Relation of Growth Hormone and Bioelements with Ischemic Manifestation in Patients with Stable Angina Pectoris

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The purpose of this study was to determine association of growth hormone (GH) and bioelements with ischemic manifestation in patients (pts) with stable angina pectoris (SAP) without coronary stenosis. For this purpose we have investigated GH and bioelements (Zn, Cu, Mn, Mg, Ca) in association with the grade of stenosis of coronary arteries (CA) in pts with SAP before and after exercise testing. The following groups of male pts with SAP (aged 40–65 years) have been investigated: 21 pts without CA stenosis; 15 pts with CA stenosis <50%; 28 pts with stenosis ≥50%, and a control group of 30 healthy men. Diagnosis of angina pectoris was established on the findings of pain, clinical manifestations, ECG, physical testing and coronarography. GH level in plasma was determined by radioimmune method. The concentration of bioelements (Zn, Cu, Mn, Mg, Ca) in plasma, erythrocytes and hair was detected by absorption spectrophotometry. GH level in plasma of pts without CA stenosis and with stenosis ≥50% was significantly decreased. The concentration of bioelements in plasma, erythrocytes and hair was dependent on the degree of CA stenosis. In pts without CA stenosis and only with a lowered GH concentration in plasma, with a decreased Zn content in erythrocytes and with an increased Mn level in hair, ischemic manifestations were revealed during physical testing. In pts with stenosis ≥50% the bicycle stress test initiated ischemic manifestations or the response was non-informative, these were independent of GH level and of the levels of bioelements. Thus, GH and bioelements have an influence on ischemic manifestations in pts without CA stenosis during physical load.

Key words: stable angina pectoris, growth hormone, bioelements (Zn, Cu, Mn, Mg, Ca), plasma, erythrocytes, hair

INTRODUCTION

Internists are more or less sure in diagnosing coronary artery disease (CAD) when there are evident essential risk factors such as typical clinic and CA stenosis. In cases when pts complain of anginal pain but there are no atherosclerotic injuries found in the CA by angiography, it is necessary to determine whether the above-mentioned complaint is caused by some other condition or it is the very first phase of CAD, manifested by a strong vasoconstriction or spasm.

At the present time it is known that the dysfunction of the central nervous system (1, 2) causing a disorder secretion of hormone plays an important role in the pathogenesis of CAD. It is a well-established fact that GH by modulating the activity of adrenoreceptors realise their action, inducing quantitative changes of biochemical substrates including bioelements, which are closely associated with heart insufficiency, cardiac rhythm and conduction disorders, as well as with sudden coronary death, including contractility of cardiomyocytes and smooth muscles (3–6). In addition to the data of Brown et al. (7), GH is required for the development of normal contractile capability of cardiac and vascular tissues, since it differentially regulates atrial and ventricular responsiveness. Administration of human GH resulted in a significant decrease in the incidence of ventricular aneurysms. Scanning electron microscopy has revealed a good preservation of the connective tissue components of the myocardium (8).

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On the other hand, repeated exercise tests are the most widely accepted means for estimation of the course of CAD and of the efficacy of treatment. The low workload established at exercise testing is one of the most informative predictors, especially if it is associated with ST-segment depression and anginal pain (9, 10). Thus, the purpose of this study was to determine association of GH and bioelements with ischemic manifestation in pts without coronary stenosis during exercise testing.

MATERIALS AND METHODS

The following groups of male pts (aged 40–65 years) with stable angina pectoris were investigated: 21 pts without CA stenosis; 15 pts with CA stenosis <50%; 28 pts with coronary stenosis ≥50%, and a control group of healthy men. Stenosis in one CA was found in 8 (19%) cases, in two arteries in 25 (58%), in three arteries in 10 (23%) pts. All subjects were examined and treated at the Cardiology Outpatient Department of Kaunas Medical University. Stable angina pectoris was diagnosed relying on the findings of clinical examination and exercise stress testing and was verified by coronary angiography. The workload at the beginning of exercise testing was 50 W and increased by 25 W every 3 min. A 12-lead ECG was recorded at rest, then at the peak of the exercise and for 6 min after its termination; a $V_{1–6}$ –lead ECG was recorded every minute during the test and during the recovery period. The ischemic response was defined as a horizontal or downsloping ST depression of >0.5 mm, occurring 0.08 s after the J point, or as a typical anginal pain. On the other hand, the bicycle stress test was considered no ischemic, if ST-segment depression and/or anginal pain did not appear and the submaximal heart rate (HR) was achieved. The bicycle stress test was considered noninformative, if ST-segment depression and/or anginal pain did not appear, and the submaximal HR was not achieved, but the testing was terminated for some other conditions such as fatigue, dyspnea, arterial hypertension, and rhythm disorders. The rate-pressure outcome was calculated using maximal HR and systolic blood pressure values recorded during exercise.

The GH level in blood plasma of pts was determined by the radioimmune method. The concentration of bioelements (Zn, Cu, Mn, Mg, Ca) in blood plasma, in erythrocytes and in hair was detected by atomic absorption spectrophotometry (503, Perkin-Elmer, USA). Details of the methods of analysis are contained in reference (6). In all group of pts the concentration of GH and bioelements was es-

Table 1. **Changes in the level of growth hormone (GH) and amount of bioelements in plasma (P), erythrocytes (E) and hair (H) depending on the grade of stenosis of coronary arteries (CA) in patients with stable angina pectoris**

Biochemical findings	Group of patients			
	Healthy	Without CA stenosis	With CA stenosis <50%	With CA stenosis ≥50%
	Group 1 (n = 30)	Group 2 (n = 21)	Group 3 (n = 15)	Group 4 (n = 28)
GH: ng/ml	1.62 ± 0.17	0.9 ± 0.18*	1.58 ± 0.25	0.36 ± 0.05**
Zn: P, μmol/l	26.6 ± 1.1	25.6 ± 1.5	31.5 ± 1.6****	27.1 ± 1.7
E, μmol/l	143.2 ± 2.9	154.6 ± 5.6	145.2 ± 10.3	158.1 ± 5.3
H, μg/g	167.2 ± 4.7	194.3 ± 8.9	209.1 ± 20.1	191.1 ± 7.0
Cu: P, μmol/l	16.1 ± 0.5	23.7 ± 0.87	26.5 ± 3.65	23.0 ± 0.81
E, μmol/l	12.4 ± 0.4	21.6 ± 0.94	19.4 ± 1.2	21.8 ± 0.89
H, μg/g	17.7 ± 0.98	18.8 ± 1.0	24.5 ± 2.6***	24.5 ± 1.97***
Mn: P, μmol/l	1.37 ± 0.05	1.76 ± 0.08	2.77 ± 0.37	2.25 ± 0.25
E, μmol/l	2.44 ± 0.16	2.23 ± 0.09	1.92 ± 0.13	2.02 ± 0.1
H, μg/g	4.45 ± 0.21	4.1 ± 0.58	4.2 ± 0.88	5.78 ± 0.68
Mg: P, mmol/l	1.16 ± 0.08	0.8 ± 0.03	0.85 ± 0.06	0.77 ± 0.03
E, mmol/l	1.86 ± 0.08	1.94 ± 0.08	1.69 ± 0.09	2.01 ± 0.08*****
H, μg/g	280.6 ± 20.4	165.1 ± 16.5	295.2 ± 48.1	150.1 ± 17.4*****
Ca: P, mmol/l	2.69 ± 0.07	2.36 ± 0.05	2.41 ± 0.2	2.32 ± 0.05
E, mmol/l	1.12 ± 0.11	0.76 ± 0.04	0.86 ± 0.03	0.72 ± 0.04*****
H, μg/g	1206 ± 104	2321 ± 220	3247 ± 210***	3279 ± 219***

Note. Values are given as mean ± SEM; *P < 0.05 *versus* GH level in Group 1;
**P < 0.05–0.01 *versus* GH level in Groups 1, 2, 3; ***P < 0.05–0.01 *versus* Cu and Ca concentration in Groups 1, 2; ****P < 0.05 *versus* Zn level in Groups 1, 2; *****P < 0.05–0.01 *versus* Mg and Ca levels in Groups 1, 3.

Table 2. **Changes in the level of growth hormone (GH) and the concentration of bioelements depending on the grade of stenosis of coronary arteries (CA) and response to exercise testing in patients with stable angina pectoris**

Group of patients and response to exercise testing	GH level in plasma ng/ml	Mn concentration in hair µg/g	Zn amount in erythrocytes µmol/l
Without CA stenosis:			
1. Ischemic response (n = 10)	A. 0.18 ± 0.08*	A. 7.9 ± 1.44**	A. 136.7 ± 9.1***
	B. 0.05 ± 0.04	B. 6.01 ± 1.91	B. 148.1 ± 15.4
2. No ischemic response (n = 11)	A. 1.47 ± 0.53	A. 4.05 ± 0.66	A. 187.1 ± 18.0
	B. 1.98 ± 0.70	B. 5.66 ± 2.49	B. 168.7 ± 21.4
With CA stenosis ≥50%:			
3. Ischemic response (n = 18)	A. 0.11 ± 0.09		
	B. 0.07 ± 0.06		
4. Noninformative response (n = 10)	A. 0.08 ± 0.03		
	B. 0.27 ± 0.14		

Note. Values are given as mean ± SEM; A – data before exercise testing; B – data after exercise testing; *P < 0.05 *versus* GH level in Group 2 A; **P < 0.05 *versus* Mn concentration in Group 2 A; ***P < 0.05 *versus* Zn level in Group 2A. The data of other bioelements are not presented, because their concentration changes were not statistically significant.

tablished before exercise testing, but in pts without coronary stenosis as well as in pts with stenosis ≥50% the level of bioelements and GH was also determined additionally after exercise.

Student's paired t test was applied when performing statistical analysis and establishing differences in p values (<0.05 was considered to be significant).

RESULTS

Independently of physical load, i.e. before exercise testing, the findings of GH and bioelements are presented in Table 1. The concentration of GH in the blood plasma of pts without stenosis of CA was significantly decreased as compared with that of healthy persons. The amount of GH in the blood plasma of pts with coronary stenosis ≥50% was significantly decreased in comparison with the respective control group, pts without artery stenosis and with stenosis <50%. Besides, the level of Cu and Ca in the hair of pts with coronary stenosis (<50% and ≥50%) was higher than of healthy persons and of pts without coronary stenosis. The content of Zn in the blood plasma of pts with coronary stenosis <50% was significantly higher in comparison with that of the respective healthy persons and of pts without stenosis. In pts with stenosis ≥50% the concentration of Ca in erythrocytes and Mg in hair was lower than in pts with coronary stenosis <50% and in healthy men, but the level of Mg in erythrocytes was higher than in pts with only coronary stenosis <50%. Investigation findings obtained after exercise testing are presented in Table 2. In pts without CA stenosis who manifested myocardial ischemia during physical load, the concentration of GH in plasma, the level of Zn in erythrocytes were lower, while the level of Mn was higher in the hair as compared with that in pts that developped no ischemia during the test. There was also a significant correlation between the decreased level of GH in plasma and of Zn in erythrocytes ($r = 0.75$; $P < 0.05$), as well as between the lowered level of GH and the increased concentration of Mn in the hair ($r = -0.81$; $P < 0.05$) of pts without stenosis. In pts with coronary stenosis ≥50% the bicycle stress testing initiated ischemic manifestations or the response was noninformative. In both cases, depending upon physical load, the level of GH in the plasma of pts did not change. There were also no changes in the content of bioelements.

DISCUSSION

A significantly decreased concentration of GH in the blood plasma of pts without coronary stenosis in comparison with the respective healthy persons and the low plasma level of this hormone in pts with coronary stenosis ≥50% (Table 1) point to a relation of GH with the development of CAD. In some phases of the disease, the level of GH may rise (as according to our data in pts with stenosis <50%), and this may have a compensatory character. Besides, the rise in the content of Zn in the plasma of pts with stenosis <50% may have also influenced the stabilisation of stenosis development in the arteries, because Zn plays a critical role in maintaining the endothelial cell integrity and can act as an endogenous protective factor against athe-

rosclerosis by inhibiting the oxidation of low-density lipoproteins in cells (11). The increased concentration of Ca and Cu in the hair of pts with coronary stenosis may be associated with the atherogenic processes, causing decreased levels of this element in other tissues, particularly in arteries. According to the data of Ustdal et al. (12), the concentration of elements (Zn, Cu, Mn, Mg, Ca) in arterial tissues of atherosclerotic pts was found to be significantly decreased. Decreased levels of Cu, Mn, Mg Ca in the thoracic aorta caused by hypodynamic stress was detected together with contractility derangements of smooth muscles (13). Besides, it is known that risk factors of CAD such as hypercholesterolemia and others enhance the production of free radicals, which damage the membrane of cells. Thus, the decreased level of Ca in the erythrocytes of pts with arterial stenosis ≥50% may be related to the increased permeability of erythrocyte membranes. The increased level of Mg as a constituent of enzymes Ca^{2+}, Mg^{2+}-ATP-ase in erythrocytes may have a compensatory character affecting the stabilisation of the membrane. According to our previous research data (14) hypercholesterolemia, smoking, overweight significantly decrease the level of Ca and increase the concentration of Mg in erythrocytes in pts with CAD. Ischemic response to physical testing in pts with stable angina pectoris depended on the grade of CA stenosis and on the level of GH and bioelements. The low level of Zn, which is a constituent of the enzyme carbonic anhydrase participating in the metabolism of oxygen in erythrocytes might have influenced the ischemic response to bicycle stress test in pts without stenosis. Besides, a decreased level of GH and an increased content of Mn in hair, and their material correlation may be related to a strong vasoconstriction in pts without CA stenosis during physical load. The findings correlate well with the data obtained from an experiment on pigs receiving GH. It was established that the concentration of Mn in the hair of pigs correlated well with the contractility force of CA (3). In addition, Mn ions at a millimolar concentration can induce cell membrane depolarization and directly activate contractile proteins in smooth muscles (15).

In conclusion, the correlation between the lowered GH concentration and the increased Mn level in hair, as well as the lowered Zn level in erythrocytes of pts without artery stenosis suggest myocardial ischemia. Normally the concentration of GH has no influence on ischemic manifestations during exercise testing in pts without CA stenosis. In cases of badly damaged CA, *i.e*. when stenosis ≥50%, the character of the response (ischemic or noninformative) to physical load was independent of the level of GH and of the content of bioelements in plasma, erythrocytes and hair in pts with stable angina pectoris. Thus, GH and bioelements are associated with ischemic manifestations in pts with stable angina pectoris without CA stenosis during physical load.

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SERGANČIŲJŲ STABILIA KRŪTINĖS ANGINA AUGIMO HORMONO IR BIOELEMENTŲ RYŠYS SU ISCHEMIJOS PASIREIŠKIMU

S a n t r a u k a

Darbo tikslas – nustatyti sergančiųjų stabilia krūtinės angina be vainikinių arterijų (VA) stenozės augimo hormono (AH) ir bioelementų (Zn, Cu, Mn, Mg, Ca) ryšį su ischemijos pasireiškimu. Tirti vyrai nuo 40 iki 65 metų: 30 sveikų ir 64 sergantys stabilia krūtinės angina (be vainikinių arterijų stenozės ir su vainikinių arterijų stenoze, išreikšta <50% ir ≥50%). Stabilios krūtinės anginos diagnozė nustatyta pagal skausmo sindromo charakteristiką, dozuoto fizinio krūvio (veloergometrijos), elektrokardiografijos ir koronarografijos duomenis. Bioelementų koncentracija kraujo plazmoje, eritrocituose ir plaukuose tirta atomine absorbcija, AH koncentracija kraujo plazmoje – radioimuniniu metodu.

AH koncentracija kraujo plazmoje sergančiųjų stabilia krūtinės angina be VA stenozės ir esant kraujagyslių stenozei (≥50%) buvo statistiškai patikimai sumažėjusi, palyginus su sveikais asmenimis ir sergančiaisiais, kurių VA stenozė buvo <50%. Bioelementų kiekis plazmoje, eritrocituose ir plaukuose priklausė nuo stenozės pasireiškimo. Ligonių be VA stenozės sumažėjęs AH kiekis plazmoje, Zn eritrocituose, padidėjusi Mn koncentracija plaukuose siejosi su ischemijos pasireiškimu dozuoto fizinio krūvio metu. Ischemijos išsivystymas krūvio metu ligoniams, kurių VA stenozė buvo išreikšta ≥50%, nepriklausė nuo AH ir bioelementų koncentracijos.

Raktažodžiai: stabili krūtinės angina, augimo hormonas, bioelementai (Zn, Cu, Mn, Mg, Ca), plazma, eritrocitai, plaukai