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LIFE AND SCIENCE

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Childhood. University. First Steps in Science

Many of my predecessors in this section of the Reviews of Biochemistry, the prefatory chapters, must have experienced the same difficulty that I feel confronting me now. One has to steer between Scylla and Charybdis—find the correct way between giving a scientific review article, summarizing the scientific contributions, and giving a strictly autobiographical sketch of the events that constituted one's life course, without overloading it with scientific details. I have tried my best to avoid the two extremes. I will be happy if I have succeeded in achieving the necessary blend.

I was born in 1894 in Moscow, not in my parent's home, but in the obstetric department of Moscow's Central Children's Hospital, where my father temporarily worked. Two months later I was taken to Jaroslavl, a district town on the Volga, where our permanent home was. Here I spent my childhood and college years, till the age of 25, so that I regard Jaroslavl as being practically my home town. As though I had some indirectly introduced genes relating me to medicine, I kept in touch with this sphere for a considerable time, in different ways, besides being born in medical surroundings. My grandfather on my mother's side was the chief surgeon and head of the local hospital of Jaroslavl. My father was the head of the department of obstetrics and gynecology of the same hospital. After college

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I entered the medical faculty of Moscow University, graduated as an MD and served two years as a military doctor. My first scientific appointment was in the Biochemical Institute of the Commissariat of Public Health, my first professorship was the chair of Biochemistry of the medical faculty of Kazan University (later Medical Institute), and my first academic nomination was as a member of the newly formed Academy of Medical Sciences of the USSR. But looking back at my life's course, I cannot remember a single case where I healed a person of any disease. The effect of medical genes seems to continue into the next generation: my younger daughter works as a scientist in the laboratory of the Oncological Center, Moscow, and one of my granddaughters is a postgraduate at the chair of normal and pathological neuropsychology of Moscow State University.

Perhaps the first signs of my inclination toward a scientific profession became perceptible at a fairly early stage. It so happened that during primary school my classmates, fond of giving nicknames, as probably all boys in the world do, honored me with the title "Wolodya-Outcheny," which means scientist, Wolodya being the diminutive of Wladimir. This probably was due to my liking for playing around with all kinds of simple apparatus—electric bells, primitive toys—and gadgets such as those that show the attraction of light objects to a rubbed haircomb or glass-rod, etc. I even recollect my first "invention:" a small test tube with a light bead of elder-marrow in it and two wires attached to the ends of the tube. I declared that this apparatus could serve as a "cunny-scope," showing the degree of cunning of a person, according to the displacement of the bead when the wires were applied to definite parts of the body. Needless to say that the effect was produced by dexterously rubbing one end of the tube with a piece of silk cloth by which the "apparatus" was held. Thus I started my scientific career as a quack. I hope that my later behaviour has amended this regrettable qualification.

The next landmark of my career was in the field of chemistry, in which I became interested in college. As every beginner I was attracted by the handling of explosive materials. Nitrogen iodide was the object of choice, for when dry it explodes when even slightly touched. I carried a piece, damp and therefore safe, the size of a wheat-corn, to enlighten my classmates. But I dropped the damned stuff near the teacher's pulpit during the lesson on orthodox religion. At the very end of the lesson, the boy in charge of pronouncing the closing prayer stepped on the ill-fated piece, producing no harm but great noise.

It was a period when attempts on the life of tzars and members of the royal family were fashionable, so the priest who gave us the lesson rushed out of the classroom to the headmaster's study shouting: "an attempt has been made on my life . . ." Efforts by my parents were necessary to prevent my being expelled from the school. But not the highest mark for conduct stood in my final diploma which I had to present when applying for entrance to the Petersburg Polytechnical School where I intended to study electrical engineering. I did not pass the competition. This was a blow to my expectations. As a matter of fact it seems that the ominous chemical experience diverted my interests from chemistry to the physical field, particularly electricity. Still at college, with no help or advice from teachers, I constructed entirely from scrap collected on the market a complete radio set, transmitter, and receiver, with a crystal detector or coherer of iron filings; it exchanged signals all through our flat, passing closed doors and walls, to the bewilderment and great admiration of my parents. A high-frequency Tesla transformer followed, with mirror galvanometer, thermoelectric battery etc. I even published in a semipopular journal, a small article describing the replacement of the usual vibration interrupter of a small Rumcorf coil by a miniature mercury switch, which doubled the efficiency. That was my first scientific publication!

Having missed the Polytechnical School, I entered the mathematical faculty of Moscow University, where the requirements were less rigid, expecting that a good knowledge of mathematics would certainly be useful if I continued as an engineer. At school I was regarded as fairly good in mathematics, but a few months sufficed to destroy this illusion. I felt completely incapable of grasping the very first principles of higher mathematics, exposed by the eminent professor Lusin. After one completely fruitless term, I started to attend courses in chemistry and even passed one of the examinations, but changed my mind once more and finally settled down as a student of the faculty of medicine.

I hardly attended the lectures or the practical courses, but spent all my days working in the biochemical laboratories of a few different chairs. How I managed to obtain my medical diploma remains a mystery to me. In the evenings I attended lectures of a more general biological character, delivered at the Popular Shanyavsky University on immunology, and especially on physico-chemical biology, by professor Nikolai Koltzov.

My true inclination developed rapidly: the study of biological phenomena by means of exact sciences such as chemistry and physics. Perhaps this was the result of two fascinating books recommended by Koltzov, and of his brilliant lectures. The books were by Fisher: *Oedema* and *Nephritis.* I not only studied in the laboratory the uptake of water by tissue slices, but during my summer vacations, when I worked as volunteer in a small hospital in Jaroslavl, I tried to apply my modest knowledge to the treatment of renal diseases. Unfortunately the results were not exactly favorable for the patients and I was severely forbidden to continue my experiments on humans.

At home, electrical apparatus gradually became replaced by a self-made thermostat, heated by a kerosene lamp, and dangerous explosives gave place to harmless test tubes with protozoa or yeast suspensions. I succeeded in demonstrating that a mistake was present in Koltzov's conclusion on the nature of the influence of pH on phagocytosis by a unicellular organism. The effect was measured by the rate of uptake of India-ink suspension, and I established that changes of the electrical charge of the lamp-black particles were responsible for the changes observed and not the behavior of the organisms. When I reproduced an experiment at Koltzov's laboratory and told him about my explanation, he was quite excited and said that this was the correct way to tackle an experimental problem, not to be afraid of disagreeing even with authorities.

Civil war ravaged the country. The first socialist state stood alone, deprived of any external supply, against the adherents of the broken-down tzarist regime, the so-called "white forces" supported by armies of many western countries. It was a struggle of ideas against military force, and the victory was at last won by the ideas.

Immediately after graduating from the University I was called for military service as a doctor, and spent two years on the Southern front as head of a field hospital of a cavalry division. I went from the Don to the Crimea, and ended in the Caucasus, after the explusion of the English occupation forces.

In those years the war was as much against typhoid fever as against military forces—"war against lice"—and medical personnel were the first to fall victims. By pure wonder I escaped the fatal end. I withstood a severe infection under most unfavorable conditions: on a stretcher in an unheated wagon, during retreat before an advancing White-Cossak army. A good friend of mine kept me alive with huge doses of camphor, the only drug available, through a week of unconsciousness. After a month's recovery I was again on active service, now, fortunately, having become immune.

In 1921 the civil war came practically to an end, I was dismissed from military service, returned to Moscow, and from that time on began my scientific career.

By happy chance I was admitted to the newly organized Biochemical Institute of Public Health. I wonder whether there was not some kind of favorable influence from Koltzov, who was at that time director of another, closely related neighboring institute. Our Institute was under professor Alexei Bach, an eminent chemist and political figure. He was well-known for his research on the enzymatic mechanisms of biological oxidation, where a central role was ascribed to the formation of peroxides as the initial, all-important step. Bach's attitude even when dealing with young beginners, was not to "lead by the hand" by everyday instructions, but to give first of all general advice, indicating the main directions in which to proceed. At the beginning, Bach suggested that I study the properties of immune antienzymes, particularly the antiphenolases. One part of Bach's advice I have always remembered. After having done reasonable work on immune antibodies against enzynes (antienzymes) I once told Bach that I had a theory concerning the nature of immunity. What did he think about my developing it? "Dear boy," he told me, "if I were paid for producing theories, I could sit all my life inventing new and better ones. Good theories come from good facts. You had better turn to your bench, to your bench, my boy."

I have never had regular training in biochemistry and related subjects. In this respect, in scientific education and studies, I have to regard myself as a self-made investigator, not having spent any time in a traditional school. Of course I have been influenced by scientists of the older generation, but by more or less fragmentary contacts rather than in a systematic fashion. More exactly, there was just one very brief period when, after several years of work at the Biochemical Institute, I spent in 1927 about two months in Peter Rona's laboratory at the Charité hospital in Berlin. Here again the atmosphere was extremely liberal. Everyone could choose the subject on which he preferred to work; Rona himself was always ready to render assistance, but no strict program was ever prescribed. His lectures were excellent, meticulously prepared and accompanied by impressive experiments. Here I became familiar with the peculiar German way of expressing admiration during some especially intricate lecture demonstration: it was by stamping the feet. I regarded it as a special kind of applause by the lower extremities, the hands being busy taking notes or drawing sketches of the apparatus.

Rona's laboratory had a worldwide reputation. I even remember a jocular interpretation of the abbreviation, FRS, as meaning "Frühere Rona's Schüler." It was a kind of breeding pool from which many future excellent researchers emerged. For many of them it was the way to a scientific paradise—some of the Kaiser-Wilhelm Institutes, the KWI as they were known. Future Nobel prize winners were among the students there, as Fritz Lipmann, Hans Krebs, Max Perutz, E. B. Chain. There one could make close aquaintance with a broad diversity of interesting and able people. I remember with great pleasure working at the same bench, side by side, with David Nachmansohn; on the upper floor Hans Weber was working on muscle proteins. Rona arranged visits to KWI for me to meet Carl Neuberg, Otto Warburg, Karl Lohmann, and Otto Meyerhof. Some of these contacts, even if initially of short duration, grew later into life-long friendships.

The work on antienzymes did not bring results of any importance from the point of view of giving new insight into the nature of enzymes or their action. But it led to a discovery, of minor importance by itself, but which nevertheless appeared later to contain elements of broader significance. I have in mind the use of what I later called "the principle of fixed partner" (1). The starting point were observations that showed that immune antibodies can produce their reaction with the antigen even when transferred from solution into a heterogeneous state, in adsorbed form on some suitable carrier such as kaolin or aluminum hydroxide. The use of this principle permitted establishment of the antigenic properties of haemoglobin. Under usual conditions, immunization with this protein did not produce antibodies; if examined in ordinary solution no precipitation or changes in its specific, oxygen-binding properties were observed. But if the serum of an animal immunized with haemoglobin was adsorbed on a colloidal carrier, this suspension had the remarkable property of binding hemoglobin and removing it from solution. Similar conditions were observed in experiments with yeast invertase. Whereas other enzymes, such as oxidases, produced antibodies that inhibited their enzymatic activity, invertase seemed to be devoid of antigenic properties. But here again, when the serum from an immunized animal was used in the form of an adsorbate, in the fixed state, complete removal of the enzyme from solution resulted after centrifugation. It was easy to show that in this bound state, after reaction with the "fixed partner," the enzyme still exhibited its original catalytic action. These experiments might be regarded as precursors of the now so popular use of "immobilized" forms of enzymes or other biologically active substances.

Discovery of Oxidative Phosphorylation

Immunological studies, attractive as they were, seemed at the time to have led to a kind of blind alley. My interest shifted to the study of metabolic processes involving phosphoric acid, which in the meantime had become a central point of significance. Its participation in anaerobic carbohydrate metabolism attracted the greatest interest, due to the fundamental work of Embden, Lohmann, Meyerhof, and Parnas after the breakthrough discoveries of Harden and Young.

In 1929 I accepted the invitation from the Kazan University to occupy the chair of biochemistry. The laboratory had to be organized completely anew, since it lacked even the simplest modern equipment. All I possessed for my personal work after a year of effort was a poor imitation of a Warburg respirometer, made in the university's modest workshop, and the simplest kind of colorimeter, an "Authenrieth" wedge apparatus, only later replaced by a small Duboscq model. But the teaching took very little of my time; I had plenty of it for working on my modest bench and for meditation.

The results were highly satisfying—the discovery, that respiration in cells can produce a synthesis of ATP. At that time it was well known that ATP is synthesized in the course of anoxidative breakdown of glucose, as a result of fermentation and glycolysis. In retrospect it appears almost impossible that nothing whatever was known about a possible participation of ATP (or of phosphate in general) in the processes of the other great predominant source of energy, namely respiration. The explanation is simple: it was the lack of appropriate experimental objects. A very limited number of biological objects served at that time as materials of choice for the study of the two fundamental sources of energy in living objects: fermentation and respiration. They were yeast, on the one hand, and muscle and liver tissue on the other. Experimenters neglected the valuable advice given by the remarkable biologist, Dr. Krogh, in his address to one of the Physiological Congresses. He said that Nature has been generous toward naturalists, by creating some special object particularly suited for the study of each of the more important problems. The condition of success for a scientist attacking a new problem is to find and use the appropriate object.

As a paradox, the first indication of participation of ATP in the processes of cellular respiration came not from experiments where respiration proceeded, but from those where it was absent.

By lucky chance it so happened that I had chosen the kind of cells for my study that might be regarded as specially suited for investigating the possible participation of ATP in respiratory processes. These were the nucleated avian blood cells. Their structure is of the simplest kind: they have only one strictly defined function to fulfill, i.e. to keep in a closed space a highly concentrated solution of hemoglobin, in which swims a large nucleus. Evidently, thanks to the latter feature, they possess a very intense respiration, in contrast to the non-nucleated mammalian erythrocytes. And most important, they have a very high content of ATP, of almost exactly the same order as muscle tissues.

This ATP content remains practically constant as long as respiration goes on. But as soon as the latter is interrupted, by cyanide poisoning or removal of oxygen by repeated evacuation and flushing with pure nitrogen, rapid dephosphorylation of ATP is observed, accompanied by a corresponding increase of inorganic phosphate. Within an hour at 37° the acid-labile phosphate has disappeared, hydrolyzed to inorganic phosphate. How was the stability of the ATP content under aerobic conditions to be interpreted? Obviously, the choice had to be made between two explanations. Either the splitting of ATP was stopped by oxidative conditions, or it did proceed, but was compensated for by a steadily proceeding reverse process: reesterification of the inorganic phosphate that had been set free.

The preference was decidedly the second alternative, which assumes a continuing "phosphate cycle," for no example was known of an inhibition of hydrolytic processes by oxygen. This would have remained an indirect conclusion, were it not unambiguously supported by the direct demonstration of esterification of inorganic phosphate, formed during a period of anaerobiosis, after reestablishment of aerobic conditions. Thus aerobic esterification of inorganic phosphate, with formation of ATP was proved beyond any doubt (2, 2a). I called the process "respiratory resynthesis" of ATP; it corresponds to the nowadays generally accepted term oxidative phosphorylation.

The experiments permitted at least a rough quantitative evaluation of the efficiency of the process, represented in the form of the P/O quotient. This value was found to be about 1.0, of the order of magnitude established later in numerous cases.

With the discovery of oxidative phosphorylation, ATP was immediately raised to a pivotal role in bioenergetics, especially when very soon the work of Wladimir Belitzer, and of Herman Kalckar established the important fact that not only the primary attack on the sugar molecule was accompanied by the binding of inorganic phosphate, but also the oxidation of several of the intermediates in the long chain of steps leading to the oxidative breakdown of hexose. It became commonplace to regard ATP as the storage form, the energetic currency by which the energy produced by fermentation and respiration is made available for fulfillment of all physiological functions. The stored chemical energy is set free by the action of the corresponding enzyme, the ATPase.

A peculiar property of ATPase should be mentioned here, which became apparent during our further studies of the ATP metabolism of nucleated blood cells. In the absence of respiratory resynthesis, their ATP content is hydrolyzed within about one hour. This means that the ATPase activity is small. But if the cells are hemolyzed, the ATP is split almost instantly, in an explosive way. When my collaborator Tatiana Venkstern and I studied this peculiar behaviour (3) it appeared that there were two kinds of ATPase activity. The smaller part is present in the internal contents of the cell, and is responsible for the slow breakdown or turnover of ATP in normal, intact blood cells. The other, many times larger part, is firmly localized on the exterior surface of the cells and its activity is directed outwards. It is this latter ATPase—we called it "ecto-ATPase"—that splits the ATP of the cells when they are lyzed. A simple experiment shows its presence. If we add some ATP to a suspension of avian bloodcells in normal saline, all the added triphosphate is rapidly hydrolyzed. But the inner content of ATP remains completely unattacked at the initial, normal level. Circumstances did not permit us to continue this study. The biological role of the ecto-ATPase represents an intriguing mystery. We have to suppose that the enzyme never meets its substrate, for there is no measurable amount of ATP in the blood plasma. Perhaps it is a peculiar ontogenetic relic from the process of erythropoiesis?

Nature of the Pasteur Effect

Cellular respiration may be regarded as possessing a dual role (7). Oxidative phosphorylation is obviously its most important manifestation, a powerful generator of high-energy phosphate bonds that serve as the immediate source of chemical energy for all physiological functions. The other role is of, so to say, vicarial significance, of less direct character. This function is represented by the so-called Pasteur effect. It governs the interrelation between respiratory and anaerobic (glycolytic or fermentative) metabolism. Under aerobic conditions the wasteful fermentative breakdown of the carbohydrate is suppressed. Thus through the action of the Pasteur effect the fate of a hexose molecule becomes decided—whether it will follow the respiratory or the fermentative pathway.

It was natural that my interest was attracted by this, at the time, hardly explored problem—the nature of the Pasteur effect. The most plausible explanation of its mechanism was to assume that the suppression of fermentation under aerobic conditions was due to the oxidative inactivation of some member of the enzymatic machinery of fermentation and glycolysis. The leading idea of our approach, which I undertook in collaboration with my postgraduate student Nikolai Sakov, was to investigate the sensitivity toward oxidation of the different enzymes that take part at the first stages of the anaerobic breakdown of glucose.

Accordingly, the effect of redox dyes of different redox potential was tested on the enzymes responsible for the initial stages of the glycolytic pathway.

Of the several enzymes tested (hexokinase, isomerase, aldolase etc) all, with one exception, showed complete insensitivity toward the redox dyes over the whole range of concentration or E'_0 values. The only exception, significantly, showed a striking difference. This was the enzyme, phosphofructokinase, which leads to the formation of the starting point of fermentation, hexose-1,6-diphosphate, by transferring a phosphate residue from ATP onto the preceding stage, fructose-6-phosphate.

Phosphofructokinase appeared to be highly sensitive to the action of redox dyes, with positive potential within the range of 0.05 and 0.250 V. Complete inhibition was obtained also with a variety of oxidizing agents: iodine, quinine, hydrogen peroxide, dehydroascorbic acid etc. All the reduced forms were without effect.

Evidently, the effect of these agents, completely alien to the normal catalytic system of the cell, even if highly suggestive, was only of an indirect kind. But an impressive proof of the validity of the findings was obtained when an exactly similar effect was found using the major physiological oxidizing system, cytochrome and its oxidase. In the presence of a suitable intermediate carrier, oxidized cytochrome by itself taken in stoichiometric amount, inhibited the phosphofructokinase. But, most important, the inhibition could be obtained with minute, catalytic amounts of cytochrome in the presence of cytochrome oxidase. In air, almost complete inhibition is observed, whereas in nitrogen no inhibition occurs. This experiment can well be regarded as the closest modeling of the Pasteur effect under the most simplified conditions.

We have to regard hexose-6-monophosphate as the turning point at which the further fate of the hexose molecule is decided. If the first C atom is phosphorylated, with formation of the Harden-Young ester, the molecule is predestined to be split into two equal parts, the trioses, and then to follow a series of fermentation steps; we called this the dichotomic pathway. If on the contrary, the first carbon atom, instead of being phosphorylated is oxidized, hexose enters the oxidative pathway, which may proceed by consecutive splitting off of single-carbon products; we called this the apotomic pathway.

Sakov was perhaps the most brilliant of my pupils; he had a keen mind and was an excellent experimenter. His lot was tragic. The work on the Pasteur effect had a deeply regretful issue. The experiments were finished in the spring of 1941. The war broke out; Sakov was soon called to military service. My family and I were evacuated from Moscow to Frunze, the capital of the Kazakh republic in Central Asia. I carried the laboratory protocols and notebooks with me, waiting for news from Sakov. They came only after a long silence, and were sad. A note from the armed forces informed me that Sakov had perished on the battlefields of Stalingrad.

The paper was published posthumously. There was no possibility of sending it to a foreign journal, and it appeared only in the Russian journal *Biokhimiya* (4) and thus remained almost completely unknown, except that some of the initial stages of this research, already showing the important points, were mentioned in a few lines in an article by Dean Burk in the Cold Spring Harbor Annual Report (5). The interpretation we gave to the mechanism of the Pasteur effect was "rediscovered" after exactly twenty years, by Janet Passoneau and O. Lowry, and published under the title "*Phosphofructokinase and the Pasteur effect* (6). I reported the work at a conference convened in Paris in 1946, in commemoration of the fiftieth anniversary of Pasteur's death. Apparently the Proceedings remained unpublished; at least I received neither reprints nor information about publication. With a lapse of over thirty years I gave an account of the main results at a conference on bioenergetics, held at the American Academy of Arts and Sciences, in Boston, in 1973 (7).

Myosin and Adenosinetriphosphatase

Obviously, muscle is the most appropriate object for the study of problems of bioenergetics. With the discovery by Einar Lundsgaard of the "alactacid contraction" it became clear that the immediate source of energy for the work of muscle is the splitting of ATP and that consequently, great importance should be attributed to the enzyme that catalyzes this splitting, adenosinetriphosphatase.

There was every reason to regard this enzyme as fulfilling a key role in the function of muscle and to study its properties. But, strangely enough, this did not happen for a considerable time. The reason was peculiar—no ATP-splitting activity was found in aqueous extracts of muscle tissue. And it was these extracts that were the favorite material of muscle biochemists. Intense work was proceeding, directed toward obtaining the different enzymes that constitute the glycolytic complex, in soluble form. "Water-soluble enzymes" formed a distinguished group; the discovery of a new enzyme belonging to it was the predominant aim of experimental efforts, a kind of hypnotic spell. But adenosinetriphosphatase was not found in these extracts and remained neglected, an enzymological Cinderella, although it was known that if ATP is added to minced muscle it is rapidly split. But the extracted residue from which all the water-soluble enzymes have been removed was generally regarded as presenting no further interest and was discarded.

In collaboration with my wife, Militza Nikolaevna Lyubimova, my former postgraduate student, we undertook the study of the apparently evasive ATP-splitting activity. We turned our attention to the residue that remains after the extraction of the water-soluble enzymes. The very first experiment, prosaic in its simplicity, brought the unequivocal answer; an extremely high enzymatic activity exists in the "insoluble" part of muscle tissue, after the rather mild conditions of aqueous or saline extraction that are usually applied.

At this stage our merit was modest. Figuratively speaking, we simply took out from the waste-bucket the stuff that others rejected as not deserving attention. Much more significant was the next step, when we tried to isolate the ATP-splitting activity, that is, to separate the enzyme from the different water-insoluble proteins to which the function of contractility belongs.

Having once violated the canons by studying the residue instead of the extract, we continued to move along a heretical way, by using, instead of water or saline, concentrated salt solutions, which were known to extract the main contractile protein, myosin. It was natural to start with the removal of this protein, which was known to be the preponderant component of the insoluble protein fraction. Great was our astonishment, when after treating the residue with solutions of higher ionic strength, as used for the isolation of myosin, we found the full enzymatic activity present in the myosin-containing extract. This result was exactly opposite to our expectations. We intended to remove the bulk of "structural" protein and find the enzyme in some minute, individual fraction. On the contrary, the whole activity was found in the myosin fraction itself; all methods known at that time for the isolation of myosin invariably yielded products carrying the whole enzymatic activity. Moreover, the well-known great heat lability of myosin was found to be characteristic also for the enzymatic properties. Unexpected as it was, and apparently unlikely, the conclusion had to be drawn that the enzymatic, adenosinetriphosphatase activity belonged to myosin itself.

In our first publication in *Nature* (8) we ventured to introduce an abbreviated name for the enzymatic property of myosin, calling it ATPase instead of the cumbersome adenosinetriphosphatase, but the editors rejected it. By now it has won general acceptance and is used here.

Naturally, the fact that enzymatic properties were ascribed to a highly specialized protein that carries out structural and mechanical functions and constitutes a large percentage of the dry weight of our body, seemed highly improbable. No wonder that, regardless of strong experimental evidence, certain sceptical authors, some very eminent, raised objections and expressed strong doubts. But all these objections could easily be shown to be untenable and the initial statement remained unaltered. We had no reason to complain on the verdict of the most exacting judge—time. At present even the precise localization of the catalytic activity within the molecule of myosin is known.

The importance of the fact that myosin possesses ATPase activity needs hardly to be stressed. It means that the structural contractile substance that performs the mechanical work of muscle, itself provides the moving force for this work, by splitting ATP and liberating its chemical energy.

One can now wonder how it happened, that for such a long time the study of muscle proceeded by two completely independent, separate lines. On the one hand was the study of the building material of muscle, the proteins, which constitute the physical basis of the living machine. On the other hand was the study of chemical processes and the corresponding enzymes, which provide the moving force for the accomplishment of mechanical work. The finding that chemical and mechanical properties are combined in myosin, the establishing of its ATPase properties, served to fill out the gap that separated the two approaches, introduced a certain kind of unity, and replaced the former dualism.

But this was not the whole story. A new aspect, of not minor importance, was introduced when it appeared that interaction of ATP and myosin is of a bivalent, reciprocal character, going beyond the simple relation between an enzyme and its substrate. Namely, it was found that ATP changes the physical properties of myosin, for which there was every reason to expect some fundamental role in the mechanical effects involved in muscular contraction. Work on this line started almost simultaneously in two places, by the group of Joseph Needham in Cambridge and in our laboratory.

Evidently, the motivation was the same in both groups: if myosin acts on ATP and liberates its energy, does not ATP in its turn act on myosin, and produce changes in its physical properties that could play a role in the performance of work?

Needham and co-workers studied the physical properties (i.e. the viscosity and flow birefringence) of myosin solutions (9). In both cases addition of ATP produced strong effects, which disappeared as soon as the added ATP became split by the ATPase action of myosin. This clearly demonstrated the bilateral character of the ATP-myosin interaction as observed at or near the molecular level. Needham aptly named myosin a contractile enzyme (10).

I started with experiments with monomolecular layers of myosin, using a Langmuir trough with a rotating disc in the plane of the surface. The measurements of the viscosity of the monolayer were very impressive, and I remember the excitement of Needham, when on a visit to the USSR, he came to my laboratory and I demonstrated the apparatus (again, as in younger years, self-made partly from scrap) to him. But the effects of ATP were poorly reproducible, and I soon abandoned this approach, switching over to an object of higher structural order, myosin threads. These can easily be prepared by squirting a concentrated myosin solution into distilled water. They possess a small, but measurable degree of strength that can be registered by a torsion balance.

ATP has a well-reproducible, strong effect that greatly changes the extensibility of the threads. This appeared to depend on the ATPase activity: after treatment of the threads with very dilute silver nitrate solution, which completely inhibits ATPase activity, no effect of ATP on the mechanical properties of the thread could be observed (11).

These observations of mine were even reflected in an auto-epigram, which I attached to an amiable cartoon drawn by a friend of mine on the occasion of the award of the State Prize, which my wife and I received for our work on myosin. The drawing represented my wife holding onto my shoulders, hanging from a thin thread, which grows, downwards, into a solid rope.

The epigram, originally in Russian, and later translated by me into English, ran thus:

Those who envy will say: "on a thinnest thread
Holds the fame of this new laureate."
And I pray to the Almighty: "My only hope
Is that the myosin thread be as strong as a rope."

The subsequent discovery, by Bruno Straub in Hungary, of the second muscle protein, actin, intimately associated with myosin, brought new details to this field. Later investigations in our laboratory and others of spermatozoa, ciliae of unicellular organisms, protoplasmic flow, etc, have led to the conclusion that similar fundamental principles apparently hold true for biological motility in general. These experiments led us to formulate the principle of mechanochemistry of muscle (and other motile objects). Its essence is the assumption of a reciprocal interrelation between the source of chemical energy, ATP, on the one hand, and the basic component of contractile structures, myosin (or actomyosin, or myosin-like proteins), on the other. ATP provides the energy, and at the same time alters the mechanical properties of the contractile protein, myosin. The latter by its catalytic, enzymatic property, liberates the energy of the high-energy bonds of ATP, and at the same time undergoes changes of its physical state, which are responsible for the production of mechanical work.

Many years earlier Meyerhof gave a very concise formulation, based on clear thermodynamic considerations, of a principle well-known and almost obvious, which must be expected to be operative in a machine where transformation of energy takes place. These are his words: "In any machine, if it is not a heat engine, the energy-producing chemical process must interfere with the structural base of the machine, in order to produce the changes which are the source of the work performed." Is it not astonishing, that this succinct statement was not applied, for more than a decade, to the phenomenon of muscle movement? The discovery of the ATPase properties of myosin provided an excellent place to test the validity of the above-mentioned principle.

Administrative Work

Years went on, and duties unrelated to research work, accumulated, of an organizational, administrative, and social character. Managing human affairs, even of a scientific nature, required increasing amounts of time that couldn't be compensated for by more work at the bench, as had been my custom. In all the investigations mentioned above I took an active part by experimenting with my own hands.

It is to Archibald Hill that I owe my introduction into the attractive, but time-consuming engagement in scientific public affairs. It was during his presidency of the International Council of Scientific Unions (ICSU) that I was invited to become a member of its Bureau, and in later years Vice-President. This gave me fascinating opportunities to meet and often come in close contact with a great variety of scientists, both in my own field of biochemistry, and in the worldwide scientific community represented in this international organization. I remember with great affection my years with the ICSU. After Hill, Rudolf Peters took over the presidency and his unforgettable charm remains with me over the years. He masterfully managed to keep an excellent atmosphere during the meetings of the ICSU Bureau, despite the sharp controversies that sometimes arose. I recollect vividly the clash of opinions between myself and Lloyd Berkner, of the USA, when I strongly opposed what I considered to be his intentions to make ICSU dependent on "Big Business," as represented by various American trusts and foundations. I was happy when my point of view became adopted and the really international and independent character of ICSU was supported by the other members. The wise leadership of Peters helped to keep up the fruitful, friendly collaboration that always permeated our work.

With modesty I feel that I can attribute to my efforts an important result achieved during these years: the acceptance of the German Democratic Republic as an ICSU member. The opposition was tacit but strong, and my struggle lasted long; it seems that I exhibited unexpected diplomatic abilities. Anyhow the result was achieved, to my great satisfaction. It was the first time that the GDR, of the socialist fraternity, became a member of an international, powerful organization.

My experimental work came to a complete standstill when the Academy of Sciences of the USSR assigned me a difficult charge relating to its diverse biological research institutes, by appointing me to the post of Academic Secretary in Biology. These were the years when Lyssenko was still in full power, and the situation of biological science within the Academy and in the country as a whole was, in many respects, in a highly unfavorable state, to say the least. For several years I carried out my duties as best I could, but evidently did not fulfill expectations. I was finally dismissed.

An excellent recompense for my dismissal was given me in charging me to organize a new institute for the study of the physical and chemical basis of life phenomena. The duties of Director of this Institute of Molecular Biology I continue to carry out. The Director's study and writing desk have replaced my laboratory bench; a skillful secretary helps me instead of a talented collaborator. I do what I can to keep abreast of the work going on in several teams of younger and middle-aged personnel of the Institute, and to provide the necessary material and human conditions for carrying out their research and to ensure a favorable atmosphere of friendship, collaboration, and mutual respect.

The organization of our Institute marked a turning point in the development of physico-chemical biology in our country. It was the first and only center of its kind at that time. We were successful in attracting a number of able chemists; biologists were easier to find, and physicists were fewer in number, but very eager to bring all the help necessary. My leading idea was to have something like a 30:30:30 ratio of chemists, physicists and biologists. We had to start from practically zero level, but the enthusiasm was great and the support from the Academy generous. The Institute organized the publication of a large series of monographs on most important aspects of molecular biology. It was a great relief to me and a happy chance for the growth and successive development of the Institute that a considerable number of excellent scientists of the intermediate and younger generations joined the staff at the early stages of its existence. Particularly fruitful was the renewed partnership with my former pupils, some of whom, as for instance Alexander Bayev or Alexander Braunstein, had been among my postgraduate students in a rather remote past, and now formed the backbone of the newly created Institute. We owed to Bayev and his group the first outstanding success in the field of nucleic acid study. It resulted in establishing the full primary structure of the second nucleic acid to become known after the first achievement in this line, the work of Holley on alanine tRNA. Two years later Bayev's group gave the structure of the valine tRNA. Continuing his work at the Institute in the field of genetic engineering, Bayev is at present Past President of the International Union of Biochemistry, under the ICSU.

The formation of the Institute of Molecular Biology occurred first under another name: "Institute of Physico-Chemical and Radiation Biology." This was a kind of camouflage, because at that time the very name molecular biology sounded somehow suspicious. Only after a few years, when the reputation of the Institute became well-established and the general situation changed for the better, did I ask our President of the Academy, M. Keldysh, on the occasion of an anniversary of some kind, whether he would like to make me a present. There was some anxiety in the expression on his face when he asked what I would particularly like to have. I told him: "Just to change the name of our Institute to what it should be." With evident relief he immediately agreed, and at the next meeting of the Academy's Presidium the matter was settled and the Institute obtained its present name.

The strained situation with even the concept of molecular biology appeared clearly during the preparation of the XVth International Congress of Biochemistry, which was held in Moscow in 1961. There was a section on problems of molecular biology, where Max Perutz and myself were the joint organizers. We intended to name it simply "molecular biology section." But when I proposed this at a meeting of the Moscow group of our organizing committee, strong objections were raised by the more orthodox members: "What is molecular biology? We do not know such a science or branch." To satisfy my opponents (I was in a minority of one!) I invented a euphemistic name, "biological functions at the molecular level." This was accepted.

The Institute is now twenty years of age. Affiliated with the institute is a "Scientific Council on problems of molecular biology." It consists of about 20 members, who represent the main centers of research in the field of molecular biology, which have grown in the Soviet Union during these last decades. At the annual meeting of the Council or of its Bureau the results obtained during the period are summed up, and plans for future activity, steps needed to avoid unnecessary duplication or to fill undesirable gaps in the treatment of studies on urgent problems that present special interest and importance are discussed. Special attention is paid to the effective planning of the publication of the corresponding scientific literature, the organization of reference editions etc. Of great significance for the progress of the young science was the organization of the specialized journal "Molecularnaja Biologia," where the scientific production of the country is published. The journal appears also in English translation in the USA.

In order not to break my bonds to concrete science, I engaged in a project named "Revertase," designed to develop on a broader base studies of the enzyme of the so-called reverse transcription. The project serves to coordinate and support research going on in research centers in Moscow, Kiev, Novosibirsk, Riga, and others in the USSR, and also in the German Democratic Republic, Czechoslovakia, Poland. Winter schools on various relevant topics are organized yearly by the Council in Moscow or other cities. Directing these schools gives me the refreshing opportunity to keep in closer contact with the youngest generation of scientists, on whom the future growth and development of our science depends.

Usually in biographical articles, even in shorter notes of a questionnaire type, some place is given to the topic of "Hobbies." I could mention only a single one, in my younger years, mountaineering. Mostly accompanied by my wife, who bravely shared the difficulties, we visited first the mountain ridges of the central Caucasus, later followed high passes and glaciers of the Pamirs, and then the mountain crests in Tien-Tchang, on the border of China. Fresh in my memory is the day, when, in company with the Swiss yachtswoman-writer-mountaineer, Ella Maillart, we stood on top of a steep ridge, over 4 thousand meters in altitude, which formed the frontier between the USSR and China. We stood with one foot on Soviet soil, the other on Chinese territory, with the hazy sky extending over the great desert of Takla Makan before us. Mountain friendships are among the strongest. I was happy when I was able to pay a visit to Ella at her chalet in a valley near the Matterhorn. I attended a conference in Arolla, the same Valais canton in Switzerland, and a friend, Guido Pontecorvo, arranged a trip by car to Chandolin, to meet Ella after a lapse of some forty years. Great was our joy!

One is well justified to regard this century as crucial for both my country and for the world as a whole. My fatherland has passed through several wars: the Russian-Japanese, when a vast but feeble empire was defeated by a small, almost negligible neighbor; the First World War, when the country was again defeated, but also liberated of the rotten tzarist regime; the civil war of the young new state against the remnants of the old regime, which were supported by armed forces of about a dozen interventionist and occupational countries: the English in the Crimea and the Caucasus, whose aim was the rich oilfields, The French in Archangel, the Japanese in the Far East, the Austrians Rumanians on the west front, and so on. And finally we suffered the terrible Second World War. We had two revolutions in 1905; that drowned in blood by Nicolas II, and the victorious Great October Revolution. This changed the world and created a new society in which, I believe, human values—brotherhood, social equality and freedom from exploitation—replaced the "anti-values" of previous generations—wealth among pauperism and profit as a dominant directive force of the whole social structure. In the new society pecuniary wealth has disappeared and has given place to social, spiritual, and intellectual values.

There have been years of great hardships and struggles, also of great triumphs. I think that both kinds of experiences helped me to develop my strong optimism. Recently my friends, cineasts, were showing a film "Faustian Tale," in which I took part. They asked me in their professional language whether, if presented with such a possibility, I would change the film of my life to something different. I replied that fate has been benevolent to me, and that I have no complaints with the scenario that it chose for me. My wish would only be for a prolongation of the lives of my mother, who died in middle age because of a street accident; and of my wife, my true and devoted life-companion, whom I lost a few years ago. For the rest I have no complaints; the reel could well be turned again.

A beautiful verse by our great lyricist, Tyutchev, is always in my mind. It is with his lines that I close my article. I could not find in Moscow an English edition of Tyutchev's poetry, and I am not a Marshak, the excellent translator of verse from English into Russian and vice versa. So I have to rely on the indulgence of my readers when I give a word for word unrhymed translation, knowing well that much of the charm of Tyutchev is lost:

> Blessed be those who visit our world
> At its crucial, fateful moments.
> They have been invited by the celestial powers
> To partake at their convivial feast.

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