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SUCROSE FEEDING PREVENTS CHANGES IN MYOSIN ISOENZYMES AND SARCOPLASMIC RETICULUM Ca^{2+}-PUMP ATPase IN PRESSURE-LOADED RAT HEART

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Summary: Pressure-overload due to banding of the abdominal aorta in rats for 10 weeks resulted in cardiac hypertrophy, redistribution of myosin isoenzymes and reduction in the sarcoplasmic reticulum (SR) Ca^{2+}-stimulated ATPase activity. Administration of sucrose in the drinking water (0.8%, w/v) to rats prevented changes in myosin isoenzymes and SR Ca^{2+}-stimulated ATPase in hypertrophied hearts. This beneficial effect of sucrose feeding with respect to remodeling of the subcellular organelles in the myocardium was not associated with any significant changes in plasma glucose or thyroid hormone levels. It is suggested that the prevention of subcellular changes in the hypertrophied hearts due to sucrose feeding may be due to a shift in fuel utilization by the myocardium.

In hypertrophied hearts due to pressure-overload, the proportion of myosin V_3 is increased whereas myosin V_1 is decreased; these changes are believed to result in a reduced speed of cardiac muscle shortening (1,2) and an increased mechanical economy (3,4). Although the altered mechanical performance of the heart is considered as an adaptation process to the pressure-overload, cardiac dilatation occurs typically at a high proportion of myosin V_3 content, if the stimulus leading to pressure-overload is not removed adequately (5). The redistribution of myosin isoenzymes is most probably not an isolated event, but rather is an example of a more general process occurring in the myocyte, because Ca^{2+}-stimulated ATPase of SR is also reduced in the compensated stage of myocardial hypertrophy (6). It was, therefore, considered important to search for interventions which possibly could prevent changes in subcellular organelles in the hypertrophied hearts. Previously, it has been shown that swimming exercise could prevent to a great extent the

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increase in myosin V_3 in spontaneously hypertensive rats (7) or renal hypertensive rats (8). However, swimming exercise by itself is known to induce cardiac hypertrophy and thus can be seen to affect heart function adversely. In this study it is shown that feeding low amounts of sucrose in the drinking water can prevent not only the increase in myosin V_3 but also the reduction of SR Ca^{2+}-pump ATPase activity without affecting the cardiac hypertrophy due to banding of the abdominal aorta in rats.

Methods

Thirty two male Sprague Dawley rats (6 week old) were fed a standard chow; half of these animals received sucrose (0.8%, w/v) in their drinking water. Each of the two groups consisted of 8 sham operated control rats and 8 rats with pressure-overload due to constriction of the abdominal aorta for 10 - 12 weeks. Fragmented SR was isolated from the left ventricle as described previously (9) and Ca^{2+}-stimulated ATPase was assayed in a medium containing 100 mM KCl, 5 mM NaN_3, 5 mM $MgCl_2$, 0.01 mM free Ca2+, 5 mM ATP and 20 mM Tris-Cl, pH 6.8. Myosin isoenzyme population in the same ventricle was determined by using the pyrophosphate gel electrophoresis according to the method described earlier (7). The plasma concentrations of thyroxine (T_4) and triiodothyronine (T_3) were determined using the radioimmuno assay, whereas plasma glucose was measured by the glucose oxidase method. Statistical comparisons were performed with analysis of variance or in the presence of unequal variances with the test of Kruskal and Wallis. The statistical significance was assumed at $P < 0.05$.

Results

The data in Table 1 indicate a significant increase in heart weight and left ventricular weight in rats with abdominal aorta banded for 10 - 12 weeks. Feeding of sucrose in the drinking water resulted in an additional daily calorie intake of only 1% of total calories consumed by the rats. In both control and aortic banded animals, the body weight, heart weight and left ventricular weight were not significantly affected by sucrose feeding; the tibia length, as a measure of body growth, was also unchanged (Table 1).

The proportion of myosin V_1 was reduced and that of myosin V_3 was increased in the hypertrophied hearts due to pressure overload (Table 2). In control rats, sucrose feeding had only a slight effect on the myosin isoenzyme population whereas in the pressure-loaded ventricles, sucrose feeding prevented the increase in myosin V_3 and decrease in myosin V_1

Table 1. Effect of sucrose feeding on food intake and growth characteristics of rats in the presence or absence of pressure-overload

	Control	Sucrose-fed Control	Pressure-loaded	Sucrose-fed Pressure-loaded
Food consumption/day (g)	24.6 ± 5.1	25.7 ± 4.8	25.4 ± 3.9	25.1 ± 2.6
Water intake/day (ml)	36.5 ± 1.8	40.0 ± 4.4	34.9 ± 6.9	45.7 ± 5.3
Sucrose intake/day (g)	0	0.32 ± 0.04	0	0.37 ± 0.04
Body weight (g)	481 ± 49	500 ± 29	513 ± 29	500 ± 28
Heart weight (mg)	1085 ± 125	1203 ± 117	1387 ± 99*	1391 ± 45*
Left ventricular weight (mg)	794 ± 90	856 ± 109	1037 ± 85*	1043 ± 62*
Tibia length (mm)	4.2 ± 0.1	4.3 ± 0.1	4.3 ± 0.1	4.3 ± 0.1

Each value is a mean ± S.E. of 8 animals.
*-significantly different ($P < 0.01$) with respect to control or sucrose-fed control groups.

(Table 2). The activity of SR Ca^{2+}-stimulated ATPase was decreased without any changes in the SR protein yield in the hypertrophied hearts (Table 3). The Ca^{2+}-pump ATPase of SR was not affected in normal rats by sucrose feeding significantly, but the reduction in the activity of this ATPase in the pressure-loaded ventricles was prevented by sucrose feeding (Table 3).

Table 2. Effect of sucrose feeding on myosin isoenzyme populations in left ventricles of rats in the presence or absence of pressure-overload

Myosin	Control	Sucrose-fed Control	Pressure-loaded	Sucrose-fed Pressure-loaded
V_1 (%)	53.7 ± 12.1	60.1 ± 14.5	35.9 ± 6.2*	50.6 ± 9.7**
V_2 (%)	27.4 ± 4.9	24.5 ± 7.7	33.0 ± 1.7	29.7 ± 3.9
V_3 (%)	18.9 ± 8.0	15.4 ± 7.6	31.1 ± 6.0*	19.7 ± 6.2**

Each value is a mean ± S.E. of 8 experiments.
*significantly different ($P < 0.01$) with respect to control group.
**-significantly different ($P < 0.01$) with respect to pressure-loaded group.

Table 3. Effect of sucrose feeding on cardiac SR protein yield, cardiac SR Ca^{2+}-stimulated ATPase activity and plasma concentrations of T_4, T_3 as well as glucose in rats in the presence or absence of pressure overload

	Control	Sucrose-fed Control	Pressure-loaded	Sucrose-fed Pressure-loaded
SR yield (mg/g)	1.15 ± 0.14	1.20 ± 0.16	1.25 ± 0.20	1.23 ± 0.12
SR ATPase activity (nmol P_i/mg/min)	145 ± 26	159 ± 25	89 ± 19*	131 ± 13**
Plasma T_4 (ug/dl)	5.2 ± 0.1	5.6 ± 0.6	5.5 ± 0.5	6.0 ± 0.4
Plasma T_3 (ng/dl)	78 ± 5	83 ± 6	73 ± 6	84 ± 7
Plasma glucose (mg/dl)	149 ± 16	158 ± 8	151 ± 6	145 ± 7

Each value is a means ± S.E. of 8 experiments.
*significantly different ($P < 0.02$) from control group.
**-significantly different ($P < 0.02$) from pressure-loaded group.

In order to gain some information regarding possible mechanisms by which the observed changes in subcellular organelles in the hypertrophied heart were prevented by sucrose feeding, plasma concentrations of thyroid hormones and glucose were determined (Table 3). No significant changes in the plasma glucose as well as T_3 and T_4 levels were seen in both control and aortic banded animals upon feeding sucrose. For demonstrating a possible correlation between myosin isoenzyme redistrileution and changes in the SR Ca^{2+}-pump ATPase in the hypertrophied hearts, the population of myosin alpha-heavy chains (HC_{alpha}), as calculated by $V_2/2 + V_1$, was plotted versus Ca^{2+}-pump ATPase activity (Fig.1). Noteworthy is that an increase in the proportion of HC_{alpha} was associated with a higher Ca^{2+}-pump ATPase activity; such a relationship suggests that trigger mechanisms for remodeling of myosin and SR Ca^{2+}-pump proteins in cardiac hypertrophy may be operating in a coordinated manner.

Discussion

The observations regarding the reduction in myosin V_1 as well as SR Ca^{2+}-pump ATPase activity and increase in myosin V_3 in hypertrophied

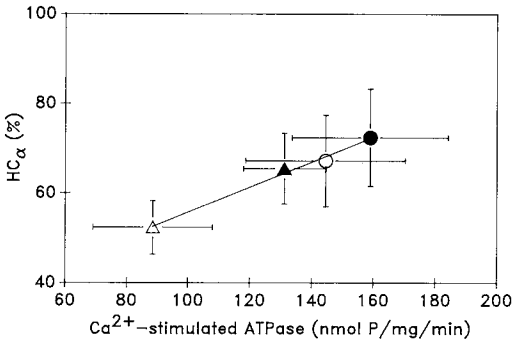

Figure 1. Relationship between changes in myosin isoenzymes and SR Ca^{2+}-pump ATPase activity for (○) control rats, (●) sucrose-fed control rats, (△) rats with pressure-overload and (▲) sucrose-fed rats with pressure-overload. A linear regression was used to calculate the line (y = 0.28 x + 27.8, r = 0.99).

hearts are consistent with the findings reported earlier (6,10,11). The data in this study demonstrate that the changes in myosin and SR in the compensated stage of cardiac hypertrophy can be prevented by sucrose feeding. In contrast to swimming exercise which greatly reversed the increase in myosin V_3 (7,8), sucrose feeding did not affect the ventricular mass and is not expected to alter ventricular geometry. This treatment could also be applied after a prolonged pressure-overload where exercise would not be advisable. Another possible approach leading to a reduction in myosin V_3 could involve treatment with thyroid hormones (12) which again would not be acceptable because of their marked metabolic effects. The treatment with sucrose is of particular interest because it affects not only the myosin but also the SR Ca^{2+}-pump ATPase and thus resulting in an increased potential for fast shortening and relaxation of the hypertrophied cardiac muscle. However, further studies with respect to the functional consequences of sucrose treatment as well as modification of the onset of cardiac dilatation and heart failure due to chronic pressure-overload remain to be carried out for making any meaningful conclusion.

Sucrose as well as fructose feeding has been shown previously to increase the proportion of myosin V_1 in the hearts of hypophysectomized

and thyroidectomized rats (13) but these treatments were reported to exert either a small (14) or no effect (15) in normal adult animals. Accordingly, it was suggested that some neuro-endocrine mechanisms may be involved in mediating the effects of carbohydrate feeding. However, the results in the present study revealed no significant changes in plasma thyroid hormone levels in sucrose-fed aortic banded animals. Nonetheless, a steady intake of sucrose by the animals is likely to initiate gluco-regulatory signals in the gastro-intestinal tract (16) and these can be seen to result in a shift of metabolic fluxes in the myocardium to favour the utilization of glucose. Such a mechanism may involve the participation of the sympathetic nervous system because sucrose feeding has been shown to increase the peripheral adrenergic activity in rats (17). The metabolic shift in fuel utilization by the myocardium may be an important determinant for the alterations in subcellular organelles in the myocardium because in chronic diabetes, where glucose utilization is markedly impaired, changes in myosin isoenzyme distribution and SR Ca^{2+}-pump ATPase were found to be similar to those seen in hypertrophied hearts (18,19). It is, therefore, suggested that a shift in the fuel utilization by the myocardium may be responsible for remodeling of the subcellular components upon feeding sucrose to rats with chronic pressure-overload.

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