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Changes of tyrosine blood concentrations in patients undergoing coronary artery bypass grafts

It is well know that tyrosine is an endogenous amino acid which is produced by the organism from phenylalanine by hydroxylation. This process is strictly dependent on tyrosine hydroxylase – the enzyme directly affecting the rate of synthesis of catecholamines and thus the use of their substrate – tyrosine. The influence of extracorporeal circulation (ECC) on the levels of the amino acid discussed is not clearly defined and the literature reports emphasize the importance of intra- and post-operative tyrosine changes (14). A multi-stage character of ECC procedures as well as intraoperative therapy used may not only affect blood tyrosine levels in a direct way but also through the adrenergic reaction, which is widely known and described by many authors (3, 4).

The aim of the study was to assess the changes in blood tyrosine levels in patients undergoing bypass procedures with ECC and normovolemic haemodilution.

MATERIAL AND METHODS

The study was approved by the Bioethical Committee of the Medical University of Lublin (No KE-0254/244/2000) and included the patients who underwent operations due to I° and II° coronary disease (according to CCS).

In the evening preceding the operation the patients were administered premedication – oral lorazepam (Lorafen, Polfa, PL) – 2mg and i.m. Promethiazine (Dophergan, Polfa, Pl) – 50mg. One hour before anaesthesia all the patients received oral lorazepam – 3mg and i.m. morphine (Morphicum hydrochloricum, Polfa, Pl) – 0.1mg/kg body wt. The patients underwent general anaesthesia with fentanyl (Fentanyl, Polfa, Pl) in the dose of 0.01–0.02mg/kg body wt., midazolam (Dormicum, Roche,) – 0.05–0.1mg/kg body wt. and etomidat (Hypnomidat, Janssen, G) – 0.1–0.5mg/kg. Muscle relaxation was obtained by injecting a single dose (0.08–0.1mg/kg body wt.) of pancuronium (Pavulon, Organon-Teknica, F). The anaesthesia was maintained throughout the procedure using midazolam-fentanyl infusion and inhalatory fractionated doses of foran (Izofluran, Abbot, USA). During the implantation of aorto-coronary by-passes the circulation and ventilation were maintained by the heart-lung machine S III(Stockert). The following substances were used for priming: Ringer«s solution (Ringer, Fresenius-Kabi,G) – 1000ml, 6% solution of hydroxyethylated starch (HAES, Fresenius-Kabi,G) – 500ml, 20% mannitol (Mannitol, Fresenius-Kabi,G) – 250ml, sodium hydroxycarbonate (Natrium bicarbonatum, Polfarma Pl) – 20ml and heparin – 75ml. Cardioplegia was prepared using 0.9% salt solution supplemented with 3g of potassium chlorine (Kalium chloratum, Polfa, Pl) and 20ml of sodium hydroxycarbonate.

Taking into consideration the complex character of procedures and intraoperative treatment applied, the patients were divided according to the treatment (first division – 1) and degree of normovolemic haemodilution (second division – 2). In the first case the patients were divided into 3 groups: group A1- the patients not requiring catecholamine infusions, group B1-those receiving dopamine in the doses adjusted to their clinical condition and group C1- those receiving dobutamine in the doses suitable for their clinical condition. According to the degree of normovolemic haemodilution, the patients were divided into 2 groups: A2- those weighing<75kg and group B2- those weighing>75kg. The degree of normovolemic haemodilution induced by constant volume of priming (1800ml) was determined on the basis of haematocrit measurements and body weight.

All the patients consumed their last meals 12 hours before surgery; immediately after the procedure they were transported to the Postoperative Intensive Care Unit (PICU), where they received a short-term infusion of 5% glucose solution with insulin. Determinations were performed in 5 stages: 1) after the radial artery cannulation and before the innitiation of anaesthesia and operation, 2) during deep hypothermia, 3) after the operation but before sending to the PICU, 4) on the 1st postoperative day, 5) on the 2nd postoperative day. Blood samples were collected from the radial artery and centrifuged (25000r/min., temp.0°C); the obtained serum was frozen at -20°C. The tyrosine levels were determined after serum deproteinization with 6% sulphosalicylic acid solution using liquid chromatography in the apparatus AAA-400 INGOS, Prague.

The results were statistically analysed using the Wilcoxon and Mann-Whitney tests in interstage and intergrup relations.

RESULTS

The examinations were conducted in 30 men aged 53–70 (63.1±5.2). Twenty three patients had myocardial infarction during the past 3 years and 26 were treated due to concomitant arterial hypertension (I° according to WHO classification). None of the patients was treated for endocrinological, neurological and other systemic diseases or was resuscitated because of circulatory arrest. The mean duration of the procedure was 215 min±41 and of anaesthesia 233min.±45. In all the patients the aorta was typically clamped and the mean closure time was 48.21min.±10.5. The aorto-coronary anastomosis was performed in shallow hypothermia whose value was 34.51°C±0.41. In all the cases the heart-lung machine disconnection was uneventful and there was no need of intra-aortic contrpulsation. In the examined group 10 patients did not require catecholamine substitution throughout the examination period (group A1); 5 patients needed dopamine infusions in the 3rd stage (group B1) in the doses dependent on their clinical state (5–15μg/kg body wt./min. - average 8.1±1.93) and 15 were administered dobutamine infusions (group C1) in the same stage (3–15μg/kg body wt./min. – average 9.32±2.71).

The analysis of blood tyrosine concentration showed its decrease in all patients in the 3rd and 4th stages ($p<0.01$) and increase in the 5th stage ($p<0.05$) (Table 1). Decreased levels of this parameter ($p<0.05$) were also observed in the group without catecholamines and in the group receiving dopamine in the 3rd stage (Fig.1, 2).

Compared to the 1st stage, the highest decrease in the stages 2,3,4 was observed in group A2 ($p<0.05$). The analysis of blood tyrosine concentration revealed its decrease ($p<0.05$) in group A2 in stages 3 and 4 and its increase in group B2 in stage 5 (Fig.3, 4). In the group of 10 patients weighing less than 75kg, six required dobutamine and two – dopamine infusions; in the group of 20 patients whose body weight was >75kg, ten required dobutamine and three – dopamine infusions.

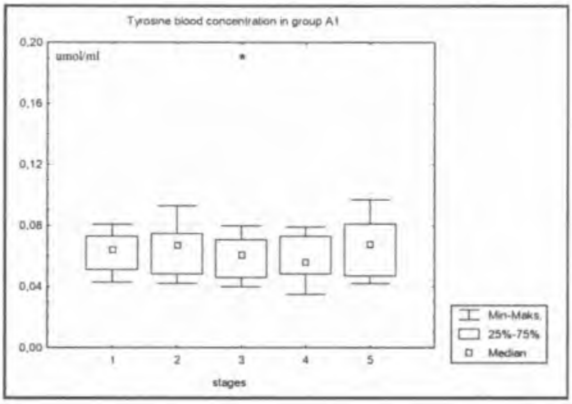


Fig. 1. Changes of tyrosine blood concentrations in patients without catecholamines supplementation

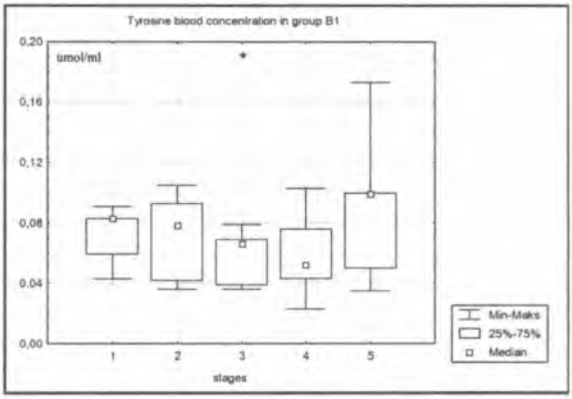


Fig. 2. Changes of tyrosine blood concentrations in patients with dopamine infusion

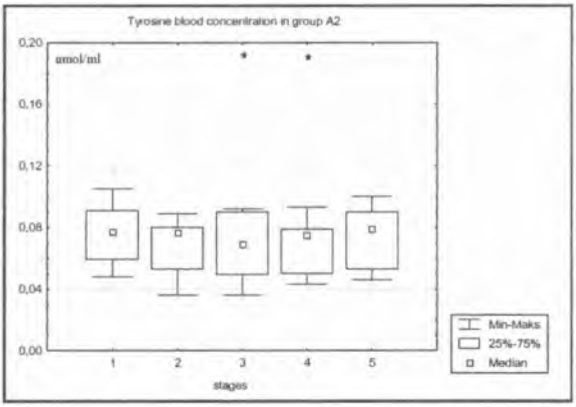
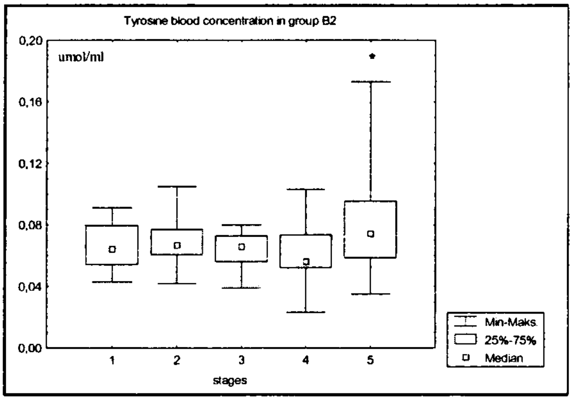


Fig. 3. Changes of tyrosine blood concentrations in patients with body mass less than 75 kg



* $p < 0.05$ comparison with stage 1

Fig. 4. Changes of tyrosine blood concentrations in patients with body mass more than 75kg

Table 1

Tyrosine blood concentration in each stages						
Stages		1	2	3	4	5
General	median	0.069	0.069	0.0675**	0.062**	0.0755*
	quartile 1	0.05675	0.05975	0.054	0.052	0.05725
	quartile 3	0.081	0.0795	0.0745	0.076	0.093
Tyrosine blood concentration in each groups						
Group A1	median	0.0645	0.0675	0.061*	0.056	0.068
	quartile 1	0.05125	0.05075	0.048	0.049	0.04925
	quartile 3	0.07225	0.0745	0.0705	0.07025	0.07975
Group B1	median	0.083	0.078	0.066*	0.052	0.099
	quartile 1	0.059	0.042	0.039	0.043	0.05
	quartile 3	0.083	0.093	0.069	0.076	0.1
Group C1	median	0.074	0.07	0.066	0.067	0.079
	quartile 1	0.0595	0.062	0.061	0.055	0.0685
	quartile 3	0.0855	0.0785	0.0775	0.0785	0.092
Intergroup relationships						
A1:B1 (p)		0.254412	0.767899	0.678655	0.859141	0.370962
A1:C1 (p)		0.19628	0.530544	0.128889	0.115134	0.177495
B1:C1 (p)		0.932792	0.800052	0.230005	0.394866	0.672472
Haematocrite level						
Group A2	median	38.85	23.2**	29.3**	35.4*	37.1
	quartile 1	37.25	22.1	28.75	34.3	35.425
	quartile 3	42.025	24.15	29.925	36.65	39.525
Group B2	median	41	27.05*	29.95*	34.15*	36.75
	quartile 1	38.025	26.875	29.325	32.1	36.4
	quartile 3	44.1	27.8	30.075	35.15	39.425
Tyrosine blood concentration						
Group A2	median	0.077	0.0765	0.069*	0.075*	0.079
	quartile 1	0.06125	0.0585	0.05025	0.05425	0.05825
	quartile 3	0.089	0.0795	0.08625	0.07875	0.08825
Group B2	median	0.0645	0.067	0.066	0.0565	0.0745*
	quartile 1	0.055	0.06125	0.057	0.052	0.05975
	quartile 3	0.07875	0.075	0.073	0.07325	0.09475
Intergroup relationships						
A2:B2 (p)		0.248419	0.558847	0.649616	0.248419	0.879765

*$p < 0.05$ comparison with stage 1, ** $p < 0.01$ comparison with stage 1

DISCUSSION

The effects of ECC on the blood tyrosine concentration have not been fully explained. The operative stress alone as well as the treatment used are likely to affect the levels of this parameter as it is widely known that this amino acid is a substrate of the cycle producing catecholamines, which seems important in hypercatecholaminemia observed during such procedures (3, 4). Moreover, many authors stress an important influence of stress not only on catecholamine secretion but also on the activity of tyrosine hydroxylase (TH) – the enzyme acting directly on the tyrosine molecule and indispensible in the production cycle of catecholamines (5, 12) since it is known that increased TH activity affects the blood tyrosine level. Therefore, it may be supposed that changes in this activity should be reflected in variations of blood tyrosine levels. The question is, however, whether the operative procedure results in TH activity changes. Examining the changes in blood adrenaline and noradrenaline concentration in patients after partial hepatectomy, Knopp et al. (8) observed a significant increase in TH activity resulting from intensified synthesis of both catecholamines. Likewise, Adams and McMillen (1), who studied the effects of stress and hypoxia on the catecholamine synthesis, observed increased activity of the above-mentioned enzyme resulting from prolonged hypoxia. Moreover, Erdem et al. (5) studying the effects of exercise on the synthesis of catecholamines noted a 35% increase in TH concentration in the adrenal glands associated with a 62% increase in the activity of this enzyme. Although it is difficult to define explicitly whether increased TH activity significantly affects tyrosine levels in blood serum, it may be supposed that this relation is not substantially disturbed in stress situations. Comparing the blood tyrosine levels in healthy women and those who underwent gynaecological operations, Gomar et al. (7) noted a significantly higher concentration of the amino acid in question in the operated group, which may suggest that stress situations affect tyrosine levels in blood serum. A decrease in the tyrosine level after using opioids and lack of any changes during operations with high doses of fentanyl are also worth stressing. Moreover, it should be added that analgesics significantly decrease the adrenergic response of the organism to stress thus decreasing blood catecholamine levels. Therefore, it is likely that high doses of opioids used in our study substantially limited the response to operative stress and the changes observed might have resulted from the ECC procedure itself. However, many authors emphasize the effects of hypothermia as well as normovolemic haemodilution on catecholamine levels, which seems to affect also the levels of the discussed amino acid (4, 6, 9). Lehot et al. (9) observed that catecholamine levels greatly depended on the degree of hypothermia. So it seems that mild hypothermia used in our study is likely to affect blood tyrosine concentration. This is confirmed by the observations of Sabban and Kvetnansky (12) concerning the effects of low temperatures on TH activity. They also noted that repeated, prolonged exposure of the organism to low temperatures resulted both in increased TH activity and increased dopamine â-hydroxylase activity. Thus, it may be thought that decreased blood tyrosine levels observed by us in all the patients were caused by its increased hydroxylation; however, the TH activity was not determined in our material and direct influence of hypothermia on tyrosine levels is not fully defined and requires further studies.

Another question is whether the level of the discussed amino acid also depends on intraoperative treatment. The examinations revealed its changes in the post-operation-before PICU group of patients who did not require catecholamine infusions and in the group of patients receiving dopamine. Thus, we suppose that decreased parameter levels observed in group A1 are likely to result from the above-mentioned reraction of the organism to operative stress (7, 14). However, it is difficult to explain a decrease in this amino acid observed by us in the patients receiving dopamine infusions. Many authors believe that the system of dopaminergic receptors is strongly involved in the regulation of TH activity, which would explain the changes observed (10, 15). However, Lindgren et al.(10) in their study analysing the effects of agonists of dopaminergic receptors on TH in the nervous system observed its decreased activity caused by the stimulation of D_2 receptors. Booth and Baldessarini (2), on the other hand, stress that the stimulation of D_1 receptors does not affect the TH activity. So it may be supposed that decreased blood tyrosine levels observed in our study during dopamine infusions were not caused by catecholamine effects on TH activity since according to the above authors the inhibition of its activity should lead to an increase and not decrease in the level of this amino acid. However, it should be emphasized that the authors mentioned above studied these relations in the nervous tissue and not in the serum. What is then a decrease in the serum tyrosine level caused by? The activity of dopaminergic system (11) and intraoperative disorders of tissue perfusion causing transient hypoxia (13) are implicated. In addition, the inductive effects of hypoxia on TH activity are worth stressing. Smitt et al. (13) observed significantly higher TH activities during hypoxia leading to increased synthesis of catecholamines. It seems, however, that the changes in blood tyrosine levels in our study do not result only from the influence of hypoxia as the changes observed by Smitt et al. (13) concerned long-term hypoxia.

It is also difficult to explain decreased blood tyrosine levels observed by us in group A2. The direct effects of intraoperative haemodilution on the levels of this amino acid are not well documented in literature. It seems that haemodilution alone is likely to decrease the level of this amino acid. Moreover, significant catecholaminemia observed by Dąbrowski et al. (4) at bigger haemodilution is also likely to be relevant. Estafanous et al. (6) studying the effects of normovolemic haemodilution on noradrenaline levels in blood serum also demonstrated a strict relation between its concentration and the degree of normovolemic haemodilution. However, it can not be explicitly stated that normovolemic haemodilution applied during ECC procedures does not disturb the tyrosine-noradrenaline relation. Moreover, the direct influence of the operation and anaesthesia on blood tyrosine changes, and the tyrosine-TH relation is not known. Therefore, precise explanation of the causes of blood tyrosine level changes observed during ECC procedures seems to be impossible and further studies are required.

CONCLUSIONS

1. ECC procedures result in blood tyrosine level changes.
2. Dopamine infusions cause decreased blood tyrosine levels.
3. The degree of haemodilution affects changes in blood tyrosine levels.

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SUMMARY

The aim of the study was to assess the changes in blood tyrosine levels in patients undergoing bypass procedures with ECC and normovolemic haemodilution. 30 male patients underwent CABG under general anaesthesia in extracorporeal circulation (ECC) in normovolemic haemodilution with 1800 ml of priming. Patients were divided according to the treatment (first division – 1) and degree of normovolemic haemodilution (second division – 2). The degree of haemodilution varied between groups because of different relation between body mass and the same priming volume in all cases. The tyrosine blood concentrations were noted in five stages: 1 – before surgery, 2 – during ECC, 3 – after surgery, 4 – on the first postoperative day, 5 – on the second postoperative day. The study showed: its decrease in all patients in the 3rd and 4th stages and increase in the 5th stage, and its decrease in 3rd and 4th stages in group with less body weight. Decreased levels were also observed in the group without catecholamines and in the group receiving dopamine in the 3rd stage. Conclusions: 1) ECC procedures result in blood tyrosine level changes; 2) dopamine infusions cause decreased blood tyrosine levels; 3) the degree of haemodilution affects changes in blood tyrosine levels.

Zmiany stężenia tyrozyny we krwi pacjentów poddanych zabiegom pomostowania naczyń wieńcowych

Wpływ procedury krążenia pozaustrojowego (ECC - *Extracorporeal Circulation*) na zmiany stężenia tyrozyny we krwi nadal pozostaje tematem dyskusji i badań. Celem pracy była ocena zmian stężenia tego aminokwasu u pacjentów poddanych chirurgicznej rewaskularyzacji mięśnia sercowego z zastosowaniem ECC oraz normowolemicznej hemodilucji. Obserwacje przeprowadzono w grupie

30 pacjentów, u których wszczepienia pomostów aortalno-wieńcowych dokonano w ECC. Ze względu na złożony charakter procedury pacjentów podzielono na: A1 – niewymagających substytucji katecholamin, B1 – otrzymujących wlew dopaminy, C1 – otrzymujących wlew dobutaminy (podział pierwszy) oraz A2 stanowili pacjenci z masą ciała < 75kg i B2 pacjenci z masą ciała > 75kg. Stopień hemodilucji wywołany podażą płynów wypełniających aparat płuco–serce (*priming*) o stałej objętości 1800 ml określano w oparciu o pomiar hematokrytu i w odniesieniu do masy ciała. Ocenę zmian stężenia tyrozyny we krwi dokonano w pięciu etapach procedury: 1) przed rozpoczęciem operacji i znieczulenia; 2) podczas ECC; 3) po zakończonej operacji przed przekazaniem pacjenta do OIOP; 4) rano w pierwszej dobie pooperacyjnej; 5) rano w drugiej dobie pooperacyjnej. Przeprowadzone badania wykazały spadek stężenia tyrozyny u wszystkich pacjentów w trzecim i czwartym oraz wzrost w piątym etapie badawczym. Mniejsze stężenie tyrozyny obserwowano także w grupie A1 i B1 w etapie trzecim. Normowolemiczne rozcieńczenie krwi powodowało istotny spadek stężenia tyrozyny w grupie A2. Wnioski: 1) procedura krążenia pozaustrojowego powoduje istotne zmiany stężenia tyrozyny we krwi; 2) wlew dopaminy zmniejsza stężenie tyrozyny we krwi; 3) stężenie tyrozyny we krwi zależy od stopnia normowolemicznego rozcieńczenia krwi.