

Role of the Autonomic Nervous System and Vascular Endothelial Factors in the Development of Primary Arterial Hypotension in Paediatric Patients



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Background

There is a dearth and inconsistency of data on the role of the autonomic nervous system (ANS) in the development of arterial hypotension. Also, little is known about the involvement of endothelial factors in the development of this disease. The aim of the study was to investigate the role of the ANS and endothelial factors or vasoregulators in the development of primary arterial hypotension (PAH) in children.

Methods

The cardiointervalography and clino-orthostatic test results of 113 children with PAH were compared with 88 healthy children of comparable age (7–11 years). Serum endothelial factors (nitric oxide and endothelins) of all children were measured.

Results

The findings revealed that children with PAH had higher activity of the sympathetic ($p < 0.001$) and parasympathetic ($p < 0.001$) divisions of the ANS at the initial (resting) position of clino-orthostatic test. The activity of these divisions of the autonomic nervous system correlated with the activity of a cardiac pacemaker. The change of position from horizontal into vertical was accompanied by a rise only in sympathetic activity ($p < 0.001$). However, there was a decline in the sympathetic nervous system ($p < 0.001$) compared to the indices of the initial (resting) position registered in the tenth minute of the vertical position. The parasympathetic division of the ANS based on heart rate variability showed high activity in all positions of the clino-orthostatic test in the patients with PAH compared with healthy children. The activity of the parasympathetic nervous system was associated with increased synthesis of endothelial factors (nitric oxide and endothelins) in blood.

Conclusions

The inadequate response of the autonomic nervous system to the clino-orthostatic test in children with PAH is associated with disorders of both divisions of the autonomic nervous system as well as vascular endothelial factors.

Keywords

Primary arterial hypotension • Autonomic nervous system • Cardiointervalography • Clino-orthostatic test • Nitric oxide • Endothelin

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Introduction

The regulation of blood pressure is accomplished by short and long-acting mechanisms. The short-acting mechanisms involve the activity of baroreceptors, whereas the long-acting regulators are the nervous and renin-angiotensin-aldosterone systems, as well as endothelial factors. The main regulator of integrative functions (including blood pressure) is the autonomic nervous system (ANS). The ANS ensures regulated interaction amongst the organs and systems of the body, maintenance of metabolic processes and interaction with the environment [1–3]. The ANS also interacts with endothelial factors through α_2 and β receptors of endothelial cells. Under normal conditions, this interaction is maintained at equilibrium [4,5]. The endothelial vasoregulatory factors can either have a dilatory or constrictor effect on the blood vessels. Thus, the sympathetic division of the ANS stimulates the release of endothelin-1 (ET-1), which has a vasoconstrictor effect, while the parasympathetic division of the ANS, mainly through acetylcholine, increases the synthesis of nitric oxide (NO), which promotes vasodilation [6,7]. Furthermore, NO attenuates the vasoconstrictor effect of the sympathetic nervous system [8]. Consequently, NO represents a crucial endothelial factor that equilibrates the sympathetic and parasympathetic activity of the autonomic nervous system [4,6]. While the involvement of the ANS and endothelial factors in the development of atherosclerosis, and coronary heart disease has been widely reported in the literature [8–11], there is a dearth and inconsistency of data on the role of ANS in the development of arterial hypotension. Moreover, there is no information on endothelial factors in the pathogenesis of this disease. Therefore, the aim of this study was to investigate the role of ANS and vasoregulators of the endothelium in the development of primary arterial hypotension in children.

Methods

Ethics and Research Committee

The medical ethics and research committee approved the study protocol before parents and children were contacted.

Inclusion Criteria

1. Only children that met the criteria for PAH were included in the study.
2. Children who participated in the study freely gave consent or assent in addition to their parents' agreement.

Exclusion Criteria

1. Children who did not show willingness or refused participation were not involved in the study.
2. Children that could not stand up actively without support were not involved in the study.
3. Patients with moderate to severe anaemia, metabolic acidosis, heart failure, hypotensive shock, electrolyte imbalance, severe kidney injury, severe cardiac disease,

tachycardia, fractures of the extremity, and coma were not involved in the study.

Patients

The children that met the inclusion criteria were treated in designated children's hospitals of the Belarusian State Medical University in Minsk (Belarus). Following ethical approval, parents of the children were approached and the purpose of the study was explained, as well as risks and benefits of participation in the study. Consent was obtained from parents without any financial inducement. Children to be involved in the study were also informed, in a developmentally acceptable way, regarding the purpose, risks and benefits of the study. To minimise discomfort, fear and any other risk, all children were told that participation was voluntary, and that withdrawal was acceptable at any point of the study. Only children who gave their assent were involved in the study. The dignity, privacy and confidentiality of all involved children were maintained at all times. Of the 211 children that were approached, 10 declined. The study involved 88 healthy children and 113 children with PAH. The age range of all groups of children was 7–11 years.

Evaluation of Autonomic Nervous System: Computer Cardiointervallography (CGI) with Clino-Orthostatic Test

The divisions of the autonomic nervous system were evaluated with computer cardiointervallography (CGI) (Briz-M software-hardware complex, developed by the Republic Scientific and Practical Center of Cardiology, Minsk, Belarus, in accordance with the recommendations of European Society Cardiology and North American Society of Stimulation and Electrophysiology) in conjunction with the clino-orthostatic test. Heart rhythm was studied with CGI and clino-orthostatic test. The test was performed in the same time of the day in a quiet room with stable temperature and humidity for all participants. They were asked to drink coffee or eat 2 to 3 hours prior to the experiment due to the possible postprandial effects of coffee or food on cardiac functions. The test started 30 minutes after the participants had entered the room. During this test, participants were asked to rest for 10 minutes in the supine position. Thereafter, they were asked to stand upright for 10 minutes. During this active standing, CGI indices were taken in the resting position (supine), first, fifth, and tenth minute and at the first and fifth minute of repeated supine position using a mercury sphygmomanometer with an appropriately sized cuff. The following indices were analysed with the CGI: Mo (seconds) – mode; AMo (%) – amplitude of the mode; ΔX (seconds) – variation range, which is the difference between the value of the largest and the smallest cardiac interval (CI); IAR is an autonomic index of rhythm; IT – index of tension of regulatory systems; and IAE is the index of autonomic equilibrium.

Investigation of Endothelial Vasoregulators: Enzyme Immunoassay

A right forearm vein was cannulised 10–15 minutes before commencing the study and blood samples for investigation were drawn in the last minute of resting position and in the last minute of upright position. The level of endothelin-1 and -2 (ET-1 and ET-2), nitric oxide (NO), prostaglandin F₂ (PF₂), bradykinin (BR) of blood plasma of all participants were investigated by the enzyme immunoassay in the biochemistry section of Central Scientific Research Laboratory of the Belarusian State Medical University using the equipment (STAT FAX 3200, Awareness Technology Inc., Palm City, FL, USA).

Statistics

Data processing was carried out using the program Statistica 10.0 for Windows (StatSoft, Palo Alto, CA, USA). Quantitative data that were normally distributed are presented as mean and error of the mean ($M \pm m$), whereas data that differed from the normal distribution are presented as median and quartiles. The reliability of the differences was estimated by calculating the Student *t*-test and Mann-Whitney test. The probability value for significance was set at $p < 0.05$.

Results and Discussion

The results of CGI investigation of the healthy children and those with PAH showed the following results in the resting (supine) position (Table 1).

Comparative analysis of the results of healthy children and those with PAH in Table 1 showed that patients with PAH had increased activity of both the sympathetic and parasympathetic divisions of the autonomic nervous system. So, in healthy children AMo was 13.1% and the variation range was 0.3 seconds, whereas children with PAH had an AMo of 17.2% ($p < 0.001$) and variation range of 0.54 seconds ($p < 0.001$). The children with PAH had lower values of cardiointervalogram indices (IT and IAR) in the patients indicated the predominance of the parasympathetic nervous system over the sympathetic nervous system.

Correlation analysis of cardiointervalogram indices in healthy children did not reveal any statistically significant relationship. However in children with PAH, there was a significant correlation between AMo and X ($r = -0.62$) ($p < 0.05$). This suggests an interference of the central regulatory mechanisms with autonomic channel of regulation (not shown in Table 1). This may be the reason for increased levels of index of tension (i.e. tension in regulation of the heart rhythm) in children with PAH even already in the resting position, indicating that the sinus node experiences high activity of both the sympathetic and parasympathetic divisions of the autonomic nervous system.

Table 1 Cardiointervalogram indices of healthy children (control) and children with PAH in the resting position.

Indices	PAH n = 113	Control n = 88
Mo, seconds	0.72 ± 0.06	0.68 ± 0.04
AMo, %	17.2 ± 0.45**	13.1 ± 0.32
X, seconds	0.54 ± 0.03**	0.30 ± 0.03
IAR	2.6 ± 0.31**	5.5 ± 0.18
IT	22.3 ± 3.7**	37.4 ± 2.8
IAE	31.8 ± 3.7*	46.7 ± 4.1

Abbreviations: PAH, primary arterial hypotension; Mo, mode; AMo, amplitude of the mode; X, variation range, which is the difference between the value of the largest and the smallest cardiac interval (CI); IAR, index of autonomic rhythm; IT, index of tension of regulatory systems; IAE, index of autonomic equilibrium.

* $p < 0.01$.

** $p < 0.001$.

The transition of healthy children from the resting (supine) to the vertical position was accompanied by a reliable decrease in the vagal activity, and an increase in the tone of the sympathetic division of the autonomic nervous system. This reaction to active orthostasis was absent in children with PAH; they only experienced a significant increase in the AMo. The other indices of cardiointervalogram such as Mo, X, IAR, IT, and IAE did not change during the first minute of active orthostasis (not shown in Table 1, but IT dynamics in Figure 1).

Comparing the indices of the cardiointervalograms of the children of the two groups in the first minute of active orthostasis showed that the mode was 0.55 seconds, amplitude of the mode was 15.3%, and variation range was 0.2 seconds in the group with healthy children. The children with PAH had higher values of these indices. So, in the group of children with PAH, mode, amplitude of mode and variation range were 0.67 seconds ($p < 0.01$), 19.3% ($p < 0.001$), and 0.49 seconds ($p < 0.001$) respectively. The other indices of cardiointervalogram (IAR, IT, and IAE) were also reduced. These results indicate increased activity of the regulatory channels of parasympathetic division of the autonomic nervous system on the heart rhythm.

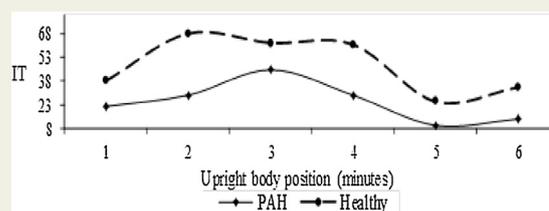


Figure 1 Dynamics of indices in children with PAH (primary arterial hypotension) under conditions of clino-orthostatic test.

The correlation analysis of the values of the indices at the upright position showed that there was a significant correlation ($r = +0.71$) ($p < 0.05$) instead of the insignificant correlation observed in the resting position. However, this correlation was negative in children with PAH ($r = -0.61$; $p < 0.05$). This result points to the fact that change in position (from supine to upright) does not affect the tension of the parasympathetic division of the autonomic nervous system in children with PAH. It also suggests that the activity of the sympathetic division is not able to provide adequate autonomic activity to regulate the activity of cardiac structures. The maximum influence on the heart rhythm is due to the activity of the parasympathetic division of the autonomic nervous system.

The analysis of the dynamics of the cardiorythm from the fifth to the tenth minute of the upright position revealed a significant change in AMo in children of both groups. While the amplitude of the mode in healthy children decreased only by 1.3% ($p < 0.05$), in children with PAH, the decrease was 7.1% ($p < 0.001$). Decrease in sympathetic activity at the tenth minute of active orthostasis compared with baseline value in children with PAH may indicate depletion of adaptive-compensatory capabilities of the sympathetic nervous system during active orthostasis. However, the tone of the parasympathetic nervous system did not change in the patients with PAH. The maximum vagal effect on the sinus heart rhythm was confirmed by a significant decrease in IAE in children with PAH from 57.4 in the fifth minute to 35.7 at the tenth minute of upright position ($p < 0.001$). In children of the control group, IAE remained the same in all positions of the clino-orthostatic test. Thus, active orthostasis in healthy children is accompanied by a stable tone of the parasympathetic nervous system, and moderate activity of the sympathetic nervous system. Children with PAH exhibit sinus rhythm dysregulation. The high tone of the parasympathetic nervous system in these patients is compensated for by the high activity of the sympathetic nervous system. However, thereafter, the adaptive-compensatory capabilities fail and the sympathetic influence on the sinus node decreases to the basal (resting) level.

Transition of children from active orthostasis to a repeated supine position led to a significant increase in Mo, X and a decrease in AMo, and IT in both groups. At the same time, the amplitude of the mode and the variation range in children with PAH during the first and fifth minute in the lying

position remained significantly increased in comparison with the healthy children. The autonomic index of equilibrium showed differences in the two groups. For example, the IAR of the children with PAH was 1.85 compared to 4.81 in the healthy children ($p < 0.001$); IT was 14.3 versus 33.7 ($p < 0.001$) respectively. The result indicates high activity of the parasympathetic nervous system in children with PAH in all positions of the clino-orthostatic test. These results suggest abnormal influence of the autonomous regulatory circuit on the heart rhythm in children with PAH in the upright position. But the increase in the activity of the sympathetic nervous system in children with PAH can be considered an adaptive-compensatory mechanism, which is insufficient for a long upright position.

During the last decade, some authors [11–15] have established a close relationship between autonomic nervous system activity and level of endothelial factors. High sympathetic activity stimulates the synthesis of endothelin, and parasympathetic activity stimulates the synthesis of nitric oxide [16,17]. Results of investigation of the level of the most significant peripheral vasoregulators of the endothelium of blood vessels in PAH children and healthy children are shown in Table 2. Comparative analysis of the levels of endothelial factors did not reveal a significant difference in the concentrations of ET-1, ET-2, PF2 α and BR. So, in the group of children with PAH, the ET-1 value was 0.72 ng/mL (0.65–0.80), ET-2–0.90 ng/mL (0.81–1.26), PF2 α – 95.0 pg/mL (59.5–110.0), BR – 5.7 ng/mL (3.8–8.4). The healthy children had the following values of vasoregulatory factors: ET-1–0.7 ng/mL (0.60–0.74) ($p < 0.1$), ET-2–0.84 ng/mL (0.76–0.96) ($p < 0.1$), PF2 α – 95.0 pg/mL (54.0–125.0) ($p < 0.1$), BR – 4.1 ng/mL (1.6–6.2) ($p < 0.1$). Nevertheless, we established an increase in the level of NO in children with PAH more than seven times compared to baseline values. Thus, the concentration of NO in the children with PAH was 11.4 μ mol/L (6.4–22.5) compared to 1.6 μ mol/L (0.9–3.8) in healthy children ($p < 0.001$). Importantly, we also observed differences in the ratio of NO/E1 in the two groups of children. The healthy children had NO/E1 value of 2.28, whereas the children with PAH had a value of 15.8. Such high concentrations of plasma NO in children with PAH, in our opinion, cannot only lead to vasodilation of the vessels (which may result in PAH), but also cause stress due to excessive release of NO in the vascular endothelium.

Table 2 Level of vasoactive substances in children with PAH, Me (25–75%).

Groups	ET-1 ng/mL	ET-2 ng/mL	NO μ mole/L	PF ₂ α pg/mL	BR ng/mL
Children with PAH	0.72 (0.65–0.80)	0.90 (0.81–1.26)	11.4 (6.4–22.5)	95.0 (59.5–110.0)	5.7 (3.8–8.4)
Healthy children	0.70 (0.60–0.74)	0.84 (0.76–0.96)	1.6 (0.9–3.8)	95.0 (54.0–125.0)	4.1 (1.6–6.2)
Significance	–	–	Z = 4.6, $p < 0.001$	–	–

Abbreviation: PAH, primary arterial hypotension; NO, nitric oxide; ET, endothelin; BR, bradykinin; PF2 α , prostaglandin 2a.

Therefore, high blood plasma nitric oxide in children with PAH is associated with high activity of the parasympathetic nervous system and stimulation of NO synthesis by acetylcholine. The excess nitric oxide causes dilatation of the blood vessels, a negative inotropic effect on myocardial contractility, and a decrease in peripheral vascular resistance, which ultimately form the haemodynamic component of PAH in children [8,18]. Thus, children with PAH may not only have dysfunctions in autonomic nervous system functions, but endothelial dysfunction, in which the formation of vasodilators (NO) predominates over the synthesis of vasoconstrictors (ET-1, ET-2, PF2 α).

Conclusion

Autonomic homeostasis in children with PAH according to cardiointervalography is characterised by high activity of the sympathetic and parasympathetic division of the autonomic nervous system in each position of active clinorhthostasis. The indices of variation range of PAH patients exceed the values of healthy children and ranged from 0.2 seconds ($p < 0.001$) in the third to 0.41 seconds ($p < 0.001$) in the sixth minute of active clinorhthostasis. The conduction of a clinorhthostatic test increases the vagal effect on the sinus node of the heart and increases X of the cardiointervalogram by 0.19 seconds ($p < 0.001$). The drop in AMo of the cardiointervalogram of PAH by 24.3% at the fifth minute of active orthostasis to 18.2% in the tenth minute of the upright position ($p < 0.001$) indicates depletion of the adaptive-compensatory capabilities of the sympathetic nervous system and may be the cause of syncope in this category of patients. Increase in blood nitric oxide level by more than seven times ($p < 0.001$), high NO/E1 ratio, but normal indices of ET-1, ET-2, PF2 α , BR in children with PAH strongly suggests that endothelial dysfunction is due to the predominance of the synthesis of vasodilators (NO) over the synthesis of vasoconstrictors (ET-1, ET-2, PF2 α).

References

- [1] Nishida Y, Tandai-Hiruma M, Kemuriyama T, Hagsawa K. Long-term blood pressure control: is there a set-point in the brain? *J Physiol Sci* 2012;62(3):147–61.
- [2] Di Rienzo M, Parati G, Radaelli A, Castiglioni P. Baroreflex contribution to blood pressure and heart rate oscillations: time scales, time-variant characteristics and nonlinearities. *Phil Trans R Soc A* 2009;367:1301–18.
- [3] Thrasher TN. Baroreceptors and the long-term control of blood pressure. *Exp Physiol* 2004;89(4):331–41.
- [4] Smirnov AV, Petrishchev NN, Panina IYU, Mnuskina MM, Ankasova VV, Rummyantsev ASH, et al. The level of endothelin-1 and the reactivity of the vessels of the microcirculatory bed of the skin in patients at early stages of chronic kidney disease. *Ther Arch* 2011;6:13–8.
- [5] Harris KF, Matthews KA. Interactions between autonomic nervous system activity and endothelial function: a model for the development of cardiovascular disease. *Psychosom Med* 2004;66(2):153–64.
- [6] Chowdhary S, Marsh AM, Coote JH, Townend JN. Nitric oxide and cardiac muscarinic control in humans. *Hypertension* 2004;43(5):1023–8.
- [7] Dessy C, Moriotte S, Ghisdal P, Havaux X, Noirhomme P, Balligand JL. Endothelial beta3-adrenoceptors mediate vasorelaxation of human coronary microarteries through nitric oxide and endothelium-dependent hyperpolarization. *Circulation* 2004;110(8):948–54.
- [8] Joshi MS, Ferguson TB, Johnson FK, Johnson RA, Parthasarathy B, Lancaster JR. Receptor-mediated activation of nitric oxide synthesis by arginine in endothelial cells. *PNAS* 2007;104(24):9982–7.
- [9] Gomazkov OA. Vasoactive peptides and growth vascular factors. Role in the pathogenesis of arterial hypertension. *Cardiovasc Ther Prev* 2003;2:11–5.
- [10] Popova AA, Mayanskaya SD, Mayanskaya NN, Berezikova EN, Khidir-ova LD. Arterial hypertension and endothelial dysfunction (part 1). *Bull Mod Clin Med* 2009;2:41–6.
- [11] Sayfutdinov RG. Role of nitric oxide in diseases of internal organs (review of literature). *Vestnik Mod Klinicheskoi Med* 2009;3:48–53.
- [12] Ivanov SN, Starovoitova EA, Ogorodova LM, Volkova TG. The role of endothelial dysfunction in the genesis of cardiovascular pathology. *Sib Med J* 2007;1:99–104.
- [13] Amiya E, Watanabe M, Komuro I. The relationship between vascular function and the autonomic nervous system. *Ann Vasc Dis* 2014;7(2):109–19.
- [14] Harris KF, Matthews KA. Interactions between autonomic nervous system activity and endothelial function: a model for the development of cardiovascular disease. *Psychosom Med* 2004;66(2):153–64.
- [15] Bruno RM, Ghiadoni L, Seravalle G, Dell’Oro R, Taddei S, Grassi G. Sympathetic regulation of vascular function in health and disease. *Front Physiol* 2012;3:284.
- [16] Xu Z, Tong C, Eisenach JC. Acetylcholine stimulates the release of nitric oxide from rat spinal cord. *Anesthesiology* 1996;85(1):107–11.
- [17] Damon DH. VSM growth is stimulated in sympathetic neuron/VSM cocultures: role of TGF- β 2 and endothelin. *Am J Physiol Heart Circ Physiol* 2000;278(2):H404–11.
- [18] Sikorsky AV. Hemodynamic disorders in children with primary arterial hypotension. *Probl Health Ecol* 2007;2:71–8.