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SHORT COMMUNICATION

Phospholipid Composition in the Rat Heart Exposed to Pressure Overload from Birth

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Summary

A pressure overload was induced in 2-day-old male rats by abdominal aortic constriction, and the phospholipid composition of the left ventricle (LV) and the right ventricle (RV) were determined. Sixty days after the surgery, body weights was lower and LV weight were higher in aorta-constricted (AC) rats in comparison with sham-operated animals. Increased ventricular/body weight ratios indicated a significant degree of hypertrophy of LV and smaller hypertrophy of RV. The concentrations of total phospholipids (PL), choline phosphoglycerides (PC), ethanolamine phosphoglycerides (PE), diphosphatidylglycerol (DPG) and phosphatidylinositol (PI) were decreased in both ventricles of AC rats. The concentrations of sphingomyelin (SM) and plasmalogen PE (PLPE) increased in LV only. The changes in phospholipid composition in the developing pressure-overloaded myocardium may contribute to altered membrane functions connected with heart hypertrophy.

Key words

Myocardium – Pressure overload – Aorta banding – Hypertrophy – Postnatal development – Phospholipids – Plasmalogens

The development of hypertrophy in a pressure-overloaded heart is connected with structural and functional alterations in cell membranes. This was well documented for both protein and lipid components of membranes in adults (Swynghedauw 1991, Reibel *et al.* 1986, Cserhati and Szögyi 1993). Even though the stage of myocardial maturity influences the course of the hypertrophy process (Perloff 1982), very little information is available concerning changes in membranes of the pressure-overloaded heart in the early postnatal period. The aim of the present study was therefore to characterize and to compare changes in phospholipid concentrations in LV and RV of rat heart subjected to pressure overload from birth. It has been shown previously that a pressure overload from birth (Černohorský *et al.* 1995) elevated the concentration of collagenous proteins and affected the proportion of collagen types and myosin light chains in the overloaded myocardium.

A gradual pressure overload was induced in 2-day-old male Wistar rats by banding the abdominal aorta. The aorta was constricted by silk suture around a steel wire (diameter 0.25 mm). At 60 days after surgery, the body weights of AC rats were lower by 17 % as compared to the controls, while the pressure overload elevated the weight of the heart (HW: 1291.8±88.5 vs 794.4±34.5 mg, $p<0.05$); the relative increase was observed not only in LV (LV/BW: 3.04±0.25 vs 2.76±0.06 mg/g, $p<0.05$) but also in RV (RV/BW: 0.99±0.11 vs 0.77±0.04 mg/g, $p<0.05$).

Samples of RV and LV from banded and control rats were frozen in liquid nitrogen, pulverized and extracted (Folch 1959). Lipid extracts were evaporated under nitrogen, phospholipids were separated by two-dimensional thin layer chromatography and the spots of individual phospholipids were analyzed for phosphorus (Rouser *et al.* 1970). Plasmalogen components of PC and PE were analyzed according to the method of Horrocks *et al.* (1968).

Table 1
Distribution of phospholipids in the left ventricle of aorta-constricted (AC) and control (sham-operated) 60-day-old rats

	Phospholipid concentration μmol P/g wet wt		% Distribution	
	Sham	AC	Sham	AC
PC	11.49±0.25	10.23±0.22*	40.67±0.23	42.02±0.37*
PE	10.25±0.18	8.78±0.26*	36.29±0.24	36.01±0.33
DPG	4.04±0.10	3.07±0.12*	14.31±0.19	12.59±0.24*
PI	1.17±0.03	0.99±0.03*	4.13±0.09	4.04±0.10
PS	0.70±0.02	0.67±0.02	2.48±0.06	2.75±0.07*
SM	0.57±0.01	0.63±0.01*	2.03±0.07	2.58±0.08*
Total PL	28.26±0.53	24.37±0.60*		
PLPC×	0.70±0.03	0.65±0.09	2.50±0.08	2.65±0.33
PLPE×	2.50±0.05	2.70±0.03*	8.81±0.15	11.12±0.28*

*PL (phospholipids), PC (choline phosphoglycerides), PE (ethanolamine phosphoglycerides), DPG (diphosphatidylglycerol), PI (phosphatidylinositol), PS (phosphatidylserine), SM (sphingomyelin), PLPC (plasmalogen PC), PLPE (plasmalogen PE). Values are means ± S.E.M. * Significant differences ($p<0.05$), $n=6$, Student's t-test was used for statistical evaluation. #PLPC and PLPE are included in PC and PE, respectively.*

Table 2
Distribution of phospholipids in the right ventricle of aorta-constricted (AC) and control (sham-operated) 60-day-old rats

	Phospholipid content μmol P/g wet wt		% Distribution	
	Sham	AC	Sham	AC
PC	11.51±0.26	10.30±0.17*	40.87±0.27	41.53±0.50
PE	10.07±0.24	8.56±0.17*	35.75±0.24	35.06±0.18*
DPG	4.03±0.14	3.30±0.17*	14.32±0.38	13.24±0.46 *
PI	1.18±0.03	1.05±0.03*	4.18±0.08	4.22±0.11
PS	0.74±0.01	0.75±0.02	2.63±0.06	3.04±0.11*
SM	0.63±0.02	0.64±0.01	2.23±0.07	2.59±0.11*
Total PL	28.16±0.60	24.82±0.57*		
PLPC×	0.67±0.05	0.69±0.08	2.45±0.23	2.77±0.27
PLPE×	2.49±0.06	2.54±0.05	8.96±0.22	10.26±0.34*

For symbols see Table 1

Table 1 shows that the total PL concentration of LV was decreased by 14 % 60 days after AC. This decrease was caused by a lower concentration of PC, PE, DPG and PI (11 %, 14 %, 24 % and 15 %, respectively). The concentration of SM and PLPE was elevated by 10 % and 8 %, respectively. The changes in phospholipid concentration in RV (Table 2) were similar to those found in LV but were less pronounced. Total PL, PC, PE, DPG and PI decreased by 12 %, 10 %, 15 %, 18 % and 11 %, respectively. There were

no significant changes in the concentration of PS, SM and both plasmalogens in RV. When the individual phospholipids were expressed as a percentage of total PL, the proportion of DPG was decreased whereas PS, SM and PLPE increased in both LV and RV of AC rats.

In our experiments, the pressure overload was induced in the early postnatal period when both hyperplasia and hypertrophy were involved in growth of the myocardium. This represents a broad spectrum of morphological, biochemical and physiological changes. Two major differences between young and adult myocardium have been described: the diameter of the foetal cell is smaller than that of the adult one and the proportion of noncontractile mass (e.g. nuclei, mitochondria and membrane structures) relative to the number of myofibrils in foetal tissue is significantly higher than in adults (for review see Pelouch *et al.* 1992). The changes in phospholipid composition which we observed in a developing pressure-overloaded myocardium may reflect either a change in the distribution of phospholipids in individual subcellular membranes, or a change in the relative proportion of one or more of the subcellular membranes in the hypertrophied heart. Undoubtedly, this is the case of mitochondrial membranes, where we found a 25 % lower concentration of DPG in LV of AC animals in comparison with the sham-operated ones. Surprisingly, a very similar decrease in DPG concentration (by 18 %) was observed in the much less hypertrophied RV ventricle. Thus, it remains to be explained which of the mechanisms involved in the development of heart hypertrophy triggers changes in membrane phospholipids in both ventricles of pressure-overloaded myocardium after birth.

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References

ČERNOHORSKÝ J., PELOUCH V., MILEROVÁ M., OŠŤÁDAL B.: Protein profiling of the myocardium exposed to pressure overload from birth. *Physiol. Res.* **44:** 139–142, 1995.

CSERHATI T., SZÖGYI M.: Interaction of phospholipids with proteins and peptides. New Advances III. *Int. J. Biochem.* **25:** 123–146, 1993.

FOLCH J., LEES M., SLOANE-STANLEY G.H.: A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.* **226:** 497–509, 1957.

HORROCKS L.A.: The alk-1-enyl group content of mammalian myelin phosphoglycerides by quantitative two-dimensional chromatography. *J. Lipid Res.* **9:** 469–472, 1968.

PELOUCH V., MILEROVÁ M., OŠŤÁDAL B., PROCHÁZKA J.: Ontogenic development of the protein composition of the right and left ventricular myocardium. In: *Right Ventricular Hypertrophy and Function in Chronic Lung Disease.* V. JEŽEK, M. MORPURGO, K. TRAMARIN (eds), Springer Verlag, London, 1992, pp. 39–54.

PERLOFF J.K.: Development and regression of increased ventricular mass. *Am. J. Cardiol.* **50:** 605–612, 1982.

REIBEL D.K., O'ROURKE B., FOSTER K.A., HUTCHINSON H., UBOH C.E., KENT R.L.: Altered phospholipid metabolism in pressure-overload hypertrophied hearts. *Am. J. Physiol.* **250:** H1–H6, 1986.

ROUSER G., FLEISCHER S.F., YAMAMOTO A.: Two dimensional thin layer chromatographic separation of polar lipids and determination of phospholipids by phosphorus analysis of spots. *Lipids* **5:** 494–496, 1970.

SWYNGHEDAUW B.: Remodeling of the heart in chronic pressure overload. *Basic Res. Cardiol.* **86:** 99–105, 1991.

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