of the experiment and reached the lowest value in the group with 6-months diabetes. It was 75.1% lower than in the control group, which is extremely significant statistically.

Regarding the activity of free katepsin D, there was recorded, a statistically significant increase for 3 months and then the activity started to decrease to reach the value 25.5% lower in rabbits with 6-months' diabetes than in the control group. The activity of connecting katepsin D primarily decreased in rabbits with 3-weeks' diabetes and then it increased by 96% in 3rd and 6th months of experiment in comparison with the control group, which is extremely significant statistically.

Table 1. The activity of free fractions of pancreatic lysosomal enzymes in rabbit during induced experimental diabetes

Fraction of enzymes	Control group	3-weeks' diabetes	6-weeks' diabetes	3-months' diabetes	6-months' diabetes
Acid phosphatase	37.66 12.05	31.79 ±10.52	31.03 ±12.82	12.77 ±2.52	14.20 ±3.05
β -galactosidase	45.84 ±15.01	27.96 ±8.25	8.90 ±2.02	8.32 ±2.03	7.66 ±1.16
NAGL	49.67 ±17.22	45.50 ±14.25	14.8 ±3.15	13.71 ±2.95	7.23 ±1.16
Katepsin D	42.00 ±16.05	57.90 ±20.05	63.70 ±22.32	56.1 ±19.85	31.30 ±12.25
Katepsin L	73.50 ±28.51	80.60 ±30.22	110.80 ±42.32	87.50 ±33.03	52.80 ±18.55
Lipase	22.54 ±6.82	42.86 ±12.85	84.30 ±45.28	71.03 ±26.25	45.61 ±17.02
Sulphatase	2.94 ±0.12	4.24 ±1.02	2.35 ±0.08	9.96 ±2.15	4.48 ±1.15

The activity of free katepsin L was higher in groups with 3- and 6-weeks' and 3-months' diabetes than in the control group and reached a statistically significant different value in the group with 6-weeks' diabetes. In the group of rabbits with 6-months' diabetes there was recorded a statistically significant decrease of this activity by 28.2% in comparison with the control group. The activity of connecting katepsin L was the highest among all tested enzymes. An extreme growth of the activity of this enzyme was noted in rabbits with 3- and 6-months' diabetes (by 90.1% and 95.3%), which is extremely significant statistically. Regarding the activity of free lipase, there was noted an increase, which reached the maximum value in the rabbits with 6-weeks' diabetes (by 274%) and 3-months' diabetes (by 215.1%) in comparison with the control group. The activity of connecting lipase primarily also showed a increasing tendency, reaching the highest value in the group with 6-weeks' diabetes. It was 181.2% higher in this group than in the control group, which is extremely significant statistically. However, the activity of this enzyme in the group of rabbits with 6-months' diabetes decreased below the value of the control group. The activity of free sulphatase was the lowest among all tested enzymes. It increased and decreased alternately in the course of the experiment and the greatest value of this activity was noted in rabbits with 3-months' diabetes. It was by 238.7% greater than in the control group, which is extremely significant statistically. The activity of connecting enzyme increased during the disease and reached the greatest value after 6 months, which exceeded by 343.4% the activity in the control group.

Table 2. The activity of connecting fractions of pancreatic lysosomal enzymes in rabbit during induced experimental diabetes

Fraction of enzymes	Control group	3-weeks' diabetes	6-weeks' diabetes	3-months' diabetes	6-months' diabetes
Acid phosphatase	85.89 32.02±	57.08 ±18.92	39.32 ±16.03	45.42 ±18.21	79.98 ±28.21
β -galactosidase	35.14 ±14.05	8.59 ±2.21	7.97 ±2.21	10.68 ±3.32	19.24 ±7.15
NAGL	27.54 ±8.15	19.84 ±6.52	8.48 ±2.21	16.43 ±6.52	6.86 ±1.92
Katepsin D	54.00 ±16.52	45.10 ±15.52	56.60 ±17.22	105.50 ±42.15	105.80 ±42.27
Katepsin L	98.40 ±38.21	103.70 ±40.28	109.90 ±43.32	187.10 ±62.11	192.20 ±63.25
Lipase	48.94 ±15.62	64.00 ±18.52	137.62 ±52.15	53.07 ±15.21	44.70 ±14.89
Sulphatase	3.19 ±0.52	4.38 ±0.82	3.40 ±0.61	8.71 ±2.32	14.14 ±3.21

REFERENCES

1. Duffy M. J.: The role of proteolytic enzymes in cancer invasion and metastasis. Clin. Exp. Met., 10, 145, 1992.
2. Gorelick F., Matovick L. : Lysosomal enzymes and pancreatitis. Gastroenterol., 109, 620, 1995.
3. Wójtowicz Z. et al.: The activity of lysosomal enzymes of rabbit's gingiva mucosa during induced experimental diabetes. Annales UMCS, Sectio D, 59, 2004.
4. Yamaguchi N. et al.: Characterization of a cathepsin L-like enzyme secreted from human pancreatic cancer cell line HPC-YP. Cancer Res., 50, 658, 1990.

SUMMARY

The investigations were carried out on New Zealand rabbits, male adults. Diabetes was provoked by means of an i.v. injection of alloxan. Segments of pancreas were taken to the investigation. The material was homogenized in the solution of saccharose. The activity of the examined lysosomal enzymes was marked with the proper substrates. Regarding free enzymes, the activity of katepsin L was the greatest. The activity of free NAGL, β-galactosidase, katepsin D, acid phosphatase and lipase was lower and the activity of free sulphatase was the lowest. Regarding connecting enzymes, there was the greatest activity of katepsin L and acid phosphatase, a lower activity of katepsin D, lipase, β -galactosidase and NAGL and the lowest activity of sulphatase. The activity of the lysosomal enzymes was variable in the course of diabetes and it was usually statistically significant.

Aktywność enzymów lizosomalnych trzustki u królika w przebiegu cukrzycy doświadczalnej

Badania przeprowadzono na królikach samcach rasy nowozelandzkiej białej. Cukrzycę wywoływano poprzez dożylne podanie alloksanu. Do badań pobierano wycinek trzustki, który homogenizowano w roztworze sacharozy. Aktywność enzymów lizosomalnych oznaczano przy pomocy odpowiednich substratów. Z enzymów wolnych największą aktywność wykazywała katepsyna L, mniejszą NAGL, β-galaktozydaza, katepsyna D, fosfataza kwaśna i lipaza, najmniejszą zaś sulfataza. Spośród enzymów związanych największą aktywność przedstawiała katepsyna L i fosfataza kwaśna, mniejszą katepsyna D, lipaza, β-galaktozydaza i NAGL, najmniejszą zaś sulfataza. W przebiegu cukrzycy zmieniała się aktywność enzymów lizosomalnych trzustki przeważnie w sposób istotny statystycznie.