RESEARCH PAPER

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Effect of the sympathetic nervous system co-transmitters ATP and norepinephrine on thermoregulatory response to cooling

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Abbreviations: ATP, adenosine-5'-triphosphate; NE, norepinephrine; SERCA1, sarco/endoplasmic reticulum Ca(2+)-ATPase; Ca(2+), calcium.

The existence of co-transmitters of the sympathetic nervous system norepinephrine (NE) and ATP implies variations in the neuromodulator mechanisms of physiological processes. The role of ATP, as a transmitter of the peripheral part of sympathetic nervous system in the formation of thermoregulatory response is not clear. Whether ATP modulates any parameters of thermoregulatory response to cold; if yes, whether co-transmitters of sympathetic nervous system ATP and NE differently modulate thermoregulatory response and on which parameters of cold-defense response the influence of ATP is more pronounced. Experiments were carried out on rats. ATP (10^{-6}), NE (10^{-3}), and their mixture introduced iontophoretically into skin. Their effects on thermoregulatory parameters (temperature parameters, total oxygen consumption, carbon dioxide release, muscle activity, respiratory coefficient) were studied in thermoneutral conditions (without cold load) and under the cooling. In thermoneutral conditions both ATP and NE enhance total metabolism through increase in metabolic rate of lipids, NE effect being more expressed. It was shown that ATP and NE influence predominantly on the different components of the metabolic response to cold. ATP affects to the greatest extent on cold muscular thermogenesis by increasing shivering almost twofold and lowering its initiation temperature thresholds, whereas NE mainly promotes increase in non-shivering thermogenesis. When introducing the mixture of these biological substances the effect of NE is more expressed and the ATP effect is weakened. The obtained results allow to suggest that *in vivo* the NE effects can be more expressed when the sympathetic nervous system is stimulated by cold. Thus, NE and ATP being co-transmitters and predominantly acting on the different processes of cold thermogenesis (ATP on shivering and NE on non-shivering) may organize the certain sequence of cold defense responses.

Introduction

It is well known that the sympathetic nervous system is involved in the responses of the whole body to acute cooling. For a long time the influence of sympathetic nervous system on thermoregulatory effects, targeted at maintenance of thermal homeostasis under cold exposure, have been conceptually associated with NE, the major transmitter of this system. There are ample data in the literature indicating that the activity of the sympathetic fibers and concentration of its transmitter norepinephrine (NE) in the blood are augmented under cold effect.[1-3] Increase in NE concentration causes the changes in the function of effector organs and modulate the activity of afferent nervous fibers including thermosensitive. Support came from researches showing that exogenous NE can modify not only the function of effector organs (muscles, brown adipose tissue, blood vessels e. c., this is well known), but also impulse activity of the skin thermoreceptors[4-8] and thermosensitive neurons in the hypothalamus.[9] Modulation of thermoreceptor activity by exogenous NE is associated with shifts of a set of regulatory parameters of the thermal homeostasis system and changes in the thermal thresholds and the magnitude of cold defense responses.[10-12]

Now, it is known, that in addition to NE, adenosine-5'-triphosphate (ATP) and neuropeptide Y are released when sympathetic nerve endings are activated.[13,14] The ATP molecule is a long known source of energy for intracellular metabolism. Its

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properties as a neurotransmitter are also beyond question today. Thus, ATP acts as a neurotransmitter in the central nervous system, ganglia, and neuromuscular junctions.[15-17] ATP has been implicated in the mechanisms regulating cardiac activity, vascular tone, and respiration through its action on purinoergic receptors.[14,18-23] The data on the influence of ATP on the impulse activity of the central and peripheral thermosensitive structures are absent but there are data on the modulating effects of ATP on sensitive C- and Aδ-fibers.[24-27] Peripheral thermosensitive afferents just belong to these types of nervous fibers.

The existence of co-transmitters suggests their different modulator impact on the thermoregulatory processes. Given the fact that ATP is a NE co-transmitter, it is important to clarify the components of the thermoregulatory response affected by each of the transmitter alone, also to see how these substances (ATP or NE) act when getting together.

The organism's response to cold is a set of different responses directed at the maintenance of thermal homeostasis. The role of the afferent signal is decisive in the determination of the pattern and sequence of the different physiological responses to any effect. The sequence of initiation of different components of thermoregulatory response is possibly defined by the sequence of different thermosensitive sensory structures activation. Under the effect of external cooling, thermal thresholds for initiation of the responses, which estimated by the skin and deep body temperature, may be indices of the physiological significance and contribution of the peripheral and central thermal afferent signal.

Here we address the following questions. Does iontophoretic administration of ATP into the skin, the location site of peripheral thermal afferents, have an influence on (1) the parameters of thermal homeostasis in normal conditions, without thermal load, and (2) the parameters of the cold defense responses of the whole body to rapid deep cooling, (3) what would be the difference between the effects of ATP and NE on the thermoregulatory parameters? (4) the effects of which biologically active substances, ATP or NE, will be more expressed when they administered as a mixture?

Materials and Methods

Male Wistar rats weighing 270–290 g were used in the experiments. They were housed in standard conditions the animal facility. All had free access to water and food. Experiments were done in conformance with the humane principles of the Instructions of the EU Directive 2010/63/EU and the Helsinki Declaration. Cooling and recording of the physiological parameters were carried out in anaesthetized rats (Nembutal 40 mg/kg) to exclude the emotional component and moving in the animals. Every animal was used only once.

Cooling

Experiments were performed at ambient room temperature 24–25°C. The initial precooling temperature of the animal was maintained by temperature controlled panel so that the rectal temperature was 38,1±0,11°C; the abdomen skin temperature was 37, 3±0,13°C. A depilated skin in abdominal area of 25 cm^2 was cooled using thermostat. The experimental model of rapid cooling with a rate of skin cooling as 0.1°C/sec was used. Cooling was continued until a decrease of rectal temperature by 3–4°C was reached.

ATP and NE application

Iontophoresis of substances ATP (Fluka, BioChemika), NE (Sigma) was applied with drug delivery system (Medical galvanizator "Potok," Russia); current intensity was 0.08 mA/cm^2 in the area of 25 cm^2 during 20 minutes. Temperature of the applied solutions was 37–38°C. Four groups of rats subjected to iontophoresis were used to detect the effects of ATP (Fluka, BioChemika), NE (Sigma) or their mixture: (1) ATP (n = 14), (2) NE (n = 13), (3) ATP+NE (n = 16), (4) without substances (control group, n = 13). In the experimental groups rats were subjected to iontophoresis of one of the substances or their mixture into the abdominal skin, the area of the subsequent application of the cold stimulus. For iontophoresis, a solution of ATP (10^{-6}), NE (10^{-3}) or their mixture ATP (10^{-6}) and NE (10^{-3}) in distillated water was used.

Thermoregulatory response

The following thermoregulatory parameters were continuously measured throughout the experiment: (1) temperature at a site remote from the cooled and isolated from the environmental and cooling effects, the auricular floor skin; this allowed us to judge how skin vessels respond to the application of drugs and cooling; (2) rectal temperature to measure core temperature; (3) intracutaneous temperature of the cooled abdominal surface to control the skin cooling rate and determine the threshold skin temperature for the cold defense responses; (4) total oxygen consumption and carbon dioxide release to estimate thermogenesis; (5) electrical activity of neck muscles to estimate shivering. The value of CO_2/O_2 was calculated to estimate the changes in respiratory coefficient. All the parameters were recorded by computer IBM PC, using the "Biopac" system. The procedures used to measure the parameters have been also described elsewhere.[28] The following changes during cooling were accepted as threshold values to estimate the start of changes: 0.1°C for temperatures, 1 ml/min kg for oxygen consumption and carbon dioxide release, and 1 μV for electrical muscle activity. The value of CO_2/O_2 was calculated to estimate the changes in respiratory coefficient. Threshold decrease in skin or rectal temperatures for cold defense response (skin blood vessel constriction, the first and second phases of oxygen consumption and CO_2 increase, electrical muscle activity increase) were calculated as a difference of the value of initial temperature before cooling and the value of temperature at which cold defense response started. The maximum values of responses were estimated as the differences between initial values before cooling and peak values during cooling. To determine the effect of substance, the parameters of the cold defense responses were evaluated during cooling in rats – without and with preceding administration of substances.

Design of experiment

Animals were anesthetized, and then all the sensors were fixed (for registration of temperatures, muscle activity, oxygen consumption and carbon dioxide release). Within 5–10 minutes the initial parameters were recorded, then at continuous recording of parameters the application of substances or solution (distillated water) was carried out during 20 min, which were followed by cold exposure. This allowed us to identify the effect of substance in thermoneutral conditions, and in cold.

Statistical analysis

Student's t-test for group comparisons was used for statistical treatment of the data. Differences were considered significant at $p < 0.05$. All values are presented as mean ± SE.

Results

Without cold load

The effects of administration of ATP, NE and their mixture into skin on the recording parameters in the initial conditions, without cold load, are given in **Table 1**. Metabolic parameters, total oxygen consumption, carbon dioxide expiration, and the respiratory coefficient changed under the effect of substances. Changes in the respiratory coefficient reflect the course of changes in oxidation substrates: increase in the respiratory coefficient evidences for increase of carbohydrates oxidation, while its decrease provides evidence that lipid oxidation is increased. ATP caused increase in oxygen consumption by 6.5%, did not change carbon dioxide expiration and, as a consequence, the respiratory coefficient decreased by 4.9% ($P < 0.001$ in paired comparison). NE produced more expressed shifts in metabolism than ATP. Administration of NE resulted in a considerable increase in both oxygen consumption and carbon dioxide expiration. Since oxygen consumption increased to a greater extent than carbon dioxide expiration, the respiratory coefficient diminished by 9.1% ($P < 0.001$ p.c.), even 2 times more compared with ATP effect. When ATP and NE mixture was administrated, the effect of NE was predominant and respiratory coefficient decreased by 9.0% ($P < 0.001$ p.c.) (**Table 1**). Therefore, under the effect of both ATP and NE, the respiratory coefficient reduced, this reduction being more expressed under NE. Thermal parameters and electrical muscle activity were unaffected by ATP, NE and their mixture in thermoneutral conditions. Iontophoresis of solvent had no effects on registered parameters.

Cooling

There is a definite sequence of thermoregulatory responses characteristic to rapid deep cooling.[29] To begin with, when only skin temperature decreases, the *first phase* of metabolic response characterized by rising respiratory coefficient develops (**Fig. 1**); then also without change in core temperature, a constrictor response of the skin blood vessels is initiated. The *second phase* of metabolic response characterized by a decrease in the respiratory coefficient enfolds when not only skin, but also deep body temperature falls, this is associated with an increase in muscle activity (shivering). This was the sequence of responses observed in our current experiments. Administration of ATP, NE, and their mixture prior to cooling was without effect on the sequence of responses; however, there were changes in a number of parameters of the thermoregulatory response to cooling.

Administration of ATP had no influence on thermal threshold and magnitude of constrictor response of the skin blood vessels causing a reduction of heat loss (**Figs. 2, 3**).

Under the effect of ATP, there were no changes in the thermal thresholds for the increase in oxygen consumption and carbon dioxide expiration in response to cooling. (**Fig. 2**), but strongly expressed changes in values of these metabolic responses, especially in the second phase, were revealed. Thus, under ATP oxygen consumption increased by 15% in the first phase (**Fig. 3**, O2 – 1 phase), and by 40% in the second phase (**Fig. 3**, O2 – 2 phase) of metabolic response compared with control cooling when no substances were applied.

The most significant was the influence of ATP on the muscle activity, a component of the metabolic response to cooling. Administration of ATP produced a reduction in the latency of this response, from 396, 8 ± 64,04 sec to 169, 5 ± 33,44 sec ($P < 0,01$). An increase in oxygen consumption and that in muscle activity occurred simultaneously in the second phase of metabolic response to cold. There was a more than twofold lowering of the threshold (by rectal temperature) for thermoregulatory enhancement of muscle activity (**Fig. 2**, bottom). The maximum of muscle activity (shivering) during cooling on the background of ATP increased by 85% compared to controls (**Fig. 3**).

Administration of NE accelerated the constrictor response of the skin blood vessels, the rate of fall of the ear skin temperature increased almost twofold – from 0.11 ± 0.028°C/min to 0.20 ± 0.021°C/min.

The following changes were characteristic of the metabolic response to cooling on the background of NE: 1) earlier

Table 1. Changes in the metabolic parameters under the influence of ATP, NE and their mixture in the initial conditions without cold load

		Changes of values at iontophoresis of substances (% to baseline values)		
Metabolic parameters	**Baseline value (100%)**	**ATP**	**NE**	**ATP + NE**
Total oxygen consumption	21,7 ± 0,41 (ml/min*kg)	6,5 ±2,10* ↑	43,1 ±4,02** ↑	42,1 ±5,01** ↑
Carbon dioxide expiration	16,9 ± 0,40 (ml/min*kg)	1,3 ±2,23	30,2 ±3,99** ↑	29,9 ±4,89** ↑
Respiratory coefficient	0,78 ± 0,007 (unit)	−4,8 ±0,76* ↓	−9,0 ±0,75* ↓	−8,8 ±1,11* ↓

Significant difference in paired comparison to baseline value of parameters: * – $P < 0.05$; ** – $P < 0.01$. Baseline value is the value of parameter before iontophoresis of drugs; it is taken as 100%.

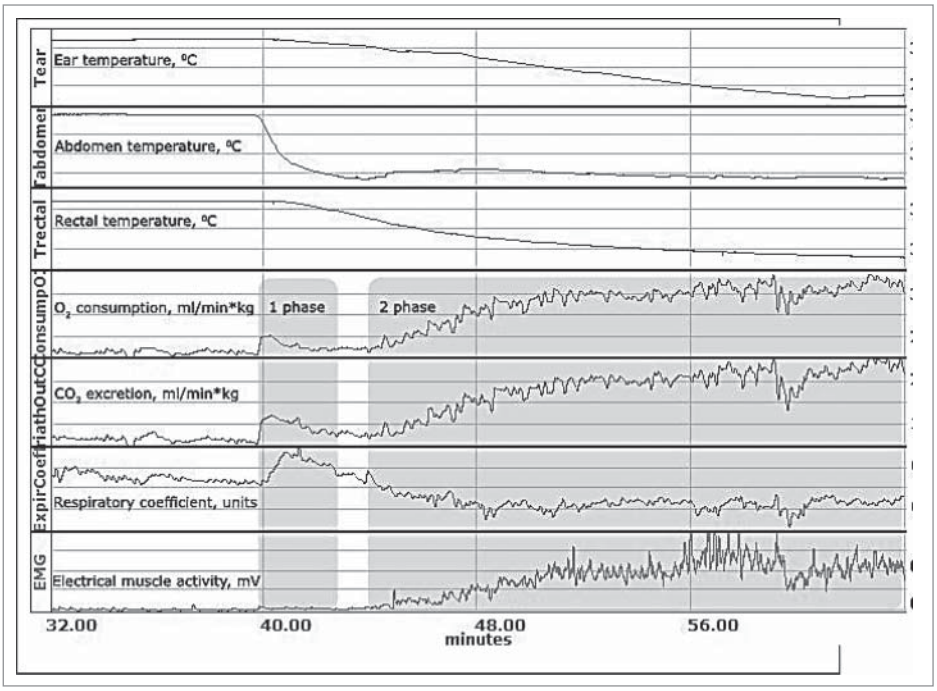

Figure 1. Typical records of thermal and metabolic parameters at rapid cooling; darker areas show the first and second phases of the metabolic response to cooling.

initiation of increase in oxygen consumption in the second phase of metabolic response to cooling (latent period was 426 ± 74 sec in the control, it was 147 ± 22 sec on the background of NE, $P < 0.05$); 2) lowering of thermal threshold (by rectal temperature) for increase in oxygen consumption in the second phase (**Fig. 2**, bottom); 3) increase in the maximum value of oxygen consumption in the first phases of metabolic response (**Fig. 3**).

It should be noted that increased maximum value of oxygen consumption in the first phase of metabolic response was due to calorigenic effect of NE already in thermoneutral conditions. As for additional increment in oxygen consumption in response to cooling in conditions of prior NE administration, it did not change in the first phase (2.4 ± 0.32 ml/min*kg – in control and 2.0 ± 0.54 – on the background of NE; $P > 0.05$), and it was threefold smaller in the second phase of metabolic response compared with that in control cooling (19.5 ± 1.80 ml/min*kg – in control and 5.7 ± 1.53 ml/min*kg – on the background of NE; $P < 0.05$). This means that itself the response to cooling was significantly reduced. Thus, increment rate in oxygen consumption during cooling was 2.7 times greater in the control than when NE was applied (0.019 ml/min*kg*sec in the control, 0.007 ml/min*kg*sec on NE background; $P < 0.05$). Changes observed for carbon dioxide expiration were similar.

The response of muscle activity did not undergo any changes under NE effect. This was in contrast with considerable enhancement and acceleration of muscle response initiation caused by ATP. Another observation merits attention: under the influence of NE in the second phase of the metabolic response the increase in oxygen consumption was much ahead of enhancement in muscle activity (the latent period of increase in oxygen consumption and muscle activity were 147±22 sec, and 634±118 sec, respectively; $P < 0.05$). Threshold decrease in rectal temperature for these responses also differed greatly, being 0.6±0.12°C for increase in oxygen consumption and 3.0±0.27°C for muscle activity.

Administration of mixture of NE and ATP. In these experiments (**Figs. 2, 3**) NE effects were the same as when it was administrated alone, but effect of ATP in the mixture was lower than that, when ATP was administrated alone. Contribution of ATP we observed only in a small decrease by 0.7°C ($P < 0.05$) in the rectal temperature threshold for the muscle activity initiation in response to cooling activity increase compared to NE effect (**Fig. 2**).

R*espiratory coefficient under cooling* on the background of NE, ATP, and their mixture. In all the cases, there was an increase in the respiratory coefficient in the first phase of the metabolic response to cooling, conversely, its decrease in the second phase. (**Fig. 4**). This supports and extends the results of our previous studies.[29] The significantly smaller increment in the respiratory coefficient in the first phase of the metabolic response on the background of NE called attention. It could be smaller presumably because of the considerable increase in lipid utilization caused by NE already in thermoneutral conditions.

Discussion

The results we have obtained provide evidence indicating that, already in thermoneutral conditions, iontophoretic administration of ATP, NE, and their mixture is accompanied by a reduction in the respiratory coefficient. This reduction may be evidence of intensified lipid metabolism. However, it should be stipulated that, in thermoneutral conditions, these changes are considerably more expressed under the effect of NE than ATP. It will be recalled that NE promotes lipid metabolism through enhanced utilization of fatty acids. The effect of NE on brown adipose tissue has been widely discussed.[29]

Thermal thresholds of the thermo-defense responses characterize the structure of the thermoregulatory response; and they are those indicators that are in the dependent on the activity of thermoreceptors, when organism exposed to cooling or heating. Thermal thresholds of thermoregulatory responses are determined by various groups of

thermoreceptors located in the skin, brain structures, and visceral organs. We used here a specific strategy for administration of biologically active substances (iontophoretic administration into the skin of a cooled area). This implies highest concentration of the substance for the skin receptors in the cooled area, so that changes in thresholds for skin temperature during formation of cold-defense response would be expected with good reason. ATP was without effect on skin temperature thresholds for all the cold-defense responses. This would seem to be evidence for no significant effect of ATP on the activity of skin thermoreceptors. But the lowering of deep temperature threshold for initiation of cold shivering thermogenesis, nevertheless, argues for the possibility that ATP may have an influence on the activity of thermoreceptors structures. As is well known, the afferent signal for initiation of effector responses is a combination of signals from skin and internal thermoreceptor structures whose sensitivity to the mediators of the sympathetic nervous system can be different. It is possible also that a small increase in the activity of skin thermoreceptors, although not displayed in the skin temperature threshold, nevertheless, does facilitate initiation of responses, and it appears at a smaller fall in deep body temperature. Changes in the thresholds of the thermoregulatory responses under the administration of NE and ATP may evidence for feedback influence of the sympathetic nervous system transmitters on thermoreceptor structures, which is consistent with our previous data with respect to NE.[8]

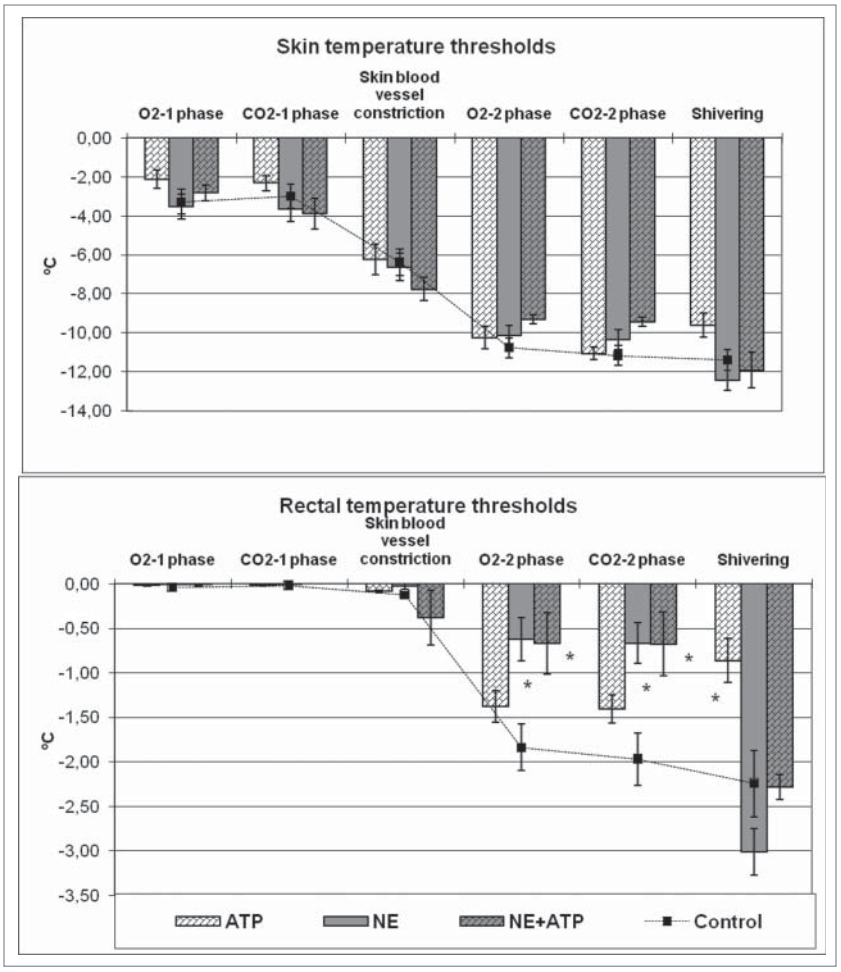

Figure 2. Threshold decrease in skin (top) and rectal (bottom) temperature for cold defense responses in control (without any drugs) and on the background of ATP, NE, and their mixture. Significant differences from control – $*P < 0.05$, Student's t-test. Significant differences between NE and NE+ATP – $\# P < 0.05$.

It appears that at deep cooling, with decreasing in heat loss, metabolic response is initiated, too, namely cold thermogenesis directed toward greater heat production. The current study demonstrates that under ATP effect both the first (emergency) phase of thermogenesis, the one associated with enhancement of carbohydrate metabolism, and its second phase, the one associated with enhancement of lipid metabolism, increase. The increase in the second phase is particularly pronounced.

The second phase of metabolic response is characterized by a rise in total oxygen consumption. It incorporates 2 components. One is nonshivering thermogenesis, which is ensured in rats mainly by heat production in brown adipose tissue; the other component is shivering thermogenesis provided by heat production during thermoregulatory muscle activity.

It is thus evident that administration of ATP influences both thermal threshold and the magnitude of shivering thermogenesis, one of the effector components of the thermoregulatory response. The mechanisms potentiating shivering thermogenesis under the effect of ATP presumably associated with both the action of ATP on the muscles as a mediator, and well-known process of ATP utilization in muscle Ca(2+)-ATPase. The recruitment of Ca (2+) for cold thermogenesis has been confirmed by many researchers. In our previous experiments, administration of agents blocking calcium channels and those blocking α1-adrenoreceptors (calcium is a secondary messenger of these receptors) leads to shivering suppression, while administration of Ca(2+) ions vice versa promotes earlier initiation of shivering and its enhancement.[30,31] There are also data regarding the influence of α1-adrenoreceptors on ATP release and Na-K ATPase activity.[32,33] The participation of SERCA1, one of the enzymes of the sarcoplasmic reticulum of muscle cells, involved in heat production, may be also suggested.[34,35] SERCA1 modulates the distribution of energy, which is liberated during ATP hydrolysis, between heat production and Ca(2+) transport in favor of the former process.

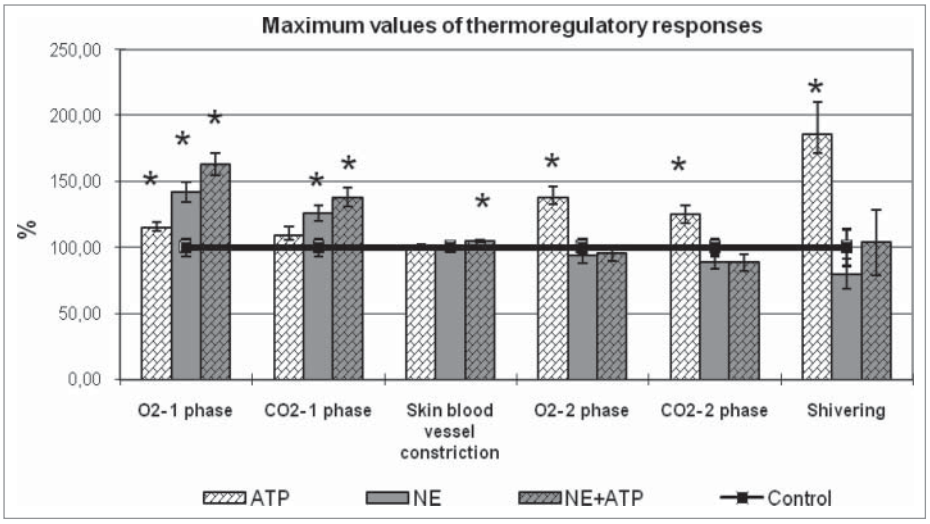

Figure 3. Effects of ATP, NE, and their mixture on the maximum values of cold defense responses. Black squares – the values of responses in control animals (without any drugs) are taken as 100%. Significant differences from control – * $P < 0.05$, Student's t-test.The absolute values of responses in controls are as follows: in the 1 phase of metabolic response O2 consumption −24.5 ±0.66 ml/min*kg, CO2 release −19.1 ±0.71 ml/min*kg; skin blood vessel constriction characterized by the decrease in skin temperature to 28.0 ±0.27°C; in the 2 phase of metabolic response O2 consumption −31.5 ± 1.60 ml/min*kg, CO2 release −20.9 ±1.34 ml/min*kg; shivering (electrical muscle activity) −16.1 ± 2.34.

Unlike ATP, NE lowers the thermal threshold of non-shivering thermogenesis, enhances the metabolic component of the thermoregulatory response by non-shivering thermogenesis whereas neither temperature threshold, nor magnitude of the activity of the muscle alter.

The results of the current study demonstrate that ATP and NE increase the metabolic response to cooling, due to different components: ATP – at the expense of the shivering thermogenesis, NE – due to non-shivering thermogenesis.**Fig. 3**

Debates waged in the 1960–1970ies over the problem of the nature of cold thermogenesis with reference to the involvement of the sympathetic nervous system in its regulation. It was reported that NE and activation of sympathetic nervous system influenced on non-shivering thermogenesis (a rapid rise in brown adipose tissue function), also on the heat effect of muscle contraction. NE potentiates the activity of brown adipose tissue and augments the heat effect of skeletal muscle contraction through β-adrenoreceptor mechanisms.[29,36] Potentiation of these processes was observed after long-term adaptation of the organism to cold.[2,37] ATP as a transmitter and regulator of these processes has not been considered. On the agenda were the discussion on the problems of change in ATP synthesis brought about by uncoupling of oxidative phosphorylation and a greater energy cost of heat production. The current study demonstrates the different application processes of the ATP and NE as co-transmitters in the regulated changes in metabolism caused by cold exposure of the organism.

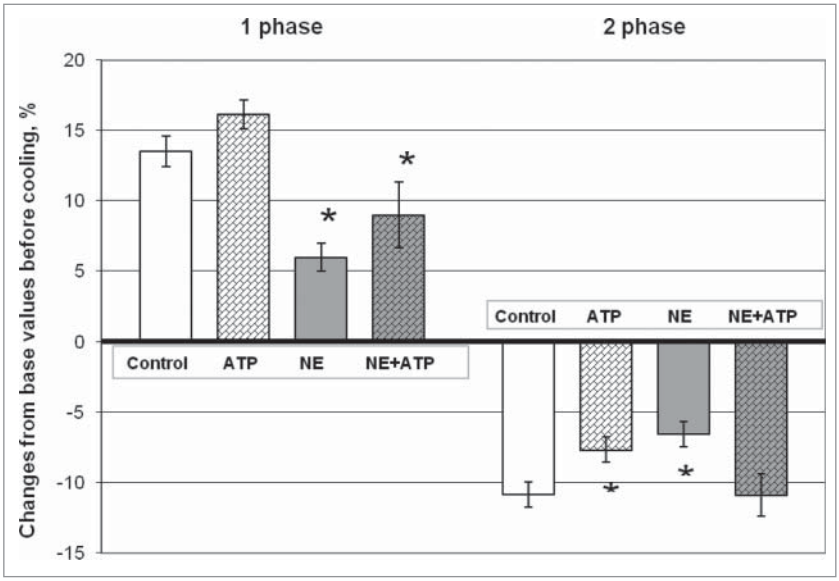

Figure 4. Changes in the respiratory coefficient in the first and second phases of metabolic response to cooling in control (without any drugs) and on the background of ATP, NE, and their mixture. Significant differences from control – * $P < 0.05$, Student's t-test. The absolute values of the respiratory coefficient (RC) changes in control animals: in the 1 phase of metabolic response RC increases by 0.09 ±0.008 ($P < 0.05$); in the 2 phase of metabolic response RC decreases by 0.08 ±0.007 ($P < 0.05$).

Summarizing, both NE and ATP are conducive to enhancement of thermoregulatory response to cold. NE is mainly conducive to activation of vascular response and non-shivering thermogenesis in the first and second phases of the metabolic response, while ATP somewhat enhancing emergency thermogenesis, affects, to the greatest extent, on muscle cold thermogenesis, increasing it almost twofold. Consequently, transmitters of sympathetic nervous system NE and ATP affect the different components of the thermoregulatory response to cooling.

When administered as a mixture (NE and ATP), the effect of NE is fully expressed but the effect of ATP is lower. It may also be so, when the sympathetic nervous system is activated NE effect dominates, and NE acts ahead, thereby producing greater enhancement of non-shivering thermogenesis and an increase in heat production per unit of muscle contraction, whereas the

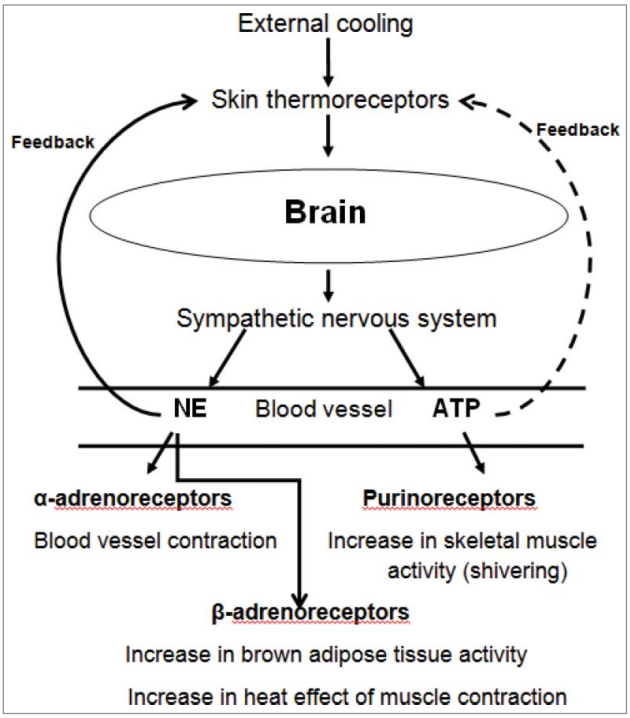

Figure 5. Scheme of ATP and NE participation in regulation of thermoregulatory responses. The presented data may testify in favor of the influence of ATP on the skin thermoreceptors, however, to be sure, the data on the effect of ATP on impulse activity thermoreceptors need. That is why the feedback effect of ATP on thermoreceptors is shown by dotted line.

enhancement of muscle contraction (shivering) is caused with the action of ATP, co-transmitter of NE. Thus, NE and ATP being co-transmitters and predominantly acting on the different processes may organize the certain sequence of cold defense responses while forming the response of the living organism to cold *in vivo*. The data may, also, evidence for the possibility of feedback influence of the sympathetic nervous system transmitters, NE and ATP, on thermoreceptor structures about which the changes in the thresholds of the thermoregulatory responses indicate.

According to previous and presented results we can suggest the following scheme (**Fig. 5**). Cold, in dependence on depth, activates or only peripheral skin thermoreceptors, or both peripheral and central thermoreceptors. Thermosensors form the signals to the brain structures which formulate the efferent information and stimulate sympathetic nervous system. Co-transmitters of sympathetic nervous system NE and ATP are released to the blood and modulate the different components of the thermoregulatory response. NE and ATP can change the blood vessel tonus, NE increases the brown adipose tissue activity and amplifies the heat effect of muscle contraction (participation of ATP in these processes is not known), ATP increases the skeletal muscle activity which we can observe as shivering (the increase in electrical muscle activity). Besides that, NE effects on the skin and brain thermal afferents and can change their sensitivity to temperature – this is feedback to afferents.[4–8] The character of ATP influence on impulse activity of thermal afferents requires the following researches.

Recent evidence suggests that keratinocytes, the major cell type in the epidermis, can communicate with sensory neurons due to ATP (38, 39). Keratinocytes can also respond to changes in temperature as they express temperature-sensitive TRP ion channels.[40,41]

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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