# ACTA BIOLOGICA ACADEMIAE SCIENTIARUM HUNGARICAE

ADIUVANTIBUS

B. FALUDI, V. FRENYÓ, B. GYŐRFFY, J. SZENTÁGOTHAI, J. TIGYI

REDIGIT

# I. TÖRŐ

# SUPPLEMENTUM 6 (SUPPLEMENTUM AD TOMUM XV) PROCEEDINGS OF THE SIXTH MEETING OF THE HUNGARIAN BIOLOGICAL SOCIETY

Budapest, May 28-30, 1964

AKADÉMIAI KIADÓ, BUDAPEST

1964

ACTA BIOL. HUNG. SUPPL.

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# Abstracts of papers read at the sessions on the biology of algae

### THE ALGAE AND THEIR IMPORTANCE

#### T. HORTOBÁGYI

#### UNIVERSITY OF AGRICULTURAL SCIENCES GÖDÖLLŐ, HUNGARY

The energy of the photons, streaming to the Earth in the radiation of the sun, makes it possible to produce organic matter by means of the chlorophyll molecules in the living structure. In agriculture we aim at a photosynthetic assimilation of as high degree as possible on a unit area and within a time unit. In the past, all such investigations took their objects from the domain of the phanerogamic plants. In these last decades attention turned towards the investigation of the unicellular algae of very simple construction because one could experiment with them as intact individuals, as against the nesessarily isolated organs of the phanerogamic plants.

The production of more nutrients of better quality became a world problem since the life of the major part of mankind is abbreviated by starvation. If we want to improve this situation we must study more thoroughly the life processes of the organisms of especially advantageous productivity and the most useful possible fixation of the energy of the sun.

In the opinion of F. R. SEN, General Director of FAO, the following two decades will be of decisive importance in the solution of food problems. In this domain a real revolution must take place. In the relation of the whole world 1.75 times as much food will be needed in twenty years as compared with the present supplay. How can this be accomplished?

According to NITSHIPOROVICH, agricultural cultivation is conducted on a round 2500 million hectares; this ratio can not be improved substantially since the overwhelming part of the uncultivated areas is either too dry or too cold, or rocky, so hardly suited for agricultural production. It became evident that the traditional terrestrial farming is not capable, even at present, to produce the protein of necessary quality and quantity for the rapidly increasing human race. In the next future the inexhaustible protein richness of the oceans, the products of the fish ponds and the mass production of the algae will supply the basis of protein and other important organic matters for the nutrition of mankind for the feeding of domestic animals and to provide for industry.

According to recent investigations the beginning of terrestrial life can be put to ab. 3 milliard years ago. As we know today, on the lifeless Earth there was a reductional environment. The athmosphere of to-day is of oxidative character. This essential change must be attributed to the microorganisms, containing chlorophyll and its accompanying compunds and this is the point where we meet the first great role of the algae. The free oxygen of the air came into being by the vital activity of the green vegetation and it is also originating now from the water necessary for assimilation.

Algae remained until our days mainly the inhabitants of the waters, although some of their forms which tolerate temporal desiccation proceeded to the continents, even to the semi-deserts. In the waters the floating plants (plankton) and 90 per cent of the lit-oral flora can be found, until a depth of 40-50 metres, i.e. in the surface layers of the waters. The sea-shore vegetation is an especially significant plant mass. According to CHAPMAN the greatest depth to which the coloured algae can live is determined by the differences in the illumination of the seasons. Particularly the mass of blue, green and diatomic algae which lives in the soil till 50 cm depth, is no negligible factor.

One of the pillars of the production of organic matter, proceeding in the water, is the energy of the sun penetrating into the water; this is the energetical basis. The other pillar is the useful anorganic salt contents of the water; this is the chemical basis. The production of organic matter of the green plants in the waters depends on these two factors. It is the green plants in which both the cosmic energy of the sun carbon dioxide and nutrient salt supply of the environment is realized. All the aqueous organisms are in an immediate or indirect. connection with the algae or seaweeds which stand on the first scale of the feed chain. The energy gets from this  $\lambda_1$  energy level further, into the three levels of the animals storing the organic matter and energy: into the herbivorous animals ( $\lambda_2$ ); by means of these into the planctonovorous animals, first of all into the fish ( $\lambda_3$ ) and, at last, into the piscivorous fishes of pray of big size ( $\lambda_4$ ).

In our waters most of the living material (biomass) and energies are found in the plants. The unicellular algae with small dimensions and large active surfaces are most suitable for the full stocking of the available space, and for the production of greatest efficiency. The alga suspension utilizes the light energy better than does the terrestrial vegetation. So it is the unicellular algae from which we can get most organic matters and energies, and the second great significance of the algae consists in this. The organisms of small dimensions are of big surface as compared with their volume, therefore their metabolism is rapid. According to the data of FELFÖLDY, 1 litre of alga suspension containing 1.5 gr of dry material has got an active surface of 20 m<sup>3</sup>. Being of small size, they will quickly filled up with their organic compounds, and they can hardly store their assimilates in their tiny bodies. It is this fact which compels the algae to multiply very rapidly; thus the number of their individuals increases, con-sequently the production of organic matter keeps on growing. The production of the organic materials can be regarded as optimal if the number of the algae does not surpass the saturation degree. We must keep their number on the most favourable level in order to obtain a large production. The density of algae, the primary producers is influenced by the secondary producing beings of the waters which store the organic matter, i. e. the animals which consume the algae. It is the fish which stands at the end of the storing organisms, quasi-directing the material and energy circulation. The great importance of the storing animals consists in that they build further the organic matters produced by the algae, and they store and keep them in circulation for a long time and in a living condition protected from bacteria and fungi. So, by consuming the superfluous amount of the algae they make possible the undisturbed and continuous production of the organic matter and, at the same time, they retain the organic materials in their bodies.

Some environmental factors (the changes of the meteorological and chemical conditions, the rise of water temperature etc.), can ensure optimal life conditions in the shallow lakes abundant in nutrients for one or more alga species, so they can appear in the biotops with an extraordinarily high, "explosion-like" number. The "water-blooms" which cause the conspicuous coloration of the water are unpleasant phenomena of the fish ponds accompanied by major minor destruction of fish as a rule. The rapid multiplication of the algae, viz., goes together with the increasing utilization of nutrients. Concurrently with this, the light conditions of the water suffer a change. Near to the surface self-shadowing takes place. Eventually the assimilation of the algae, settled in deeper regions, may cease, and they produce carbon dioxide only. The water gets contaminated which is still aggravated by the important destruction of individuals and by the poisoning products of decomposition. The algae may block up the gills of the fish. The sedimented destroyed organisms fully cover the roes of fish. All this results in a mass decay of fish which, according to the studies of Rezső Vámos, is part of a bigger complex of phenomena.

It is especially the blue algae which cause a very dangerous "water-flowering" because they can excrete certain toxic materials as the studies of GRAHAM, BENNETT, SHILO, PANKOV, GORHAM et al. inform us.

The experiments, directed to the utilization of the "water-blooms" are very remarkable On the Indian Algological Symposion M. T. PHILIPOSE rendered account of artificially induced waterflowerings. A similar communication was made by DAVIS. By means of bringing forth special algal-flowerings at a proper measure, incalculable chances are before us in the domain of the increased utilization of fish ponds.

It is the utilization of the sewage water for algal production and the use of algae for the cleaning of the sewage water that is more and more stressed in these last years. The sewage waters, as a consequence of the extraction of organic compounds by the algae, and the oxygen produced by them, become clearer, and the algae readily multiply in the sewage water. So we can achieve a double utilization, a hygienic and an economic one. The third great importance of the algae appears in the cleaning of the sewage water. According to the data of LIEBMANN, if we dilute the sewage water, cleared with sedimentation, by pure water 3-6 times and then conduct it into the fish pond, it suffers a load of 2000 persons per ha and it will be cleared to a good efficiency degree. The ponds of sewage water are suited for cleaning the sewage water of the households and the food industry. The stagnant waters are much better suited for the cleaning of the sewage waters than the rivers, as the living beings of the stagnant waters are much more abundant in number. In FISCHER's opinion, one hectare of a carp pond bears ten times as much sewage than a hectare of river surface.

In order to precisely establish the sewage - water - cleaning, the indicator organisms can be taken as a good help because their presence and individual number reflect the given biological condition of the water. The cleansing of the sewage waters resp. and the establishment of the efficiency degree of the clarifying plants can also be carried out with the aid of laboratory algal cultures. In these last years, especially in America, the sewage waters are cleaned with special sewage ponds (oxidative ponds), and in these ponds no fish is bred. The renouncement on the economic utilization offers the advantage that the water surface can be more charged with sewage water. CALDWELL in 1946, and MYERS in 1948 were the first to suggest oxidative ponds in order to reduce the easily decomposing organic matters to stable compounds, by means of the use of the symbiotic character of algae and bacteria which occur together. As for the use of the maritime algae of big size for human food it is a several thousand years old idea. Early Chinese literature already deals with these algae. We can read about them in the Grecian and Roman works as well, although in VIRGIL's opinion "Nihil vilior alga" i.e. nothing is more worthless than the algae. First of all, it was the maritime algae of big size and easy to collect which aroused man's interest as for food, manure and cosmetics; for about 300 years they have also played a part as industrial raw material. According to 'SZEJTLI's data, more than 850,000 tons of fresh marine algae are processed yearly in 15 countries at present, and the countries, possessing proper seashores make efforts to increase the quantity of the products prepared from algae. In the first place feeds and chemical fertilizers are produced from algae but they also enter into consideration as raw material of the biochemical industry. The marine algae, processed industrially are predominantly brown algae. The role of the widely spread red algae is minor because they live in deeper waters, their dimensions are smaller, their collecting is more expensive. The maritime green algae are not significant.

The ash constituent of the brown algae, related to dry matter is 20-45% that is much more than that of the terrestrial plants; in addition they contain especially much Na, K, Zn, J and nearly 60 different microelements. Nowadays it is the alginate, consisting of the complex compounds of the polymers of A-mannuron acid which is the most important algal product; it was discovered in 1883 and has been used by many branches of industrial production. As for its amount, it is the alga meal that seems to be most important; it is used, first of all, for feeding purposes but, at a small rate, it takes part among the human foods as well. It is an excellent source of vitamin — and mineral matter, and it is cheaper then any other animal feed. It is an excellent chemical fertilizer.

In the course of the history man utilized innumerable plants; at first within the framework of his collecting manner of life, later by means of their introduction into cultivation. This process also keeps on going nowadays, and people start cultivating these microscopically examinable, tiniest fresh-water plants for industrial, feeding and nutritional purposes. The equipments for their breeding can be established and operated whenever and anywhere: in the deserts, on the tops of the houses, on unusable fields, in the open air and in closed rooms. We can claim together with Mothes that the breeding of the tiny fresh-water algae expels the worry for the overmultiplication of mankind.

With this revolutionary changement our inland researchers also became connected.

At the Zoophysiological Department of the Research Institute of Animal Breeding alga-producing experiments were started in 1953. At the Medical Chemistry Department of the University Medical in Szeged industrial algological studies are conducted. At Tihany, in the Biological Research Institute of the Hungarian Academy of Sciences the theoretical bases of algal mass production are investigated.

It is an important property of the algae that they are able to assimilate radioactive elements quickly and in a big quantity, and to build them into their bodies; therefore they can be used for the production of isotope amino acids. It was GILEVA who pointed out that using this property of the algae we can remove the detrimental materials from the waters contaminated by radioactive matters.

As for the significance of the algae as a food, we can only measure it if we compare their properties with our cultivated plants. Our agricultural plants are of huge dimensions as related to the algae but as a rule only a minor part of them (root, foliage, young sprouts, fruit, seed) constitute a valuable human food or animal fodder. In unicellular algae, however, the whole body is utilized, and there are less indigestible or hardly digestible matters in them; their cellular walls are thin, or they are covered with plasmatic membranes only.

Our cultivated plants use very much organic matter in order to build up their bodies, to sustain their life processes indisturbed, to protect themselves from the unfavourable, extreme effects of the continental conditions. There is a great amount of cellulose and such constituents in them which are less valuable or totally useless for us. Their material movement is of exceedingly great dimensions. They must convey water and nutritive salts of the soil, taken up by the roots to the leaves, each cell demands both the organic compounds, produced in the leaves and the water. For the sake of the new food uptake they must evaporate much water. Storage takes place far from the leaves. The transport courses are huge. Light utilization extends only to some cellular layers of the surface. Their energic economy, as for the products which are useful for us, is wasteful; their productivity, related to micro-organisms living in water, is low.

The algae, as a contrast, produce a great deal of organic matter as related to their volume with little expenditure of total energy. Their light utilization is optimal, their movement of materials is minimal. While the cultivated flowering plants use 0.1-0.5% of the sun's radiation energy, the algae utilize 2-3, or even a higher per cent, according to some workers. The light saturation, decreasing the photosynthesis, can be eliminated relatively without difficulties in them. So it is possible to produce, by them, more and better quality protein and organic matters, in addition: more easily, with a higher efficiency than in the case of the most developed terrestrial plants. Their production of organic matter is of a big volume, we need not await the ripening of fruits, the formation of tubers, the thickening of roots. The whole alga can be used as food. Even if there are useless matters in it they are restricted to the cell-wall, e.g. in the case of the autosporulational multiplication of Scenedesmus. If the feeds: carbon dioxyide, nutritive salts, water and light energy are available, their multiplication is very rapid. The algae accomplish assimilation as early as from the very moment of their birth. The alga cells are surrounded by the nutritive solution, they are so-to-say bathing in it, their environment, in comparison to the air, is richer in CO2, and it is much easier to keep the CO<sub>2</sub> of the water on a favourable level than that of the air. So the technologic conditions of an increased cultivation are much more advantageous than in terrestrial plants; on the other hand it is rather difficult to obtain the floating alga cells from the nutrient solution. In the case of the terrestrial plants the assimilates are accumulating gradually, and at a determined moment they are taken away (harvest, vintage, etc.). In the water the production of the organic matters is continuous, therefore they can be carried away continuously. The dynamics of the organic matter production of the water totally surpasses that of the terrestrial production.

Up-to-date agricultural production tries to obtain a bigger mass of products of organic matter by means of a directed photosynthesis. In these last decades two methods of such kind have been realized although to a little extent. One of them is continuous plant cultivation on an irrigated and permanently grazed or mown field. The other method consists in the permanent production and parallel permanent filtration of the microscopical fresh-water algae. According to very cautious estimates, the protectin production of the controlled photosynthesis fluctuates between the tenfold and hundredfold of the usual agricultural production.

The high productivity of organic matter in algal production is the combined result of several factors. In algal production the algae are kept constantly in the most rapid growth phase. If a fish pond is properly populated, the situation there is the same. But the quick growth of the agricultural plants lasts for a short time, then the rapidity markedly decreases, and at the ripening time it ceases altogether. The algae - provided the necessary light energy is available – produce much more safely than the phanerogamous plants do, and they produce in innumerous layers. Our agricultural plants, however, are placed in one layer only. The algae do not suffer from the lack of water and nutrients, they are not influenced by drought, hail, wind, fire, flood; they are much more independent of the weather. The innumerous animal and fungus parasites of the agricultural plants are insignificant as against the algae. In the algal production the frost, the light-decreasing fog and some microscopical animal parasites are the inhibiting factors. If we take all these into consideration, the average organic matter production of the algae is at least half of the possible maximum production. Again, the average production of our agricultural crops falls far behind the maximum possibilities. The light utilization of the agricultural crops is much less then that of the micro-organisms, and the latter require a lower amount of nutritive salts. The average yield of wheat is barely 20% of its maximum, the production of fruit takes place at stages, fixed by the vegetative period. On the other hand, there is no theoretical obstacle to make algal production continuous and independent of periods and seasons.

The dry material of the algae consists of protein to more than 50%, and they contain all the essential amino acids which are of vital importance. Their amino acid set is more perfect than that of many phanerogamous plants; tryptophan, lysin and methionin are in algae at a minimum. Amino acids of low molecular weight are rather predominating and, as a consequence of this, the proteins of the algae are easier to digest. Their vitamin content is diversified and high (A-provitamin, thiamin, riboflavin, niacin, panthotenic acid, follic acid, B<sub>12</sub>, C, D, E, K).

Of all the plant types it is the algae which are least dangerous for man. We know about only two furrowy algae which, indirectly, can cause a lethal poisoning. If edible shellfishes consume a great amount of *Prorocentrum micans*, the poisoning matter of this alga species can be injurious for man, indirectly by the shell-fish. Similarly, Gonyaulax catenella can cause a lethal poisoning by the mediation of edible molluscs.

Afgae have a great importance in space investigations. As a result of the closed and relatively narrow environment of the space ships, and because of the special conditions of the space, few plants respond to requirements. Such plants can come into account only which endure the fluctuation of temperature, the extreme radiation conditions, the consequences of the big velocity, the acceleration, the weightlessness, the oscillations well enough, besides are of rapid development and multiplication, of small size, unpretentious and resistant to bacteria and fungi. From all this it follows that our traditional cultivated agricultural crops cannot enter into consideration — because of their demand on the soil, big size, significant weight, slow process of development in the space ships. As to the requirements mentioned above, it is the algae which respond most. They are not only quickly multiplying and resistant but, by changing the environmental factors, the quantity and quality of their assimilates can also be influenced.

Nowadays we efforce tremendous energies, bound to the tiniest parts of the matter to our service, and by means of them we conquer the space. At the same time we utilize the microscopically dimensioned plants of the living world, the algae, — which bind the cosmic radiation energy of the sun coming to us from the space — as a new source of organic matter which is the richest among all those we have become acquainted with in the course of history.

## QUELQUES REMARQUES SUR LA SYSTÉMATIQUE ET LA PHYLOGÉNIE DES ALGUES

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L'importance de la Systématique est souvent méconnue et parfois même les biologistes pensent que le systématicien ne fait pas oeuvre scientifique. On oublie trop souvent qu'en algologie par exemple, le systématicien fait appel à la biochimie, à la cytologie, à la morphologie, à l'écologie, à la physiologie afin de donner en une synthèse harmonieuse une description correcte et complète des organismes qiu'il observe. Le systématicien ne cherche pas seulement à décrire et nommer les algues, mais à les intégreer dans une classification naturelle dont l'ensemble suit pas à pas le lent cheminement de l'évolution et retrace l'histoire de la genèse des organismes.

Le travail du systématicien, une fois l'organisme décrit avec le plus précisions possibles, sera de confronter cette description à celle des algues qui semblent voisines et de trouver ainsi sa place dans une série naturelle. S'il rencontre une diagnose qui correspond parfaitement à la description de son algue, le problème est résolu.

Parfois l'algue étudiée offre un ou plusieurs caractères originaux qui n'ont jamais été signalés, il peut alors se présenter deux possibilités:

1 — les diagnoses des formes affines connues sont simplement incomplètes ou mêmes erronées;

2 - il s'agit vraiment d'une nouvelle unité systématique.

Le premier cas est insoluble en l'absence de matériel type de référence ou de type d'herbier. Il se pose de plus le problème de l'importance des caractères et de leur hiérarchisation. Là intervient un élément subjectif très important: ainsi un auteur considère que chez Trachelomonas la structure de la logette, c'est-à-dire son ornementation est plus importante que la forme de cette logette. Pour un autre algologue, la forme de la logette passera avant sa structure. Le premier mettra dans le même groupe tous les Trachelomonas à logettes épineuses tandis que le second placera dans la même série les espèces sphériques, tant lisses qu'épineuses.

Prenons un autre exemple: dans les Chrysophycées flagellées solitaires à thèque (genres Kephyrion, Stenocalyx, Pseudokephyrion, etc) un spécialiste indique comme coupure générique le nombre de flagelles, tandis qu'un autre considère ce caractère comme secondaire et fonde ses coupures génériques sur la structure et l'ornementation de la thèque.

Comment donc hiérarchiser les caractères: il n'y a, hélas, aucune règle, cela est laissé à l'initiative et à l'intuition de chacun et seule une grande pratique des algues permet de donner une importance plus ou moins grande aux caractères observés. La constance d'un caractère est souvent dans la nature bien difficile à vérifier; dans ce cas les cultures (pures ou cloniques) peuvent rendre de réels services. Seules ces cultures peuvent indiquer les marges de variations des caractères systématiques et renseigner ainsi sur la hiérarchisation de ces caractères. Mais en cultures certaines algues montrent un polymorphisme exacerbé que l'on ne retrouve jamais dans la nature. Il semble alors que les conditions de cultures sont si éloignées des biotopes naturels que les organismes sont complètement déséquilibrés et perdent leur aspect normal. Les résultats obtenus en «maltraitant» les cultures d'algues par des milieux glucosés par exemple, nous semble difficilement utilisables en systématique.

Rappelons aussi que l'importance d'un caractère peut changer lorsqu'on passe d'un genre ou d'une famille à une autre: ainsi le nombre de pyrénoides est fondamental pour le genre *Chlamydomonas*, il devient très secondaire dans les Ulothricales ou les Chaetophorales.

Si l'on est en présence de caractères encore inconnus, caractères qui semblent importants, il s'agit alors d'un nouveau taxon. Que faire? tout d'abord donner une diagnose trèsprécise et une ou plusier figures de façon qu'il soit possible à tous les autres algologues qui retrouveront cette algue de la reconnaître avec certitude. Doit-on ensuite en faire une nov. var, une nov. sp., un nov. gen.? Expérience, intuition, goût personnel conduisent alors l'algologue. Dans certains cas l'organisme observé, par ses caractères originaux bien tranchés se montre très nettement éloigné des taxa voisins. Par contre dans certains groupes, Desmidiées par exemple, si l'on rassemble les espèces d'un genre, puis celles des genres voisins, on voit qu'il se forme ainsi une chaîne continue, presque homogène, où chaque taxon est entremêlé, relié aux taxa voisins par les tentacules de son polymorphisme. Cela est d'ailleurs fort rassurant car c'est la preuve que nous nous trouvons devant un phylum naturel.

Mais il faut pourtant scinder ce phylum continu en petits fragments qui seront des taxa différents. Les cultures pourront rendre des services, mais comment distinguer dans des algues sans sexualité, les caractères particuliers à un clone de ceux d'une espèce? Là chacun coupera, taillera ses taxa suivant son expérience ou son «flair». Il y aura des «pulvérisateurs» pour qui les genres seront nombreux et ne grouperont que quelques espèces et au contraire, les «rassembleurs» qui feront des genres plus importants riches de nombreuses espèces. En tous cas les limites des taxa, que ce soit, familles, genres, espèces, seront presque toujours artificielles. Seules les espèces ou genres très spécialisés, fin de phylum en général, complètement isolés par disparition des bases évolutives, seront des taxa naturels parfaitement définis. Mais bien souvent l'«entité» élémentaire, espèce ou variété, est engagée si étroitement dans le processus évolitif qu'elle se raccorde à la fois au taxon dont elle dérive et à celui qui découle d'elle.

En l'absence de toute donnée paléontologique, la phylogénie algale est un pur exercice intellectuel. Il éxiste ainsi plusieurs manières de reconstruire le monde algal, toutes aussi logiques, mais toutes incontrôlables. On peut aussi bien placer les Flagellés à la base du tronc algal que les formes pleurococcoides. Convergence, néoténie, absence de documents géologiques, rendent ce problème de la phylogénie insoluble. De ce fait la définition d'une espèce naturelle nous semble impossible chez les Algues.

Seule une meilleure connaissance des algues, de leur chimie de leur cytologie, de leur physiologie, de leur biologie, de leurs cycles vitaux permettra d'apprécier la valeur des caractères distinctifs et de tracer avec plus de précision les cadres d'une saine systématique.

#### PILOT PLANT EXPERIMENTS ON ALGAL MASS CULTURE AT TIHANY

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An equipment of a total capacity of 2 m<sup>3</sup> and of 10 m<sup>2</sup> total surface was constructed at Tihany. The units of this construction are glass walled containers of a dimension of  $20 \times 100 \times 100$  centimeters (= 200 litres). The suspension was aerated with 1.5-3% carbon dioxide in air. The cells in this narrow and high water column are stirred up by the bubbles sufficiently for maintaining an adequate light supply. The strain used was: 5618. Scenedesmus obtusiusculus CHOD. The pilot plant was operated during two seasons (1. August -15. November 1962 and 1. June-15. August 1963). The operation was half continuous, inasmuch as two vessels were halved at every 5th day. The suspension remaining in the containers after harvest served as inoculum. The cells were separated from the nutrient solution with supercentrifuges: types FS45 and FC70 ("Zuglói Gépgyár, Budapest", Hungary). The yields in the half month periods investigated are as follows:

	Dry	Yield kg dry					
Time	g/litre	g/m³·day	matter/20m <sup>2</sup> . half month				
1-15.6.1963	1.33	26.6	4.0				
16 - 30.	1.20	24.0	3.6				
1-15.7.	1.21	24.2	3.6				
16 - 31.	1.23	24.6	3.9				
1-15.8.	1.06	21.2	3.2				
$1\!-\!15.8.1962$	1.21	24.2	3.6				
16 - 31.	1.24	24.8	3.7				
1-15.9.	1.26	25.2	3.8				
16 - 30.	0.90	18.0	2.7				
1 - 15.10.	0.74	14.8	2.2				
16 - 31.	0.73	14.6	2.2				
1-15.11.	0.45	9.0	1.4				

Supposing that 14 and 24  $g/m^2$ . day is produced in April and May respectively, then the average daily yield produced by the photosynthetic activity in the frost-free season between 1st April and 31th October would be 21.5 g dry matter/m<sup>2</sup>. day, which amounts to 46 metric tons pro hectare during this period.

# STUDIES ON PHYSICAL, CHEMICAL AND BIOLOGICAL CHANGES IN THE NUTRIENT MEDIA OF THE ALGAL PILOT PLANT AT TIHANY

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In the study of the liquid phase of algal mass culture three different factors are of main importance: (1) The composition of nutrient solution before inoculation, and the technology of its treatment. (2) The upper limit of cell concentration. (3) The properties and handling of the "exhausted" nutrient solution separated from the algal cells. (Waste water problems of algal mass cultivation.) In the first group two viewpoints are dominant: the nutrient solution should contain all substances necessary for maximal growth of algae and it must be inexpensive. In these experiments Péti-só, an artificial fertilizer  $(NH_4NO_3)$  manufactured in Hungary was used as nitrogen source. Phosphorus was fed in the form of phosphoric acid and potassium carbonate. Though in the preparation of the nutrient solution tap water and chemicals of technical quality were used, it was regarded necessary to add iron (ferrous sulphate) and trace elements (boron, manganese, zinc, copper, molybdene), because the natural trace element content of the suspension gets soon exhausted by the algae. The composition of trace element solution used in these experiments is not of general validity, because allowance must be made to the chemical properties of the water at disposal. The density of suspension, the upper limit of dry matter: water ratio is one of the most important fundamental questions in the mass culturing of unicellular algae. For sake of economy an effort was made to produce a dry matter concentration as high as possible. Two phenomena related to cell density were observed here: In the course of culture dry matter content did not increase over a certain level, irrespective of the density of the initial suspension. This upper limit (maximum dry matter concentration), where the slope of growth curve turns into horizontal direction is 1.2 g dry matter/litre when using open air containers. Many attempts were made to increase this value, but without lasting success. The other fact is that the amount of organic, so-called extracellular substances produced by the algal cells and getting into the liquid phase increases parallel with the concentration of algal cells (partly by way of diffusion, partly by way of lysis of empty cell wall rests or dead cells).

The amounts of hydrogen and hydrocarbonate ions are influenced by the intensity of photosynthesis, the amount of carbon fed and by the cations present in the solution. This latter regulates the buffer capacity of the solution, consequently the amount of carbon able to get bound chemically. Growth is favoured by potassium and sodium, however, if they get accumulated they alkalinize the exhausted nutrient solution. Sodium is expecially harmful in this respect, because it is not utilizable by the algae. The presence of calcium and magnesium ions is more favourable in the further treatment of exhausted nutrient solution (their carbonates are insoluble in water). In the suspension a most peculiar behaviour of these substances was observable. It is assumable that the Ca and Mg ions used for the purpose of maintaining buffer capacity and of binding the carbon dioxide bubbled through got lost as free ions for the suspension by partly getting absorbed to the hydrate membrane of the algal cells, and partly entering into complex linkages with the organic substances excreted by the algae. Our own experimental results and the experiences obtained during study-tours abroad both suggest that it is not suitable to the purpose in view to increase further the density of suspension.

## ANALYSIS OF THE PRODUCT OF THE ALGAL PILOT PLANT AT TIHANY

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An as yet unsolved technological problem of algal mass culture is the separation of algal cells from the liquid phase of suspension. It is regarded necessary to use a sedimentation technique before starting the energy consuming process of centrifuging or separation. In the course of experiments dealing with the surface phenomena of algal cells it was observed that sedimentation is favourably influenced by changing suddenly the hydrogen ion concentration of suspension. The harvested suspension was led into sedimentation containers and alkalinized with 1 ml/litre calcium hydroxide solution. The supernatant was simply siphoned, and the suspension concentrated to about 1/10 centrifuged with supercentrifuges (types FS45 and FC70. Zuglói Gépgyár, Budapest). Pigment and sterol analyses were carried out on the living material obtained. The remaining material was dried at 105°C and pulverized before analysis. The chemical composition of algal cells is not equal with the composition of the product obtained by centrifuging, because the latter is influenced by the treatment used for sedimentation. By adding calcium hydroxide solution to the harvested suspension not only calcium carbonate is produced, but other dissolved inorganic salts and organic substances get also precipitated. The disturbing effect of these substances in the evaluation of analytical results may be eliminated by ash content determinations. The chemical composition of the product separated without using the sedimentation procedure is similar to that of washed algal cells: average protein content 52.3%, ash 8.3%. The amount of protein ash-free dry matter is about 57%. The ash content of the material obtained with sedimentation treatment may reach values as high as 19.3-30.6 and even 47%, while protein content decreases in reverse proportion to ash content, namely to 46.7-34.8%. Protein content in the ash-free dry matter basis is 48.6-75.8%, in average 61.1%. In order to examine the nutritive value of algal product feeding experiments were performed on hundred baby chickens in 1962. Dried and milled algae were willingly eaten by the chicks, and no deficiency disease was observable on them. Their growth remained scarcely below that of the group of chicks fed on standard chicken food, though due to the restricted amount of algae available the total protein content of their food was slightly less than in the standard food. Feeding experiments on raw living algae did not give satisfactory results. The flesh of chicks fed on alga-meal tasted good. Their feet, bill, skin, fat and nerve fibres had a bright yellow colour, in contrast to the white or pale pigmented color of the control group.

#### PHENOL TOLERANCE OF SCENEDESMUS OBTUSIUSCULUS CHOD.

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Algae may have different roles in sewage biology: they may act as indicators or as participants in the self purification of contaminated waters, they may be used as tools in artificially clarifying sewage and may finally be utilized as test organisms in determining the efficiency of various clarifiers. Only few experimental data are known on the phenol tolerance of algae. The No. 5618 Scenedesmus obtusiusculus CHOD. strain in the collection of algae of the Biological Institute at Tihany was not destroyed even by a phenol concentration as high as 500 mg/litre. The growth inhibiting effect of phenol is in direct proportion to its concentration.

# UTILIZATION OF VARIOUS SUGARS BY UNICELLULAR GREEN ALGAL STRAINS

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The utilization of organic substances by unicellular green algae is the specific property of the various strains. This property has been used by many workers to establish systematic distinction between morphologically less differenciated algal species. Two strains of the collection of living algae at Tihany were investigated in this respect (7K Chlorella vulgaris BEYER. and 5618 Scenedesmus obtusiusculus CHOD.) The comparative investigations as to fifteen different sugars pointed to marked differences between these two strains. The uptake of various organic substances depends both on the physiological age and the momentary condition of the suspension. To avoid errors that may arise from these factors the degree of sugar utilization is expressed by a relative value i.e. in units of glucose. This "relative glucose unit" may be used also for establishing numerically the degree of utilization of other organic substances.

## GLYCINE UPTAKE OF SCENEDESMUS OBTUSIUSCULUS

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The uptake of labelled glycine- $C^{14}$  of *Scenedesmus obtusiusculus* was investigated in relation to the incubation period, the intensity of light and changes in the concentration of external solution. The time curves established in light and in dark show that the glycine uptake period of *Scenedesmus obtusiusculus* can also be divided into two phases: the first rapid physical period and the second active one. The stimulatory effect of the light was observed in the first five minutes and continued during the whole uptake investigation period. The glycine uptake was stimulated even by a minimum of light. Further increased intensity of the light depends on the concentration of external glycine solution; at low concentration it is a low level but when the concentration of the external solution is 10 mM, the light increases the uptake by 100%. In spite of the wide range of external concentration saturation effect of the concentration curve can hardly be observed.

# PROBLEMS OF QUANTITATIVE ANALYSIS IN ALGA-COENOSES OF POTAMIC PLANKTON AS DEMONSTRATED ON THE EXAMPLE OF THE RIVER TISZA

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Thorough quantitative analysis is needed to recognize such dynamic changes that proceed in the plankton alga-coenoses of watercourses in space (along the stream) and time (under different seasonal and weather conditions). The methods to determine the quantity of plankton can be considered reliable that present a true and real picture of the full coenosis

- including the nannoplankton. Such are the sedimentation methods, out of which the UTERMÖHL technique can be considered, in every respect, to be the most adequate one. Author has carried out, since 1962, the analysis of the alga-coenoses of the plankton of the river Tisza and its tributaries with UTERMÖHL's method from the following points of view =(1) Concerning the coenoses every taxonomical group is taken into consideration, i.e. all efforts are made to give an objective picture of the complete alga-coenosis. (2) The samples containing considerable amount of floating mineral particles are first subjected to detailed qualitative examination using also net-collected plankton samples. In the sedimented quantitative samples the determination of the algae sometimes partly covered by mineral particles can more reliably made after a preliminary qualitative orientation. (3) Beside the samples taken from the main current and characteristic of the mass of the river-water ("eupotamos") samples have been taken also from the lenitic littoral-water in order to study the direct effect of the affluent waters (accessory waters, cleaned and uncleaned sewage waters). (4) The precise periods of sampling are fixed according to the changes of the water-level or water-level conditions, decisively influencing the potamooikos. (5) Author's efforts in fixing the sampling places and working up his material aim to make good use of his experimental material, beside the general research object of the potamolimnology, also for the saprobiology. Author demonstrates his most characteristic results concerning the quantitative composition of the coenoses of planktonalgae of the river Tisza. From these it is evident that the plankton production of the river fluctuates between the minimal winter production characterized by a few thousand  $\Sigma$ -ind/l values and the maximal late summer production characterized by a few million  $\Sigma$ -ind./1 values i.e. within a very wide range. Further it is seen from the data that the ind./l value of certain species is sometimes significantly increased in the length of the river, which is a very important date for the general interpretation of the "potamoplankton". It was found remarkable that plankton-alga coenoses due to certain seasonal and river-conditional complex effects, appeared from year to year when these complex effects appeared anew. These repeatedly occurring coenoses of aspect feature are well characterized by the more or less rising mass-production of some algal organisms or at least by their relative predominance. The quantitative plankton analysis by individual counting offers directly utilizable data from the coenotical and saprobiological view-points, whereas for the estimation of the biological productivity these data furnish only approximative information. To correctly evaluate the latter other methods must be made available too.

# THE ALGOLOGICAL CONDITIONS OF THE RESERVOIRE IN THE RAKACA-WALLEY

# Z. Szabó

#### BUDAPEST

Abstract not received.

## STUDIES ON THE FACULTATIVE KRYOBIONTISM OF THE ALGAE

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Algae occurring in mass productions at the surfaces of snow in mountains are stenothermic, i.e. they propagate only at temperatures closely over the freezing point. The majority of the algae are eurythermic. I observed as an extreme form of eurythermia in several cases algae, which form algal bloom under the ice cover in winter, to pass into the ice or into the snow that covers the ice and to form mass production on the surface, or in these layers. Thus a kryobioseston colouring is produced, resembling the colouring due to genuine kryobionts. This phenomenon may be designated as facultative kryobiontism in contrast to the genuine kryobiontism, referring to organisms which need a low temperature just a few degrees over zero centigrades and cannot multiply at higher temperatures. I found facultative kryobionta mass productions so far only on ice cover of waters rich in decaying organic matter as yellowgreen, grey-green or brown stripes or spots. The ice always was slightly elevated, it "stuffed" out of the surface in the coloured areas indicating that the volume of the ice also increased where the invasion by micro-organisms starting from the water occurred. The elevation is less expressed on snow surfaces. If a "bloom" of the snow covering the ice is observed, the ice and the underlying water are also coloured by the bioseston, accordingly the algae forming the mass production originate from the water. This phenomenon was due mostly to *Chlamydomonas* and *Euglena* species known to be thermophil-eurytherms, which can produce mass productions in spring or in warm summer periods too. Lately I observed similar mass productions of *Pieromonas*, *Hormidium* and chlorobacterium species in the south of the Great Hungarian Plain. Our meteorobiological analyses had shown that facultative kryobiontic mass productions are mostly evoked by sunny prefrontal weather constellations when there was a rise of temperature. The unicellular algae coming from the water and the ice are usually not damaged by freezing, on the contrary, the vitality of some cultures rather increases after a certain time. If *Spirogyra* was frozen in summer its protoplasm was completely destroyed. Similar treatment damaged also the cells of *Chlamydomonas reinhardi* and of *Euglena polymorpha* taken from the natural algal blooms. On basis of these results it seems probable that the mentioned eurythermic organisms gradually acclimatize to the frosty environment par. allel with the decreasing of temperature.

# THE FORMING OF THE POISONOUS CHAIN $(H_2S-SO_2-NH_3-HCN-TOXINS)$ IN FISH-PONDS

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In the ponds whose water is rich in mineral nutrients the algae become very numerous-During the periods of great rises in temperature in summer, blooming of the water appears as a result of climatic influences and this is followed by the destruction of fish in smaller or larger numbers. Examining these phenomena we have found that owing to the fall of temperature a layer of mud containing ferrous sulphide (FeS) becomes oxidized and the sulphuric acid liberates  $H_2S$ . The  $H_2S$  reduces the sulphuric acid, and  $SO_2$  and colloidal sulphur form. The  $SO_2$ enhances the permeability of cell walls. Under the effect of  $H_2S$  and  $SO_2$  hydrolysis, instead of synthesis, takes place in the alga cells. The alga cell so demaged gives off sugar and amino acids into its environment. Since the  $H_2S$  blocks the iron-containing terminal oxidases but not dehydrogenase, the algae can produce  $H_2$ . This  $H_2$  is utilized by the sulphate-reducing bacteria in the water. The effect of  $H_2S$  becomes intensified. Owing to loss of specific weight the algae rise to the surface and a good deal of them die. The anaerob decomposition of the disintegrating alga bodies gives off more ammonia and HCN into the water, and we have been able to demonstrate putrescine in the water. This means that bacterial toxins may also have been present. The bacteria that multiply in the water, the surface layer of algae, and the oxidation of  $H_2S$  cause a complete lack of oxygen and the fish die. So it is not only the  $H_2S$  forming in ponds with acid peaty soils or the ammonia content of alkaline ponds which is responsible for the destruction of fishes, but also the members of the above-mentioned poisonous chain collectively, in different degrees depending on the pH values of the water and the mud.

# A MICROKINEMATOGRAPHICAL STUDY OF THE CHANGES IN VISCOSITY OF THE CYTOPLASM AND ITS RELATION TO THE CELL WALL IN CLOSTERIUM

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The cytoplasm of *Closterium acerosum* (SCHRANK) EHR. is easily separated from the cell wall in the case of plasmolysis with  $KNO_3$ ; convex plasmolysis is observed. This indicates that the cytoplasm is but slightly viscous and it hardly sticks to the cell wall. During deplasmolysis the speed of the movement of the cytoplasmic cell organelles (spherosomes, mitochondria) is markedly increased as shown by microkinematography. Apparently, the viscosity of the

cytoplasm is reduced. When the cells are plasmolyzed again immediately after deplasmolysis, contrary to expectations, the cytoplasm is not easily separated from the cell wall, typical concave plasmolysis is observed and a high number of Hecht-treads are formed. This is explained by the fact that the cytoplasms after deplasmolysis, due to its low viscosity, establishes an intimate contact with the cell wall; probably the cytoplasm even penetrates the cell wall to some extent. It is likely that the fine pores visible in the cell wall of *Closterium* under the light and electronmicroscope play an important role in this phenomenon. When this connection with the cell wall is established, probably a rapid increase in viscosity takes place at least in the region of the cytoplasm bordering the cell wall. The highly viscous cytoplasm penetrating the cell wall resists the forces which tend to separate it from the cell wall. It seems certain that the close connection between cell wall and cytoplasm is established very fast, perhaps within a few seconds.

# Abstracts of demonstrations

# THE ULTRASTRUCTURE OF INTER-Z BRIDGES IN THE INSECT FLIGHT MUSCLE

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In 1924 TIEGS mentioned the possibility that the Z-membranes would fulfil the function of the transverse impulse conduction inside the muscle fibre. Electronmiscroscopic investigations did not support the existence of the "Grundmembran", therefore the intracellular impulse conduction is generally ascribed to the transverse tubular system of the sarcoplasmic reticulum. On the ground of our earlier experiments we described interfibrillar bridges which were recently found in ultrathin sections, too. Our pictures show them to be running immediately from one Z-line to the other one and to belong commonly to the two Z-lines on the same level of the neighbouring myofibrils. The Z-lines of the peripheric fibrils are connected with the cell membrane by means of similar bridges. Due to the fact, that in the insect flight muscle the transverse tubular system became extremely reduced and that the external appearance of the Z-bridges does not resemble to the sarcoplasmic reticulum, we regard the Z-bridges as cell components different from the sarcoplasmic reticulum. The Z-lines of the myofibrils, connected by interfibrillar bridges, form uninterrupted — although not membraneous — transverse networks, which result in an immediate connection between all the myofibrils of a muscle fibre.

# IDENTIFICATION OF THE SECRETORY GRANULES IN THE ADRENAL MEDULLA

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In the first part of our experiment the adrenal gland of snakes (Natrix natrix) was studied histochemically; and the cells containing adrenaline were separated from those containing noradrenaline. The electronmicroscopic examinations completing the histochemical ones demonstrated that in the noradrenaline containing cells showing rough granulation by light microscopy the diameter of the secretory granules ranged from 1500 to 3500 Å, while that of the adrenaline cells, having fine granulation, varied between 500 and 2000 Å. The difference in size and in specific weight of the secretory granules containing adrenaline or noradrenaline was verified by fractionation in the ultracentrifuge. After centrifugation at 3000 imes g for 30 min. 80 per cent of granule fraction of the dog medulla contained noradrenaline, whilst the fraction centrifuged at  $24,000 \times$  g for 30 min. contained 96 per cent adrenaline. The adre-naline nature of the small granules in rats was supported by the experiments using insulin treatments. The injection of insulin was followed by an 86 per cent decrease in the adrenaline content of the gland. The electronmicroscopic picture of these medullary cells containing the remaining granules, showed that their diameter was over 2000 Å in 90 per cent. From these results it may be concluded that the small granules mobilized by insulin contain adrenaline. Another type of granulation could be observed beside the above mentioned adrenaline and noradrenaline granules. The diameter of granules ranged from 0.5 to 1.5  $\mu$ . They were bounded by a distinct membrane and showed a fine inner structure. According to the separation by centrifugation and chemical determination it may be assumed that they are precursor granules containing dopamin.

# ELECTRONMICROSCOPIC STUDY OF MORPHOGENESIS OF SECRETORY GRANULES IN THE PARATHYROID OF THE FROG

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The secretory product is accumulated both in normal and in hyperfunctional conditions in secretory granules in the parathyroid of *Rana esculenta*. The secretory granules are surrounded by a membrane in normal functional condition and they contain a relatively homogeneous substance. The inner density of the granule is rather different, it depends on what phasis of the granulogenesis is represented by it. According to our electronmicroscopical observation, the genesis of the secretory granules take place in more steps. The first process is the formation of many empty vesicules in the Golgi apparatus, which moves away from this zone after the formation. The condensation of fine granulated components were observed around the empty vesicules in a later phasis of the granulogenesis. These components get into vesicules, where they assumed to develop into lamellar elements. Since ergastoplasm can often be observed around immature secretory granules, we might suppose that they are of ergastoplasmatic nature. The inner homogeneity of the maturated secretory granules is the result of dissolution of these granular and lamellar components.

# SUBMICROSCOPIC CORRELATION BETWEEN THE CELL TYPES AND SECRETORY GRANULES IN THE ADRENAL MEDULLA OF THE FROG

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In the adrenal medulla of the frog (Rana esculenta) three cell types have been observed with the electronmicroscope. A very close morphological relationship was found between the cell types and the formation of the secretory granules. (1) The so-called "dark" cells represent the phase of ingestion, of which the presence of smooth surfaced endoplasmic reticulum and a few intravesicular catecholamine containing granules are characteristic. (2) In the "intermediate" cells there is a diminution of the electron density of the cytoplasm and nucleus, and also the number of smooth surfaced endoplasmic reticula and of the precursor dopamine granules is reduced. At the same time there is a great increase in number of immature catecholamine granules which are situated in the cisternae of the ergastoplasm. It is assumed that the adrenaline and noradrenaline granules are formed in the "intermediate" cells. (3) Some of the differentiated secretory granules in the "light" cells are surrounded by a membrane, while others are not. They represent different phases of storage and release.

# AN ELECTRONMICROSCOPIC STUDY OF SOME STAGES OF DEVELOPMENT OF THE ASCARIS LUMBRICOIDES VAR. SUIS OVUM

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In the present work we have studied different stages of development of the oocyte of *Ascaris lumbricoides* var. *suis.* Our experiments were carried out on developing eggs from different parts of the reproductive system and on dividing oocytes having first blastomers. Oocytes taken from different areas show significant morphological differences. The oocytes from the upper portion of the oviduct are elongated, showing the formation of yolk granules. The lipid inclusions are of irregular appearance, smaller in the lower portion of oviduct, have few mitochondria, and the structure of their nucleus is characteristic. These cells are capable of fertilisation. The oocytes in the uterus are fertilized, the yolk and lipid inclusions are situated on the cortex of the oocytes. The yolk granules are vacuolized, the number of mitochondria

is increased. In the first blastomers the number of mitochondria continues to increase. The number of yolk and lipid granules is smaller, they show irregular distribution. In general, the ergastoplasm is dilated at all stages of development, the Golgi-apparatus appears in simple form. These electronmicroscopic observations are evaluated in connection with the stages of development.

# ELECTRONMICROSCOPICAL EXAMINATION OF THE BASKETS AROUND THE PURKINJE CELLS

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Light microscopical examinations revealed that the body of the Purkinje cells and the initial portion of the axons originating from them are surrounded by a strongly impregnating basket rich in fibres. According to logical conclusions basing on both the interneuronal connections in the cerebellar cortex and recent electrophysiological observations this basket ending is of the inhibitory type; a thorough electronmicroscopical analysis is therefore justified. According to our electronmicroscopical examinations the fibrous basket surrounding the body of the Purkinje cell is formed by relatively thick  $(1-3 \mu)$  basketaxon-endings, the characteristic feature of which is unusual abundance in neurofilaments and relative poorness in synaptic vesicles. It is only with the inferior part of the Purkinje-cells that these endings establish direct synaptic contacts. These synapses belonging to Gray type II are found almost exclusively on the cellular surface corresponding to the so-called "emerging cone of axon". Some larger basket axon-endings establish contacts with Purkinje axons even at their initial portions, forming here "axo-axonic" synapses. The initial — still unmedullated — portion of the Purkinje axon is formed by a dense network of basket axon-endings, the processes of which have extremely fine diameters  $(0.1-0.5 \ \mu)$  and show characteristic finger- or almost horse-tail-like ramifications. The Purkinje axon is almost embedded in this plexus. These specific fine fibres do not contain neurofilaments any more, some synaptic vesicles, however, may be still observed. These fibres do not form any true synaptic contact surfaces with each other, or with Purkinje axons. From outwards the superior coarser fibrous basket and the inferior axonbasket-sheath with its fine fibres are surrounded by Bergmann's glia, forming an almost perfectly closed sheath, that through the terminal branches of the fibrous basket intrudes in some places as far as the Purkinje cell body or the beginning of the axon.

# THE EFFECT OF DECAPITATION ON THE ULTRASTRUCTURE OF THE EMBRYONIC CHICK THYROID

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Earlier radioautographic experiments pointed out, that the embryonic chick thyroid begins to accumulate  $1^{131}$  on the 11th day due to the onset of TSH-secretion. However, with the light microscope no structural changes were observed following this fundamental functional alteration. The present electronmicroscopical study attempts to identify such a structure and thereby tries to prove the TSH-dependence of thyroid development. The thyroids of 8-, 11and 14-days old embryos were elaborated. The 8-days old showed a rather poor cytoplasmic pattern, with characteristic large mitochondria, Golgi-zones and rarely distributed rough surface vesicles. In case of the 11-days old embryo an increased quantity of lamellar type ergastoplasm is apparent in the thyroid cells; in the 14-days old this structure is even more marked. Beside the ergastoplasm follicles limited by numerous cells are formed. Decapitation (the elimination of the hypophysis, diencephalon and prosencephalon) results that the development of the above structures does not occur and the thyroids of the 11- and 14-days old decapitated embryos are correspondent to the intact 8-days old gland. These data are indicating, that TSH is essential in the functional and structural differentiation of the thyroid and this hormone is activating the gland by forming a lamellar type ergastoplasm in the thyroid epithelial cells.

# COMPARATIVE STUDIES OF DIFFERENT IMPREGNATION METHODS ON THE NERVOUS SYSTEM OF FRESH-WATER MUSSEL

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During the last years some new methods were described suitable also for impregnation of the nervous system in Molluscs. In Hungary ABRAHAM and MINKER succeeded in impregnating the nervous elements of the adductors in Lamellibranchiates. These works did not include the impregnation of the central nervous elements in Lamellibranchiates. Investigations were carried out on the ganglia of Anodonta cygnea L. The Jabonero-quick, Jabonero-Bielschowsky, Bielschowsky, Bielschowsky-Ábrahám, Cauna, Rowell, Cajal I., Cajal IV., Weber, Golgi-quick, Cajal-Faworsky methods were applied. All these methods were carried out according to their original descriptions. The methods of Bielschowsky, Jabonero, Jabonero-Bielschowsky, Cajal-Faworskiy, Golgi-quick, Weber and Cajal IV. proved to be fruitless. Some success was obtained with the methods of Bielschowsky-Abraham, Cajal IV., Cauna and Rowell. These latters were modified by us and after the modification we succeeded in obtaining good results with some of them. The Rowell-method was modified in changing the pH of the AgNO<sub>3</sub> and we made also a gilding at the end. The Cauna method was modified by prefixation with AFA but also by the duration of the fixation, the concentration of AgNO<sub>3</sub> and the number of the formalin-baths was also changed. When the Cajal I (Romeis) was modified by pretreatment with 2-4% FeSO<sub>4</sub> and gilding at the end, it gave also a good result. By these modifications we succeeded in impregnating the frozen and paraffin-embedded sections, both of the nerve cells and the nerve fibres of the fresh-water mussel, and also the total ganglion impregnation can carried out successfully.

#### RENAL VASCULAR STRUCTURE STUDIED BY CORROSION METHOD

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The corrosion technique has long since been employed to demonstrate the vascular tree of the different organs. At present these methods can give information indicating circulatory functions under both physiological and pathological circumstances. In general the kidney is one of the best organs to carry out the above injection technique. The adequate substance used was a 5% polyvinylchloride solution. It has a viscosity and quality by which the normal capillary bed can not be filled. In order to obtain specimens of comparative value the techniques have been standardized, especially the pressure employed. Casts of normal kidney in man, dog and rabbit have been demonstrated. There are marked differences in these various species regarding the diameter and arborization of the vessels. The changes in the individual age are also remarkable. In advanced age less complete filling of the vessels, various diameter of the glomeruli and an irregular vascular diameter could be observed. In chronic renal failure in man a greater number of aglomerular medullary vessels has been found than in the control casts. In various kind of experimental shocks performed on dogs casts indicating juxtamedullary circulation and in other instances cortical shunts without juxtamedullary filling were seen. The effect of shock was studied in dogs after denervation of the left kidney. Casts of both kidneys showed more severe changes on the innervated side and therefore the influence of the nervous system on the vessels under shock condition has been reinforced.

# Abstracts of free contributions

## PHENO-ECOLOGICAL INVESTIGATIONS ON SUMMER CEREALS

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An ecological research work on field crops by recognizing the fact of individual amplitude new vistas were opened to determine the behaviour of varieties and species. Earlier investigations revealed that the dispersion of phenological phenomena manifesting themselves on different specimens depends on environmental factors even in homogeneous stands. The more unfavourable the weather during the vegetation period, the greater is the individual dispersion of phenological phenomena in stands. I could also be established that in winter cereals the phenological phenomena of "periodical crops" (obtained by delayed sowing from September to December) show a regular dispersion and the values of the individual amplitude change like those of an "optimum curve". Earlier investigations with summer crops (maize, hop) did not reveal a dispersion similar to that in winter cereals. Due to the long vegetation period of the species examined the investigation could not be performed during the unfavourable summer period, because ripening would have become uncertain. Therefore the range of individual amplitude was studied on summer cereals of short vegetation period. In 1963 pheno-ecological experiments were conducted with summer barley and oat varieties in Tápiószele. Surveying the stands of "periodical crops" displayed that individual amplitudes of earing, panicle de-velopment and waxy ripeness show a dispersion similar also to that of an "optimum curve". Under the much warmer conditions of the year 1963 poor in precipitation summer barley varieties of arid feature gave proof of minor susceptibility than oats of higher water demand.

# RELATIONSHIP OF PLANT COMMUNITIES AND SOIL TYPES ON THE NATURE CONSERVATION AREA OF CSÉVHARASZT

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On the basis of working up and mapping of the plant communities as well as that of local and laboratory investigations of the soils the following can be stated: (1) Under the original stands of Festuco-Quercetum populetosum albae and under its cultivated stands of acacia there are soils of two layers: partly carbonaceous sand poor in humus (humus content under 1%), partly brown forest soil with carbonate relicts, (humus content above 1%) resp. transitious between these two types. (2) In the soil profile of Festuco-Quercetum brachypodietosum the primitive form of the rusty brown, compactly sandy carbonaceous accumulation horizon can be recognized. One part of the culture stands with acacia is similar, but poorer in humus (under 1%). Their C horizon is in general a very carbonaceous loess sand. These soils of three layers can be included in the type of brown forest soils with carbonate relicts. Another part of the culture stands of acacia can be drawn to the variation of the same soil type without accumulation horizon, resp. to the sand poor in humus. The well-growing stands of the profiles of two layers are a result of the near ground water and of the buried humuslayers. (3) Under the original stands of Convallario-Quercetum and its culture stands of acacia and pine (silvestris, nigra), in general on loess substratum, a profile of rusty brown forest soil has been formed, practically free of carbonates in its surface layer. (4) As a result of inundations and fen accretions in wet depressions the succession has got as far as to the subclimax community of the oak-elm forests (Querco-Ulmetum populetosum nigrae). Their profiles can be included in the type of meadow forest soil. Soil formation. The profiles of Scirpo-Phragmitetum, Caricetum elatae, Caricetum acutiformis-ripariae, Deschampsietum caespitosae and Calamagrosti-Salicetum cinereae can be drown to the meadow fen soils, as grades of the fen soil series (from swamp through fen to carr) close to each other. The sand series is leading from the cover sand (Festucetum vaginatae fumanetosum) through the carbonaceous sand poor in humus (Festucetum vaginatae stipetosum) to the formation of chernozem-like sand (Astragalo-Festucetum sulcatae). The formation of the brown forest soil with carbonate relicts and within this that of the accumulation horizon indicates a further development. From the carbonaceous profiles the washing out effect of precipitation forms the most developed rusty brown forest soils, carbonaceous in the substratum.

#### CYTOGEOGRAPHICAL PROBLEMS IN PLANT COENOLOGY

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The genetical and evolutional importance of polyploidy for the vegetation has been recognized some decades before. First the polyploidy-relations of the flora of certain regions only, but later on the plant communities as well have been analyzed. PIGNATTI brings - as contrasted with the former ecological aspect - some news in cytogeography. He studies the role and importance of polyploidy not in respect of the ecologycal conditions of the different plant communities, but from the point of view of regularities existing in species combination of coenological units. His diploidy-index aims to present – in contrast to the former  $P_{M}^{o}$  – the quantitative characteristics of the vegetation too. In his opinion diploidy-index is a number characteristic of every plant association, from which we can conclude the part of the polyploid species in different successional processes, and the relative age of the plant communities as to their development centre as well. Author conducted - chiefly in beechwood associations with the application of the diploidy-index comparative cytogeographical investigations, and made the following statements: (1) Diploidy-index cannot be considered as a number characteristic of the plant associations, because in many cases the coenological units are cytogeographically inhomogeneous. (2) It is useless to search for direct connection between the age of plant associations and frequency of the diploid species and this is why the diploidy-index can be considered — regarding to the development centre of associations — only as an informatory date; but it can be never taken in consideration as a proof. (3) A serious defect of the diploidyindex is that it reflects the quantitative conditions of the species only, but not the qualitative differences of the vegetation, and that is why it does not give a real picture of the cytogeo-

graphical conditions of the coenological units. Instead of diploidy-index  $DI = \frac{d_p}{P_p}$  it is more

exact to use the formula  $\frac{d}{P} \cdot \frac{d_p}{P_p}$  in which qualitative differences can stand out as well. (d indicates the number of diploid, P the number of polyploid species to be found in the association;

Pp is the presence-sum of polyploid species; resp. the number of them to be found in the coenological table.)

#### CHROMOSOME STUDIES ON HUNGARIAN MOSSES

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In the past ten years while chromosome numbers of moss population have been surveyed in constantly increasing number, even now only about 600 described moss species are known cytologically. So far in Hungary no cytotaxonomical attempts were even made to study the variation of bryophytes and explain variational discontinuities and relationships in terms of cytology. The present investigation was undertaken as a part of a broader bios ystematical study on the Hungarian bryophytes including cytogenetical and population-genetical approaches as well as culture and transplant experiments. All cytological studies were made on spore mother cells at meiosis, employing the squash technique. Chromosome counts were made of 13 species of acrocarpous mosses representing 11 genera and 6 families. The chromosome numbers of three species are reported apparently for the first time, these are Hymenostomum microstomum (HEDW.) R. BROWN, n = 13, Mildeella bryoides (DICKS.) LIMPR., n = 15, and Funaria hungarica BOROS, n = 56. From morphologically distinct populations of Phascum cuspidatum HEDW. two were found to have the chromosome number n = 21, and a third population proved unquestionably to have n = 42. These counts are of significance, because the numbers n = 26, and 52 are reported for this exceedingly variable and wide-spread species. Thus the number n = 21 is the lowest for this species. Previous counts were confirmed by the present study of Hungarian populations of Ceratodon purpureus (HEDW.) BRID., n = 13, Pleuridium subulatum (HEDW.) LINDB. n = 13, Weissia crispa BR. germ. n = 13, W. controversa HEDW. n = 13, Physcomitrium piriforme (HEDW.) BRID. n = 36, Funaria hygrometrica HEDW. n = 14, Bryum argentum HEDW. n = 10, Fissidens minutulus SULL., ssp. pusillus (WILS.) WIJK et MARC., n = 10 and Buxbaumia aphylla HEDW., n = 8.

## DEVELOPMENT OF MORPHOLOGICAL AND CONTENT PROPERTIES OF GENERATIVE POPPY HYBRIDS

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Relying upon the results of earlier experiments with poppy hybrids, the author has produced in 1961 reciprocal hybrids with individuals of pure lines of five poppy varieties, each of different morphological properties, phenological character and alcaloid-spectrum. The first hybrid generation was raised in 1962 ( $F_1$ ), and  $F_2$  in the following vegetative period; each of the parents and hybrids where represented in four series, by 30, respectively 50 plant individuals per plot. On this experimental material the author examined the appearance or the lack of somatic, reproductive and adaptive heterosis and furthermore, the hereditary features of the varieties chosen as parents, as distributed and changed in their descendants. The comparative evaluation of the test results has not only definitely demonstrated the heteroses (absolute improvement of 20-40% as compared with the better parent), but the highest values could be shown to have appeared in those hybrids, whose maternal parent partner was the Bulgarian opium poppy, distinguishing itself by earliness and lack of anthocyan. Regularities have been found moreover in the development of the substances discolouring the sprouts, the petals and the seeds, as well as in that of the alcaloid-spectrum. For the latter, the author has found not only quantitative accumulations, but also qualitative differences in the majority of the hybrids, which permits to conclude on maternal inheritance. These differences provide a starting point for further examinations of the inheritance of alcaloids; they are also remarkable, because the progeny of these hybrids offer the possibility to raise a new selection, which would meet the latest requirements of pharmaceutical industry and be of outstanding value in regard to the different co-alcaloids.

# THE EFFECT OF DIFFERENT MUTAGENS ON THE FREQUENCY OF CHROMOSOMAL ABERRATIONS IN MAIZE

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The frequency of chromosomal aberrations (anaphase with bridge and fragment) which had been induced by various mutagens was studied for investigating physiological and genetical mutagenic sensitivity in the inbred lines (C5 and T18) of maize. The cytological analysis was carried out on first mitosis cells of the root-tips. The air-dried grains were treated with X-rays (7500 r and 15,000 r), neutrons (720 rad and 1440 rad) and ethylmethanesulphonate (EMS; 0.25 percent and 0.50 percent). In the inbred line T18 X-radiation gave rise to most chromosomal aberration, EMS treatment induced the lowest number. The neutron-irradiation occupied an intermediary place. Significant difference has been found between dosages, while between concentrations there was no difference. The comparison of two lines on the basis of chromosomal anomalies showed that line T18 had responded to radiation more susceptibly than to EMS treatment. The line C5 showed approximately the reverse situation: it was effected

sligthly by radiation and greatly by EMS treatment. The mutagenic sensitivity of inbred lines (C5 and T18) is in close relationship with the relative size of the embryo. The line T18 has 21.03 per cent embryo/endospermium ratio, 16-18 per cent protein content, 9-10 per cent oil content, 3-4 per cent oil content. Negative correlation was observed between the frequency of chromosomal aberrations which had been observed in the mitosis, but by anomaly of metabolism induced by mutagenic traetment. After X-irradiation storage of grains for half a year increased the frequency of chromosomal aberrations in the line T18. In the control (untreated) we had observed an increase (of 3.22), too, but it was not significant (SD5% 6.06). In principle this observation may be explained by the so-called "potential break" which had come into being by irradiation in the chromosomes. This break may be transformed into a real chromosomal break with greater probability after a longer period in the presence of the oxygen of the air.

# THE EFFECT OF DEOXYRIBONUCLEIC ACID ON THE SURVIVAL OF ESCHERICHIA COLI POPULATIONS IRRADIATED WITH X-RAYS

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Macromolecular DNA, — both isologous and homologous, in a concentration of 50  $\mu$ g/ml, irradiated together with bacteria — increased the proportion of surviving colony building centres in 2 prototroph and 1 methionin-requiring strains of *E. coli*. After irradiation the cultures were incubated in media containing isologous or homologous DNA. Small doses (5 kr), increased, high doses (10 kr) decreased the proportion of surviving cells. Within the limits of  $10-100 \ \mu$ g/ml concentration the quantity of DNA used did not influence the results significantly. On the other hand, the incubation of non-irradiated bacteria in minimum medium with DNA led to the decrease of colony-building centres. Our experiments proved the radioprotective role of biologically active DNA, the effectivity depending on the actual circumstances.

# EVALUATION OF THE REPLICA PLATING METHOD IN THE INVESTIGATION OF CARBON AND NITROGEN SOURCES OF YEASTS

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LEDERBERG's replica plating method was recommended by SHIFRINE et al. to the taxonomists for the investigation of carbon source assimilation patterns of yeasts. Superiority of the method is thought to be based upon the possibility of simultaneous inoculation and study of about 25 strains on various media. With the above method the assimilation of 10 organic acids as carbon sources and 11 amino acids as carbon, nitrogen and simultaneous carbon and nitrogen sources were investigated. It was established that the method is suitable in carbon source investigations only for screening, but in nitrogen source investigations it is often unsuitable because of the high control values, and it is not suited for diagnostic carbon source investigations either. The high values of nitrogen-free controls can be explained by the wellknown fact that in general the fungi require nitrogen-poor carbon sources. The results of carbon source and simultaneous carbon and nitrogen source tests of amino acids generally agreed with each other, although in some cases the growth in the former type of experiments was stronger than in the latter one or vica versa. In the former case this may be due to the better assimilation of the respective amino acid (as a carbon source) in presence of  $\rm NH^+_+$  ions, and in the second case probably the presence of  $\rm NH^+_+$  ions may inhibit the uptake or decomposition of some amino acids in carbon source experiments. These results imply that the method in question is suitable for screening of interesting data for further physiological investigations too. The differences of carbon source data of the investigated yeast-species were discussed from the taxonomical point of view.

# ACTIVITY OF ACID PHOSPHATASE AND PROTEASE IN TETRAHYMENA PYRIFORMIS GL OF DIFFERENT STATES OF NUTRITION

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Earlier cytochemical studies suggested an intimate relationship of acid phosphatase activity of *T. pyriformis* GL with the nutritional state of the cells. In order to test this assumption two hydrolases were determined in cells disrupted by repeated freezing an thawing. Acid phosphatase activity was assayed with p-nitrophenyl phosphate at pH 5.1 according to HOLTER and LOWRY and protease activity was measured with urea denatured haemoglobin at pH 3.5 according to ANSON. The activities were expressed on the basis of cell number and of protein content. Cells grown axonically on a tryptone-yeast extract medium show high activities of both hydrolases. When cells starve in Prescott solution the activities show a rapid drop during the first two days and do not change significantly for six more days. When cells starved for one or two days are again replaced for one or two days into the nutritive medium, increased enzyme activities are again observed. The activity changes are consistently greater than changes of total cell protein. This observation favours the assumption of a specific decrease or increase of enzyme activities depending on the nutritional status of the cells. The elucidation of the mechanism of this effect needs further studies.

# LABILE STORAGE OF CATECHOLAMINES IN THE CENTRAL NERVOUS SYSTEM

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The central nervous system (CNS) of cats and rats has been studied by means of FALCK's fluorescence technique to locate catecholamines histochemically. Cryostat sections obtained from fresh material were used in lieu of frozen-dried specimens, proposed originally by FALCK. It was found that the cerebral cortex, hippocampus, lateral geniculate body and spinal cord, when removed in vivo by means of neurosurgical techniques, contained numerous fluorescent fibres of the same size and shape as those described by FALCK in the hypothalamus. Yet there were no or only occasional fluorescent structures to be found at these sites when collecting the samples from decapitated animals, whereas the hypothalamic system did not show such a high sensitivity with respect to post-mortal alterations. The only locus where labile catecholamines were found in the somata of nerve cells was the substantia nigra. The fluorescent substance proved to be norepinephrine or some closely related compound since it could readily be depleted by intraperitoneal injections of reserpine (5 mg/kg) or alphamethyl-meta-tyrosine (400 mg/kg). Bilateral extirpation of the superior cervical sympathetic ganglion did not induce degeneration of the nerve fibres that contained catecholamines in that labile form (excludes the possibility of sympathetic origin), nor did the bilateral enucleation of both bulbi (excludes the possibility of retinal origin). Fluorescent fibres in the cerebral cortex remained intact even in surgically isolated small cortical areas (method of the "remaining elements" by SZENTÁGOTHAI), proving that the origin of these fibres must be located in the isolated area itself. Accordingly, and taking into account the increasing number of physiological data suggesting the inhibitory role of catecholamine substances in the CNS, the hypothesis appears for us acceptible that inhibitory functions in the CNS, responsible for the selection of circumscript areas of excitation, are operated by axons or axonal collaterals, containing catecholamines in an extremely labile form of storage.

# HISTOCHEMICAL INVESTIGATIONS CONCERNING THE MORPHOLOGICAL DIFFERENTIATION OF THE BURSA FABRICII AND THE THYMUS IN CHICK EMBRYOS

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In addition to the thymus in birds a so-called "rectal thymus", viz. the bursa Fabricii is also found. As observed in 5 day-old chick embryos, this organ has a lumen lined with a few rows of undifferentiated columnar epithelial cells. Morphological and histochemical investigations have revealed that lymphocytes arise at an early embryonic age from these indifferentiated entodermal epithelial cells of the bursa Fabricii. Significant changes occur on the 15th day of the incubation, especially in the PAS-positive polysaccharides granules of the lymphocytes. Certain thymocytes containing a diastase-resistant PAS-positive substance were observed in the thymus of 5 to 15 day old chick embryos. This substance forms granules which stain well with alcian blue and gallocyanin; they are localized on the nuclear membrane and in the nucleolus. Fat begins to accumulate in the cells of the reticular epithelium next to the nuclear membrane. It is in the last phase of embryonic development that Hassall's corpuscles arise in the epithelium of the thymus. The present investigations have yielded the following results. (1) The bursa Fabricii is probably of epithelial origin; opinions in this respect are not unanimous. (2) At the borderline between epithelium and follicles cysts were found in the bursa Fabricii of 14 to 16 day old chick embryos. Available references mention only postembryonic cysts in this organ. (3) There is a close similarity between the thymus and the bursa Fabricii as regards synthesis of nucleic acid. (4) Mucopolysaccharides have identical localization in the cells of Hassall's corpuscles and in those of the cysts in the bursa Fabricii, a further indication of the functional similarity of the two organs. The assumption that a close functional correlation exists between the two organs during embryonic life is also substantiated by a similar localization of their mucopolysaccharides.

# IMPORTANT MORPHOGENETICAL STEPS IN THE EARLY DEVELOPMENT OF CHICK EMBRYO

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Cinematographical observation of normogenesis is an important tool in studying the pathological changes of early morphogenesis. A new technique for preparing and filming the early chick embryo was worked out by the author which made such observations possible. This method seems to be suitable for pathological, pathophysiological and pharmacodynamical investigations in addition to its didactic value in embryological education. The cinematographical observation is a part of our complex method for investigating the early morphogenesis of the chick embryo. This complex method consists of histological, histochemical, phasecontrast- and fluorescence microscopic examinations in addition to cinematography.

# DETECTION OF PRESUMPTIVE AREAS IN EARLY EMBRYOS WITH JANUS GREEN B, AND ITS TERATOGENIC SIGNIFICANCE

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Under the action of JgB, applied in different concentrations on the blastoderm of chick embryo in the early phase of morphogenesis, mitochondrion-succinodehydrogenase areas develop which correspond to the actually existing stage of development. The areas are invariably found in well defined parts of the blastoderm. They can be demonstrated long before the visible appearance of the presumptive morphogenetic changes or that of the primordial organs. Dehydrogenase activity is indicated by the reduction of the stain. This method seems to be suitable to determine the distribution and the functional activity of mitochondria during the early periods of morphogenesis. From the difference between the rate at which the stain is bound and that at which it is reduced in the early stages of development we may conclude on a three-stepped course of morphogenesis. A morphologically visible change is always preceded by a two-stepped change on submicroscopic or molecular level, namely the formation of functionally inactive mitochondria (the stain is bound but not reduced) and the development of mitochondrial function (reduction of the stain). Higher concentrations of JgB give rise to well definable malformations. The phase-specificity of the malformations corresponds to the presumptive areas of organs or organ systems indicated as such by the JgB. The JgB is attached to a specific lipoprotein of the mitochondria which plays a central role in the biologica oxidation, and exerts its action on energy-consuming biological processes. JgB is known asan uncoupling agent. The uncoupling of the oxidative phosphorilation produces a pathologic shunt which is responsible for the teratogenic effect.

# THE STRUCTURE OF THE FNLCCRINE SYSTEMS IN THE CENTRAL NERVOUS-SYSTEM OF THE WATER-BEETLE (DYTISCUS MARGINALIS)

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In the protocerebrum of the water-beetle three cell groups were found whose members show a neurosecretory activity. The first group is located in pars intercerebralis, the second is beside the medial mushroom body and the third at the basis of the lateral mushroom. body. The cells of the first group are large and unipolar. In the cell body as well as in the thick and straight cell process granules could be seen appearing red stained with paraldehyde fuchsin and dark violet, pale violet or red using the chromhaematoxilin-floxin technique. To get a real picture concerning the function of the cells and the factors influencing them, the animals were lighted with electric light, treated with ultrasonic, with alternating current and some of them became blind. On the basis of the experiments it was established that the cells produce great. amounts of neurosecretum which passes on the cell process and in some places of the cell surface into the neuro-pil. The greatest part of the secret-granules are dispersed here without any special pathways, the smaller part enters the two nervi cardiaci interni via the two pathways originated from the secretory cell groups. The productivity of the cells is increased by the ultrasonic as well as by the electric excitement. The highest hypersecretion was found when the animals were exposed to a constant and strong electric light. The neurosecretum appearing dark violet, pale violet or red produced by the same cell is a glycoprotein which shows different staining reactions in the various transformation phases. The production of the neurosecretum is the highest at 12 o'clock, at 24 and at 2 in the morning. The cells of the second group are of larger and smaller size, their granules could be stained only with paraldehyde fuchsin and they do not leave the cell body. The cells of the third group are all of smaller size, their granules pass from the cell body and form thin columns between the cells. or the nerve fibres respectively.

#### ANTIBODY FORMATION IN FROGS

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Frogs (Rana esculenta) were immunized with soluble (ovalbumin, rabbit  $\gamma$ -globulin) or corspuscular (Salmonella typhosa) antigen at various ambient temperature. The sera of frogs kept at  $+8^{\circ}$  C contained no or very low antibody level but a relatively high, specific titer was produced in frogs kept at  $+22^{\circ}$  C. As least eleven serum components were determined by immune electrophoresis. The specific antibodies are localized on the field of  $\beta$  and  $\gamma$ -globulin fractions. The  $\gamma$ -globulin component of sera of immunized frogs has a sedimentation coefficient of 6.5-6.8 S. The same in the sera of control frogs was 8.0-8.4 S.

# ELECTROPHYSIOLOGICAL INVESTIGATIONS CONCERNING THE FUNCTION OF THE FROG'S LYMPHATIC HEARTS

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The author - dealing with the controversies in the literature about the electrical activity of the frog's lymphatic hearts - demonstrates that the slow activity described by PAPP et al. consists of a pulsation artifact. On the basis of own experimental data author confirms earlier observations according to which the pulsation of the lymphatic hearts is accompanied by rhythmic, approx. 25 cps electrical activity. From the electrophysiological analysis it is concluded that the systolic contractions of the lymphatic hearts do not conform to the "all or nothing" law and therefore their functional properties are much more similar to respiratory muscles, than to those of systemic heart. The central regulatory system of the lymphatic similarly to the respiratory centres - is automatic in character; it is organized on hearts the spinal level. Each lymphatic heart has its own generator in the given area of the spinal cord. The spatial and temporal interconnections of these generators may be different. Between the homolateral centres were observed the most permanent connections resulting in homolateral synchronization or in 1:2 viz. 1:3 cervical-coccygeal rhythm. It was established, however, that in spite of previous reports the symmetrical centres are not absolutely independent of each other, since totally synchronous contractions are not unfrequently seen. Under given circumstances this synchronization may be dominant, but it can be replaced by homolaterally synchronized contractions in response to different stimuli. On the ground of the reported observations it is concluded that the homolateral interneuronal pathways are more resistant against external inhibition, while those of commissural nature are very sensitive to external stimuli. This may be the reason why earlier investigations failed to show the existence of the commissural-type interneuronal synchronization.

# DIRECTION OF STIMULATING CURRENT AND THE EXCITABILITY OF EMBRYONIC MUSCLE FIBRES

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The author stimulated through fluid the larvae (glochidia,  $0.3 \times 0.7$  mm) of fresh-water mussel (Anodonta cygnea L.). A single square impulse evoked a phasic contraction of embrionic adductor muscle. The experimental conditions permitted to precise spatial relation between the stimulating electrodes and the animal. The threshold voltage and strength-duration curve regularly depended on the angle between the direction of the muscle fibres and that of the current. If the direction of current and the longitudinal axis of the glochidia were parallel, the threshold voltage was minimal. At 90° angle it was very high. The results show that the molluscan embryonic muscle fibres are the less excitable with cross-directed current. The function between threshold and angle follows RUSHTON's reciprocal-cosinus relation. The small deviation from this relation and the secondary changes in the slope of the strength-duration curves suggest that the motoric innervation of adductor muscle fibres is problematical.

# A COMPARATIVE STUDY OF THE DEGREE OF SEXUALIZATION FROM THE MESOLITHIC TO THE LATE MIDDLE AGES

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The morphological bases of secondary sexual characteristics of the skeleton are genetically determined. Their expressions become manifest at the time of the production of sex hormones. As regards sexual dimorphism the individuals and populations show very characteristic differences. If these peculiarities are examined in light of microevolution, gracilism, taxonomy and adaptation they provide a dependable basis for the biological reconstruction of ancient populations. Thirty qualitative and quantitative features were used to determine the sex of the anthropological material of fifteen series from between the Mesolithic and the late Middle Ages and the application of a five-grade scale established the degree of sexualization of each sex (-2 = hyperfeminine, -1 = feminine, 0 = indefinite, +1 = masculine, +2 =hypermasculine). The fifteen series originating from historical times included 2000 adults. The following conclusions were made from their sexualization. (1) The populations between the Mesolithic and Aeneolithic Ages are characterized by sharp sexual dimorphism and may be regarded as grobic. In this case the males are not only hypermasculine, but generally are above the +2 value and may be considered ultra-hypermasculine (+3) as for instance the mesolithic population of Taforalt. (2) From the beginning of the Bronze Age the "semi-gracile population" is typical, that is to say, there is little sexual dimorphism: females have masculo-feminine and the males femino-masculine characteristics. The semi-gracile population is found up to the late Middle Ages. One of the finest examples of this is the Khazar series unearthed in the Soviet Union at Sarkel. (3) Finally, sexualization proceeded quite a considerable degree towards femininity and resulted in gracile populations which are also characterized by great sexual dimorphism. For the most part the males approach the 0 value while the females near the upper limit of, or surpass hyperfemininity. Excellent illustrations of this are certain series from the early Arpád period in Hungary. Sexualization may be an excellent factor in determining life conditions and taxonomical questions. The task of future research is the elaboration of this question not only for the Europoid type but also for the Negroid and Mongoloid ones.

# CHANGES IN GROWTH OF CHILDREN FROM MEZŐKÖVESD DURING 50 YEARS

## M. FEHÉR

BUDAPEST

Abstract not received.

# Abstracts of papers read at the sessions on integrating mechanisms in animals and plants

# A STUDY OF PHOSPHOROUS FRACTIONS WITH P<sup>32</sup> IN PLANTS TREATED WITH GROWTH INHIBITORS

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Abstract not received.

# INVESTIGATIONS ON SOMATIC TWIN-FORMATION, BENIGNANT AND MALIGNANT TUMOURS IN THE SPECIES DUGESIA TIGRINA (PLANARIIDAE, TURBELLARIA)

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Authors have made observations and experiments on Dugesia tigrina clones cultivated for over one year and a half. This species reproduces exclusively in asexual way (in the population imported into Hungary) and shows an interesting phenomenon which is termed by the authors as spontaneous "somatic twin-formation". Present investigations have pointed out the hereditary character of this phenomenon and have elucidated its developmental rules as well as histologic and organologic features of its morphogenesis. It has been pointed out that this "somatic twin-formation" is connected with a hereditary lability of the integrating mechanisms in the organism and - first of all - with that of the nervous integration. The spontaneous but organized hyperdevelopment of the central nervous system leads to a regular duplication, respectively multiplication of the main organs and body parts. This process results in a transformation of the adult organisms into double or multiple twins. The twinorganisms formed in such a way retain their normal capacity for asexual reproduction by crossdivision. Organization of the descendants forms itself, in each of the cases, depending on that given in the adult body parts developing into whole organisms. Further observations have shown that the hereditary tendency for somatic twin-formation is closely related with that to form spontaneously benignant or malignant tumours. These tumours can well be distinguished on the basis of their morphogenesis, histology and pathological features. The malignant tumours, in contrast to the benignant ones, grow relatively quickly and with an infiltrative character; they destruct the normal tissue and organ structures and influence the morphogenetic processes of the regeneration. These tumours differ from the chemically induced "malignant" tumours since do not change their pathologic character when influenced by morphogenetic processes of the regeneration, i.e. they do not "normalize". However, the nerve regeneration can temporarily inhibit their development.

# INVESTIGATIONS ON THE EXCITATORY EFFECT OF CARDIOREGULATOR NERVES IN THE HEART OF THE SNAIL (HELIX POMATIA)

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It was observed that on the heart of the snail according to the chosen parameter stimulus not only inhibiting but also excitatory effects can be evoked. The threshold is for the excitatory fibres higher and the appearance of the excitatory effect is influenced also by season-
al changes, that is to say it could not be evoked before the winter sleep. The excitatory effect produced by electrical stimulation of the intestinal nerve can be transferred by humoral way to a non-stimulated recipient heart, suggesting that this effect is based on the release of specific chemical agents. Identical effect to the stimulation can be evoked by applying 5-HT in  $1\cdot 10^{-10}$  g/ml concentration. Antiserotonin drugs (BOL, Chlorpromazine) prevent the effect both of stimulation of the excitatory fibres and that of the serotonin introduced from outside confirming indirectly the chemical character of the transmitter agent. The results are in agreement with the data for Lamellibranchiates of WELSH (1957) and LOVELAND (1963) and affirm that serotonin may play a role in the excitatory mechanism in the heart of the gastropod *Helix pomatia*.

# OXYGEN LEVEL AS AN UNUSUAL REGULATOR IN THE RHYTHMIC ACTIVITY OF FRESH-WATER MUSSEL (ANODONTA CYGNEA L.)

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It is accepted that the periodicity of the life activity in the living organisms is in correlation with the periodicity of the environmental factors, though their direct regulating role is in several cases discussed (endogen rhythms). Recently the author, continuing his earlier work investigated the importance of the oxygen content of the water in the regulation of the biological rhythm in fresh-water mussel. It was found that by changing the oxygen supply of the animal the frequency of the periodic activity can be influenced, i.e. at a low ( $< 1 \text{ mg O}_2/l$ ) oxygen level of the surrounding water, the alternation of the active periods with the rest (both lasting for hours) is frequent, but at a high  $(5-10 \text{ mg O}_2/1)$  oxygen level it becomes rare. Presumeably under physiological conditions it is exactly the oxygen level that plays the most important role in the regulation of the frequency of the periodic activity; the biological importance of the latter may be explained with the adaptation of the animal to the oxygen deficiency. It was stated that by cutting the nerves supplying the syphos and gills, the periodic activity was not influenced by the oxygen deficiency, showing that the periferial structures must have a very important role in the control of the regulating mechanism of the central nervous elements. The lactic acid content of the investigated tissues does not show any significant differences when the oxygen level is changed. It can be concluded that the regulation of the periodic activity in the fresh-water mussel is not controlled by a periodic alternation of any external factor, but that it is a result of an internal regulation which is influenced by a given level of the external oxygen supply.

# ALKALINE AND ACID PHOSPHATASE ACTIVITY IN THE CENTRAL NERVOUS SYSTEM OF ANODONTA CYGNEA L. AND ITS CORRELATION WITH SEASONAL NEUROSECRETORY CHANGES

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Author observed in earlier investigations that the secretion of a Gomori-positive substance by the cells of the central nervous system in the fresh-water mussel was subject to seasonal fluctuations. In the present experiments the alkaline and acid phosphatase activity in these cells was studied in view of the said changes. It has been found that the two phenomena are correlated. Increase in the neurosecretory activity and the metabolic processes is accompanied by increased acid phosphatase activity in the examined species. It is in summer that the ganglia contain the greatest amount of Gomori-positive matter, and it is likewise in summer that their acid phosphatase activity reaches a maximum. Both the secrete and the acid phosphatase activity diminish at the end of summer. In winter, neither Gomori-positive substance nor acid phosphatase activity was observed. Alkaline phosphatase activity changes in the opposite direction: it is practically or completely absent in summer, moderate in autumn and spring, but very pronounced in winter. It is suggested that acid phosphatase is involved in katabolytic processes whereas alkaline phosphatase plays a role in synthetic processes. The Gomori-positive substance appears in an active form in summer, is broken down along with the increased metabolism of the animal, and the activity of the acid phosphatase is most pronounced at this time. In winter, when the synthesis of the Gomori-positive substance takes place (presumably in the form of prosecretion), the alkaline phosphatase shows increased activity.

# DISINTEGRATING EFFECT OF CHORIO-GONADOTROPIC HORMONE AS EXERTED ON EARLY EMBRYONARY DEVELOPMENT OF AMPHIBIANS

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As a starting point for our investigations served one feature of the chorio-gonadotropic hormone (HCG) i.e., the disintegrating effect by which it separates the ripe ovum from the ovary (FSH effect). We regard all effects decreasing the correlative co-operation of the unit of organism or stopping it as disintegrating effects. They are at the same time bound to hinder embryonary morphogenesis, since disintegration is a process contrary to differentiation and integration. Our tests proved that HCG checked or completely hindered the embryonic morphogenesis of amphibians. By quantity of 266  $\mu$ g/ml it hindered the gastrulation of Bufo bufo ova. Under this effect the blastula cells don't wander into the blastocoel but settle on the surface of the blastula. In a 133  $\mu$ g/ml quantity the gastrulation takes place abnormally, however, even in the best case it results only in deformed neurula. The exclusion of volk from the morphogenesis occurs frequently. Bombina bombina eggs can better endure the damaging influence of 266  $\mu$ g/ml HCG than the ova of Bufo bufo. The furrowed Bombina eggs seemingly suffer no injury, the detrimental effect of the HCG, however, manifests itself later on in the organogenesis. The eyes and other organs of the head don't differentiate, and the pigmentation also is different from those of the control-larvas. Even the vitelline substance is not used up during the development. The tail crooks dorso-ventral below and before the yolk sac. The swimming-fins of the tail are well developed. Under the effect of 10 and 20 mg/ml even the furroving of ova stops. The eggs of Pleurodeles waltlii, Pelobates fuscus, Xenopus levis and Rana esculenta suffer, more or less, the same injury because of HCG as the eggs of Bufo bufo. HCG is produced by the trophoblast cells of the embryos of mammals. In our view the primary function of HCG must have been the disintegration of the endometrium and myometrium of the uterus. HCG is therefore a considerable factor for the active implantation of the embryo. Under normal circumstances HCG does not return into the embryo, therefore it cannot exert any disintegrating effect. In the course of evolution mother organisms adapted themselves to the effect of HCG, so they responded to it with increased mitoses, and on the other hand, the hormone entered into the mechanism of integration for sustaining gravidity with mother organisms (LH effect).

# DISINTEGRATING EFFECT OF URETHAN AND CHORIO-GONADOTROPIC HORMONE ON THE REGENERATION OF PLEURODELES LARVAS

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We consider regenerative morphogenesis as a phenomenon related to embryonary morphogenesis, since both of them are featured by the differentiation of cells and their increasing integration. We assumed that factors which are effective against embryonary morphogenesis — as chorio-gonadotropic hormone (HCG) and urethan — must exert a checking effect on regenerative morphogenesis, too. For a test we examined the regeneration of tail and legs with *Pleurodeles* larvas. We had cut off 5 mm of the tail-end and the legs at the joints. The employed solutions were: 240  $\mu$ g/ml and 120  $\mu$ g/ml; urethan 1000  $\mu$ g/ml. We entirely exchanged the solution every 48 hour. Our observations were made once in every 24 hour by means of binocular microscope with citoplast system. In case of tail regeneration: in 24 hours an epithel will be sprouting on the wound, by 48 hours the regenerating blastema can be observed with the control-media, while in the case of HCG and urethan no regeneration is found. After 120 hours the regeneration of control-larvae was practically finished, the regenerated

tail-end is, however, different from that of the starting condition, it is not pointed and the laps are swollen. By the use of HCG the wound has been completely epithelized, although the regeneration has not yet started. Using urethan the blastema-formation on the axial part begins. In the instance of leg-regeneration: in 96 hours the regeneration with the control-media is due to start. Using HCG and urethan, however, only the epithelization of the wound-surface can by observed. By 216 hours the control-medium keeps on regenerating, while in the case of HCG and urethan we meet no further changes. 288 hours later the control-medium continues regenerating but the renewal is developing in some cases more intensely on both sides of the legs and in other cases on the axial part. Its extent may reach half of the thickness of the leg. In the case of HCG and urethan we find no changes. We obtained the same results with the tests of tail-end regeneration with *Rana viridis* larvae. Our experiments lead to the conclusion that disintegrating factors are able to check regenerative morphogenesis which implies that regeneration is also suitable for the study of factors of disintegration.

# DEVELOPMENT OF HETEROPLASTIC MOTOR END-PLATE, INVESTIGATED BY LIMB TRANSPLANTATION BETWEEN CHICK AND DUCK

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The wing and leg anlages of 4 days old duck embryos were transplanted to the place of the removed right wing anlage of chick embryos. In an other series the wing anlages of 4 days old chick embryos were transplanted on the 5th day of incubation into duck donors. The heteroplastic motor end-plates formed this way were studied with histochemical and impregnation methods. Normally in chick embryos the motor end-plates of the limb muscles are showing cholinesterase (CHE) activity on the 12th-13th day of incubation, while in duck embryos this reaction appears later, on the 15th-16th day. According to the present experiments the duck motor end-plates, when the muscle is innervated by chick motoric nerves are earlier CHE active. On the other hand, the heteroplastic chick motor end-plates are showing this activity a few days later. As the impregnation pattern indicated the innervation of the heterotransplantated limb muscles were found to be normal. There was a close temporal correlation on chick as well as on duck embryos between the appearance of CHE activity of the spinal motoric cells and the CHE activity of the limbic motor end-plates. This data is suggesting that the developmental stage of the spinal motoric nerve cells is determining the formation of the motoric end-plate and postsynaptic membrane.

# CHANGES OF GOMORI POSITIVE PERIVENTRICULAR GLIAL CELLS IN RATS OF DIFFERENT AGES

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Numerous glial cells containing Gomori positive granules are to be found in the central nervous system. These cells constitute a periventricular system around the cerebral ventricles, they are present in greater number around the third ventricle and Sylvian aqueduct. The significance of this system is unknown so far. In our present work we have studied the changes of periventricular glial cells in rats, newborn, 3 weeks (sucker), 2 months (adolescent), 5 months (adult) and 15 months old. The number of cells increases in the age of sexual maturity, their number is abundant in the brain of old animals. The localisation and distribution of these Gomori positive glial cells is characteristic of the different parts of the brain. We suppose that the changes of the Gomori positive glial cells, due to different ages, are connected to respective changes in functions of the nervous system according to different ages.

# CONTRIBUTIONS TO THE NEURAL REGULATION OF THE NUCLEIC ACID METABOLISM IN THE HEART MUSCLE OF MAMMALS

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Referring to the effect of the somatic nervous system on the nucleic acid content of skeletal muscles many data are known in literature. However, the effects of the vegetative nervous system influencing the nucleic acid metabolism in the heart muscle have not been yet studied. The present study was undertaken in order to determine the action of thoracic sympathectomy, vagotomy and total denervation upon the nucleic acid concentration in the heart muscle of the cat. The sympathetic denervation of the hearts was done in two phases according to the method of CANNON and BRITTON. In the case of parasympathetic denervation the chest of the animals was opened between the fourth and the fifth ribs on the right side and the n. vagus was cut through on both sides. In the case of total denervation nn. vagi were cut through in the second operative phase of sympathectomy. The nucleic acid concentration of the myocardium (in the left and right ventricle) was determined by means of U. V. spectrophotometry using the method of TSANEV and MARKOV on the 15th, 30th, 45th day respectively on the 60th and 75th postoperative days in the case of sympathectomy. From the results of the experiments it can be established that neither the partial nor the total denervation does not any significant change in the deoxyribonucleic acid (DNA) concentration of the heart muscle ( $\vec{P}$  0.1-0.05). The results show furthermore that sympathectomy has no effect on the ribonucleic acid (RNA) concentration in the muscles of ventricles, while the quantity of RNA is decreasing after vagotomy and total denervation. The decreasing of the highest degree can be observed about the first month (-25.18 per cent). The effects of total denervation are similar to those seen in the case of vagotomy.

# THE EFFECT OF LONG-LASTING DARKNESS AND ILLUMINATION ON THYROID HORMONE FRACTIONS CIRCULATING IN THE BLOOD OF ALBINO RATS

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We investigated in our experiments whether upon long-lasting light or darkness a change is to be noticed in the iodine level bound to serum protein as compared to the control animals kept under changing illumination according to the local time. We studied also the change occurring on the effect of the same factors in the distribution of the 131 J containing substances of the blood. It was stated that the protein bound iodine content of the three groups did not significantly differ. But it could be proved by analysis with radio-paper-chromatography that the change of the iodine content in blood appears in the form of shifting with in the iodine hormone fractions. So in animals kept in permanent darkness the thyroxine containing iodine isotope showed up to a greater extent, iodide, however, showed low values. The long-lasting illumination, on the other hand, decreased the activity of thyroxine to nearly one half, while the activity of inorganic iodide increased to more than the double. On the effect of long-lasting darkness the activity of monoiodotyrosine and diiodotyrosine greatly decreased, but their value increased twofold in permanent illumination. It was remarkable that triiodotyronine did not react to the changes of light. We suppose according to SCHREIBER that impulses arising from light effect in the retina of the eye, evoke an inhibiting reaction, when reaching the adequate hypothalamic area, on the synthesis of the "thyreotropin releasing factor" (TRF). Accordingly the tonic influence of the TRF on adenohypophysis decreases and so does the thyreotrop hormone secretion of the adenohypophysis, too. Therefore the iodine concentrating capacity of the thyroid gland diminishes and every process of the thyreogenic function will be reduced. As a result the plasma thyroxine level decreases and because of the insufficient function of the dehalogeneses di-and monoiodotyrosine appear in the circulation to an increase degree. The long-lasting darkness presumably has an activating effect on the pituitary function of the rat, the tonic influence of the TFR grows, so does the TSH secretion of the adenohypophysis and every process of the thyreogenic function increases as well.

# Abstracts of papers read by title only

# DATA ON THE ACCLIMATIZATION OF POLAR BEARS WITHIN THE TEMPERATE ZONE

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It was noted for many years, that polar bears in the Budapest Zoo just as in severa other Zoos within the temperate zone take hardly a bath during the winter season. After investigations conducted for a winter season it appeared that the bathing activity is closely related to the temperature of the water and the air: a greater bathing activity appears only when the weather thaws; by cold weather it ceases, or at least will be restricted to the slightest degree. This behaviour of polar bears seems to justify that these animals have to acclimatize themselves to the weather conditions of our temperate zone to a significant measure.

# INVESTIGATION OF PHOTODYNAMIC EFFECTS ON PROTOZOAN CYST SENSIBILIZED WITH DYES

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Strong visible light reduces or arrests the life activity of most cells and its harmful effect has been observed in Protozoa too. Platyophrya lata KAHL encysted in suitable culture medium after a longer period usually ceases to show any reaction under the effect of stronger light (50,000 lux); aggregates appear in the protoplasm and the animal dies. Anyway, the unstable protective and reproductive cysts show a peculiar response to light stimulus of 5,000 to 50,000 lux intensity; often a lively mitochondrial movement starts and protoplasmic flow is well observable; this is followed by the intrathecal movement of the animal in varying directions. The protoplasmic components of the otherwise translucent animal transform, according to AGROTTHUS DRAPPER's law, the molecularly absorbed light energy into chemical energy of mobilizing effect. In cysts stained with vital dyes (neutral red, methylen blue, Janus green-B etc.) this effect is much more pronounced, since the dyes as photosensibilizers intensify the photodynamic effects in an interesting way. The energy transferred from the dyes to the protoplasm shortens the latency period which is necessary to provoke the movements mentioned. Simultaneously it also changes the time of survival within the cyst. According to investigations carried out so far, the intensity of the light energy used, the quality of the dye absorbed and its molecular structure are important but not decisive factors. The age of the cyst (its sol-gel state) and its physiological state play a more important part in this respect.

# CYTOLOGICAL AND NEUROSECRETORY INVESTIGATIONS ON THE OPTIC TENTACLE OF PULMONATA

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The investigations were made on the species Helix pomatia, Cepea hortensis (Stylommatophora), Fagotia acicularis, Fagotia esperi, Galba truncatula and Limnaea stagnalis (Basommatophora). Especially the collar cells, the lateral oval cells and the lateral cells of processes of the optic tentacle were studied. Impregnations and special staining methods rendered possible to demonstrate the nervous character of these three cell types. These are bipolar neurons containing neurofibrils in their processes and Nissl-substance in their perikaryon. In *Helix pomatia* the neurits of the lateral oval cells and the lateral cells of processes are in close contact with the nerve fibres extending under the dermo-muscular layer of the tentacle. With paraldehydefuchsin and chrome-hematoxylin-phloxin stainings the authors demonstrated the neurosecretory activity of these cell types. In further investigations, the cytology, histochemistry, the cyclic character of the neurosecretion as well as the transport and giving off of the secretion were studied. The investigated Basommatophora species contain no similar cells in the eyeregion, or in the oral tentacles.

# DIFFERENTIATION OF THE TYPES AND LOWER CATEGORIES OF THE ALKALI SOILS OF THE SOUTHERN PART OF THE AREA BETWEEN THE RIVERS DANUBE AND TISZA ON THE BASIS OF THE VEGETATION

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In the course of the program of investigation of the alkali soils of the southern part of the area between the Danube and the Tisza author collected phytocoenological and soilecological data. Evaluation of these data made it possible to determine the alkali and semialkali soil types with the aid of their vegetation. Between the soil types, subtypes, variants and the phytocoenoses developed on them the following relations were demonstrated:

- 1 a) Very humid and deeply salty juvenile meadow soil: Astero-Agrostetum Eleocharis facies.
  - b) Humid and only on the surface salty juvenile meadow soil: Astero-Agrostetum normale.
  - c) Slightly humid and slightly salty juvenile meadow soil: Agrosti-Caricetum distantis Trifolium fragiferum facies and Festuca arundinacea facies.
  - d) Slightly humid and only on the surface salty meadow soil: Agrosti-Caricetum distantis Plantago maritima facies.
- 2 a) Humid muddy solontchak with a low hydrocarbonate content: Crypsidetum aculeatae normale
  - b) Humid solontchak with a high hydrocarbonate content: Suaedetum maritimae hungaricum
  - c) Humid solontchak with a high sulphate and chloride content: Salicornietum europeae hungaricum.
- 3 a) Humid solontchak-solonetz with a low salt content: Bolboschoenetum maritimi Agrostis alba subass. (Frequently transitions to the solontchak type.)
  - b) Humid solontchak-solonetz with a high salt content: Bolboschoenetum maritimi Puccinellia subass.
  - c) Drying up solontchak-solonetz with a moderate salt content: Lepidio-Puccinellietum Asterpannonicus subass.
  - d) Drying up solontchak-solonetz with a high salt content: Lepidio-Puccinellietum normale.
  - e) Dry solontchak-solonetz with a high salt content: Lepidio-Camphorosmetum normale, (Frequently transitions to the solontchak type.)

# SOME CHARACTERISTICS OF THE ABSORPTION OF HALIDE IONS BY WHEAT AND BARLEY

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The uptake of halides and their mutual effects in the uptake was studied in short time experiments on wide concentration range with excised roots and intact seedlings. From the experiments with excised roots the authors came to the conclusion that at least three concentration zones may be differentiated in the absorption of ions. (I) High concentration zone: 0.5-30 mM. In this range the concentration curve of uptake rises after a relative saturation in zone II. The competition is diminishing or completely ceasing among the similar ions, probably because of the participation of a non-active absorption process. (II) Medium concentration zone: 0.03-0.5 mM. The levelling part of the concentration curve with clear competitions among the related ions. (III) Low concentration zone: 0.001-0.03 mM. The concentration curve is steeply rising in this range and the competition abruptly comes to an end in the direction of the more diluted solutions. The raising of the plants and the experimental conditions deeply influence the concentration curve and the rate of competition in the three zones but only in a quantitative way; e.g. the K<sub>m</sub> of carrier system(s) involved in the uptake of Br<sup>-</sup>; varies between 0.013-0.058 mM. Special attention was devoted to the role of exchange adsorption processes and the gradient of absorption along the root. Finally on the basis of the result obtained with excised or segmented roots, the bromide absorption of intact plants was investigated in the three concentration zones.

# CONTRIBUTIONS TO THE RADIOLOGICAL INVESTIGATION OF THE BODY COMPOSITION OF MAN

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The soft X-ray radiographs make possible the radiological differentiation of the bones, adipous tissue and musculature on the basis of their different absorption capacity. The quantitative determination of the musculature and adipous tissue can best effectuated on the radiographs of the extremities, where the mineral content of the bones can also determined by the measurement of the breadth of the corticalis of the bones. We found that: kg of adipous tissue of the man = breadth of the adipous tissue in the height of the crista ilei in mm + 5 = 1.25breadth of the adipous tissue in the height of the trochanter major in mm + 7 = 6.0 breadth of the adipous tissue of the inner side of the thigh in mm + 5. We could determine a lot of similar equations for the same purpose. The variability of the body structure of different people and groups of people can naturally change the relations between the breadth of the adipous tissues on different regions of the human body. So we found different relative values in the Hungarian population as related to the Swedish data of LINDEGARD. The gain and loss of body weight changes especially the breadth of the adipous tissue. The mass of the musculature can be best determined with the aid of radiological investigation and the mass of the musculature of the whole body can be relatively simply measured and calculated. The changes of the musculature following sports and working activity can be best followed with radiological investigation. We found that the radiological investigation gives more accurate values than the generally used kreatinin-excretion urine test. The integration functions manifest themselves generally in the relative changes of the masses of the different tissues. The substratum of the metabolic processes are the quantitative relations of the different tissues. The propagation of the quantitative methods for the detection of the body composition, a method till now not utilized in Hungary also in the other fields of biology secures the unity of the standardized and unified biological investigation methodics and gives a relatively simple routine method for the determination of the fat and meat of the cattle in cattle-breading.

# EXAMINATIONS OF ION EFFECT ON ISOLATED HEARTS OF EDIBLE SNAIL. (HELIX POMETIA L.)

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Author studied the effect of  $BaCl_2$ ,  $CdCl_2$  and of  $NH_4Cl$  on the isolated hearts of edible snail. On the basis of these examinations it was found that an intensive pulse height, a dropin frequency and a rise in the tone occur due to the effect of the Ba ions. Initially the Cd ions, like the Ba ions, result in greater amplitude and an intensive frequency decrease whereupon the function of the heart ceases. The tone decreases due to the effect of the Cd ions. The  $NH_4$  ions produce a decrease in the pulse height together with a slight drop in the tone. Comparing these data with the results of other authors obtained from snails' intestine in the case of the same ions it can be stated that the antagonism of BaCl<sub>2</sub> and CdCl<sub>2</sub> concerning the change of the tone exists also in the case of the heart. The slight tone reducing effect of  $NH_4$ Cl contrary to BaCl<sub>2</sub> proves effective only temporarily. Taking into consideration the change of frequency and pulse height the antagonism of BaCl<sub>2</sub> and  $NH_4$ Cl is pronounced, however, no such thing can be noted as regards the heart in the case of CdCl<sub>2</sub>.

# EFFECT OF CONTINUOUS LIGHT AND DARKNESS ON THE 17-KETOSTEROID OUTPUT OF TESTICLE AND ADRENAL ORIGIN IN ALBINO RATS

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We observed during our experiments that the 17-ketosteroid output of the urine of albino rats kept in continuous light decreases considerably in the first days of the treatment compared with the control group kept under light conditions according to the local time. It reaches the mean value of the controls on the sixth day, increases gradually from that time on till the 18th day (25 per cent on the average), and keeps this increased level during 30 experimental days with slightly declining trend. In the animals kept in continuous darkness a considerable decrease could be observed in the ketosteroid output on the second day of the treatment: on the third day the increase has not reached the control values and from that day on a gradual decrease followed till the 16th day. In this period the mean value was about 30 per cent lower than the initial one. This reduced value increased slightly till the 30th day of the experiment. Analyzing the 17-ketosteroid output qualitatively and quantitatively by paperchromatography it was found that in animals kept continuously in light or darkness the quantity of the fractions of 11-hydroxyandrosterone, 11-B-hydroxyethiocholanolone, dehydroepiandrosterone, epiandrosteron, i-androstanolone and 5-androstene-3- $\beta$ -chloro-17on of adrenal origin has not changed. The quantity of 11-ketoethiocholanolone, ethiocholanolone and androsterone fractions of testicle origin increases significantly on continuous light effect, while during continuous darkness the quantity of the same fractions decreases as compared to the control groups. It can be concluded that in the regulation of androsterone production of the adrenals light does not play any part but influences greatly the function of the testicle-

## LICHENCOENOSIS ALONG THE RIVER TISZA

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#### SZEGED

In the course of potamobiological investigations of the River Tisza the author deals with the lichen-flora and epilith, epigae and epiphyton lichencoenose. The investigations were carried out both on the Hungarian and Yugoslavian flood-area of the Tisza.

Along the upper-, middle- and lower course of the river the height above sea level as well as the microclimatic factors of the environment under microclimatic conditions also cause essential differences in the structure of lichencoenosis. The coenose will show quantitative and qualitative mutations on account of humidity conditions and changing of light which may be evaluated in the structure, dinamics, total development and fragmentation of the associations, further in the appearance of vicarial species and in the varied difference of the number of characteristic species. New lichen associations are the *Endocarpetum pussilli* and the *Lecanoretum albomarginatae*. The loes favouring *Endocarpetum pussilli* coenosis is from the upper course of the Tisza in the area of Tokaj. The *Lecanoretum albomarginatae* grows on silicate containing stones, especially on andesites and on buildings made of concrete or cement. In both associations are prevailing the crustaceous lichen species. In the course of his investigation author took into consideration the growth of coenosis and the relations to succession. On the powland area of the investigations the lichen coating of the damms covered with andesit, bricks or cement were particularly studies. The following lichencoenose are worth mentioning: Aspicilietum calcareae, Caloplacetum citrinae, Caloplacetum murorum, Lecanoretum albomarginatae, Parmelietum conspersae, Physcietum teretiusculae, and Verrucarietum nigrescentis.

# ORGANIZATION CONDITIONS OF THE INFERIOR PISTIL OF THE SUNFLOWER (HELIANTHUS ANNUUS L.)

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In general, authors consider the organization of the inferior pistil to be of appendicular, axillary or intermediary nature. For such determination there are but few literary data available on the pistil of the family Compositae. We have examined the conditions of development of the inferior pistil of the sunflower, from the appearance of the flower primordium to the fully developed age. The flower primordia appear in a centripetal arrangement on the gradually enlarging, plate-like inflorescence axis. The first to appear on the edge of the young, flat flower primordia are the petal primordia, followed by the stamen primordia and, finally, by the carpel primordia. At the organization of the pistil, the style and the stigma primordia protrude, while the part corresponding to the ovary gradually sinks into the axial part, because, during the differentiation of the floral leaves, the axis (receptacle) becomes strongly distended due to the intensive divisions taking place in its upper third and includes the ovary cavity of the pistil. As far as the differentiation of the flower's fascicular system is concerned, we have observed that two main fasciculi get into the axial part that includes the ovary and are meeting in an annulus above the ovary. The conveying fasciculi of the floral leaves are starting upwards from this line, while downwards the ovary part of the pistil is covered with a network of several recurrent fasciculi.

On the ground of the above described organization, we consider the inferior pistil of the sunflower to be of axial origin.

## LOCAL HISTAMINE – ANTIHISTAMINE ANTAGONISM AND STORAGE

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Storage of intravenously administered dyes increases in the site of irritated tissue. Tissue irritations, tissue damage is accompanied by histamine release. Histamine transforms the endothelium of the small vessels into phagocytes which phagocytize the heterogeneous substances. Local storage of dyes is enhanced by intracutaneous histamine, stain is marked on the injection site of histamine. This phenomenon may be impeded with antihistamine substances. A white spot is present on the place of the intracutaneously or subcutaneously given Suprastin, following 72 hours after the intravenous injection of the dye. According to MELCZER local inhibition of storage is related with biophysical properties. He points to the fact that on the place of the administration of different agents causing anodic and cathodic block total inhibition of dye storage was observed. A white spot encircled by a red margin was seen on the injection site 72 hours after the simultaneous administration of i. v. congo red and s. c. Suprastin in rabbit. This proves that the formation of the white spot is not a static fact but the result of a dynamic occurrence. The red margin is the consequence of the peripheric presence of a greater amount of histamine displaced from the centre which is effectuated by antihistamine. The speed of storage, the intensity of the resulting spot, and the mentioned local phenomenon of histamine-antihistamine competition differ according to animal species. E.g. the effect of histamine is greater in guinea pig than in rabbit. The nature of the histamine induced phenomenon depends on the histamine content of tissues, the degree of histamine sensibility, the intensity of histamine breakdown, and the counter regulation of the organism. Likewise, local histamine-antihistamine competition is influenced by occurrences in the organism. Thus giving local Suprastin in acidification to rabbit produces a white spot, while in alkalinization an additional tissue proliferation may be observed.

# CORRELATION OF QUADRAT SIZE AND SPECIES DISTRIBUTION IN OPEN DOLOMITE ROCK SWARD (FESTUCETUM PALLENTIS HUNGARICUM)

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The examined material was sampled in the Kisszénás mountain group situated above Pilisszentiván, where the endemic plant Linum dolomiticum BORB, found exclusively in this region is living. The coenological survey comprised an area of about four square metres (m<sup>2</sup>). in which all individuals of each species were registered. Calculations, however, were confined to the following most frequently appearing five species: Linum dolomiticum, Festuca pallens, Fumana procumbens, Alyssum tortuosum, Anthericum ramosum. Subsequently the occurrence frequency of these species was established, first in the network of basic  $10 \times 10$  cm quadrats and referred then - by increasing the unit area - to basic quadrat networks of 200, 400, 800 square centimetres (cm<sup>2</sup>). The fidelity and the distribution pattern of the values thus obtained was established according to THOMPSON (1958). Anthericum and Fumana showed first random distribution turning into regular as a consequence of increased quadrat size. This points to an aggregation of individuals which may be due to spreading by rhizomes in Anthericum and in Fumana to accumulation of seeds by rainfall on the banks of steep slopes. As against, by increasing quadrat size the distribution of Linum turned from random first to regular and became later anew a random one. From this to two forms of aggregation can be concluded, probably because this plant may be propagated and spreads respectively by seeds and sprouts. Aggregations differ not only as to their origin but also in time. Alyssum and Festuca represent a transition between both plants previously mentioned. Disregarding the possibility of accumulation on slope banks the high seed yield explains in itself the aggregation of *Alyssum*. The reason of the two aggregation forms of *Festuca* may be found in the two age groups of individuals: the number of aged speciments producing high seed yield is low as against young ones inhibited in their development by erosion. Though the supposed correlation should be proved by further investigations, the results evidenced that data of coenological surveys are influenced both by requirements prescribed for ourselves and by environmental factors.

# HISTOCHEMICAL AND ULTRASTRUCTURAL FEATURES OF THE INTERCALATED DISCS IN MAMMALIAN CARDIAC MUSCLE

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Recent electronmicroscopical investigations prove the non-syncytial character of the cardiac muscle. It has been shown that the intercalated discs are specialized junctions between cellular units of the myocardium. Electrophysiological investigations published by SPERELAKIS suggest that there are no low resistance pathways for spreading of excitation between adjacent cardiac cells, thus the heart muscle can not be regarded as a functional syncytium either. In our previous works it has been shown that the intercalated discs display a well-defined acetyl-cholinesterase activity, probably associated with the conduction of excitation in a similar way as in the myoneural junction. In the present study acetylcholinesterase activity, PAS-positivity and a "lead-reactive substance", all characterizing peripheral synapses operating with a neurohumoral transmission mechanism, could be demonstrated histochemically in the intercalated discs both of auricle and ventricle muscles. The ultrastructure of the intercalated discs shas been investigated under the polarization microscope by means of Romhányi's precipitation anisotropic staining. It was found that the arrangement of protein and lipid parts of the intercalated discs of ventricular muscles. Further investigations revealed that the more complicated molecular organization of the intercalated discs in the ventricular muscle develop dinamically under the postnatal life by mechanical effects exerted by myofibrils.

# EXAMINATION OF THE CONDITIONS OF INFLORESCENCE AND FLOWER ORGANIZATION OF CORNUS SANGUINEA L., WITH SPECIAL REGARD TO THE PROBLEM OF THE INFERIOR OVARY

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Although several comprehensive works have already been published on the Cornaceae, the histology and histogeny of the flower of Cornus sanguinea have been analyzed but in a small number of papers. Even these were not unambiguous, particularly as far as the development of the inferior ovary was concerned. The author of the present paper has made her examinations on the usual microtomed serial sections, and in a stereo-microscopic visual field, with so-called clarified preparations. Due to this clarifying technique and to the staining of the flower with toluidine blue, the spatial arrangement of the conveying fasciculi and their correlation with one another became most lifelike and easy to be examined. The organization of the flower and, in close connexion, of the inflorescence has been observed from the phase of the reproductive shoot apex (June-August) to the full development of flower (May next year). It was found, that the three pairs of protective squamae covering the inflorescence bud at the beginning are transformed into assimilating leaves in next spring (naked bud). The basic type of the fully developed inflorescence is the corymb, further ramifications of which are ending in double and then in single umbrels. However, many combinations are possible within this pattern. The different floral regions (calyx, petal, stamen, pistil) are differenciating in acropetal order. The development of the pistil region begins within  $8-8\frac{1}{2}$  months after the beginning of the flower's reproductive phase. The carpels are of peltate organization, with a conspicuous crosszone (Querzone). In its growth, the latter keeps level with the dorsal sides of the carpels. The seed initial is differenciated out of the cross-zone (Querzone) and is of apotropic type. As for the much discussed problem of the inferior ovary (appendicular, receptacular theory), the author has examined, among others, some characteristic histological particularities, such as transverse fasciculi organized above the ovary. Compared with several characteristic marks, the latter can be considered as nodal level and is thus indicative of the axial origin of the tissues surrounding the ovary.

# HISTOCHEMICAL INVESTIGATIONS ON THE THYMUS OF LOWER VERTEBRATES

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Investigations have been conducted for the demonstration of mucopolysaccharides and nucleic acid in the thymus of fishes, amphibians, birds and mammals. Diastase-resistant, PAS-positive mucopolysaccharides appeared in the so-called atypical corpuscles, Hassall's bodies and cysts already in lower vertebrates. PAS-positive granules were encountered in the thymocytes, and electronmicroscopic studies revealed such granules in the epithelial cells of guinea pigs. Other investigations have shown that the amount of DNA and RNA in the thymocytes increases toward higher classes. It was found *in vitro* that it is the cells of the reticular epithelium which are mainly involved in the formation of cysts. The formation of Hassall's corpuscles begins with the detachment of macrophages of epithelial origin; meanwhile, a PAS-positive substance appears in the cytoplasm of the detached, rounded, swollen cells.

# THE SIGNIFICANCE OF THE HORIZON-CONSTRUCTION EXAMINATIONS IN THE ELUCIDATION OF THE LIGHT ENERGY CHANGES IN PLANT ASSOCIATIONS OF SEVERAL STOREYS

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In the elucidation of environmental factors the role of light presents a particularly serious methodical problem. The significance of this factor in the realization of the vital functions of the plants is thoroughly known but the measure, the rhythm of its manifestation in the plant associations could not be determined with direct measurements in a reliable way and to be generalized. This question must be still more carefully handled in the plant associations of several storeys because in these the effect of light is much more complex. When analyzing the light energy conditions we must measure the period during which direct radiation reaches the interior of the stock and that during which only diffuse radiation gets there, further the part of the radiation energy reaching in each aspect the lower (shrub, lawn, moss) storeys of the stock. Developing further the methods evolved in meteorological research work in Hungary and abroad we attempted to draw a parallel between the annual periodical changes of the vegetation and the light energy changes in the interior (different storeys) of the stocks. On a complex diagram we determine for the middle of each month the per cent values signifying the density of stocks and multiply them with the respective per cent values belonging to them according to a scale based on experience. The products are added to thus we obtain the portion of the sunshine duration possible in the open area or the radiation energy respectively which can reach the lower storeys upon the effect of the real horizon and of the shadowing impact of the crown and stems and branches of the trees, respectively. The method was tested in flood plain forest associations (Salicetum triandrae, S. albae-fragilis, Querco-Ulmetum hungaricum) as examples, expressing also the duration of sunshine getting into the individual stocks (hour) and the amount of radiation energy (Kcal/m<sup>2</sup>). In this way we can compare the plant associations with satisfactory accuracy also from the view-point of the annual change of the radiation supply.

# THE EFFECT OF NEUTRAL RED ON THE CELLS OF THE MALE ACCESSORY SEX GLANDS OF MICE

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Shortly (30 min.) after the administration of neutral red (0.25-0.50 mg/g) dye-containing granules appear in the cells of seminal vesicle and anterior lobe of prostate. The formation of dye granules is more intensive in the former organ than in the latter one. Their size, quantity and localization are similar to those of the lysosomes at the early stage of staining. In the cells of epididymis only a diffuse staining with neutral red can be observed. 2-4 hours after the administration of neutral red an RNA-containing material (the crinom) appears in the dye granules of the cells of seminal vesicle. The size of crinom granules increases gradually reaching a maximum in 8-16 hours. Later on, the crinom substance, together with the dyestuff disappears from the cells. The crinom formation in the prostatic cells begins later and is less intensive than in the seminal vesicle. The cells of epididymis are unable to form crinom. Besides RNA, the crinom substance contains a PAS positive material. Parallel with the accumulation of neutral red, the activity of acid phosphatase (detected by Gomori's method) in the lysosomes decreases. Likewise, there is a reduction of esterase activity in the epithelial cells of the seminal vesicle. The localization of dye granules, the PAS positivity of crinom substance and the decrease in the activity of acid phosphatase permit us to suppose that the lysosomes take part in the storage of neutral red in the cells of seminal vesicle and prostate.

# THE EFFECT OF PAPAVERINE ON THE TRANSPORT OF IONS AND WATER IN THE ISOLATED FROG SKIN

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Electrical potential and ionic currents were measured by the method of USSING and ZEHRAN, in the isolated frog skin (Rana temporaria, Rana esculenta). It was demonstrated that the addition of papaverin hydrochloride to the Ringer's solution bathing the inside of the skin, in varying concentrations (50, 100 and 200  $\gamma$  per ml) diminished the electrical potential. A correlation was found between the concentration employed and the decrease of potential. Ionic current in the skin, which is an identicative of active ion transport, showed a decrease parallel with the diminishment of transmembrane potential. The compound tested was found not to affect water transport: movement of water attained 2.37  $\pm$  0.10  $\mu$ l per hour per cm<sup>2</sup>, of skin surface in control experiments, the addition of 100  $\gamma$  per ml of papaverine to the medium resulted in a net transport of 2.80  $\pm$  0.76 and the addition of 200  $\gamma$  per ml was followed by the movement of 2.42  $\pm$  0.73  $\mu$ l of water. Oxygen consumption of the isolated frog skin was mesaured in Warburg's vessel and it was found that the addition of 100  $\gamma$  per ml of papaverine hydrochloride to the medium reduced oxygen consumption from 0.289  $\mu$ l to 0.120  $\mu$ l per hour per mg. The results obtained indicate that papaverine to affect water transport points out that the transport of ions and water in the skin is secured by different mechanism.

# COMPARATIVE INVESTIGATIONS OF THE EVALUATION POSSIBILITIES OF THE PIGEON CROP-SAC MICRO-TEST

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We compared the following possibilities of the evaluation of the pigeon crop-sac micromethod for quantitative determination the prolactin-content.

(a) Determination by measuring the extent of the reacting area (diameter or planimetrical measurement).

(b) Measuring the weight of the area showing the reaction.

(c) Measuring the proliferating epithelium of the crop-sac wall.

(d) Increase of the mitose number.

(e) Increase of the <sup>32</sup>P isotope intake of the reacting area. We stated according to determinations carried out on more than 400 pigeons that the labourintensive methods (c) and (d) demanding histological preparation, as well as (e) requiring isotope technique give in no way more exact results than the simple diameter measurement. We developed a new solution for the planimetrical measurement of the reacting crop-sac part. The crop-sac spanned always in the same way is dried and so a very easily measurable (by planimetry or weighing), at any time controllable, comparable and demonstrable preparate is obtained. We worked out calibration curves with the aid of the method and used standard prolactin-preparates for determining the unknown prolactin content. The determination could only be quantitatively evaluated if numerous adequate parallel measurements were made, otherwise it can be only used as a semiquantitative method.

# THE COMPACTNESS OF ULTRASTRUCTURE AND THE MECHANISM OF STAINING IN PLANT CELL WALLS

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The compactness of ultrastructure of the plant cell was studied by the use of staining methods. It has been assumed that the efficiency of staining depends not only on chemical binding but also on steric conditions: the size and form of intermicellary spaces of the cell wall play an important role in the incorporation of the dyemolecules. Results obtained with about 30 stains on *Tilia* cross sections are summarized. (1) Triphenylmethane derivatives. Staining good; selectivity is decreased when the molecule contains more than one primary amino group (fuchsin B). Staining is poorer with secondary and tertiary amines (malachite green) and the staining can be even completely removed during dehydration (methyl green, iodine green). The intensity of staining is reduced when a larger radical is coupled to the triphenyl methane group (e.g. phenyl, naphthyl radical: isamin blue, methyl blue). The good staining property is greatly impaired by the incorporation of a sulphone group into the molecule (fuchsin S). The lignified parts of the cell wall have a great affinity towards the small molecule of auramin. (2) Azo-derivatives. These compounds are preferentially incorporated into the cell walls of the pith. The presence of a sulphone group decreases the intensity of staining both in the lignified and in the cellulose wall. The naphthyl and hydroxyl groups of the molecule have the same effect (thiazin, Woodstain scarlet). The staining ability of the large molecules of azo-benzidine derivatives is poorer (Chlorazol-black). The primary amino groups improve the binding of the dye (Bismarck-brown). (3) Heterocyclic compounds. Of the pyran (dibenzopyran) derivatives pyronine B is an excellent dye for the lignified cell wall. Even the cellulose cell wall is stained by Rhodamin B, containing one more carboxyl group. Acridine orange and methylene blue, compounds of a smaller molecular weight stain the same elements with the same intensity. Safranin (phenosafranin) of the pyrazin group stains much more intensively than magdalared (naphthosafranin) which has the same composition. It is a general phenomenon that the elongated, chain-like molecules stain less intensively than the smaller globular ones (safranin, pyronin). The results indicate that the radicals of the molecule have a role in the incorporation of the dye but the shape and size of the molecule have a significant influence.

# THE EFFECT OF CHRONIC ATROPINE TREATMENT ON THE NUCLEIC ACID CONTENT OF DOG SUBMAXILLARY GLAND

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Adult male dogs were used for these experiments. Prior to the chronic atropine treatment, the left glands were removed and used as a control for quantitative nucleic acid assay. Atropine sulphate was injected subcutaneously twice a day over a period of 11 days. The dose given was increased gradually from 1 mg/kg to 11 mg/kg of body weight. The quantity of nucleic acids was determined with the aid of a U.V. spectrophotometer, at two wave-lengths. After chronic atropine treatment the wet and dry weights of the glands increased. There was 14 per cent increase in the RNA content (related to 100 mg of wet tissue) of the atropine-treated glands. The DNA content did not change considerably. The nucleic acid content of the whole gland showed the following changes: the RNA content of the atropine-treated glands exceeded that of the contralateral ones by 16 per cent; although there was a tendency for the DNA content to increase in the atropine-treated glands, the increase rarely exceeded 5 per cent. After chronic atropine treatment there was a significant increase as related to controls in the total nitrogen concentration (13 per cent) and in the total nitrogen content (21 per cent). In our earlier studies we have reported 32 per cent decrease of the total RNA content after transection of the chorda tympani. The present work is at variance with EMMELIN and his co-worker's observations according to which the chronic atropine-treatment may produce all the changes as follow the transection of the chorda tympani.

# HISTOLÓGICAL INVESTIGATIÓNS ÓN THE ADDÚCTÓR MUSCLES OF FRESH-WATER MUSSEL IN DIFFERENT FUNCTIONAL CONDITIONS

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The ability of the adductor muscles of Anodonta to both phasic and tonic contractions stood in the field of interest of the physiologists since PAVLOV. They were considered usually as smooth muscles and were investigated in order to find out the development and the maintenance of the tonic contraction. At the same time data on the histological structure of these muscles are contradictory, - some authors dealing with them as smooth muscles some others taking them for cross-striated ones but there exist also data suggesting that they are of spiral structure. Authors investigated the adductor muscles of Anodonta in different functional conditions. Their structures seemed to be in relaxation and isometric contraction like those of the smooth muscles, but in some relation also differed from them. When fixed at the beginning of the isotonic contraction they seemed to be of spiral structure and if they are fixed at an advanced stage of isometric contraction a periodicity of dark Q and light I bands can be observed, although they are not identical with the typical cross-striations. In authors' opinion these changes in the structure of the muscles correlating with the different functional-conditions serve as explanation for the origin of the contradictory opinions in the literature. Taking in consideration these properties the adductor muscle may not be ranged either among the smooth or among the cross-striated muscles, but also the name "spiral muscle" not justified. Authors propose to nominate it "polymorph muscle" and suggest that also in physiological investigations it should not be interpreted as a smooth muscle.

# THE OCCURRENCE OF CROCUS TOMMASINIANUS HERB. IN HUNGARY

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The Crocus discovered 1941 in the Querceto-Carpinetum near the village of Gyulaj, Tolna county (South'Eastern Transdanubia) was examined for years by A. HORVAT and S. JÁVORKA in an attempt to clarify its relations with the Crocus species occurring in Hungary in a very small number. At first they were of the opinion that it belonged to *Crocus vittatus* SCHLOSSER et VUKOT., later it was tought to belong to the C. albiflorus KIT.; eventually at the "Botanical Days" of 1949, JÁVORKA presented this beautiful crocus with light purple flower as a new variety under the name Crocus Heuffelianus HERB. var. Csapodyae HORVAT et JAV. a plant the three external laciniae perigonii of which look almost white from outside. Since the position of Crocus "vernus" (C. vernus WULF. + var. neapolitanus KER.-GAWL., C. albi-florus KIT., C. Heuffelianus HERB.) was far from being clarified and seemed rather complicated from the systematic point of view, JÁVORKA himself did not consider his description as final, and in the 1950's he continued to examine along with the author of this paper the crocus of Gyulaj together with the species of alpine C. "vernus", C. albiflorus and C. Heuffelianus of the Carpathians. It became clear only after the death of JÁVORKA that the crocus of Gyulaj does not belong either to the C. albiflorus or the C. Heuffelianus, but it is identical with the Illyrian species Crocus Tommasinianus HERB. This species which, owing to its beautiful flowers, is cultivated in a number of European gardens, can be found growing wild on the Dalmatian coast and occurs in Montenegro, Bosnia and Serbia. The discovery of the plant, in addition to the sub-Mediterranean Sedum neglectum TEN. ssp. Sopianae PRISZTER described recently from the Mecsek mountains by the present author, has increased the already high number of Mediterranean-Balcanic flora elements of the Southern Transdanubian floristic region (Praeillyricum) and herewith the particular southern character of the area referred to has become even more stressed.

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# IDENTIFICATION OF 3-HYDROXITYRAMINE CONTAINING GRANULES IN THE ADRENAL MEDULLA OF THE DOG

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In experiments with the identification of adrenaline and noradrenaline containing granules, we often found — in particular in the adrenal medulla of the grass snake and the dog – large granules  $0.5 - 1.5 \mu$  in size, with a fine microgranular structure, and surrounded by a membrane: in the majority of cases their electron density was smaller than that of the adrenaline and noradrenaline containing granules. Considering that in the inner part of these large granules there were often smaller granules of the size and density of the adrenaline and noradrenaline containing granules, and that in some cases they were just in the process of extrusion from the large granules, the genetical relationship between them seemed to be evident. It is assumed these large granules contain a precursor of adrenaline and noradrenaline. To prove this assumption the homogenate of the adrenal medulla of the dog was centrifuged after the sedimentation of the nuclear fraction, first with 3000 g for 30 minutes, and then the supernatant fluid with 24 000 g for 30 minutes. The electronmicroscopical control pictures demonstrated that 76 per cent of the large granules in question were sedimented in the first fraction. Biochemical and paperchromatographyc quantitation showed that 63 per cent of the total dopamine content of the gland was concentrated in the first fraction. On the basis of the facts described above it is suggested that these large granules contain dopamine.

# THE ONTOGENETIC AND ALLOMETRIC CHARACTERISTICS OF THE FORAMEN MAGNUM IN DOGS

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In the course of the systematic revelation of the cranial allometry in Canids author has examinated 950 dog- and 200 wild-canid skulls of the *Corneliana* as well as 350 dog- and 100 wild-canid skulls from other collections.

foramen magnum	ABSOLUTE MEASURES IN mm					
	width	mean	frequency	height	mean	frequency
adult	10-28	19	17 - 21	11 - 20	15	13-17
neonate and sucking	5 - 14	7	6-8	5 - 17	6	6

The comparative examinations proved the following:

Ratio of the two sizes 3:2-1:1, it occurs in dogs 2:3 (= bottleshape). The same in neonates 1:1-6:5 resp. 2:3!

The shape of the foramen magnum is very variable, basic forms are the horizontal ellypsis, the round-cornered rhombe, the circle, resp. the characteristic bottleshape. In neonates it is mostly circle, circle-like ellypsis, resp. the bottleshape which does not change much up to the wearing. The final form begins to develop only after 2 months age and is reached by the completion of the permanent teeth. The foramen magnum concerning its shape, absolute and relative sizes, proportions does not have either specific, or generic characteristics, not even sexual differences could have been stated. The only exception is that of the miniature dog-races where the bottleshape often occurs. The ossification of the bones forming up the foramen magnum happens presumably in two layers. Primary form is that of the bottleshape, since every foramen magnum looks from the braincase like that. In the course of development the desquamate ossification on the outer surface of the nuchale covers shield-like this inner embayment, closes it up from the outside and forms the final contour. This fact supports the theory of the juvenile standstill-stage in skulls of dwarf-dogs. There was not possible to demonstrate any allometric correlation between the foramen magnum and the skull as a whole, or the single bones building it up. It is the task of further investigations to detect the possible correlation between the absolute volume of the braincase and the cross section-area of the foramen magnum.

# EFFECT OF THE TREATMENT WITH ADENOSINE TRIPHOSPHATE ON THE NUCLEIC ACID CONTENT IN THE DENERVATED SKELETAL MUSCLES OF RATS

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It has been described by many authors that the nucleic acid and the protein content, as well as the quantity of high energy phosphate compounds show some changes in the denervated skeletal muscles. The interruption of the peripheral innervation is followed by a reduction of the quantity of adenosine triphosphate (ATP). In the present experiments the relation between nucleic acid and ATP content was studied in the denervated skeletal muscles after administration of Na-ATP. Albino rats (Epimys rattus var. albino) of either sex, weighing 120-150 g were used for the experiments. Under aseptic conditions a 10 mm part of the sciatic nerve was removed unilaterally to achieve the denervation of the gastrocnemius muscle. The contralateral m. gastrocnemius served as control. One group of the operated animals was injected with Na-ATP (i.m., 12 mg/day) once daily, the other group (control) was treated with isotonic NaCl solution for 20 days. The nucleic acid content of the denervated and the contralateral muscles was determined using U. V. spectrophotometry and that of ATP by means of viscosimetric micro-method on the 5th, 10th, 15th and the 20th postoperative days. The experiments have shown that the ATP dose employed by us does not give any change in the rate of muscular atrophy. The injected ATP enhances, however, the deoxyribonucleic acid content of the skeletal muscles, in this connection it increases the effect of the denervation, on the other hand it slows up the reduction of the quantity of the ribonucleic acid, thus it moderates the effect of denervation. In explaining these observations it must be taken into account that the adenosine triphosphate takes part in the synthesis of nucleic acids of both types. It is known from the assays done with radioactive isotopes that ATP enhances the incorporation of the labelled thymidine into the deoxyribonucleic acid, furthermore it can serve as an in vivo precursor for the ribonucleic acid synthesis.

# ZUR DIFFERENZIERUNG DER WACHSTUMSGESCHWINDIGKEIT BEI KINDERN UND JUGENDLICHEN UNTERSCHIEDLICHER ETHNISCHER HERKUNFT

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Unser Wissen über die körperliche Entwicklung im Kindes- und Jugendalter ist unzureichend. In die Breite gehende Vergleichsforschung auf Grund von Längsschnittuntersuchungen steckt in den Anfängen. Entwicklungskundliche Forschung wird nicht ihrer selbst halber betrieben, sondern um tätig in das Entwicklungsgeschehen eingreifen zu können, soweit es von außen her einer Steuerung bedarf und zugänglich ist. Das Verhalten der Geschlechter in den Gestaltwandelphasen erfordert besondere Aufmerksamkeit. Mit Hilfe eines Index, dessen Komponenten die Körperhöhe und das biologische Lebensalter in Monaten sind, hat Rajkai eine Bewertung der erreichten Körperhöhe in der Entwicklungszeit vorgenommen. Vom Verf. wird der Frage nachgegangen, ob sich mit Hilfe dieses Index gegenüber einer bloßen Betrachtung der sich ändernden Körperhöhenwerte vertiefte Einblicke in das Entwicklungsgeschehen bei Meßgruppen ethnischer, regionaler und sozialer Heterogenität gewinnen lassen. Dabei findet der Sexualdimorphismus besondere Berücksichtigung. Zum Vergleich werden zwölf Mittelwertstabellen hrangezogen, von denen eine allerdings nur männliche Werte enthält. Von diesen Mittelwertsreihen stammen vier aus Europa, drei aus Amerika, vier aus Asien und eine aus der Südsee. Die aus den entsprechenden Vergleichen abgeleiteten Schlußfolgerungen werden mit ausführlicher Begründung des eingeschlagenen Weges vorgetragen. Zugleich wird die Behauptung Portmanns widerlegt, daß erst der Pubertätsschuss die auffälligen Größenunterschiede hervorbringe, die ein wichtiger Teil der menschlichen Rassenmerkmale sind.

# COMPARATIVE EXAMINATIONS CONCERNING THE STRUCTURE AND INNERVATION OF THE HARDER GLAND

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The Harder gland of different species in reptiles, birds and mammalians showed many anatomical differences in their forms, but as regards the function in our opinion the great divergences of their size is more significant. We could find very large glands in the water snakes, pondtortoises and among water-fowls. The size of the glands is not proportionate with that of the third eye-lid to which the gland is anatomically connected. Seemingly the functional correlation is stronger to the way of life in particular to the water life resulting in large glands and as a consequence in greater product of gland secretion.

The histological structure and the innervation of the gland were examined with routine staining methods and silver impregnation according to BIELSCHOWSKY – ÁBRAHÁM. The structure of the secretory and chamber the quality and quantity of production as well as the pigmentation of their tissue differ in the examined species. On the basis of comparative examinations we drew the following conclusion: the serose Harder glands (in most birds especially birds of pray) shows the compound tubular type and the fat-like content of the gland is produced in compound alveolar ends (most part of reptiles, water-birds, rodolents, pigs). The quality of the product is connected with the circumstances of life and the quantity with the age of the animals and the daily and seasonal rhythm. Pigmentation of the glands is general in reptiles, birds, except the domestic species but very rare in mammalians.

The nerve fibres of the Harder glands originate from the sympathetic ganglia situated in the connective tissue capsule of the glands. The fibres are unmyelinated and form a very rich plexus in the interlobular tissue and around the secretory cells. As neither the anatomical nor the histological examinations could verify the entrance and participation of the parasympathetic oculomotoric nerve, we assume the secretory innervation of the gland only to be sympathetic. The blood vessels especially the main artery of the gland possess an own nerve supply mostly thin (sympathetic and parasympathetic vegetative) and a lesser number of thick (sensory trigeminic) fibres in it. We found a close nervous connection between artery and gland-duct.

# ANTHRACOTOMIC INVESTIGATIONS ON CHARCOAL REMAINS FROM A PALAEOLITHIC FIRE-PLACE FOUND AT BODROGKERESZTÚR (HUNGARY)

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In autumn 1963 the remains of a palaeolithic fire-place were discovered by L. VÉRTES on the Henye-Hill (about 350 m above sea-level) near Bodrogkeresztúr (Hungary). From examinations of the excavated flint implements L. VÉRTES could state these to be of the Gravettien-type, with some Aurignacian characteristics. Part of the fossils (bones) found have been identified by M. KRETZÓI as the relics of mammoth, giant deer and horse bones. The charcoal remains (altogether 154 pieces) have been examined microscopically by the present author and found to be the remains of the category *Larix-Picea*. This double denomination has been used by the author for a number of years since he had shown that there was up till now no evidence proving a reliable xylotomic distinction to exist between these two genera. The pure *Larix-Picea* charcoal set points to the fire-place to originate from one of the glacial periods; from cross-checks on the archeological and palaeontological information it would seem safe to put their origin to the Würm<sub>2</sub>-period.

# ELECTRO-SHOCK AND ELECTRO-NARCOTIC SYMPTOMS ON EXPERIMENTAL SLAUGHTER ANIMALS

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The stunning with electric current of the slaughter animals (horse, cattle, hog) was experimented with for decades. Till now the applied instruments and methods (low voltage current) did not give satisfactory results, because they caused only electrical blows of various strength. This method has several drawbacks (multiple extravasates; spasmodic, only slowly resolving muscle rigidity) and it is also inhuman. In 1963 the experiments with cattles already pointed to the significance of the amount of the electric charge, dependent upon the body-resistance of the animals. Namely also the repeated, "accumulated" electric-blows with the moderated, subtransformed current-energy caused shock-like effect appearing only after longer time and very irregularly. The experimental appliance according to FOLTÁNYI-SZÜCS-BOHN, which makes possible a change on a broad range of the components (intensity, current-voltage, periodicity, wave-form and duration, etc.) of applied electric-energy, on the abt. 10,000 test-hogs used up to now has caused successful electro-shock (ES) respectively electro-narcotic (EN) symptoms. From these experiments it may be stated that: (1) with animals (cattle, hog) similarly as with man it is necessary for sufficient ES respectively EN to use enough electric charge, which may be determined - up to a certain limit - also in Watt/second. (2) the ES and the EN may be brought about on the animals also with a sufficient charge-mass and with a higher periodic-rectangular-formed current. (3) the ES is essentially the excitement of the central nervous system (spasmodic effect) caused by a moderate electric blow. (4) EN is a blocking or disconnecting respectively, which hinders the normal spreading of the action- (biologic-) currents in the brain, for a while. This disconnecting manifests itself in total unconsciousness, insensibility and in total relaxation of the muscles.

# SOYBEAN SEED ALBUMINS AS TRYPSIN INHIBITORS AND THEIR EXAMINATION BY PAPER ELECTROPHORESIS

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Out of defatted soybean meal a protein solution was made with an aqueous extraction using distilled water and at pH 4.5 glycinin was precipitated, while the solution thus left was purified with salting out (using ammonium sulphate), dialysis and electrodialysis. During electrodialysis some precipitation was obtained, this was centrifuged and rejected. From supernatant solution the albumins were precipitated at  $-10^{\circ}$ C with acetone and the precipitate was dried with acetone and ether. The electrophoretic migration was made on a 5 cm wide Macherey-Nagel 214 filter paper (300 V, 1-2 mA/strip). As electrolyte a veronal buffer (pH 8.6) was used, jonic strength was 0.03. A hydrodynamic liquid flow was created — against electrophoresis — by way of raising the buffer level of the anode vessel. The preparations of soybean albumins were separated into 9 fractions (I-IX). The paper electrophoretic separation of albumins is promoted by the liquid flow that works against the electrophoretic particle-movement. The protein zones migrate against an opposite flow which is continually increasing. The albumin fractions impede the proteolytic activity of trypsin. The electrophoresis performed after the incubation of trypsin and albumins corroborates that albumins react with trypsin. The paper electrophoretic investigation of the reaction of trypsin and albumins is made possible by the circumstance, that in an alkaline buffer (pH 8.6) the trypsin has positive electric charge, i.e. moves towards the cathode. In such circumstances the albumins move in the opposite direction, towards the anode. If the molecules get bound, the product thus becomes conspicuous. These products are albumin-trypsin compounds. Three albumin-trypsin compounds appeared on the electropherogram and are located among the free components which move towards the cathode and anode.

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 $i \in \mathbb{N}$ 

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