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Different Views on the Tissue Thermogenesis of Organisms

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Abstract The review discusses thermogenic mechanisms of animal tissues, including mitochondria, which provide the endothermic state of the organism. Previous views on thermogenesis were largely based on the idea of low efficiency of biological processes. Later, the data gradually accumulated about the possibility of a sufficiently high efficiency of these processes. Otherwise, there would not be cold-blooded organisms having a low level of metabolism and a high efficiency of work. Currently, the role of Na, K-ATPase and proton leakage of mitochondria is being intensively elucidated in thermogenesis. Uncoupling proteins (UPC) were isolated from the inner membrane of mitochondria animals' tissues, which increase the proton leakage of membranes, reducing the efficiency of the respiratory chain. Membrane anion-translocators can also participate in the proton leakage, which as a whole can increase the basal metabolism from 20 to 50%. In another group of studies showed, that a large number of substrates oxidize in mitochondria, coupled with ATP synthesis, as well as uncoupled pathways. It is supposed, that uncoupled respiration is realized in separate subpopulations of mitochondria and adapted to thermogenesis. The authors showed the nativeness of coupled respiration, since it was also detected in isolated rat cardiocytes. Uncoupled respiration proceeds more intensively in tissues of endothermic animals than in ectothermic organisms. In general, this review critically analyzes the problem of low efficiency of biological processes and examines alternative ways of tissue thermogenesis.

Keywords Thermogenesis, Endothermicity, Efficiency, Tissue mitochondria, Proton UCP, Uncoupled respiration

1. Introduction

The subject of our review is the thermogenesis mechanisms in warm-blooded organisms. To maintain a warm-blooded state, mammals and birds must produce heat all their life regardless of the temperature conditions of the environment. This process is associated with of large energy expenditure of the organism. Therefore the study of which is one of the fundamental problems of modern biology. This prerequisite could be served as a basis for the research of mechanisms of tissue thermogenesis, providing the warm-blooded state of the organism [1-5].

Bioenergetics has important successes in elucidating the mechanisms of energy conversion and its utilization at the cellular and intracellular levels. The discovery of oxidative phosphorylation in mitochondria in the last century allowed to subsequently study the role of these systems in physiological processes, i.e. on the tissue level and the whole organism. It should be noted that along with the achievements of that period, in our opinion, some issues remained weakly developed. It must be said at that time the main object of research was often warm-blooded organisms, in which the metabolism is high [6-9]. This factor could affect the individual conclusions of the researchers, in particular, relative effectiveness of energy processes (or efficiency), what was assessed as low value. With this opinion, thermodynamic calculations were also agreed. However, the determination of metabolism in cold-blooded organisms demonstrated a low level of metabolism [10-12], and the locomotion efficiency was higher [11, 13-18] than endotherms. These data differed from the results obtained on warm-blooded animals and shown the possibility of low energy cost of life processes. Meanwhile, certain bioenergy approaches are being developed a concept that, at the tissue and mitochondrial level there are special energetic mechanisms for ensuring the warm-bloodedness of organisms [19-23].

In this review, the main attention is paid to the heat production in the organism under thermoneutral conditions. Issues, related to thermogenesis under cold conditions were considered earlier in the works of V.P. Skulachev [8]. His works of free uncoupled respiration was the first concrete mechanism of mitochondrial thermogenesis. Further studies led to the establishment of uncoupling proteins in the inner mitochondrial membrane as a possible proton leakage base

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[24-25] and uncoupled thermogenic respiration [26, 27].

Warm-bloodedness of the organism and metabolism. According to available data, the intensity of heat production of vertebrate organisms at the optimal ambient temperature, differ significantly [28-30]. The ectothermic animals have approximately 10 or more times lower metabolism and a variable body temperature than endotherms. These achievements can radically change the current understanding of the cost of life processes and the efficiency of biological processes and cause a new breakthrough in this direction. In modern literature, the concepts of "warm-bloodedness and cold-bloodedness", "homoiothermy and poikilothermy", "endothermy and ectothermy" are widely used to classify animals according to their temperature status [28, 29, 31, 32].

However, considerable effort was required to understand to some extent the tissue mechanisms of the differences between cold and warm-blooded organisms. The obtained data in this aspect showed that the investigated groups of organisms have certain differences in tissue energetics. Further studies in this direction have allowed forming certain views on the nature of tissue thermogenesis, as will be discussed below.

The role of body weight and locomotion in the metabolism. It was found that the intensity of metabolism among endothermic and ectothermic organisms depends on their body weight and decreases with increasing body weight, with the slope of the regression curve being approximately the same for these groups of animals [10-12]. This regression is expressed by the formula $V_{O2} = k \bullet M^{0.72}$, where V_{O2} is the metabolism intensity, M is the body mass and k is the proportionality constant. The dependence of metabolism on the mass of the organism is generally accepted as valid for heat and cold-blooded vertebrates [11], although considerable time was devoted to its separate aspects [31]. Meanwhile a concrete mechanism of dependence of metabolism on a body mass remains unknown.

According to the above said, at comparing cold-blooded with warm-blooded, one should take into account the equality of their mass. In the intensity of heat production, endo-and ectothermal vertebrates differ in the same body temperature by a factor of 5-10 [11, 18, 32, 33, 35] at an optimum temperature of ambient conditions (about 20℃). The difference between them reaches 40-50 times [10, 11]; at 10℃ the ambient temperatures differ approximately 100-fold [18].

Separate studies are devoted to the analysis of the energy cost of locomotion (movement speed/kg of body weight) in endo- and ectotherms. According to the data obtained [11, 13-18], the price of locomotion is 2-10 times higher in endothermic than in ectothermic organisms.

As can be seen from the above-said, the most significant difference between ectothermic and endothermic animals is the ability of the former to spend significantly less energy resources on vital activity at rest, and at activation of physiological processes. Therefore, if we start from the studies of cold-blooded organisms, vital activity appears to be an energy less wasteful process and proceeding with a high level of efficiency. However, at considering warm-blooded animals isolated from cold-blooded animals, providing vital activity seems to be associated with high-energy costs. In the evolution of organisms, apparently, certain changes in the mechanisms of energy supply of the basic functions of vital activity, in particular, the appearance in the organism of a new energy function ensuring the warm-bloodedness of the organism, could occur. This circumstance could become one of the main causes of the qualitative difference between endo- and ectothermic organisms in the intensity of metabolism.

The results of the physiological study of ecto- and endothermic animals served as a prerequisite for the development of biochemical bases of biological thermogenesis. As a result, a number of viewpoints were proposed on the origin of endothermic organisms. In particular, we will consider such aspects of the problem as the role of low efficiency of energy processes, the amount and composition of mitochondria, as well as the uncoupling mitochondrial proteins, proton leakage and uncoupled respiration.

Efficiency of the respiratory chain and thermogenesis. For a long time among researchers, the point of view on the important role of low efficiency of energy reactions in thermogenesis was very popular.

Based on the value of the standard free energy of hydrolysis of ATP (ΔG^o), which corresponds to approximately 7 kcal/mole, and the real free energy (ΔG) of ATP under the conditions of the cell is approximately 13-16 kcal/mole (36). Therefore, many researchers believed that approximately half of the energy of substrates oxidation at the stage of ATP synthesis in mitochondria is dissipated as heat, and energy-dependent processes utilizing ATP disperse about half of the remaining energy. In total, the loss in the form of heat could be about 70-80% of the initial energy of the oxidation substrates. Only about 20% of energy was stored in useful biological processes [6, 7, 9, 36-39].

The free energy (ΔG), determined on the content of ATP, ADP and phosphorus in tissue, was significantly higher than the standard free energy of ATP hydrolysis (ΔGo) and approximately corresponded to the values of 13-16 kcal / mole of ATP [36, 39-44]. These results indicate that the organization of the cell energy system of is adapted to maintain a high level of free energy in the synthesis and hydrolysis of ATP. At the cellular energy values of ATP hydrolysis (13-16 kcal/mole) and the ratio of reagents near to equilibrium, the efficiency of the respiratory chain can be more than 80%. Therefore, the synthesis and hydrolysis of ATP may not be an important source of thermogenesis. In addition, for the correct use of the thermodynamic approach in bioenergetics, it is also necessary to involve data on free energy in the tissues of ectothermic organisms, which is little used in this aspect. It is necessary a certain reorientation in this area, taking into account the existing achievements in this aspect.

Wilkström and Krab [45], studying the energy transfer on

a model system with liposomes, received data that the respiratory chain can function with an efficiency close to 100%. The possibility of a high efficiency in the energy transfer of at the ATP synthesis is also under consideration now. In particular, new molecular models of the energy transfer in the mitochondrial respiratory chain may be functioned with an efficiency of up to 100% [3, 46]. In addition, the theoretical prerequisites do not exclude the possibility of the presence in the tissues of such reactions, which take place with the absorption of heat (endothermic reactions). In particular, Hard [47] suggests that in the synthesis of ATP in mitochondria should absorb heat from the medium, because under these conditions the electrochemical potential produces chemical work. Thus, the question of the effectiveness of energy reactions is currently confronted with unexpected aspects. The above mentioned data suggest that the efficiency of these reactions can be much higher than 50%, and therefore the oxidative phosphorylation system and energy-dependent processes may not be the main source of heat in the organism.

It must also be remembered that these energy processes also occur in the body of cold-blooded organisms, in whom these processes occur at low level of metabolism, which contradicts the concept of low efficiency of biological processes discussed above. Under the condition of occurrence of biological reactions with low efficiency, it is unlikely that there were cold-blooded organisms would have a high level of metabolism. Therefore, now, researchers are conducting an analysis of energy processes in biological tissues, taking into account current trends in that area [22, 23, 48, 49].

The phenomenon of aerobic metabolism activity. Dolnik and Bennett, Ruben [18, 32], at interpretation of the nature of homoeothermic organism, attach great importance to the phenomenon of "activity" of metabolism. In particular, the authors believe that the warm-bloodedness is due to high aerobic metabolism, the level of which is up to 5-10 times higher than in cold-blooded animals, at testing the body temperature of the animals compared under the same conditions. According to their opinion [18, 32], aerobic activity is the basis for increasing the body's energy needed for the rapid movement of mammals [18] and for bird flying [31]. Discussing this issue, the evolution of blood circulation, the respiratory system in vertebrates, as well as an increase in the concentration of mitochondria in tissues [18, 51]. It must be said that "aerobic activity" in warm-blooded organisms is most likely associated with an increase in energy consumption for vital activity and an increase in heat production in endotherms. The basic metabolism, including thermogenic processes, obviously take place at the mitochondrial level. In our opinion, the evolutionary transformations of mitochondria led to increasing heat production and becoming of endothermic organisms.

The role of transport Na, K-ATPase in tissue thermogenesis. Considering that ATPase is a membrane unit of all biological cells, it was studied its role in the energetics of tissues of endothermic and ectothermic animals [52, 53]. According to the obtained data [54, 55], respiration of tissues dependents on sodium transport that is much higher in endotherms, and the effect of uabain (Na^+, K^+ -ATPase inhibitor) is also more pronounced in endothermic animals. The authors suggest that the sodium pump can be the main factor of heat production in the body of endothermic animals. A similar point of view is also shared by Hulbert and Else [34]. They showed that the respiration rate in the tissues of mice is 2-5 pas higher than in reptiles, which can cause Na^+, K^+ -ATPase.

The results of the energy contribution study of the sodium pump are diverse. According to Van Rossum [56], transport-coupled respiration with Na^+, K^+ -ATPase functioning on liver sections accounts for only about 8% of the total energy expenditure of tissue. Approximately the same energy values of Na^+, K^+ -ATPase are given in other works [57-59]. According to the results of Ismail-Beigi and Edelman [54] this process can occupy about 40% of the total energy expenditure.

Along with the analysis of energy processes, much attention is paid to the protein-phospholipid composition of tissues in different vertebrates. The results of the studies showed the prevalence of proteins and polyunsaturated phospholipids in membranes of endothermic animals than in ectothermic ones. According to the opinion of authors, polyunsaturated phospholipids activate the mobility of Na^+, K^+ -ATPase enzymes and enhance the flow of sodium through the cell membrane [35, 59-61]. These data suggested that the work of the sodium pump is associated with an increased expenditure of tissue energy and is a pacemaker of cellular metabolism [60, 61]. A number of determinations of energy inputs for the functioning of the sodium pump have been made [62-65], which showed the possibility of its unequal contribution, to the energetics of different tissues. According to the calculations, the sodium pump consumes about 20% of the basal metabolic rate in endothermic animals [62, 64, 65, 66].

The sodium pump in cold-blooded organisms has a low activity [53], the energy expenditure is significantly less than in warm-blooded animals and its efficiency is about 8% [66]. Ectothermic organisms have been little studied in terms of the energy efficiency of tissue processes. Further studies are needed in this plan, since the existing data have a certain scatter.

The amount of mitochondria, their structural features and activity of the enzymes of the main respiratory chain in endo- and ectothermic organisms. The shown low activity of metabolism in cells of ectothermic animals may depend on the number and structural organization of mitochondria in different animals. An electron microscopic study of skeletal muscle cells in endo- and ectothermic animals showed that the latter had a bulk density of mitochondria 2-3 times lower than in endothermic ones [67]. In addition, it was found smaller surface area of the liver mitochondria of reptiles than in endotherms [19, 35]. These features of endothermic animals to a certain extent could serve as a cause of low metabolic activity in ectothermic

organisms. However, as it turned out, the enzymes of the respiratory chain mitochondria in quantitative terms differ little in the homogenates of the tissues of lamprey and reptiles [68, 69]. Moreover, the amount of mitochondrial protein determined by their marker cytochromes are the same in different groups of animals per 1 g of crude liver mass [68, 70].

According to the available data [71, 72], in mitochondria of ectothermic organisms, there is a low rate of oxidation of exogenous NADH, whereas in mitochondria of endothermic organisms, oxidation of exogenous NADH proceeds at a high rate [73-81]. Low NADH oxidase, in our opinion, may be a prerequisite for the evolutionary development of uncoupling mitochondrial respiration [77] as the main thermogenic mechanism of endothermic vertebrates.

Protein and phospholipid composition of mitochondria in endo- and ectothermic organisms. A substantial reorganization of the protein and phospholipid composition occurred in the evolution of vertebrates as evidenced by the results of studies in this direction. Thus, up to 60% of the increased protein content and the amount of phospholipids in endothermic tissues are detected, in contrast to ectothermic vertebrates [56]. Among the phospholipids, semi-unsaturated forms predominate, which are associated, in particular, with the functioning of Na^+ -K^+, ATPase and increase the permeability of the cell membrane to monovalent cations [35, 66]. Composite rearrangement occurs at the mitochondrial level [35], which could be a prerequisite for activation of proton leakage.

The effect of the protein and phospholipid composition is shown on the cell membrane, because of which the permeability of the membranes to sodium is increased. As a result, the sodium cycle is activated, and the metabolism of the cell is enhanced [60], which ultimately raises tissue thermogenesis. The sodium cycle, together with other cycles, is considered as pacemaker of the metabolism [23, 60].

The adenine nucleotide pool as a factor in the regulation of energetics in organisms. Based on the results of M.V. Savina [68] an adenine nucleotide pool (the total content of ATP, ADP, AMP) can play an important role in tissue respiration. The author develops this idea based on their results obtained on the lamprey tissues, which is in the low-energy state for a long time at the period of a migration. It was shown that under these conditions there is a significant decrease in the adenine nucleotide pool in tissues, although the amount of mitochondria in the muscle remains higher, than in warm-blooded animals. The results of these experiments emphasize the important role of the adenine nucleotide pool in the regulation of the low-energy state of organisms, which is also important in the study of the hibernation or the estivation of endothermic and ectothermic animals [78].

The obtained data on the large number of mitochondria in the lamprey tissues combined with the low level of metabolism in these organisms indicate that mitochondria are a key to understanding such important problems as the mechanisms for metabolism regulation and the efficiency of energy processes, as well as a key an on-off of low-energy states. However, regulatory mechanisms of efficiency of energy processes can considerably differ in organisms.

Proton leakage, uncoupling mitochondrial respiration and thermogenesis. One of the promising approaches in this field is a study of process mitochondrial proton leak. It is caused by specific membrane proteins (uncouple proteins - UCPs) located in internal membranes of mitochondria. Several species of UCPs have been identified in different tissues, which are designated as UCP1, UCP2, UCP3, UCP4 [79]. They contribute to the dissipation of the proton gradient on mitochondrial membrane, which was created by the energy of oxidation of substrates. Dissipation of the proton gradient is a source of biological heat that provides endothermy of the organisms [25, 59, 63, 80].

The conducted comparison in phylogeny of different groups of animals showed that in reptiles the proton leakage in hepatocytes and their mitochondria is lower than in mammals, although the difference is insignificant [82]. These data, in conjunction with others [83] were poorly consistent with the idea of the thermogenic significance of UCPs and the maintenance of homoiothermy of the organism. Perhaps, therefore, the anion-translocators, in particular, the adenine nucleotide translocase (ANT). Thus, at comparison endo- and ectothermic organisms, it has been shown that the value of proton conductivity correlates well with the number of mitochondrial anion transporters, in particular, with the amount of ANT [84, 85], which may account for about 75% basal proton leakage. The authors believe that the ANT cannot determine the entire basal proton leakage and therefore the participation of other anion-translocators of mitochondria is possible. These representations are substantiated by an experimental comparison of the proton conductivity of mammalian and reptilian hepatocytes. In the latter, the rate of oxygen consumption is 4-5 times weaker, which correlated with a 4-fold lower level of ANT in the reptilian [24, 84].

It should also be noted that the whole population of mitochondria participates in the proton leakage process. Moreover, UCPs are found in ectothermic and other organisms, with a few exceptions [86].

It is considered to be possible the participation of UCPs in thermoregulation [23, 82, 87, 88], obesity, disease, aging, and superoxide level regulation [89, 85].

A number of works indicate that the contribution of proton leakage to the main tissue metabolism is about 20%, and skeletal muscle level reaches 50% [85]. Researches in this respect continue, since this contribution of UCPs proteins does not always explain the difference in metabolism between endo- and ectothermal organisms that differ in the level of metabolism within ten or more times.

It should also be noted that in studies in this direction, succinate is mainly used as a substrate for oxidation at mitochondria. To obtain a more complete proton leakage characteristic, it would be desirable to use different substrates [90, 91, 92, 48].

Proton leakage intensity was compared at use of

pyruvate+malate and succinate as oxidation substrates in mitochondria. In these studies, oxidation of succinate occurs with less coupling than in the oxidation of pyruvate + malate. Therefore, it is noted [48] that succinate can be oxidized in a different way than NAD-dependent substrates.

At present, new scientific methods are being introduced to study the energy of thermogenesis [48, 93, 94]. At the same time, there are some conditions that one can approach the questions of proton leakage and tissue thermogenesis, which we will discuss below.

In other series of researches devoted to the study of thermogenic respiration of mitochondria the additional possibilities for heat production have been identified [26, 95, 96], which can occupy a significant part of mitochondrial respiration. To judge the native state of the results, the part of experiments were performed on a cellular preparation (cardiocytes) [27] using different oxidation substrates and mitochondria of endo- and ectothermic animals [26]. It must be said that in our experiments the substrates of oxidation were also succinate and exogenous NADH (+cytochrome c) and ascorbate (+ cytochrome c). The last two substrates are not popular among bioenergetics and they are little used in experiments. The study of their oxidation together with other substrates under our experimental conditions is useful for analyzing the uncoupled mitochondrial reactions.

It is interesting to note the properties of oxidation of the used substrates. So, on succinate, the rate of respiration of the mitochondria of the heart (or other tissues) occurred at an increased rate in state 4 and with low respiratory index (V_3/V_4). This widely known fact is interpreted differently in literatures [96, 97]. We, unlike the literature, suggested that this is a manifestation of uncoupled oxidation and that in the isolated mitochondrial suspension some of these organelles have high membrane permeability that can increase the respiration rate in state 4. Uncoupled mitochondria have a particularly strong effect on the oxidation of succinate, since this process does not need in the addition of coenzymes and the electrons from the succinate are transferred directly to the respiratory chain of the mitochondria. Therefore, in the oxidation of succinate, two subpopulations of mitochondria - coupled and non-coupled - can immediately be involved, which eventually causes a decrease in the mitochondria oxidative phosphorylation parameters. Recently, other authors have also examined the unusual oxidation of succinate in mitochondria [48] that confirm our above-mentioned concept.

In our experiments on mitochondria, NAD-dependent substrates - glutamate, pyruvate + malate (without NAD and cytochrome c in incubation medium) were oxidized in the used tissue preparations with large respiratory index and ADP/O, which corresponds to numerous literature data.

Uncoupled mitochondria do not affect the NAD-dependent substrates oxidation, so these substrates first reduce the coenzyme of NAD then NADH enters the respiratory chain. However, the water-soluble coenzyme NAD leaves the high permeable subpopulation of mitochondria that inhibits the use of NAD-dependent substrates. To activate their oxidation along the non-coupled pathway, it is necessary to add into the medium coenzyme NAD (1 mM), which actually increases the rate of respiration of mitochondria in state 4 [26, 73, 95]. Typically, researchers do not always use this combination of substrates for obtaining the better coupled respiration.

Among our used substrates, the oxidation of the NADH substrate under our experimental conditions is of certain interest. In our experiments [26, 73] EDTA, BSA, and other ingredients were used for the isolation of mitochondria. These conditions influence on NADH oxidation to a lesser extent. As well it turned out that exogenous NADH (+ cytochrome c) is oxidized intensively by preparations of isolated mitochondria on the rotenone-sensitive pathway. Ascorbate (+cytochrome c) oxidation in cardiac or adipose tissue mitochondria was inhibited by sodium cyanide. The obtained data have shown, that uncoupled oxidation of substrates occur on the main respiratory chain of mitochondria.

Along with uncoupled oxidation of substrates, Mg^{+2}-ATPase was found in mitochondria [98, 99], which additionally confirms the presence of non-coupled mitochondria are the other enzymes.

In the literature, the phenomenon of intense and non-coupled oxidation of NADH in the mitochondria of the heart and muscles has been described for a long time [73, 74, 76] as a native property of mitochondria and a regulator of the cytoplasmic level NAD/NADH.

Under our conditions, it has been shown that the oxidation of NADH also proceeds without the phosphorylation of added ADP, and oligomycin or carboxyatractyloside does not have a significant effect on the dynamics of their oxidation. Moreover, mitochondrial respiration in a rat heart is inhibited by rotenone (1 μg/ml) on NADH, approximately 90%, and sodium cyanide (1 mM) on both substrates. Consequently, their oxidation occurs along the main respiratory chain of the mitochondria, but is not associated with the ATP synthesis. In rat liver mitochondria, the oxidation of NADH is only partially sensitive to the rotenone [77], and this part of oxidation proceeds along the main respiratory chain. The rotenene-insensitive fraction that proceeds along the redox chain with cytochrome b5 [8, 100].

It must be said that the two substrates, used by us (NADH and ascorbate) are usually rarely used in experiments with mitochondria. Their intensive and uncouple oxidation by many researchers is considered as a sign of damage of isolated mitochondria, i.e. as an artifact of research [6, 7]. To show the low probability of the artifact in our studies, we conducted a study of uncoupled respiration on an intact cell preparation - cardiocytes, mitochondria of which are not exposed to damaging factors of isolation conditions [27].

On isolated cardiocytes, as well as isolated mitochondria, it has been shown the functioning of uncoupled mitochondria, along with coupled ones. Isolated cardiocytes also intensively oxidized exogenous NADH and ascorbate in the presence of cytochrome c [27]. The cardiocytes also had a higher respiration rate in state 4 during the oxidation of

succinate, whereas at NAD-dependent substrates mitochondrial respiration in state 4 is low. Isolated cells actually duplicate isolated mitochondria by the nature of respiration on different substrates that we consider as one of the main native criteria. In the isolated mitochondria of the heart under a light microscope, after application succinate and rhodamine 123 into the medium, two mitochondria groups can be seen; one of which has a membrane potential, and the other has no potential [77].

It should be noted that study of the thermogenic organ – mitochondria of brown fat showed that in them NADH and ascorbate (+ cytochrome c) are oxidized very intensively in the uncoupled pathway [101]. Uncoupled respiration in the kidneys and skeletal muscles is also intense, and there is moderate activity in other tissues [26, 73, 77]. To further justify on the native state of this process, we conducted a comparative study of the mitochondria tissue of warm-blooded rats with cold-blooded frogs, turtles and agamas [78], which showed a low activity of uncoupled respiration in the tissues of cold-blooded organisms.

The experiments carried out in this respect showed that in the mitochondria of the lake frog tissues, the uncouple oxidation of both NADH and ascorbate (+ cytochrome c) occurred at a low rate, and succinate and especially NAD-dependent substrates were oxidized with high couple with ATP synthesis. Consequently, for mitochondria of cold-blooded, a low level of uncoupled respiration is characteristic, which can cause a low activity of the metabolism in the frogs' organism. The corresponding results were obtained on the tissues of desert tortoises and agamas [78].

The results discussed above can confirm the uniqueness of the oxidation processes, including uncoupled respiration during oxidation of exogenous NADH and ascorbate. Based on these results, it can be assumed that nature itself has designed uncoupled mitochondria in tissues for the sake of creating warm-blooded organisms. Because of this evolution of the energy system, organisms had the opportunity to reduce the efficiency of the energy system and sharply increase the intensity of metabolic processes, which in turn caused origin of warm-blooded mammals and birds [77].

2. Discussion

Concerning of the connection between uncoupling proteins [24, 84] and uncoupled respiration [26, 95], in our opinion, it can consist in the localization of these uncoupling proteins in the membrane of uncoupled (high permeable) mitochondria, but this question, undoubtedly needs experimental research. In addition, this issue directly elucidates the heterogeneity of the mitochondria, which still has many uncertainties and few physiological solutions to arising problems.

Much data have been obtained in different classes of vertebrates. It must be said that the differences in the metabolism level between organisms are quite high [32, 29, 68, 11] - from three to a hundred fold and more. Under the condition of low efficiency of energy processes, organisms with a low level of metabolism could not exist. The low level of metabolism is not consistent with the low efficiency. The movement of life processes, most likely, involves a high efficiency of energy utilization and its origin. In addition, under extreme conditions, a further decrease in the intensity of metabolism occurs in a number of organisms, in particular, in hibernating animals [78] or in fish with prolonged migration [68]. These data are little consistent with the ideas of low efficiency of biological processes in the organism. On the contrary, effective ways of using energy with its unique mechanisms can prevail in the living world. The application of the idea of energy conversion with low efficiency in biology came from the thermodynamics of entropy as an integral part of any energy conversion process [39, 50]. However, the biological world has the capacity for self-improvement and for millions of years could develop an ideal variant of energy transfer from one reaction to another, i.e. with high efficiency. Many biologists do not always take into account this circumstance and consideration the thermodynamic principle on the low efficiency of energy transformations as a universal phenomenon, including the biological sphere. However, by the present time a number of data have been accumulated which do not agree with this situation [3, 46].

It must also be taken into account that maintaining of the endothermic state costs the organism an enormous portion (above 70%) of the metabolism. To provide such a large flow of energy nature could invent a separate cellular system in the form of high permeable mitochondria. This factor could have a progressive significance for the cell.

For the future, new methodological approaches are being introduced that will allow us to analyze the controversial issues that can arise in this direction [48, 50, 93].

3. Conclusions

Issues of biological thermogenesis are considered in various processes based on modern estimations. In tissues, however, these processes occur near the equilibrium state, and evaluation of their thermal effect needs correction from thermodynamic positions.

The mitochondrial side of this issue is considered in more detail, where some new details of the proton leakage mechanism are described as the main thermogenic system of a biological cell.

Analysis of a proton leakage problem is begun with the long-known question - why oxidation of succinate occurs with less coupling than NAD-dependent substrates in mitochondria. This analysis leads to the need to modify the existing proton leakage model. According to available data, in the cells of warm-blooded (mammals and birds), there are two mitochondrial subpopulations. One of them is common and synthesizes ATP. Another subpopulation of mitochondria, that have UCPs in the inner membrane, have a

high membrane permeability to oxidation substrates, NADH, nucleotides and are regulated by GDP. The nativeness of uncoupled mitochondria is shown by studying of isolated cardiocytes respiration using different substrates and inhibitors.

Thus, in this review, the problem of low efficiency of biological processes is critically analyzed, and a new consideration is proposed on mitochondrial thermogenesis for maintaining of warm-bloodedness of an organism.

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