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Formation of Tonic Effects of the Autonomic Nervous System Parts on the Developing Heart.

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ABSTRACT

This paper deals with the study of formation of extracardiac neural effects on cardiac activity of dogs and rats in the postnatal ontogenesis performed through vagotomy and administration of blocking agents in the intact and sympathectomized animals. Experiments on growing animals showed that the tonic effects of the vagus nerve on a pacemaker develop gradually as the sympathetic tone develops. The experimental data obtained in growing dogs and rats indicate that the contribution of the heart rate or stroke volume to the heart adaptation can vary at different stages of ontogenesis as regulatory effects of the autonomic nervous system on these parameters develop in heterochronic manner. We have found that the tonic effects of the sympathetic part of the autonomic nervous system on myocardial contractility appear later than in relation to the heart rate. Experiments on sympathectomized growing animals revealed that the positive chronotropic effect occurring at vagus nerve transection does not depend on the intactness of the sympathetic nervous system. Nevertheless, the findings bear evidence that both parts of the autonomic nervous system in vivo take part in the regulation of cardiac activity, and the relations between the sympathetic and parasympathetic influences form the final effect.

Keywords: heart, tone, vagus nerve, sympathetic nerve, ontogenesis, sympathectomy.

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INTRODUCTION

Tone (lat. *tonus*) – a tension. In physiology, this is a long-term excitation of either nervous system or muscle tissue not accompanied by fatigue. We may consider it proved that the centers of extracardiac parasympathetic nerves (PN) in human and, to different extent, in many animals (dogs, rats, rabbits, cats, etc.) have a tonic excitement, and there are constant pulses conducted through efferent nerves to the heart. Transection of the vagus nerves leads to increase in heart rate (HR). The presence of tonic effects is proved by their surgical, pharmacological transection or registration of nerve bioelectric potentials. According to V.V. Frolkis [1], background pulsation in the centrifugal fibers of the vagus nerves is detected even when the above listed methods can not detect the manifestation of tonic effect of this nerve. However, according to M.G. Udelnov, the effect of heart rate acceleration after vagotomy is of sympathetic origin due to activation of the sympathetic-adrenal system when excluding the flow of afferent pulses in the vagus nerve [2]. This issue in case of M.G. Udelnov is an open problem. An inhibitory effect from the stimulation of the parasympathetic fibers is due to the number of excited nerve fibers ("quantitative principle"). There is a sympathetic effect at low vagus nerve (VN) stimulation, while increased stimulation and involvement of a large number of NV nerve fibers are accompanied by a parasympathetic effect.

There are also different opinions on the tone of the sympathetic nerve (SN) centers. Thus, A.G. Ginetsinskii and A.V. Lebedinskii [3] believe that the centers of the sympathetic innervation have no clearly expressed tone. Transection of the sympathetic nerves have no major impact on both heart rate and heart force. The book "Fundamental and clinical physiology" [4] indicates that both parts of the autonomic nervous system have tonic effect on the area of a pacemaker. However, V.M. Smirnov argues about the absence of SN tonic effects on heart relying upon data on the pharmacological blockade of the sympathetic nervous system in rats and guinea pigs [5].

In our experiments, we used propranolol to block β -adrenergic receptors in adult dogs, which reduced the heart force and decreased heart rate [6, 7]. The subsequent administration of atropine as well as vagotomy resulted in the increased heart rate and myocardium force. Therefore, the tonic effects of both SH and PN manifest themselves in the contractility and heart rate. We totally agree with the opinion of Academician K.M. Bykov [8] that "considering the force of heart contractions is as important as the heart rate" [7].

Experiments on dogs and rats revealed that both SN and PN tonic effects have asymmetry. Right VN transection induces more pronounced increase in HR than the left one, while the left vagotomy leads to the increased cardiac contractility [6, 9].

Formation of the tonic effects of extracardiac nerves in the ontogenesis is of particular interest. It is known that the formation of the nervous regulation of the heart is largely determined by the structural maturation of the individual units of the reflex arc. Peripheral structures are already formed, but they have not yet been used by the central regulating mechanisms. Barcroft figuratively defined the situation as "the machine is ready, but it still does not work". [10]

A huge number of studies has been devoted to this issue [11, 12, 13, 14]. These studies were mainly performed on dogs, but nowadays the most common object of research are small rodents (mice, rats, guinea pigs), and studying the formation of tonic effects on heart rate and myocardial contractility is of practical and theoretical interest [15, 16, 17, 18].

Objective of this paper was to study heart rate and stroke volume in growing rats during postnatal ontogenesis with different variants of vagotomy, as well as the time of formation of tonic effects on heart rate and cardiac contractility in dogs under administration of blocking agents.

RESEARCH TECHNIQUE

We carried out the experiments on 143 male and female laboratory inbred albino rats at the age of 143, 21, 28, 42, 56, 56 and 120 days. The animals were anesthetized with 25% urethane solution i.v. 800 mg per kbw.

Anesthetized rats were fixed on the operating table and had dissection of both vagus nerves (VN) under a binocular microscope MBS-2. After stabilization of the heart rhythm either sequential or simultaneous vagotomy was performed at 60 minute interval. There were the following types of experimental actions conducted:

- Right vagotomy;
- Left vagotomy on the background of the right vagotomy;
- Left vagotomy;
- Right vagotomy on the background of the left vagotomy;
- Single-stage transection of both vagus nerves.

To analyze the heart rate throughout the experiment we were simultaneously recording the differentiated rheogram and electrocardiogram at rest, after dissection and up to stabilization of the heart rhythm and after transection of the corresponding nerve.

Registration and analysis of cardiac activity were carried out with an integrated electrophysiological laboratory with the program «Conan», installed on the computer JBM-486. ECG analysis was conducted by the method of R.M. Baevskii [19], stroke volume was calculated by Kubicek equation [20].

Statistical analysis and identification of the reliability of differences in the results of research by Student test were performed in Microsoft Excel editor.

Sympathectomy was carried out by daily administration of guanethidine sulfate solution based on 10 mg/kg of body weight for 28 days after birth [21].

Cardiogram of the dogs of all ages (the first group - 16-18 days, the second group - 2-2.5 months., the third group - older than 3 months, and mature animals) under hexenal anesthesia (1 ml/kg of body weight) was recorded, and propranolol and atropine were administered to block β -adrenergic receptors and m-cholinergic receptors, respectively.

RESULTS AND DISCUSSION

Right vagotomy in mature rats leads to a rapid, transient increase in heart rate (Table 1) without significant changes in stroke volume. Heart rate restores gradually to its initial level by 30 minutes. After the left vagotomy on the background of intact right VN there is a short-term reduction in stroke volume and heart rate observed at the beginning, probably of reflex origin occurred due to the influences from the right VN. Further, stroke volume increases. Consequently, the VN tonic effects on the heart of rats are quite clearly expressed and asymmetric in terms of heart rate and contractility.

Right vagotomy in 2, 3 and 4-week rats causes no significant changes in heart rate. Heart rate increase was observed in 2 and 8-week animals, however less expressed than in mature animals. Right vagotomy caused no changes of stroke volume in 2-week rats (Table 1), and led to a slight increase in stroke volume in 3 and 4-week animals. According to T.A. Anikina, G.A. Bilalova and F.G. Sitdikova [22], this is the age when the effect of SN stimulation on cardiac activity starts manifesting itself. Eight-week animals in their puberty period show another reaction such as reduction in stroke volume and increased heart rate.

Left vagotomy on the background of intact VN causes no changes in heart rate in 2, 3 and 4 week rats. In prepubertal and pubertal age (6 and 8 week rats) there was a decrease in heart rate and increase in stroke volume after the left vagotomy.

To exclude some certain reflex effects on the heart at transection of one VN we conducted the experiments with simultaneous bilateral vagotomy (Table 2).

Both mature rats and growing animals over 4 weeks of age showed an increased heart rate and stroke volume after such intervention. According to M.G. Udelnov, the similar effect is due to the increased

activity of sympathetic-adrenal system. To test this hypothesis, we also performed bilateral vagotomy in sympathectomized animals.

Bilateral vagotomy in both intact and sympathectomized animals shows increase in heart rate and a slight decrease in stroke volume. Decrease in stroke volume is compensated by higher values of heart rate. This data allows speaking about tonic activity in the centers of sympathetic nerves.

According to our data obtained in dogs of all ages, the tonic effects of the sympathetic nervous system on myocardial contractility appear later than on the heart rate (Table 3). It follows therefrom that only mature dogs have reduced amplitude of cardiac contractions caused by the blockade of β -adrenoceptors with obsidan. In this case, all the age groups show reduction in heart rate (Table 3). Further administration of atropine for deprivation of influence of parasympathetic nerves on heart rate led to a significant increase in heart rate most expressed in puppies from the 3rd age group and mature dogs. Hence, the VN tonic effects on a pacemaker develop gradually too. These experiments indicate that the positive chronotropic effect of vagotomy does not depend on the intactness of the sympathetic-adrenal system. This was confirmed during the experiments on sympathectomized growing rats (Table 4). As follows from this data, only 14-day-old rats have no changes in heart rate caused by vagotomy. In other age groups of the sympathectomized animals the vagotomy led to a quite expressed increase in heart rate.

Table 1: Dynamics of stroke volume (ml) in rats upon sequential right and left vagotomy in the postnatal ontogenesis

Age	Initial values	Right vagotomy				Left vagotomy			
		1 min	5 min	30 min	60 (initial)	1 min	5 min	30 min	60 min
14	0.0113 ±0.0007	0.0111 ±0.0009	0.0124 ±0.0009	0.0115 ±0.0009	0.0116 ±0.0009	0.0117 ±0.0009	0.0128 ±0.0012	0.0118 ±0.0006	0.0120 ±0.0008
21	0.0168 ±0.0012	0.0163 ±0.0012	0.0180 ±0.0017	0.0185 ±0.0018	0.0191 ±0.0018	0.0193 ±0.0020	0.0213 ±0.0021	0.0237 ±0.0021	0.0249 ±0.0021*
28	0.0216 ±0.0019	0.0213 ±0.0020	0.0256 ±0.0021	0.0256 ±0.0023	0.0277 ±0.0019*	0.0264 ±0.0028	0.0304 ±0.0022	0.0321 ±0.0011	0.0329 ±0.0018*
42	0.0304 ±0.0027	0.0325 ±0.0023	0.0369 ±0.0024	0.0402 ±0.0037*	0.0402 ±0.0041	0.0387 ±0.0023	0.0405 ±0.0028	0.0446 ±0.0027	0.0467 ±0.0032
56	0.0587 ±0.0037	0.0551 ±0.0046	0.0646 ±0.0041	0.0700 ±0.0057	0.0626 ±0.0069	0.0659 ±0.0075	0.0790 ±0.0104	0.0744 ±0.0094	0.0787 ±0.0097
120	0.1319 ±0.0039	0.1201 ±0.0058	0.1505 ±0.0060*	0.1439 ±0.0119	0.1477 ±0.0114	0.1436 ±0.0110	0.1611 ±0.0082	0.1438 ±0.0073	0.1443 ±0.0082

Note: The significance of differences between the initial and the further indicators * ($p < 0.05$)

Table 2: Dynamics of stroke volume and heart rate in rats upon single-stage bilateral vagotomy in postnatal ontogenesis

Age	Stroke volume, ml				HR, bpm			
	Initial values	Bilateral vagotomy			Initial values	Bilateral vagotomy		
		1 min	5 min	60 min		1 min	5 min	60 min
14	0.0104 ±0.0005	0.0099 ±0.0004	0.0109 ±0.0004	0.0109 ±0.0003	362 ± 5.28	357 ± 4.86	347 ± 5.46 *	303 ± 2.27 ***
21	0.0162 ±0.0021	0.0154 ±0.0019	0.0178 ±0.0017	0.0219 ±0.0017	432 ± 9.58	433 ± 8.84	434 ± 9.23	354 ± 12.07***
28	0.0192 ±0.0018	0.0210 ±0.0027	0.0242 ±0.0024	0.0252 ±0.0026	441 ± 16.07	459 ± 10.70	474 ± 7.02	433 ± 14.96
42	0.0357 ±0.0040	0.0348 ±0.0031	0.0377 ±0.0039	0.0444 ±0.0041	422 ± 5.35	434 ± 4.45	442 ± 6.37*	364 ± 12.04**
56	0.0754 ±0.0036	0.0704 ±0.0034	0.0829 ±0.0049	0.0828 ±0.0052	409 ± 5.53	435 ± 15.13	446 ± 12.72*	373 ± 6.51**
120	0.1301 ±0.0054	0.1466 ±0.0046*	0.1474 ±0.0047*	0.1347 ±0.0013	360 ± 10.86	385 ± 6.14*	369 ± 7.19	319 ± 12.52*

Note: The significance of differences between the initial and the further indicators * ($p < 0.05$); ** ($p < 0.01$); *** ($p < 0.001$).

Table 3: Heart rate change in dogs of different age after blocking β -adrenergic receptors and m-cholinergic receptors

Age group	Initial HR per 1 minute	HR after obsidani.v. administration	Chronotropic effect	HR after atropine administration
Puppies of group 1	183±8 ³	116±5 ^{2,3}	66±8 ^{1,2,3}	146±7
Puppies of group 2	158±12 ⁴	115±8 ⁴	43±6 ^{1,4,6}	165±7
Puppies of group 3	128±7 ⁵	97±7 ²	30±4 ^{2,6}	163±16
Mature dogs	97±113.4 ⁵	75±12 ^{3,4}	22±5 ^{3,4}	153±5

Note: Numerical symbols indicate the significant of differences in the performance between: 1 - puppies of first and second age groups; 2 - puppies of first and third age groups; 3. puppies of first group and mature animals; 4 - mature dogs and puppies of second group; 5 - mature dogs and puppies of third group; 6 - puppies of second and third age groups

Table 4: Heart rate change in sympathectomized rats of different age after vagotomy

Age (days)	Initial HR (bpm)	HR (bpm) after vagotomy
14	387±5.2	384±10.2
21	435±4.1	454±8.8
28	437±1.6	457±2.8
42	401±4.6	428±8.4
56	396±7.6	436±10.6
70	382±5.1	404±5.3
120	372±6.4	393±6.2

SUMMARY

Based on the above stated we can conclude that the SN tonic effects in dogs and rats occur earlier in terms of heart rate than heart force. Positive chronotropic effect occurring upon vagotomy is determined by removing the VN tonic effects and has no sympathetic origin.

CONCLUSION

Both parts of the autonomic nervous system in vivo take part in the regulation of cardiac activity, and the final effect is formed by the relations between the sympathetic and parasympathetic influences. Our findings indicate that the contribution of the heart rate or stroke volume to the heart adaptation can vary at different stages of ontogenesis as regulatory effects on these parameters develop in heterochronic manner.

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REFERENCES

- [1] Frolkis V.V. Cardiac nerve stimulation effects // Physiology of blood circulation. Cardiac physiology, 1980. - p. 350-368.
- [2] Udelnov M.G. Cardiac physiology. M.: Publ. house of MSU, 1975. - p. 363
- [3] Ginetsinskii A.G., Lebedinskii A.V. Foundation of human and animal physiology. - Leningrad: Medgiz, 1947. - p. 746
- [4] Kamkin A.G., Kamenskii A.A. Fundamental and clinical physiology. - M.: Akademiia, 2004. - p. 1073
- [5] Smirnov V.M. Tone of sympathetic nerves and regulation of heart activity // Bulletin of Experimental Biology and Medicine. - Volume 130, Issue 10, 2000, Pages 930-933.
- [6] Sitdikov F.G. Mechanisms and age-related peculiarities of heart adaptation to the long-term sympathetic influence: Diss. ... Dr. biol. Sciences. - Kazan, 1974. - p. 312
- [7] Sitdikov F.G. Formation of extracardiac effects in dogs during ontogenesis// Evolution of biochemistry and physiology. -1981. Vol.17, No.6. - p. 569-571.
- [8] Bykov K.M. Physiology text-book. - M. Pub. house Medgiz, 1954. - p. 896
- [9] Zefirov T.L. Nervous regulation of the heart rate in rats in postnatal ontogenesis: Author. diss. ... M.D. - Kazan, 1999. - p. 39

- [10] Barcroft J. Researches on prenatal life, Oxford, Blackwell, 1946, 1.
- [11] Arshavskii I.A. Essays on age physiology. - M.: "Meditsina", 1967. - p. 476
- [12] Rozanova V.D. Essays on experimental and age pharmacology. - L.: Meditsina, 1968.- p. 223
- [13] Adolf E.F. Development of physiological functions. - M. - 1971. - p. 192
- [14] Kulaev B.S. Reflexogenic cardiac zones and blood flow self-regulation. L., 1972. p.225
- [15] Kulaev B.S., Boursian A.V., Semenova Yu.O., Sizonov V.A. Secondary rhythms of cardiac activity within early ontogenesis: Effects of blocking of adreno- and cholinoreceptors in rats. // Neurophysiology. - Volume 36, Issue 2, March 2004, Pages 126-131.
- [16] Sitdikov F.G., Gilmutdinova R.I., Minnakhmetov R.R., Gizzatullin A.R. Effect of electrical stimulation of vagus nerves on cardiac activity in sympathectomized rats during postnatal ontogeny. // Bulletin of Experimental Biology and Medicine. - Volume 135, Issue 6, June 2003, Pages 534-536.
- [17] Zefirov T.L., Sviatova N.V. Age-specific features of vagus regulation of chronotropic function of the heart in sympathectomized and intact rats // Bulletin of Experimental Biology and Medicine. - 1997, Vol. 123, No.6. p. 703 - 706.
- [18] Zefirov T.L., Ziyatdinova N.I., Khisamieva L.I., Zefirov A.L. Effect of α_2 -adrenoceptor stimulation on cardiac activity in rats. // Bulletin of Experimental Biology and Medicine. - Volume 157, Issue 2, June 2014, P.194-197.
- [19] Baevskii P.M. Cybernetic analysis of cardiac rhythm control processes // Current problems of physiology and pathology of blood circulation. - M.: Meditsina, 1976.- p. 161-175
- [20] Kubicek W.G. The minnesoz impedance cardiograph – theory and applications // Biomed. End. 1974. Vol. 9. P. 410 – 416.
- [21] Rodionov I.M., Iarygin V.N., Muhammedov A.A. Immunologic and chemical sympathectomy. - M.: "Nauka", 1988 - p. 150
- [22] Anikina T.A., Bilalova G.A. Sitdikov F.G. Effect of electrical stimulation of the right stellate ganglion on cardiac activity of rats in the postnatal ontogenesis // Bulletin of Experimental Biology and Medicine, 2002. - Vol.133. - No.4. - p. 377-379