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SOMMAIRE

PETRU M. BĂNĂRESCU, Vladykov's contribution to the knowledge of the fish fauna of the Danube river basin	91
VICTORIA TATOLE, Données comparées sur le polymorphisme chromosomal à l'intérieur du genre <i>Glyptotendipes</i> Kieff	97
VIOREL ȘTEFAN, Some studies on the relations between Enchytraeidae and soil microflora	101
I. ROȘCA, V. BRUDEA, ELENA BUCUREAN, FELICIA MUREȘAN, I. ȘANDRU, ANGELA URSEA and M. VOICU, Researches on the behaviour of <i>Ostrinia nubilalis</i> by the use of pheromone traps, as related to sterile insect release technique	105
N. MIRANCEA and DORINA MIRANCEA, The ultrastructure of preimplantational embryos used in the biotechnology of embryotransfer	117
ȘTEFAN AGRIGOROAIE and ION NEACȘU, Considerations on the mechanism of the cell membrane stabilization	123
J. MADAR, NINA ȘILDAN, A. D. ABRAHAM and ANA ILONCA, <i>In vitro</i> effects of procaine and procaine-related drugs and metabolites upon the glucose uptake by rat brain slices	129
P. ROTINBERG, SMARANDA KELEMEN, AL. SAUCIUC and [P. JITARIU], The biosynthesis antibiotic preparation A 37.4 — a new active cancerostatic agent	135
LILIANA VASILIU-OROMULU, MIHAELA MĂNESCU, V. SANDA, A. POPESCU, GABRIELA FIȘTEAG, MIHAELA PAUCĂ-COMĂNESCU, AURICA TĂCINĂ, M. FALCĂ, [VICTORIA CĂRAÇAȘ], VIORICA HONCIUC and C. ARION, Characterization of the main biocenotic components of some natural and planted forestry ecosystems from the Letea bank (Danube Delta)	139

DORINA NICOLESCU, L'évolution multiannuelle du plancton bactérien dans les écosystèmes aquatiques du Delta du Danube et du secteur prédeltaïque	151
VICTOR ZINEVICI et LAURA TEODORESCU, L'évolution de la structure gravimétrique du zooplancton dans les écosystèmes de type lacustre du Delta du Danube sous l'impact du processus d'eutrophisation (1975—1987).	155
GH. BREZEANU und R. POPA, Beiträge zum Kennen der Struktur der Jungfischpopulationen aus spezifischen trophischen Zonen des Donau-Deltas	169

VLADYKOV'S CONTRIBUTION TO THE KNOWLEDGE OF THE FISH FAUNA OF THE DANUBE RIVER BASIN

PETRU M. BĂNĂRESCU

V. Vladykov has worked only for three years in the Subcarpathian Russia, a comparatively small area of the Danube River basin; his contribution to the knowledge of the fish fauna of this province, and of that of the Danube basin in general was essential, especially in respect of lampreys (*Eudontomyzon danfordi*), *Leuciscus souffia*, the genus *Gobio* (four species) and Cobitidae (description of the genus *Sabanejewia*). Much of what he wrote on these taxa retains also now its validity.

The well-known Canadian ichthyologist V. D. Vladykov began his scientific activity in Ruthenia or the Subcarpathian Russia, then a province of Czechoslovakia, now a part of the Ukrainian Soviet Republic; he studied in the University of Prague where he graduated with a *Rerum Naturalium* Doctor degree in 1925. Three years later he moved to Paris, then to Canada. Although he lived only for three years in the Subcarpathian Russia, that is drained by the upper Tissa River (the main tributary of the Danube), his contribution to the knowledge of the fish fauna of that province, and in general of the Danube River basin, was essential and retains also now its validity. Twenty of his 290 published papers and manuscript reports (17) deal with the fish fauna of the Subcarpathia, three of these, among which a major monograph (36) having been elaborated or finished when he was already in France.

For becoming aware of the value of Vladykov's contribution to the knowledge of the fish fauna of the Danube it is necessary to give a general review of the history of ichthyological investigations in that basin.

After the pioneering work of Marsiglius (25), Kramer, Schaeffer and Grossinger (36), an active period of investigations began in the early 1800s, due mainly to the Austrian and partially Hungarian authors; this first period culminated with Heckel and Kner's well known book on the fishes of the former Austrian empire (20) and with the more critical one of Siebold on the fishes of whole Central Europe (30). A second period lasted from 1864 to 1908, during which only minor contributions have been published; the most prominent Austrian ichthyologist of that period, Steindachner, worked mainly on extra-European fishes and on West-Balkan ones, having published a single important contribution to the Danube basin fish fauna: the description of *Leuciscus polylepis* from subtributaries of the Danube in Croatia (23). During the following period (1909—1924) the dominant figure in the Danube basin ichthyology was Antipa, whose book on the fishes of Romanian (1) was one of the first in the world literature which gave not only descriptions, but also comprehensive data on the biology of fishes and first-quality illustrations.

During the inter-war period (1922—1940) the most prominent students of the Danube basin fish fauna have been Vladykov in the Subcarpathia, Drensky and especially Chichkoff in Bulgaria. These were the first to adopt the modern nomenclature for genera such as *Rutilus*, *Leuciscus* a.o.; Vladykov and Chichkoff the first ichthyologists in the Danube countries to base their descriptions on biometry, measurements and counts from specimens of the investigated area, Vladykov the first to use the ternary nomenclature.

Vladykov's contribution to the Danube basin fish fauna concerns only the fish fauna of the Subcarpathian Russia, a rather small area, encompassing only 12,653 km², e.g. about 1.6% of the surface of the entire Danube River basin. It is drained by the upper Tisza River and ten of its tributaries. The fish fauna of few other areas in the basin of the Danube has been as thoroughly investigated as that of the Subcarpathia; in his main work (36), Vladykov mentions the occurrence, or absence, of each species in the Tisza River, in each of the 10 large tributaries and in the Serniye swamps. The fish fauna of the area is rich; it consists of 47 native species (a lamprey and 46 bony fishes); Vladykov mentions a single allochthonous species, *Salmo irideus* (right name: *S. gairdneri*); nine or ten others may have been introduced later. The area is covered by large forests, the Tisza and its tributaries have a large discharge of water; both facts concur in favouring the existence of several inhabitants of the upper reach of rivers, such as *Hucho hucho*, *Thymallus thymallus*, *Leuciscus souffia* and both *Cottus* species. These are restricted to the eastern part of the province. On the other hand, the Tisza being a large slowly running river, the inhabitants of the lowland rivers were able to ascend along the Tisza to the Subcarpathia: *Acipenser ruthenus*, *Pelecus cultratus*, *Abramis sapa*, *Gobio albipinnatus* (listed by Vladykov as hybrid), *Stizostedion (Lucioperca) volgensis*, *Gymnocephalus (= Acerina) schraetser*: in few if in any other areas of the Danube basin do these lowland species reach as far upstreams as in the Subcarpathia.

Vladykov's systematic and faunistic contributions concern lampreys, three genera of Cyprinidae (*Rutilus*, *Leuciscus*, *Gobio*), one of each Cobitidae (*Sabanejewia*), Percidae (*Perca*) and Cottidae (*Cottus*) Lampreys. Vladykov was the first author working in the Danube basin countries who realized that none of the two western-central European species, *Lampetra fluviatilis* and *L. planeri* is present in the Danube basin and that the native lamprey belongs to another species which he described in 1925 (31) as a new one: *L. bergi*; he stated later (36) that this is a synonym of *Eudontomyzon danfordi*, described in 1911 by Regan after specimens in the British Museum. Vladykov has also been the first ichthyologist from the Danube countries who became aware of the description of this species; Chappuis (15, 16) has been the second. *E. danfordi* is unanimously accepted now as a Danube basin endemic, its range actually encompassing only a restricted part of this basin: the basin of Tisza River and of a few other Danube tributaries close to the Tisza. The genus *Eudontomyzon* comprises some further species, one or two of which are present in the Danube basin (*E. mariae* and *E. vladykovi*, some authors consider the latter a synonym of the former) and in those of other rivers flowing into the Black and Baltic seas (it is worth men-

tioning that not in the rivers of the Caspian Sea encatchment area), one in southern and western Greece and the last one in Korea; *Eudontomyzon* is the only genus of lampreys displaying a western/eastern Palaearctic disjunction (14).

Rutilus. Vladykov (35) assigns the Subcarpathian populations of roach to a new subspecies, *R. rutilus carpathorossicus*, characterized by a lower number of scales in lateral line: 39—42, rarely 43 (as against 44—45, rarely 41, 46 or 47 in the western European nominal subspecies); the same low number of scales is present throughout the Danube basin (2, 9, 21); *carpathorossicus* is therefore accepted as a valid taxon by many authors (2, 14, 9), except Holcik and Skorepa (21), who concluded that the average number of scales is subject to a strong, rather irregular and clinal variation and it is not possible to delimit formal subspecies. However, the character found by Vladykov occurs throughout the Danube River basin.

Leuciscus. Vladykov (36) ascribed the *L. leuciscus* populations of the Subcarpathia to a distinct geographic race, *natio roulei*, assuming that this is present throughout the Danube basin. Actually, the differences he found between the specimens from the Subcarpathia and those from the Baltic Sea basin cannot be generalized for the populations from the Danube tributaries in Romania; *roulei* is presently not ascertained as a valid taxon (6).

Vladykov's main contribution to this genus is to have established the occurrence of *L. souffia* in the upper Tisza and its tributaries. The occurrence of this species in the right tributaries of the Danube from Bavaria, Austria and Yugoslavia has been known for more than a century; the species is however absent from the right tributaries in eastern Yugoslavia, Bulgaria and from the left tributaries in Slovakia, Hungary and most of Romania. Vladykov (36) mentions its occurrence in the upper Tisza and five of its tributaries; the Subcarpathian specimens are characterized by 53—58 scales in lateral line, as against 50—56 in those from the upper Danube. The occurrence of *L. souffia* in the right tributaries of the upper Tisza River in the Subcarpathia has been confirmed by more recent authors (18, 27); it has been found also in two of the three left tributaries from Romania (and is probably present also in the third one (11, 22, 9). The Romanian specimens have 52—61 scales. The differences in number of lateral line scales justify the erection of a new subspecies for the populations from the tributaries of the upper Tisza, both the right ones (Subcarpathia) and the left ones (Romania). This will receive the name *Leuciscus souffia vladykovi*, while the populations from the right tributaries of the Danube (Bavaria, Austria, Yugoslavia) retain the subspecific name *L.s. agassizi*.

Gobio. Vladykov was the first author in the Danubian countries who realized that there were four (not two) species of gudgeons in the basin of this river (although he identified one of them as a "hybrid"); he contributed to the systematisation of each of them and also made general comparative remarks, which retained their validity up to now. In the key of the species, he mentioned, as distinctive character, the occurrence of epithelial keels on the dorsal scales in the "hybrid *G. gobio*

G. persa carpathorossicus" (actually *G. albipinnatus*) and in *G. persa carpathorossicus* (actually *G. kessleri*) and their absence on those of *G. gobio* and *G. uranoscopus*. It has later been found that these keels are present not only in the three western Palaearctic species (*G. albipinnatus*, *G. kessleri* and *G. persus*) but also in three eastern Asian ones; this is the main distinct character of the subgenus *Romanogobio* (7, 12). Vladykov has also shown that *G. uranoscopus* is not closely related to *G. kessleri* as accepted by most earlier authors; he delimited a "proles" comprising *G. uranoscopus* and *G. ciscaucasicus* (this corresponds to the subgenus *Rheogobio* in the recent acceptance) and another comprising *G. persus* and *G. carpathorossicus* (i.s. *G. kessleri*) — the present-day subgenus *Romanogobio*.

Vladykov described the *G. gobio* form in the Subcarpathia as a new subspecies, *G. g. carpathicus*, pointing out differences between this form, and the upper Danube one (*G.g. obtusirostris*). Later studies have demonstrated that the species is subject to strong local variability in the Danube basin and a single subspecies can be accepted, *carpathicus* being a synonym of *obtusirostris* (7).

Vladykov described in 1925 (31) a new gudgeon species, *G. frici*. He realized later that it was only a geographical form (natio) of *Gobio uranoscopus*, differing from the nominal subspecies in its smaller eye and shorter barbels. The other populations from the basin of the middle and lower Danube basin are similar to *frici*; this is now accepted as valid subspecies (5, 10).

In 1925 Vladykov also described the new subspecies *G. uranoscopus carpathorossicus*; he realized later (36) that this was unrelated to the true *G. uranoscopus*, being much more similar to *G. persa* from eastern Transcaucasia and especially to *G. kessleri* from the Dnjester; he lumped these three nominal subspecies in a single polytypic species, using the name *G. persa carpathorossicus* for the form of the Subcarpathian Russia; he considered that all Danube basin populations belong to this subspecies. It has been later shown that *kessleri* and *carpathorossicus* differ from *persa* not only in having eight (as against seven) branched dorsal rays, but also in the number and shape of circum-peduncular scales; this fact justifies the acceptance of a distinct species for the Danube and Dnjester basins forms. It has been shown that the species is not homogenous in the Danube River basin, the populations from the easternmost tributaries in Romania are identical to the the Dnjester form (typical *kessleri*), those from the south-western Romania are ascribed to a distinct subspecies, *G. k. banaticus* while those from the Subcarpathia and north-western Romania, corresponding to Vladykov's *carpathorossicus* are intermediates between *k. kessleri* and *banaticus*, being closer to the former (4, 7, 12); *carpathorossicus* is considered now a synonym of *k. kessleri*, although it is not quite identical to this.

Vladykov's main contribution to the Danube basin gudgeons is the description of a presumed hybrid between *G. gobio* and *G. p. carpathorossicus* (= *G. kessleri*). He has asserted that this hybrid is quite frequently met with, reaches sexual maturity and spawns; he formerly believed this to be a distinct species ("nous pensions pouvoir le considérer comme une espèce distincte" (36, p. 287), but because it is morpho-

logically an intermediate between its two presumed parents he finally described it as hybrid. This form has later been raised to specific status by Fang (19), while Bănărescu finally (2) proved that it is conspecific to *G. belingi* from the Dnjeper (actually a synonym of *G. albipinnatus* from the Volga basin). The Danube basin populations are now ascribed to a distinct subspecies, *G.a. vladykovi* (2, 7, 12). This is the most frequent species of gudgeons form inhabiting the main channel of the Danube, from Vienna downstreams, (37), the middle and lower Tisza and the lower reach of their large tributaries in lowlands (3, 7, 9).

Cobitidae. Vladykov described in 1925 (32) a new species of loach, *Cobitis montana*; he ascribed it, later on, to a new genus, *Sabanejewia* (34), after having mentioned a peculiar type of sexual dimorphism in this species, quite different from that found in *Cobitis* (33); he also realized that *montana* is a junior synonym of *Cobitis balcanica* of Karaman and that this is closely related to the Trans-Caspian and Transcaucasian *S. aurata* (36). More recent authors realized that *aurata* and *balcanica* actually are conspecific, the first name having a priority. The species *S. aurata* is subject to a strong geographical variation throughout its wide range and especially in the basin of the Danube; four subspecies are present in this basin, some of them are connected by intergrading populations and include morphological distinct groups of populations (13). The populations of the upper Tisza and its tributaries in the Subcarpathia, which have been described by Vladykov as *C. montana* are identical to those from the rivers of western Romania flowing into the Tisza, being to a certain degree intermediate between the *balcanica*-populations from the Transylvanian Plateau and the subspecies *bulgarica* that lives in the Danube proper and in the middle and lower Tisza. This Subcarpathian-western Romanian group of populations is also almost identical to the *S. aurata* population from the Vardar River, in Macedonia, the type locality of *balcanica*; hence, *montana* can in no case be accepted as a subspecies.

Sabanejewia has been considered by some authors (14, 9) as subgenus of *Cobitis*; it is now almost unanimously accepted as a valid genus (28, 29), being the only genus of the family restricted to the western Palaearctic. It comprises, besides the polytypic *S. aurata*, a second species in the Danube basin, two in the Caucasus and one in North Italy (24). Its main distinctive character is the sexual dimorphism, first described by Vladykov (33).

Perca. According to Vladykov (36), the perches of the Danube basin belong to a distinct geographical race of second order: *P. fluviatilis* natio *vulgaris* Schaeffer. This is presently not accepted as a valid taxon, although there are some differences between the populations from the Danube basin and those from western Europe (Dr. B. B. Collette, in lit.)

Cottus. Vladykov (36) ascribed the Subcarpathian populations of *C. gobio* to a new geographical race, natio *pellegrini*. Their presumed distinct characters are not present in other populations from the Danube basin (8) and *pellegrini* cannot be accepted as a valid taxon. A more important contribution of Vladykov to sculpins is the finding of *C. poe-*

cilopus in the northern (right) tributaries of the upper Tisza; the species has been found later in the southern (left) tributaries (22). *C. poecilopus* was formerly known only in the part of the Danube basin east of the Carpathians.

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DONNÉES COMPARÉES SUR LE POLYMORPHISME CHROMOSOMIAL À L'INTÉRIEUR DU GENRE *GLYPTOTENDIPES* KIEFF

VICTORIA TATOLE

A partir de l'inventaire des restructurations chromosomiales connues (données originales et fournies par la littérature) chez plusieurs espèces du g. *Glyptotendipes*, on met en évidence dans quelle mesure le critère caryologique répond aux exigences imposées d'une manière différenciée par les catégories délimitées dans la hiérarchie taxonomique: espèce, genre, catégories couramment utilisées en diagnose.

La citodiagnose d'une espèce s'achève seulement après que les caractéristiques du caryotype standard ont été complétées par les aspects du polymorphisme chromosomal décelés chez les différentes populations des confins de l'espèce. C'est pourquoi nous avons considéré opportun d'entreprendre une analyse caryologique comparée, qui a eu comme point de départ l'inventaire des restructurations chromosomiales connues chez plusieurs espèces du genre *Glyptotendipes*: *barbipes*, *glaucus*, *paripes*, *gripekoveni*, *viridis*.

Dans cet ordre d'idées, par la comparaison de nos données (5, 6) à celles offertes par la littérature (1, 2, 3, 4), nous avons essayé de mettre en évidence dans quelle mesure le critère caryologique répond aux exigences imposées d'une manière différenciée par les catégories délimitées dans la hiérarchie taxonomique: espèce, genre, catégories utilisées couramment dans les diagnoses.

Le tableau I comprend des inversions chromosomiales établies dans les quatre chromosomes polyténiques des glandes salivaires des larves du IV^e état appartenant aux cinq espèces du genre *Glyptotendipes*. D'une

Tableau 1

Types d'inversions chromosomiales décelés à l'intérieur du g. *Glyptotendipes* Kieff

<i>Glyptotendipes</i>							
<i>barbipes</i>				<i>glaucus</i>	<i>paripes</i>	<i>gripekoveni</i>	<i>viridis</i>
URSS*	Canada*	Germany*	Romania***	URSS**	URSS**	Romania***	Romania***
I S1	I S1			I R6	I ?	I B17	IID 9-C13
II L1,2	II L1,2	II L1,2	II C1-2		II R10-11	II C9-11	IV G5
III S1	III L3	III L1	IV G1-2	III R7-8	II L4-5	III F8-9	
III L1	III S1,2		IV G5	III L2	III R6	IV G6	
IV L				IV L1	III R6-9		
				IV L3			

Miseiko et al. 1971
Belianina, 1982

** Miseiko et al. 1974

*** Tatole, 1980, 1985

SOME STUDIES ON THE RELATIONS BETWEEN ENCHYTRAEIDAE AND SOIL MICROFLORA

VIOREL ȘTEFAN

Four species of Enchytraeidae: *Enchytraeus albidus* Henle, *E. bigeminus* Nielsen & Christensen, *Lumbricillus lineatus* Müller and *L. rivalis* Levinsen were used in our experiments. The observations have been carried out on living material. The rate of speed at which food passed through the worm's gut was tested. The worms were bred in aquarium-like glasses and oatmeal was their main food. Simultaneously some worms were bred in sterilized conditions on agar where microorganisms such as *Pseudomonas* sp. and *Rhizobium* sp. were developed in cultures. Bacterial heaps have been never selected by worms; if they occasionally entered the gut, the bacterial cells are not damaged during their passage through the worm's gut. Moreover, they were concentrated in the last part of the worm's gut and remained active after elimination from the gut. So Enchytraeidae play an important role in the bacterial dissemination and concentration into a limited space. This phenomenon is very important in biological processes of organic matter decomposition.

The importance of Enchytraeidae in the biological processes in soil has been of a great interest for a long time. There are very few observations on the feeding of Enchytraeidae. The importance of soil Enchytraeidae prompted this investigation of their food consumption. Jegen (1920) showed that they feed on plant debris, which they mix with mineral particles. Clark (1949) found in Australian woods Enchytraeidae feeding on plant debris with high quantities of fungi mycelia.

Zachariae (1963) found in coniferous litter that the Enchytraeidae consume Collembola excrements, which consist of litter, but sometimes they fragment plant debris which lacks cellulose. Zachariae (1964) asserts that Enchytraeidae consume litter inhabiting bacteria, but they have no importance in bacterial dissemination. Reynoldson (1939) observed that some Enchytraeidae species such as *Lumbricillus lineatus* Müller, *Lumbricillus rivalis* Levinsen and *Enchytraeus albidus* Henle, consume large quantities of Cyanobacteria (*Phormidium*), and are very important in their decomposition.

The Enchytraeidae in laboratory conditions are able to reduce fresh water plants to an amorphous dark-brown mass; the largest part consists of worms faeces. Ivleva (1969) fed *E. albidus* Henle with potatoes and noticed that the food was not completely digested, during its passage through the gut. This author pointed out that Enchytraeidae consume large quantities of bacteria, too.

This paper presents some studies on Enchytraeidae and the soil microflora.

MATERIAL AND METHODS

In the experiments we used the following Enchytraeidae species: *Enchytraeus albidus* Henle, *E. bigeminus* Nielsen & Christensen, *Lumbricillus*

rivalis Levinsen and *Lumbricillus lineatus* Müller. The worms were bred in the laboratory conditions in special aquarium-like glasses on coarse sand as substratum. The food consisted of oatmeal "a porridge" applied on large pieces of glass, placed on the top of the sand. Sometimes oatmeal was mixed with potatoes. Microorganisms could enter freely and develop on the cultures. Simultaneously, other worms were kept in pure agar cultures of *Pseudomonas* sp., *Rhizobium* sp. and blood breeding bacteria. The worms were studied "in vivo" under coverslip with optical microscopy. The gut contents and faeces were studied, too. At the same time the oatmeal was searched under coverslip for bacteria and fungi, with optical microscopy light. The formalin fixed worms were included in paraffin, sectioned and histochemically stained. The oatmeal used as food was treated in the same way. The following histochemical stain methods have been used: (1) Lillie's Azur A Eosine B, for fungi and bacteria, (2) Brown-Brenn, for Gram positive and Gram negative bacteria, (3) Mallory's variant of Weigert method for bacteria and fungi, (4) Gram stain, (5) Ziel - Nielsen, Malachite-green for sporulating bacteria. The sections were examined with optical microscopy. Estimates of the time required for oatmeal to passage through the gut were made.

RESULTS

1. OBSERVATIONS ON LIVING WORMS

During the microscopical study of worms, we noticed that the gut was filled with oatmeal. Periodically, the worms were eliminating some faeces which showed a large number of living microorganisms, mostly bacteria, yeasts and only a few fungal hyphae. The bacteria released from the gut seem to be unharmed and they are able to move very actively under coverslip. The presence of living bacteria in such a high number in the faeces led us to the question: are bacteria consumed by worms or not? Therefore, this gut content study was desirable as well as that of food movement and its passage through the gut. Firstly, the natural bacterial fluorescence was tested and proved positive. The passage through the gut of the bacteria could not be observed in fluorescent microscopy, because the chloragogen cells have high natural fluorescence as compared to bacteria.

According to our microscopical study of living worms a membrane covering the faeces has been demonstrated for the first time. This membrane holds faeces together for a time after the elimination (Figure 1 a). It is possible that this membrane initially lined the gut. In an attempt to demonstrate the origin of the membrane we used Pass Alcian blue stain. On the other hand, the speed rate of food passage through the gut was investigated. In this respect we tested more than 200 worms (Table 1) belonging to the four species. Charcoal was used as marking material mixed up with oatmeal, which entered the gut in the feeding process. Its movement along the gut could be observed in the living material under coverslip. Figure 1 b shows the charcoal in the gut. During the testing period we observed that the food passes very quickly through

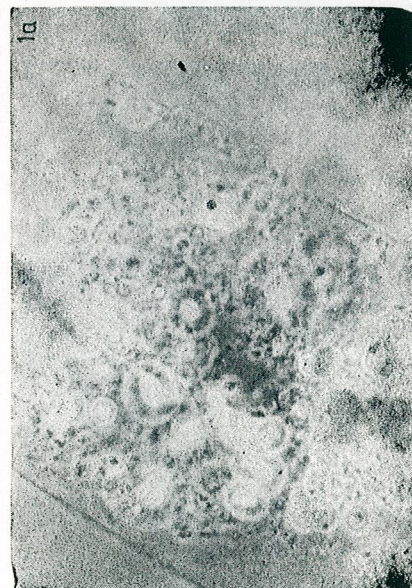
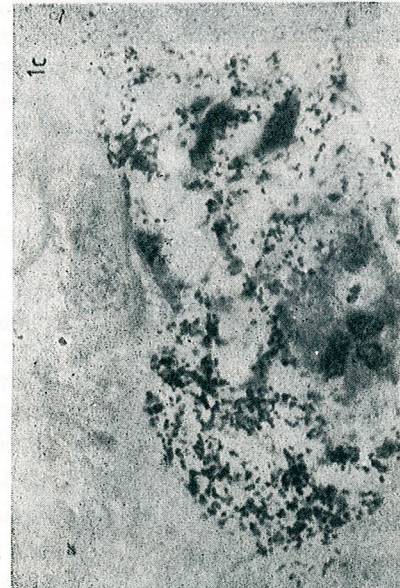
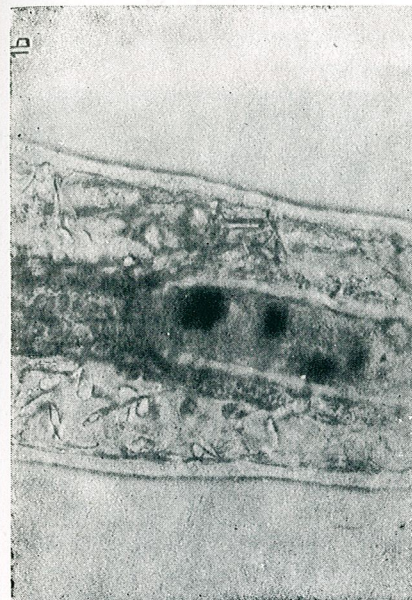


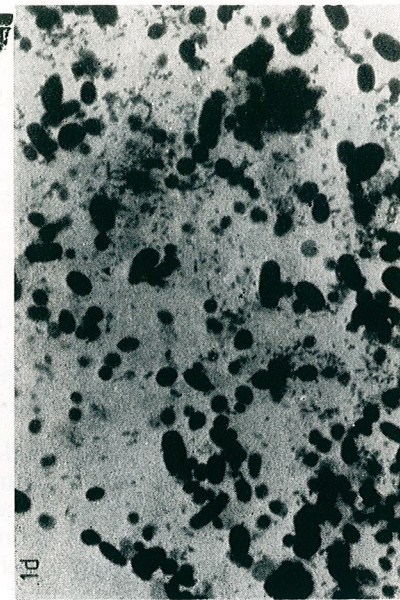
Fig. 1 a. — Faeces covered by membrane (V. Ștefan);



c. — Oatmeal containing bacteria in the histochemically stained sections of gut (V. Ștefan);



b. — Bits of charcoal in the worm's gut, used for testing the speed of passage of food (V. Ștefan);



d. — Yeast and bacteria in histochemically stained sections of oatmeal used as food (V. Ștefan).

the first part of the gut. The movement becomes slower and at its end portion (last ten segments) the content being concentrated. The speed of the food passing through the gut is shown in Table 1.

Table 1

The passing of food through the gut

Species	No. of body segments	Time of passing food in minutes
<i>Enchytraeus bigeminus</i>	24-31	37-70
<i>Enchytraeus albidus</i>	31-41	35-95
<i>Lumbricillus lineatus</i>	35-42	35-90
<i>Lumbricillus rivalis</i>	50-60	85-165

Daily time for starting the experiments was 10³⁵ h.

2. STUDY OF THE FOOD AND GUT CONTENT IN HISTOCHEMICALLY STAINED SECTIONS

Examination of histochemically stained material showed the presence of a high number of Gram negative and Gram positive bacteria, a lot of yeasts and a small number of fungi hyphae. The frequency of bacteria in the gut seems to be nearly the same as in food and food cultures. In this respect we consider of great importance the fact that bacteria are not damaged during passage through the gut. Figures 1 c and 1 d show strained material from food cultures and from food in the gut.

The concentration of the bacteria in the food in the last ten segments (Figures 2 a, 2 b and 2 c) suggests that Enchytraeidae are of a great importance in the bacterial dissemination in the soil.

The concentration of the yeasts is different; they are in a very high number in the food, but low in the last part of the gut and in the faeces.

3. BREEDING WORMS IN BACTERIAL AGAR CULTURE CONDITIONS

In agar culture conditions, *Pseudomonas sp.* and *Rhizobium sp.* grow very well, they could be seen even with the naked eye, forming little heaps on the agar surface. The repeatedly washed worms, which were put into the cultures and studied for a long period, give us a new idea about bacteria as food for worms. We noticed that the worms never select preferentially bacteria as food. The worms penetrating the agar substratum near its surface fill their gut with it; at the same time a few number of bacteria occasionally reach into their gut. Observations on the living worms from the agar cultures demonstrated the elimination of living bacteria in the faeces patches. Bacteria in small number in the worms' gut are concentrated in the last segments of the worms' gut. This becomes evident when worms were sectioned and histochemically stained (Fig. 2 d).

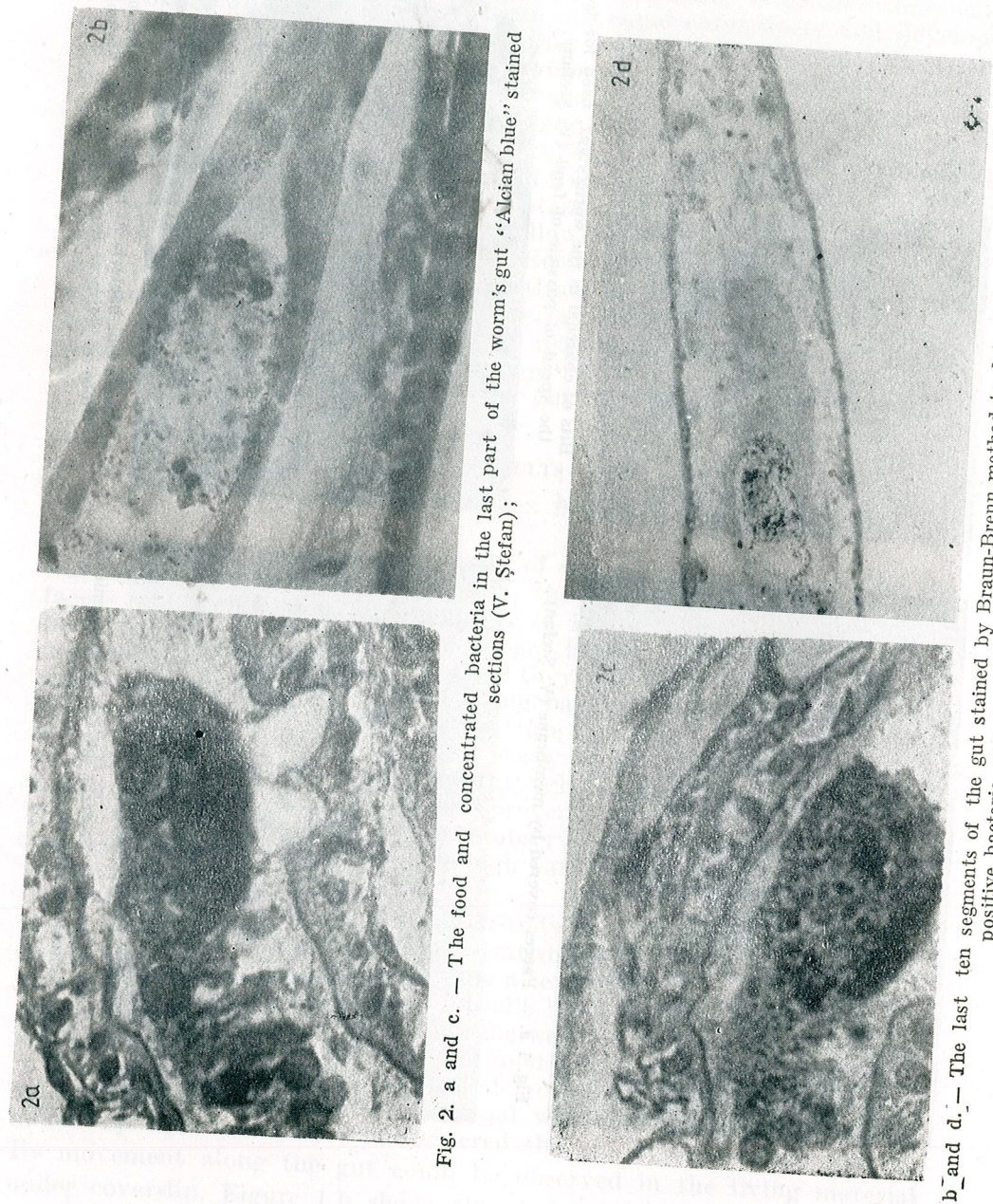


Fig. 2. a and c. — The food and concentrated bacteria in the last part of the worm's gut "Alcian blue" stained sections (V. Ștefan);

b and d. — The last ten segments of the gut stained by Braun-Brenn method to detect Gram negative and Gram positive bacteria, worms grown in agar cultures (V. Ștefan).

4. DISCUSSIONS

There are a lot of bacteria, yeasts and fungi hyphae in the worm culture glasses. They enter the worms gut together with the oatmeal they feed on. We have never observed worms especially selecting the bacteria.

After a long term study on food and worm gut content we noticed that the faeces always contain a high number of living bacteria. As concerns yeasts, they are in a high number in oatmeal, but very few in the gut and the faeces. Microorganisms which are eliminated in faeces are living and active. At the last part of the gut, microorganisms are in a high concentration, much higher than in the anterior or middle gut. This phenomenon could be noticed, both in living, or sectioned and histochemically stained worms, in both oatmeal or agar cultures. We consider of a great importance the fact that Enchytraeidae concentrate microorganisms in their gut, which are active after being eliminated. Therefore, worms are feeding on food not on bacteria inhabiting there. This fact seems to be of a great importance in the bacterial dissemination. These processes play a high role in the concentration of a large microorganisms species coexisting in a limited space. Therefore the worms, in this way, hasten decomposition of organic matter.

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RESEARCHES ON THE BEHAVIOUR OF *OSTRINIA NUBILALIS* BY THE USE OF PHEROMONE TRAPS, AS RELATED TO STERILE INSECT RELEASE TECHNIQUE

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Lures tested showed attractiveness to *Ostrinia nubilalis* Hb. males. Among our variants tested, the greatest number of captures were recorded with E₅. In Romania, the pherotypes *cis* (Z) and *trans* (E) of *Ostrinia nubilalis* Hb. exist.

The pheromone lures for the maize borer can be used for drawing up flight curves of this pest.

The European corn borer (*Ostrinia nubilalis* Hb.) is a major pest in all maize cropping countries. In Romania it is considered the most important pest of maize after the panicle appearance, being extended to all maize cultivating zones in the country. Populations of *O. nubilalis* of 14,000-40,900 larvae/ha were recorded, followed by a yield decrease between 1.3 and 17.7%, in dependence upon year and zone (16), however losses can sometimes reach 40% (15).

Under the ecological conditions prevailing in Romania this insect exhibits a unique generation per year, except for the southern areas, where a second partial generation occurs, noted at present to an extent of some 20% from the population of the first generation (8).

To control ECB application of an integrated control system shows importance, a particular part being played by cropping practices and application of some chain-loops of the biological control. The measures suitable for restricting the outbreak of this pest refer to cropping some resistant hybrids (2, 3, 4, 5, 6, 7); likewise, stress is laid on enlisting the auxiliary fauna, having possibly an outstanding role in reducing the pest populations (18, 19), at the same time performing investigations on the use of microbial products (9, 21), or of the parasite *Trichogramma* (13, 14, 17).

Special attention was paid during the recent years to the study of synthetic sex pheromones (20), and since 1988, when research related to the sterile insect release technique was approached, the pheromones have been used in investigations on the pest biology and its dynamics.

Since the isolation of the sex pheromone of *O. nubilalis* by Klun (11), all subsequent works showed that ECB males respond to a mixture of *cis* (Z) and *trans* (E) isomers of 11-tetradecenyl acetate (12, 22).

Previous field experiments carried out in various European countries led to the assumption that *cis* (Z) pherotype of *O. nubilalis* is also present in Romania (1).

At present, the main problem facing us is to find some specific and effective pheromone variants and to establish the pherotype or pherotypes of *O. nubilalis* occurring in Romania.

REV. ROUM. BIOL.-BIOL. ANIM., TOME 35, N° 2, P. 105-115, BUCAREST, 1990

MATERIAL AND METHODS

Investigations started in 1982 with various mixtures of Z:E isomers at various ratios, produced by the Institute of Chemistry Cluj-Napoca. F₁-type traps (10) were used in replicates, 50 m apart each other. The adhesive based on polyisobutylene produced by I.C.C.N. was employed.

The pheromone lures were removed at intervals of 1 week to 1 month, while their adhesive parts were changed weekly, when the number of *O. nubilalis* males/trap was also recorded.

RESULTS AND DISCUSSION

Following the preliminary results obtained in 1982-1986, 4 pheromone variants were established E₅; Z 11-14 OAc + E 11-14 OAc at a 97/3 ratio, H, and J; Z-11-14 OAc at a 97/3 ratio, and I; Z 11-14 OAc + E 11-14 OAc at a 3/97 ratio + tetradecenyl acetate, which showed promise.

The results in Tables 1, 2, 3 and 4 exhibit the relatively high number of *O. nubilalis* males caught during the whole experiment period, this number varying in dependence of locality, year and pheromone variant

Table 1
Number of *Ostrinia nubilalis* males caught/trap in 1987

Locality	Period	Type of pheromone			
		E ₅	H	J	I
Lovrin	13.V - 8.IX	12.25	17.75	12.5	30.5
Fundulea	4.VI - 17.IX	30.0	24.5	20.5	21.25
Valu Traian	4.VI - 17.IX	28.75	31.5	48.5	23.0
Podu Iloaiei	24.VI - 3.VIII	10.0	14.25	15.5	0.5
Turda	1.VI - 25.VIII	4.0	1.75	7.25	-
	Σ	85.0	73.75	104.25	75.25
	X̄	17.0	14.75	20.85	15.05

Table 2
Number of *Ostrinia nubilalis* males caught/trap in 1988

Locality	Period	Type of pheromone			
		E ₅	H	I	J
Lovrin	6.V - 2.IX	15.35	9.66	5.33	3.00
Fundulea	6.VI - 6.IX	28.00	35.00	28.00	3.5
Valu Traian	2.VI - 28.IX	57.00	43.33	13.66	8.33
Oradea	2.VI - 25.VIII	36.00	37.00	34.33	37.66
Podu Iloaiei	8.VI - 15.VIII	20.66	3.66	4.33	0.66
Turda	20.V - 29.VIII	5.66	1.66	1.33	0.66
Suceava	16.VI - 1.IX	7.66	3.00	1.66	0.66
	Σ	170.31	133.31	88.64	54.47
	X̄	24.33	19.04	12.66	7.78

Table 3
Number of *Ostrinia nubilalis* males caught/trap in 1988

Locality	Period	Type of pheromone			
		E ₅	H	I	J
Lovrin	13.VI - 12.IX	32.5	-	32.25	-
Fundulea	24.V - 14.IX	50.75	40.25	28.5	5.25
Valu Traian	17.VI - 13.IX	37.75	38.5	9.5	12.5
Oradea	13.VI - 31.VIII	27.00	26.00	22.00	11.5
Podu Iloaiei	1.VI - 17.VIII	42.5	-	42.0	-
Turda	22.V - 11.IX	101.66	86.33	27.33	4.0
Suceava	15.VI - 31.VIII	11.35	9.00	4.5	0
	Σ	303.41	200.00	166.08	33.25
	X̄	43.34	40.0	23.72	6.65

Table 4
Average number of *Ostrinia nubilalis* males caught/trap in 1987-1989

Locality	Type of pheromone			
	E ₅	H	I	J
Lovrin	20.02	13.70(*)	16.69	16.75(*)
Fundulea	36.25	33.25	25.66	10.0
Valu Traian	41.16	37.77	23.88	12.19
Oradea	31.5(*)	31.5(*)	28.16(*)	25.58(*)
Podu Iloaiei	24.39	8.95(*)	20.61	0.58(*)
Turda	37.1	29.9	11.97	1.55
Suceava	9.45(*)	6.00(*)	3.08(*)	0.33(*)

(*) - two-year results

used. It is to stress that the occurrence of both pherotypes of this species *cis* (Z) and *trans* (E) was ascertained for the first time in Romania, the latter being absent in the central and north-eastern part of this country.

Occurrence of pherotypes in Romania has raised the problem of detecting presence of some hybrid populations, and of zone delimitation between pherotypes. Generally, the *trans* (E) pherotype includes a less numerous population, even in the zones where it is present.

Among the *cis* (Z) variants, the pheromone variants H and J manifested an oscillating attractiveness, in dependence of year and locality, whereas the E₅ variant ranked constantly in the first place as to the number of target-males captured.

Specificity of pheromone formulations was not optimal, as in traps the species: *Tortrix viridana* L. in afforested zones, *Etiella zinckenella* Tr. and *Emmelia trabealis* Scop. in the other zones were frequently caught, however these are easily distinguishable species from the target-species with a little experience.

The pheromone traps record the flight of *O. nubilalis* males, and their flight curves can be drawn up both in the first and second generation (Figs. 1, 2 and 3).

With a view to enhancing pheromone effectiveness, in the first step the pheromone lures were removed at 1, 2, 3 and 4 weeks intervals. From data in Table 5 it is to note that the number of captures dropped parallelly with ageing lure. Since 1989, lure removal at two-week intervals has started in the whole research network.

Table 5

Influence of duration of use of pheromone lure on the number of males captured at Fundulea (E_5 pheromone variant)

Lure removed after :	No. of males/trap	
	1987	1988
1 week	53.75	40.25
2 weeks	50.5	31.25
3 weeks	30.5	13.5
4 weeks	28.25	15.25

As to the influence of the trap height on the number of captured males, Table 6 shows that a 2 m height led to lessening captures. Stockel (23) stated that a 2.4 m height is adequate to trace the second generation of the pest. Our results are, possibly, due to the fact that the second generation generally appears in Romania at the end of August and throughout September and is devoid of significance, being reduced numerically, on the one hand, and during this period maize begins to ripen, on the other hand.

Table 6

Influence of trap height on the number of males captured at Fundulea

Trap height	Pheromone variant	No. of males/trap	
		1988	1989
1 m	E_5	28.0	50.75
	H	35.0	40.25
	J	28.0	28.5
	I	3.5	5.2
2 m	E_5	7.5	9.25
	H	6.0	8.75
	J	17.0	3.5
	I	1.5	0.25

Trials with males labelled with alkali fuchsin or fluorescent dyes, though successful in laboratory rearing cages, failed in the field. Of the 600 males labelled with alkali fuchsin in 1987 only 10 were recovered and could be identified in the 12 traps oriented to north, south, east and west, at 25, 50 and 75 m from the releasing point, while in 1988 only 7 males were recaptured from the 400 ones labelled with red fluorescent dye.

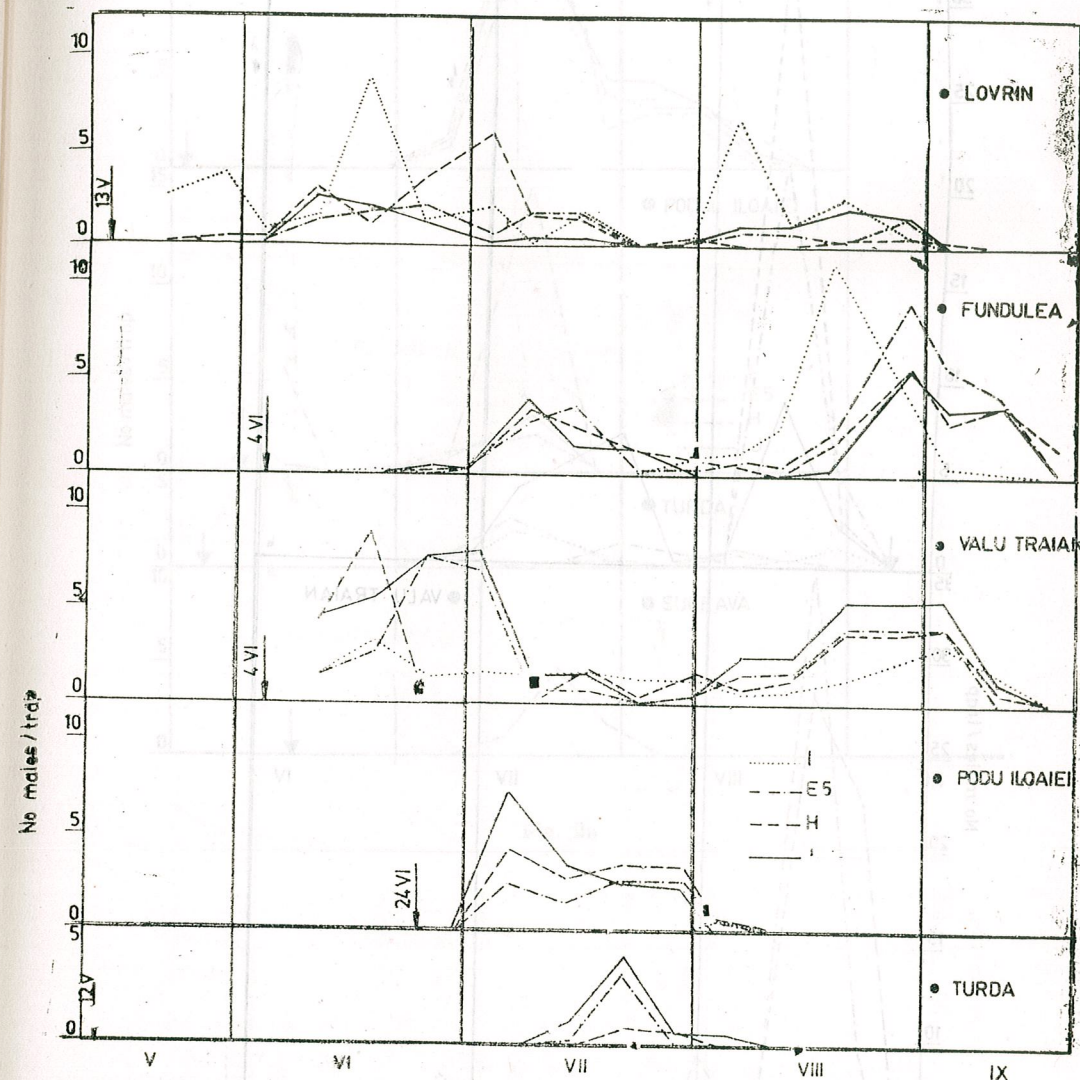


Fig. 1. — Flight dynamics of *Ostrinia nubilalis* Hb. males, as revealed from captures with pheromone traps at Fundulea in 1987.

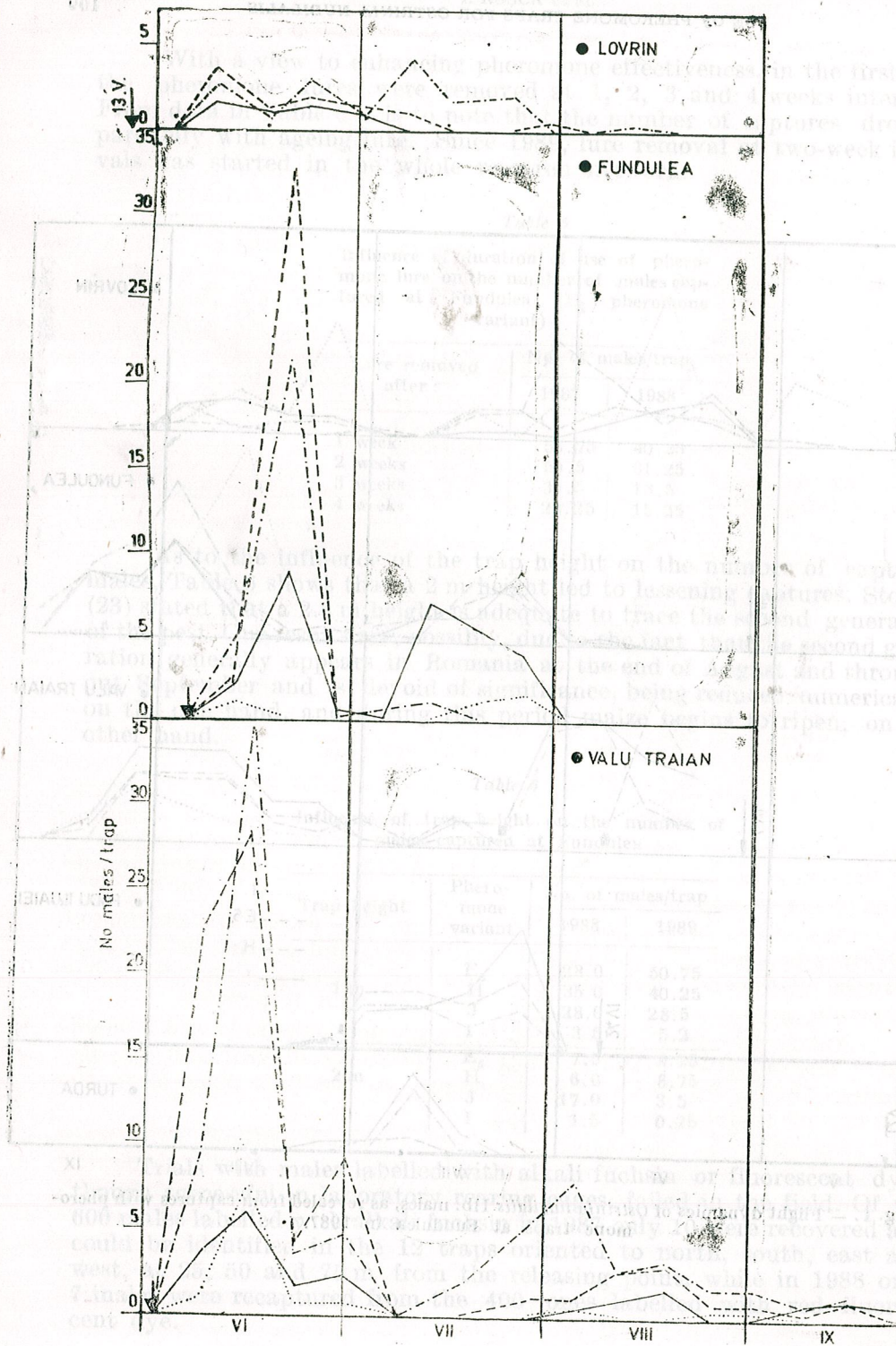


Fig. 2a

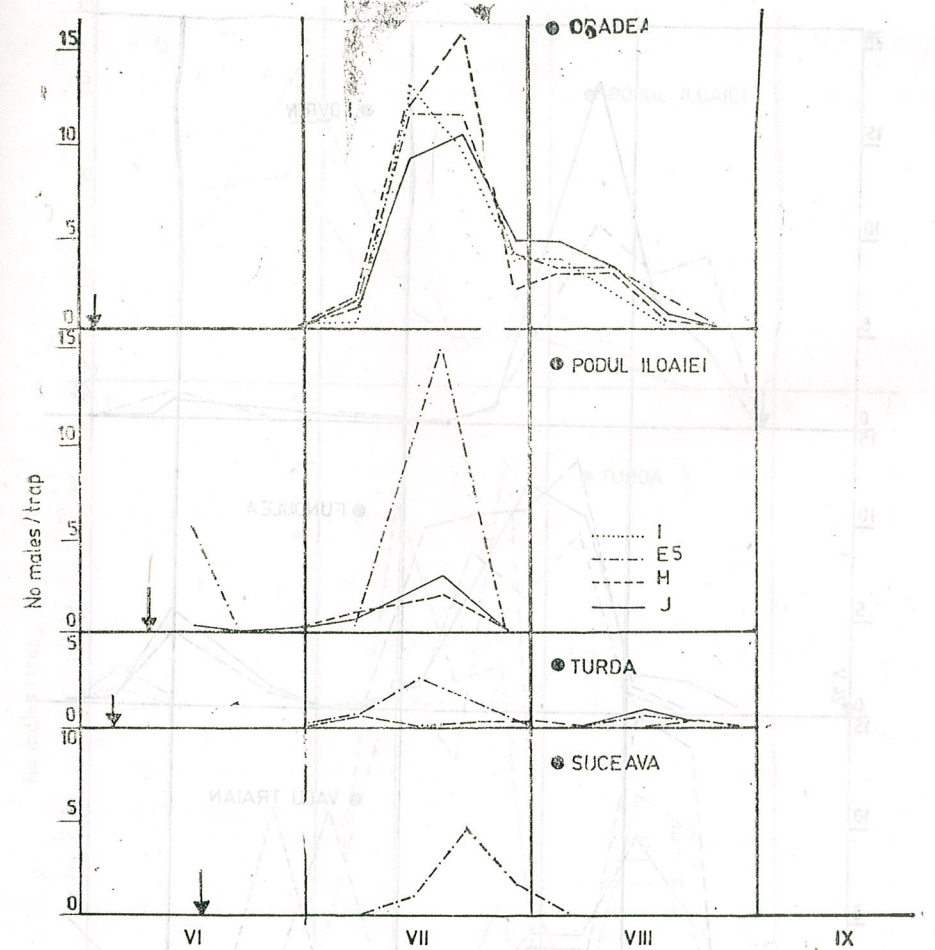


Fig. 2b

Fig. 2. — Flight dynamics of *Ostrinia nubilalis* Hb. males, as revealed from captures with pheromone traps at Fundulea in 1988.

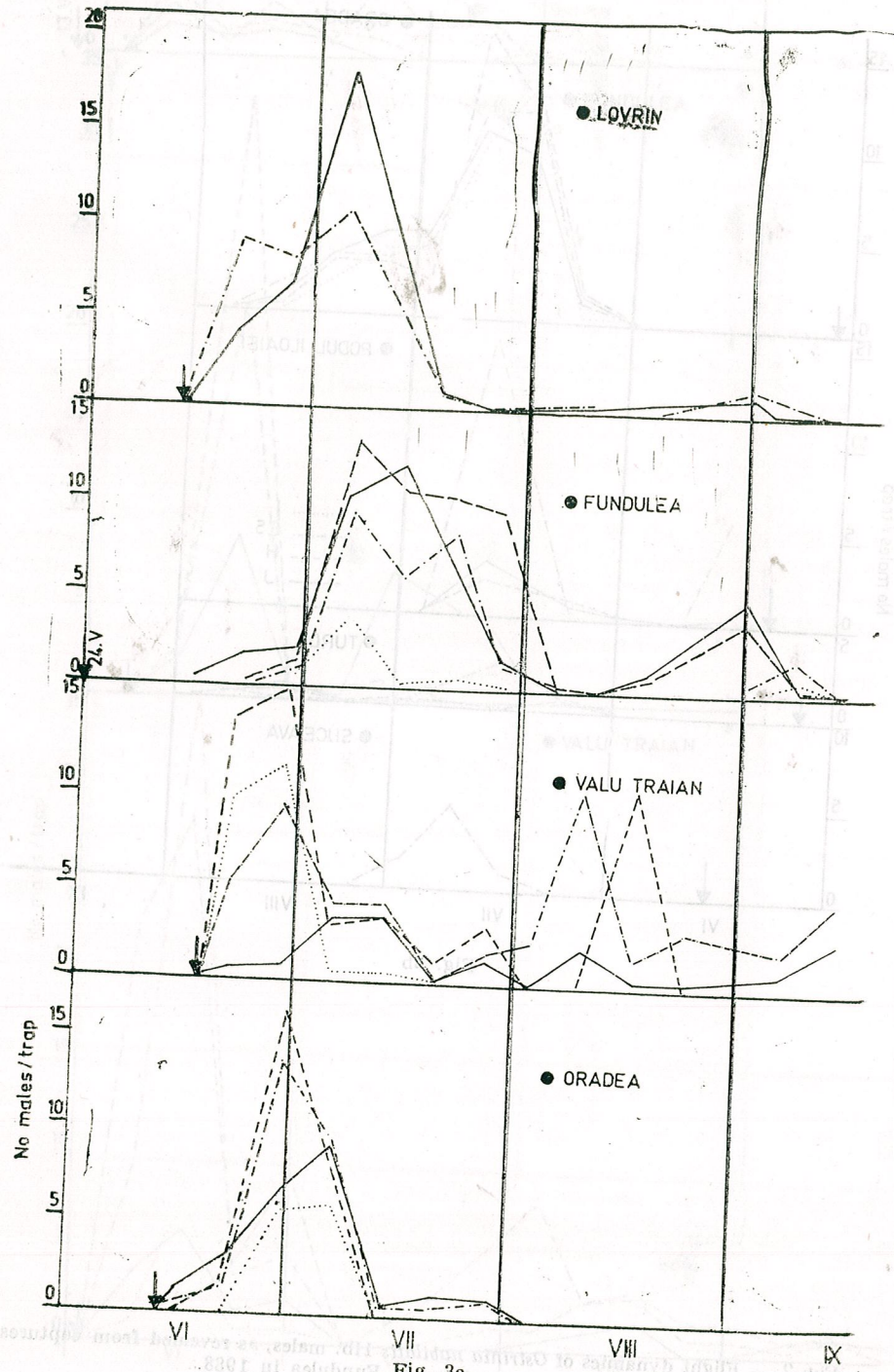


Fig. 3a

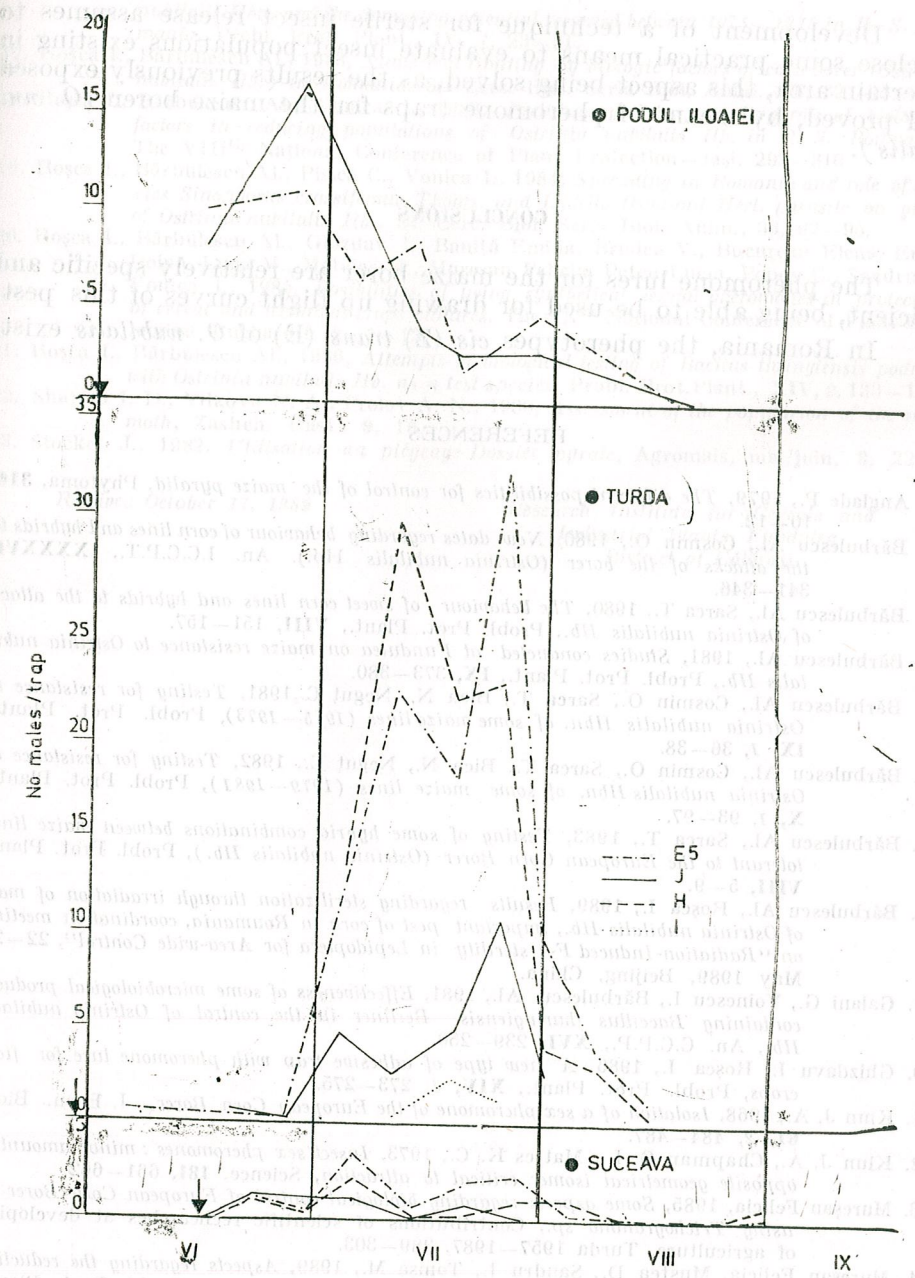


Fig. 3b

Fig. 3. — Flight dynamic of *Ostrinia nubilalis* Hb, males, as revealed from captures with pheromone traps at Fundulea in 1989.

Development of a technique for sterile insect release assumes to disclose some practical means to evaluate insect populations existing in a certain area, this aspect being solved, as the results previously exposed had proved, by means of pheromone traps for the maize borer (*O. nubilalis*).

CONCLUSIONS

The pheromone lures for the maize borer are relatively specific and efficient, being able to be used for drawing up flight curves of this pest. In Romania, the phenotypes *cis* (Z) *trans* (E) of *O. nubilalis* exist.

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THE ULTRASTRUCTURE OF PREIMPLANTATIONAL EMBRYOS USED IN THE BIOTECHNOLOGY OF EMBRYOTRANSFER

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In this paper we present some ultrastructural aspects which characterize the process of morphofunctional differentiation of the cells in the early ontogenesis of embryos used in the embryotransfer biotechnology. In this respect we present our opinion on primary cellular lineage achievement and the analysis concerning the peculiar way of passing the nuclear material from the nucleus into the cytoplasm during the syncytiotrophoblast stage — forerunner embryo implant into the uterine mucosa — aspect which marks the moment in which the genome becomes transcriptionally most active.

The electronmicroscopic study of the molecular and cellular events which take place in early ontogenesis offers the possibility of knowing the intimate mechanisms that can be implied in the cytodifferentiation and the space and time organization of the cells which will lead to the structuring and functioning of the multicellular superedifice (2), (4), (9), (10), (14), (15), (18).

The embryos transfer in mammals and in man, inclusively, requires the solving of theoretical and practical problems. The improvement of embryotransfer biotechnology offers the possibility to effectuate some genetic manipulation using the intracellular microinjection method (3), (6), (7), (12), (13) and that of cellular aggregation and fusion (6), (7), especially with the purpose to achieve a controlled ontogenetic development. In this respect the electronmicroscopic investigations come to help and refer to the knowledge of ultrastructural alterations which join the movement dynamics and the intercellular connections that take place in the first stages of embryo development (2, 6, 7, 15).

MATERIAL AND METHODS

The *Sus scrofa domestica* embryos in different stages of evolution (2, 4, 8, 16 blastomeres, morula and blastocyst) used with the purpose of their transfer to receivers were surgically obtained from females formerly submitted to a follicle-stimulating treatment with serum gonadotrophin (Folligon — 1,500 IU) followed three days later by administration of corionicgonadotrophin (Chorulon — 750 IU). The biologic material was remade with the purpose of electronmicroscopic examination. Thus the embryos were fixed in 2.5 % glutaraldehyde and 2% osmium tetroxide and inserted into Epon. The ultrathin sections contrasted with uranium acetate and lead citrate were examined with the help of a Phillips electron microscope at 50 kV.

RESULTS AND DISCUSSIONS

The main morphostructural events which take place as a result of recombination of the genetic material initiated by amphimixis up to the moment of syncytiotrophoblast constitution which announces the embryo-implant into the uterine mucosa can be thus summed up: concomitantly with the cellular proliferation by repeated mitotic divisions the cellular differentiation takes place and is characterized by a distinct morphofunctional polarization of these and is followed by cellular movements which will lead to the achievement of primary cellular lineage by the embryo- and trophoblast constitution.

After the first and second division of segmentation the resulted blastomeres are big and each equal with a large nucleus sometimes nucleolated. The cytoplasm is prevalingly occupied by heteromorphic vesicular structures and by a rich particulate contents. Periblastomerically a rich glycocalyx is distinguished and constituted by an amorphous mass in which microvesicular structures of different dimensions and microvilli belonging to the blastomeres which penetrate the glycocalyx are to be seen. We have noticed that ultrastructurally at this beginning stage of embryo development, the pellucida membrane is weakly osmiophile and appears constituted from concentric lamellar structures among which an amorphous substance is interposed.

In mammalians, the follicular cells which surround the preovulatory oocyte and constitute corona radiata send cellular processes through the pellucida membrane to contact the oolemma and establish intercellular junctions between the two heterogeneous cellular types. It has been experimentally demonstrated that the ionic couple follicular cells-oocyte is present before the ovulation but this is not obviously postovulatory (5). Within the electromicroscopic investigations made by us upon the early embryos (with preserved pellucida membrane) we have not noticed the existence of some cellular processes at the level of pellucida membrane.

In a study accomplished on oocytes and pig early embryos, Brown and Cheng (1) have noticed that the development of pellucida membrane from inside the follicle up to the embryo stage 2-4 cells includes the acquirement in its structure of glycoproteins present in the oestral oviductal follicle. It is considered that the acquirement of oviductal components made by the egg plays a most important role in the developmental control of early embryos which explains the limitation of embryos cultivation success. That is why the pig embryos of 2-4 cells obtained by the maturation of oocytes and the fertilization "in vitro" need the before hand oviducts placing of embryos for their surviving and development.

Initially, the pellucida membrane acts as a selective sieve for fertilization and subsequently as a capsule in which the young embryo is transported at the implant spot.

It is interesting the fact that after the first division of the zygote the plasma membrane of the first resulted blastomeres is no more in direct relation with the pellucida membrane as it happened in the oocyte case. Also, the trophoblastic cells establish no direct contact with the pellucida membrane while this is still under preservation.

The prolongations of trophoblastic cells achieve a filiform aspect and partially superpose taking over the function of protecting the young embryo function that was fulfilled by the pellucida membrane. It is not yet well established if the pellucida membrane has an exclusive or a prevalent part of protection of the young embryo. It is well demonstrated that in the transfer attempt of the sectioned embryos from donor to receiver the embryo fragments placing into the inside of pellucida membrane is necessary. These membranes were obtained by their previous vacuum by the own embryos.

The pellucida membrane gradually degrades but the process is improved only after the elongation of trophoblastic cells and the establishing of intercellular junctions among the trophoblastic cells as well as among these and the subjacent embryoblastic ones.

Which are the mechanisms which induce the first morphofunctional differentiations of the cells in the early stage of ontogenetic development? During the last years an important part in the explanation of this complex is due to the cell interactions with the extracellular matrix via the cellular surface [11], [16]. Synthetically, we might say that the concentration gradients of the intra- and extracellular substances (consequence of the matter changes among the cells and the extracellular medium and of the biosyntheses) together with the alteration of the relations between cell and cell (consequence of the movements of blastomeres) generate messengers which — via plasma membrane — will act upon the genome and will generate quantitative and qualitative changes of the cellular surface (for example the alteration of the types and of distribution areas of the receptors and of some specializations of the cellular surface, such as the microvilli, the intercellular junctions, and will determine new gradients belonging to the extracellular matrix subsystems with which the cells interact. This phenomenon is selfmaintained within the limits of the standard of the adaptive reaction of the respective cells.

On the basis of the observations accumulated, it is considered that a crucial event in the early ontogenetic development is represented by the compaction which is a consequence of the cellular organization and of the interactions that appear in the 8 cells stage at mouse embryo (4), (14). The compaction implies major changes within the reactions among the cells such as the constitution of intercellular junctions and the early signs of the extracellular matrix depositing. The process of cellular polarization is adjusted by the intercellular interactions and implies the rearrangement of the cellular majority components both that of its surface and from the inside of the cell, the cytoskeleton inclusively.

Within the mouse ontogenesis, the blastomeres normally resulted by repeated cleavage in the 8 cells stage have a polarization axis perpendicularly directed on the contact points cell — cell. The interactions among the cells are mediated at least partially by the adhering system Ca^{2+} (CDS) depending cell — cell, determine a focus for the intercellular events implied in the cell polarization.

It is not yet well established which is the intimate mechanism of the compaction. Johnson et al. (10) consider that during the intercellular compact formation via CDS the local depolarization of microtubules takes place.

It has been established that, as a result of monoclonal antibodies treatment to ECCD and with cytochalasine D, the cellular flattening is inhibited and the random orientation of the polarization axis takes place. Cytochalasine D destabilizes the cellular microfilaments. Maro and Pike-ring (cited by Johnson 10)) have shown that the stabilization of microtubules tends to delay the compaction events while the destabilization has a synchronising effect. Johnson et al. (10) consider that during the intercellular compaction via CDS, local depolarization of microtubules takes place. The initiation signal of polarization is won in the stage of 8 cells and this signal can be implied in the abolition of a factor which may have inhibitory effects upon the compaction (10).

Taking into account the observations in accordance with which after the removal of the inhibitory action of some substances such as ECCD the intercellular junctions which establish among the subsequent embryo stages do not permit normal reorientation of cellular polarity axis and the blastomeres cannot form the inner cells mass as they constitute in a stratum of cells with epithelium aspect (trophoectoderm) (9), (10). Therefore, we consider that the intercellular contact points initially determined among the blastomeres have a fundamental part in the spatial organization and in the synchronization of cellular polarization representing the first step in achieving the primary cellular lineage (15). Johnson and Ziomek (9) consider that the consequence of polarization represents the foundation of primary cellular lineage of the blastocyst: the inner cells mass-committed cells — and the trophoblast — differentiated cells. The two cellular populations will carry out different functions in the development of subsequent events.

The blastomeres of early embryo of suine up to the 8–16 blastomeres stage are ultrastructurally alike. A most important moment in suine embryo ontogenesis is represented by the blastocel formation. Our electronmicroscopic investigations show that the blastomeres directed towards the pellucida membrane differentiate morphostructurally in a different manner from those which will constitute the inner cells mass. Thus, the peripheric cells will send cellular prolongations with filiform aspect. The nuclei are oval shaped and the cytoskeletal microfilaments are fewer than those from the embryoblastic cells. The desmosomal junctions which establish among the trophoblastic cells are a lot more reduced in number than those which establish among the embryoblastic cells. The embryoblastic cells send several microvilli from which some, as they find themselves in stages previous to compaction, participate to the achievement of some intercellular junctions with the purpose of solidarizing the cells among themselves [15]. Afterwards, when the blastocoelian cavity is constituted, the number of microvilli will reduce and the intercellular solidarization will be achieved by numerous desmosomal junctions.

We consider that the great number of microvilli which the apical pole of embryoblastic cells expose towards the blastocoelian liquid and the changes of substances of the respective cells with the blastocoelian liquid accentuate the morphofunctional polarization gradient between the apical pole and the bazolateral cellular domains.

The nuclei of embryoblastic cells are approximately big, spherical-shaped and usually occupy a central position in the cell. Inside the nuclei

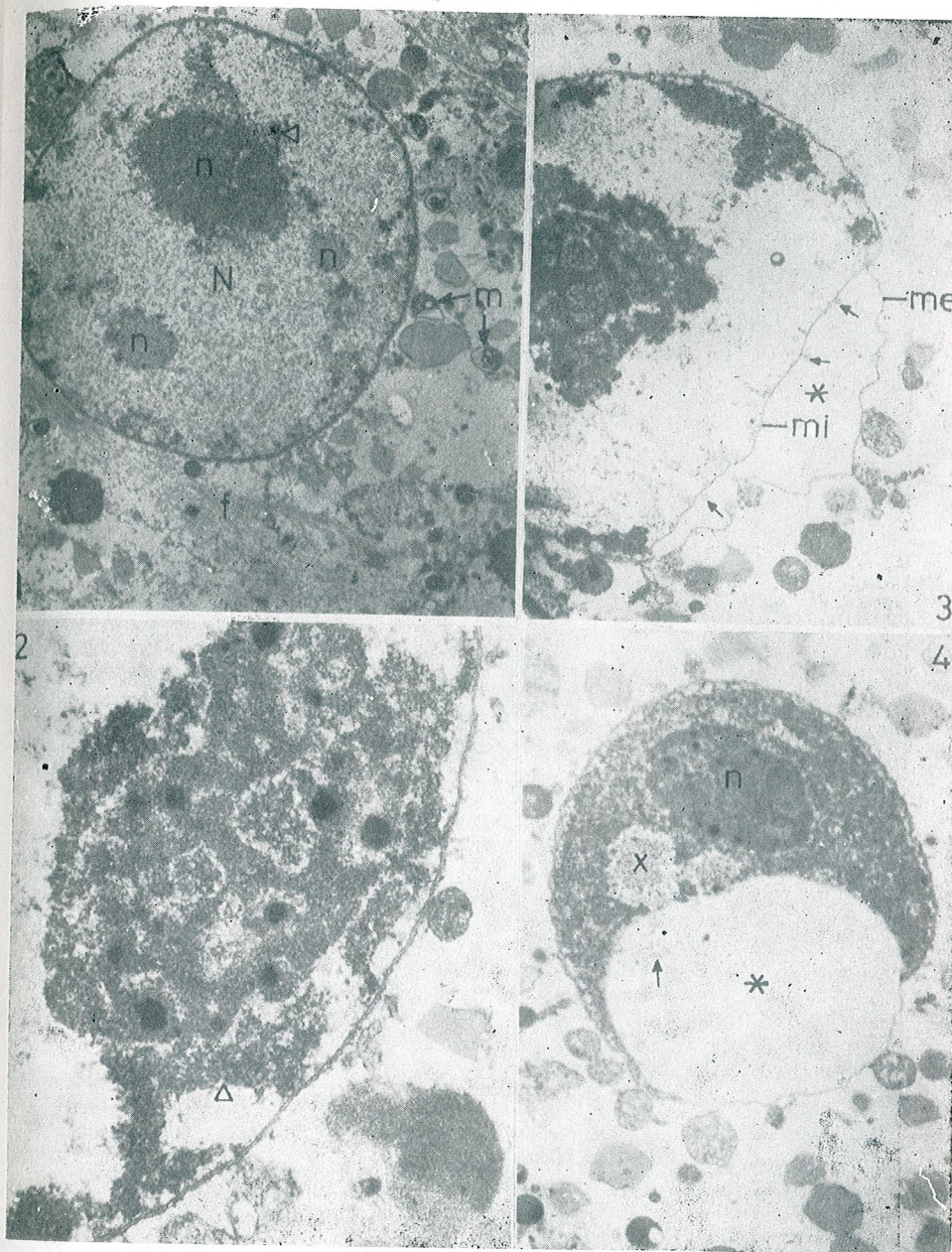


Fig. 1. — Embryoblastic cell from the inner cells mass. Nucleoli (n) are to be seen into the nucleus (N) from which the greatest presents nucleolo-associated heterochromatine (▷). The euchromatine predominance is remarkable. We may see some vacuolized mitochondria (m) in the cytoplasm inclusions with amorphous contents and cytoskeletal-like filaments (f). (×10,000).

Fig. 2. — Nuclear sector in which we may see a big nucleolus from "pars amorphosa" and "pars fibrosa" constituted. We notice the existence of some particulate electronopaque and nucleolo-associated heterochromatine structures (▷). (×14,000).

Fig. 3. — The ultrastructure of the nucleus of an embryoblastic cell in which the big nucleolus occupies a central position. On a certain sector (polarized) the perinuclear space is dilated (*); a nuclear material passed into the perinuclear space is seen (→). The internal (mi) and external membranes (me) of the nuclear envelope are wholly preserved (×8,000).

Fig. 4. — Aspect resembling the previous one. The nucleolus (n) is large. In the neighbourhood of the excessive dilation of the perinuclear space (*) the nuclear material has a particulate structure (X) and some fragments of it (→) may be seen in passage towards the perinuclear space. (×7,000).

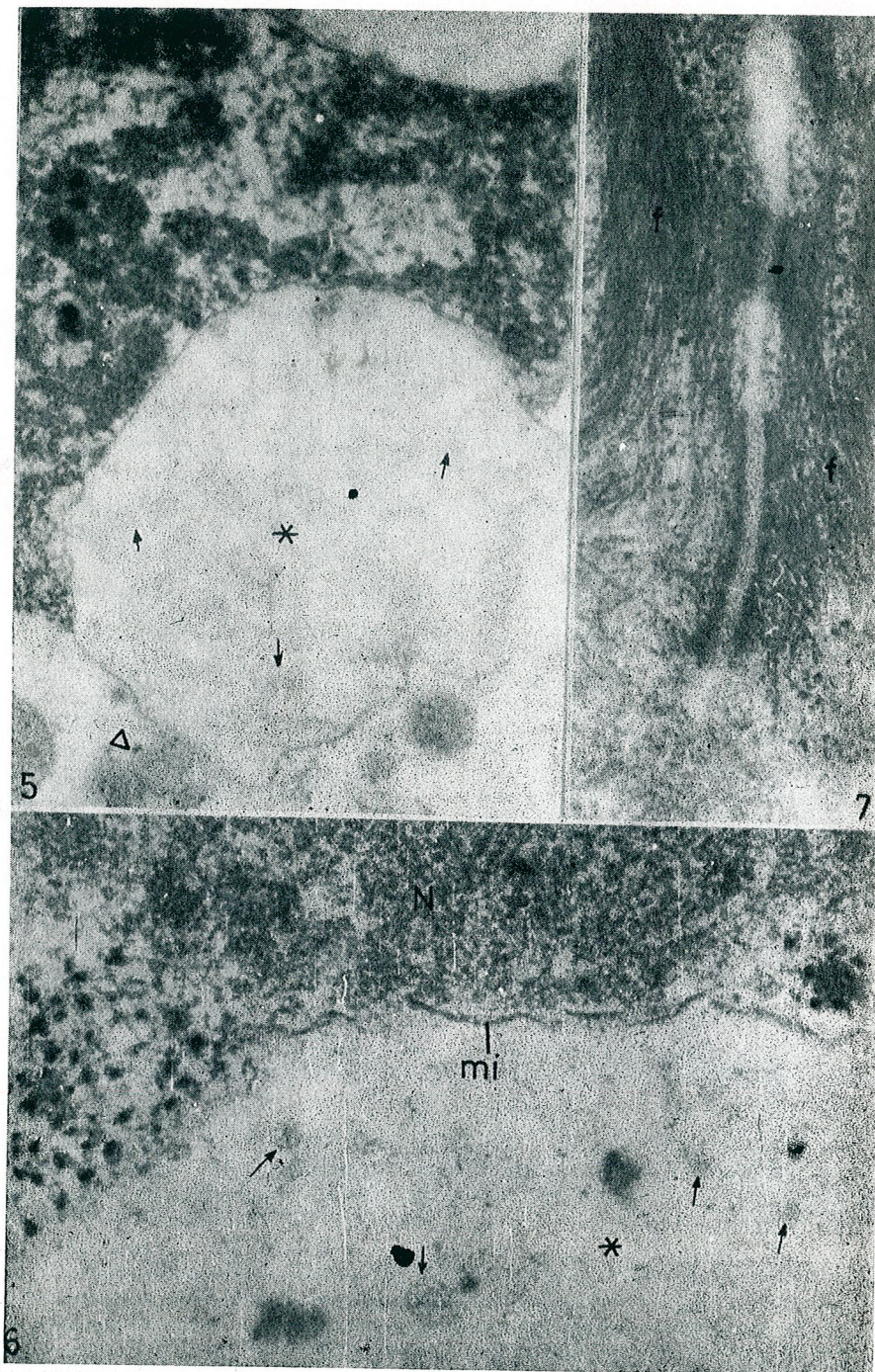


Fig. 5. — Nuclear sector with bipolar dilations of the perinuclear space (*) wholly occupied by a fibrillo-granular material of nuclear origin (→) which passed into the cytoplasm (▷). (× 15,500).

Fig. 6. — The internal membrane of the nuclear envelope (m i) is partially preserved and appears discontinuous in the left half of the image. In the perinuclear space (*), a fibrillo-granular material (→) passed from the nucleus (N) may be seen (× 20,300).

Fig. 7. — Desmosomal junction which strongly solidarizes two embryoblastic cells. The desmosomal plate can be seen and the richness of the cytoskeletal filaments (f) found in direct relation with junctional structures is noticed (× 60,000).

euchromatine prevails; heterochromatine is little and attached to the nuclear envelope or nucleoloassociated (Figs. 1—3). The nuclei of embryoblastic cells present 1—3 nucleoli. The cytoplasm of these cells is poor in organites (electronmicroscopic vacuum cytoplasmic spaces are to be seen). The mitochondria are better represented most of them being perinuclearly disposed. The presence of cytoskeletal structures is remarkable as they are prevailingly juxtannuclear distributed (Fig. 1) and at the level of junctional structures of desmosomal type (Fig. 7). For the rest, the cytoplasm is occupied by free ribosomes, heterogenous inclusions as dimensions with amorphous contents and seldom vesicular structures. The mitochondria are polymorphous with cristas disorderly directed. Mitochondrial cristas often have tubular aspects. Sometimes, we can see big intramitochondrial vacuoles (Fig. 1) with vague electrondense contents. A frequently met aspect is the mitochondria distribution near the nuclear envelope especially in the extrusion place of the nuclear material in the cytoplasm with which the mitochondria come into close relation. The great majority of mitochondria are committed in a degradation process by autolysis; the successive stages of the process are to be seen. Initially, a polar area of mitochondria is vacuolised. More seldom, the process of vacuolization advances and in the end the whole mitochondria loses the cristas and appears like an amorphous mass included in a discontinuous membrane (probably remainings belonging to the external mitochondrial membrane) with cholesterol-like aspect. Such structures with cholesterol-like contents were described in the cells of 4-blastomeres stage [15].

In the stage of preparation of the embryo for implant into the uterine mucosa, we can see nuclei with a particular aspect (Figs. 3—6) in some embryoblastic cells ultrastructurally normal. Initially, at a certain distance of the nuclear envelope there appear dilations of the perinuclear space, dilations which excessively enlarge. The consequence of the centripetal pushing of the nuclear material by the internal membrane of the nuclear envelope is the carrying out of a calotte shape of the nuclear material (Fig. 4). When the excessive dilations of the perinuclear space are bifocal (usually diametrically opposed), (Fig. 5), the nuclear material has a biconcave lens shape. The maximum number of dilation focal points of the perinuclear space studied by us is three. Quite often, in the next neighbourhood of these dilations there is a nucleolus. In the perinuclear space thus dilated, a fibrillogranular material of nuclear origin detected in its passage towards the cytoplasm is to be seen (Figs 3—6). Sometimes, this is attached to the internal membrane of the nuclear envelope. Usually, the internal membrane of the nuclear envelope is continuous at the level of excessive dilations (Fig. 3—5). Sometimes, this appears discontinuous (Fig. 6) which allows the nuclear material to massively pass in the perinuclear space which it can integrally occupy under the shape of some fibrillogranular structures (Fig. 5 and Fig. 6).

In the cytoplasm, in the next neighbourhood of the external membrane of the nuclear envelope and attached to it, a rich fibrillogranular material of nuclear origin can be seen (Fig. 5).

The electronmicroscopic images suggest that certain areas of nucleolo-associated chromatine represent the spot of formation of the infor-

mational substratum which may reach by accumulation the nuclear envelope and is then expelled at this level towards the cytoplasm. Within the studied literature we have not met the description of an ultrastructural detail to this resemblance to produce itself in the stage of blastocyst before the implant into the accepting uterine mucosa. The massive extrusion phenomenon of the nuclear material into the cytoplasm is frequently met in the prophase of oogenesis of amphibians and fishes [8]. This aspect represents a result of the amplification of ribosomal gene as a response to the necessity of a great accumulation of ribosomes, necessary in protein biosynthesis of egg segmentation time.

Which is the justification and the significance of this particular ultrastructural aspect we have noticed into the cells from the inner cell mass in early ontogenesis of suine? We suppose that this aspect is a response to the keen necessity in the respective moment of transfer of a great quantity of informational substratum from the nucleus into the cytoplasm under the shape of some copies of repetitive DNA (genic amplification) and/or messenger RNA. The high rate of cellular divisions which characterize the moment of embryo preparations for implant into the uterine mucosa (after the constitution of syncytiotrophoblast) imposes the intensification of biosynthesis rhythm so that it may supply cellular material for the plasma membrane and the membranous and particulate organites of the new cells resulted by rapid succession of blastomere cleavage. It seems that at this moment the genome becomes transcriptionally most active.

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CONSIDERATIONS ON THE MECHANISM OF THE CELL MEMBRANE STABILIZATION

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Structural phospholipids have been admitted as playing a principal role in bioelectrogenesis and permeation, by taking into account their ion exchange properties, the possibility of conformational modifications of their molecules and their supramolecular organization into laminar and globular micellae structures, which can be shifted, by phase transitions, from one form to another, depending on the ratios established by each membrane layer with adjacent phase (especially with certain ions from the phases). The normal structural stability of the membrane is attained by phenomena of selective adsorption of ions on the surface of phospholipidic, lipophobic sols. Membrane stabilizers of the cationic local anesthetic type exert their specific blocking effect by a super-stabilization phenomenon, implying both the molecule's electric charge and its lipophilia. This superstabilization effect is based both on an action at the levels of the external lipid layer and of the membrane internal one. Their mechanism and localization assumes complex characteristic interactions with ions of sodium, potassium and calcium. The adopted membrane model and the proper mechanism for the action of local anesthetics permit the correlation of a multitude of experimental facts that could not have been given a unified explanation otherwise.

Although membrane proteins play an important role in maintaining its structure as well as in some transport phenomena (by specific receptors), in order to explain the membrane bioelectric potential and the effects of a great number of substances characterized by other authors as showing non-specific action (34), we have admitted that the phospholipids in the membrane structure play an essential role (1), (2), (3), (30).

Many papers have demonstrated that phospholipids possess ion exchange properties (12), their behaviour being either similar to that of cationites or to that of anionites. It was also suggested or shown that the lipidic membrane layer is also implied in water and ion transport through the membrane (15), (23), (42).

Flowing of water, electrolytes or non-electrolytes through the lipidic leaflet would be caused, according to some authors, by certain packing "flaws" of the chains of fat acids from phospholipids (29) or by statistical, dynamic pores, possessing variable dimensions and a certain life time (8). Other authors explained them as depending on the phospholipids structural organization into laminar, tightly packed micellae and globular micellae, with quite large spaces among them, these two types of micellae being shown as coexisting in the membrane while being able to shifted from one another by phase transitions, under the action of some physico-chemical agents (pH, Ca^{2+} and K^{+} ions a.s.c.) (17), (19), (21). Phospholipids have also been shown to exist in two different molecular conformations, either with polar group in the opposite directions of the hydrophobe chains (the p_{ex} conformation) or with polar group oriented between the two chains of fat acids (the p_{in} conformation) (19).

REV. ROUM. BIOL.—BIOL. ANIM., TOME 35, N° 2, P. 123-128, BUCAREST, 1990

As regards the membrane transverse structure, its nonsymmetry has been suggested by Robertson already (32), who explained this fact by the difference between the proteins of the external layer and those of the internal layer of the membrane. Subsequently, it was demonstrated that not only proteins but also phospholipids have a different distribution within the two membrane layers (6), (9). The nonsymmetry of the membrane layers led to the idea of a relative functional independence of each of them, direct relations being established by them firstly with adjacent extramembrane phases (1), (18).

On the other hand, according to latest membrane models (37), (44), the proper structural proteins ("fundamental proteins") are known to relatively occupy only a small portion of the membrane surface, large spaces ("the lipid sea") being represented by lipids which are in immediate contact with the aqueous extracellular and intracellular phase respectively. From this, there results a small amount of specific proteic receptors and, respectively, of molecules fixed on membrane in the case of specific transports (of the order of unities, tens and sometimes hundreds receptors on a surface of $1 \mu^2$, depending on the substance nature). Having in view that, in the case of water, ions and "nonspecifically" transported substances, it has been estimated that a surface of $1 \mu^2$ contacts some millions of molecules. We have therefore to accept them as interacting with the large lipidic spaces and not with the rare protein islands ("icebergs").

In fact, conceived only as a simple dielectric material, membrane lipids, with their large surface, must play an essential part in the separation of the electric charges and in their continuous distribution on the membrane's surfaces (which is a main role in the generation of the membrane electric potential).

For explaining bioelectrical phenomena, we have firstly adopted a membrane model in which lipids of the two layers have mixed ion exchange properties — mainly cation-exchange ones in the external layer and anion-exchange in the internal one — the membrane layers being able to manifest themselves independently (each of them depending on the ratios established with the corresponding adjacent phase).

Within the external layer of the membrane, cationic lipids, which are most numerous, are organized into laminar micellae, structured by the calcium ions (i.e. the "structure making" role of the Ca^{2+}). These laminar micellae may be shifted into globular ones possessing anionic properties, structured by the potassium ions (the K^+ effect of "structure-breaking" on laminar micellae, when its concentration in the extracellular phase increases). The sodium ions from the extracellular medium accumulated (adsorbed) into the sphere of electrical protection of cationic laminar micellae from the external layer determine the normal stability of these lyophobic sols.

In the internal layer of the membrane, anionic lipids, which are most numerous, are being stabilized by certain small anions from the intracellular medium (possibly the phosphoric ion). Their structuring ion is not known, yet the Na^+ ion is found to exercise a destructuring effect, transforming them into cationic systems with a looser structure.

This model allowed a better explanation regarding the membrane electrical charge, its passive depolarization and hyperpolarization phenomena, the reversible subliminal electrical responses and the action potential of excitable fibers as well (1), (2), (3), (30).

Our standpoint on cellular membranes has been largely confirmed by subsequent investigations due to H. A. Kolb and G. Adam (18). These authors also state an asymmetrical distribution of the fixed electrical charges on the two membrane surfaces, underlying the importance of the fixed negative charges in the external layer, carried by phospholipids, in the membrane's interactions with K^+ and Ca^{2+} ions. They also stress the role of phospholipids in ion transports within the membrane, based on their ion exchange properties. The idea of phospholipidic membrane re-arrangement (phase transition!), induced by modifications in the composition of the ionic medium, is also considered. The special importance of the K^+ and Ca^{2+} ion ratio from the extracellular medium in the determination of the membrane subunit structures — induction of "closed" forms by Ca^{2+} binding of "open" forms by K^+ binding — is particularly emphasized. Modifications in the membrane selective permeability depend on the structural state of the membranar subunits, which are dependent (in their external layer) on the $\text{K}^+/\text{Ca}^{2+}$ ratio from the extracellular phase and independent on intracellular ionic concentrations.

If one accepts the idea that Na^+ ions do stabilize cationic laminar micellae in the membranar external layer, through the protection sphere of lyophobic sols, sodium substitutes (choline, tetraalkylammonium ions at the beginning of the series, etc.) are also found out as capable of stabilizing the external layer, when present in the extracellular medium (10), (14), (22), (39), (41). Nevertheless, inasmuch as their molecule shows not only a positive electrical charge but also lipophilic properties, the stabilizing effect becomes increasingly stronger, lipophilia actually leading to a super-stabilization of the cationic micellae from the external layer (1), (2).

The agents that, much to the contrary, facilitate the destructuring action of certain elements, induce an opposite effect, of membranar destabilization, their name being that of labilizing agents (36).

Local anesthetics, generally known as membrane stabilizers and as impulse blocking agents, exert however a more complex action, as compared to sodium substitutes.

Worth mentioning is that, depending on the pH of its medium, these anesthetics may be found in a dissociated or undissociated (neutral) form, or, to a certain ratio, in both forms. On the other hand, their molecule also shows significant hydrophobic properties, which are also at play in the blocking effect. Finally, as observed in the case of anesthetics with positive charge (procaine, etc.), one has also to take into account the specific interaction way with the structural components of the membrane (the capacity of substituting or of being substituted by membranar calcium).

As regards the action mechanism of local anesthetics, several hypotheses, often incomplete or even contradictory, have been worked out (7), (11), (13), (16), (20), (31), (33), (35), (38), (40), (43).

Based on the membrane model presented in our paper and on several experimental investigations in which procaine was used as a local anesthetic, we suggested an action mechanism for these agents, which can furnish a unified explanation to the complex of specific interactions with cell membrane (4), (5), (24), (25), (26), (27), (28).

When the membrane is at rest and the extracellular medium possesses a normal pH (7.2), procaine induces a slight hyperpolarization of the membrane, as observed by several authors, yet never correlated with the blocking phenomenon. We have considered it as part of the general action of the anesthetic, inducing an increase of about 15% (not at all negligible) of the depolarization threshold needed to initiate the impulse. As the membrane hyperpolarization assumes, under the conditions stated, an extension of the cationic laminar zones of the external layer, we have admitted it as being determined by the phase transition of the globular, K^+ -structured micellae, into laminar micellae, usually Ca^{2+} -structured, whose role is now played by procaine (substitution by cationic procaine, which is to be found in large amount at such a value of the environment's pH, of the K^+ in the structure and, respectively, imitating of the calcium role).

Actually, the main phenomenon of the general action of impulse stabilization and blocking is not hyperpolarization, but another effect, which does not modify the membrane electrical charge.

It is generally known that the onset speed of procaine blocking effect is greater with increasing external pH, which suggests its dependence in the concentration of the neutral (nondissociated) procaine form. The cationic form of procaine cannot penetrate towards the membrane internal layer, possessing anionitic properties. The neutral form of the anesthetic is, however, capable of easily penetrating into the internal layer, due both to the lack of electrical charge and to its high lipophilia.

Such a property of the neutral procaine induces a high stabilization (super-stabilization) of the internal anionitic layer, at whose level it is fixed by hydrophobe forces. An over-increase of the internal layer stability does not induce, under these conditions, any change in the membrane resting potential, yet evidently imposes an increase of the sodium concentration required for initiating the upward phase of the section potential after the application of a stimulus (the threshold increase with the destructuring action of sodium ions on the internal layer, the intense inhibition of the increase in the sodium active conductance).

Thus, this effect represents a main constituent of the procaine stabilizing and blocking action. Yet, the two above-mentioned effects of procaine are not sufficient for explaining all aspects regarding the impulse blocking. Indeed, the procaine-treated membrane shows also, in conditions of a stimulus application, a very intense inhibition of the increase of K^+ active conductance.

Such a procaine effect is based on another action of the anesthetic; namely of the cationic form. It concerns the substitution of the Ca^{2+} ions in the laminar, cationic micellae of the external layer and the prevention, in this way, of the destructuring action of K^+ ions upon them. Evidence of this action was the reduction effect of the passive depolarization induced by external high- K^+ , too. It also contributes to the de-

crease of the increasing rate of the ascending phase of the action potential (respectively, the reduction of its amplitude) and to the prolongation of its descending phase (preventing the attack of K^+ coming out of the cell on the membrane external layer).

The impulse blocking by cationic local anesthetic is thus obtained by several effects, installed even in the resting state of the membrane, some of them occurring at the level of its external layer (the interaction of the dissociated form with K^+ in globular micellae and with Ca^{2+} in laminar micellae), while others arising at the level of the internal layer (interaction of the neutral form, based on its lipophilia with anionitic micellae, usually destructured by Na^+).

The membrane model we have started from allows thus a unified explanation of the mechanisms by which cationic local anesthetics exert their specific effect on the membrane, interacting differentiatedly both with phospholipids from the external layer and with those from the internal one, in both cationic and neutral forms.

The complex effect of the impulse blocking (local anesthesia) actually means the over-normal increase of the stability of the membrane phospholipid structures, an important role in this superstabilization being played by the lipophilia of the molecular structure of these agents.

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IN VITRO EFFECTS OF PROCAINE AND PROCAINE-RELATED DRUGS AND METABOLITES UPON THE GLUCOSE UPTAKE BY RAT BRAIN SLICES

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The *in vitro* effects of procaine-HCl, tetracaine, dibucaine, lidocaine, Gerovital H₃, Aslavital, 4-paraamino-benzoic-acid and 2-diethylamino-ethanol were investigated upon the glucose uptake by normal rat brain hemisphere slices from glucose containing Krebs-Henseleit bicarbonate solution (16.7 mM glucose; pH = 7.4) under aerobic conditions (gas phase = 95% O₂ + 5% CO₂). The data suggest that in equivalent concentrations with procaine-HCl (10 micrograms/ml medium) the procaine related substances, drugs and metabolites exert quantitatively different stimulatory effects upon the rate of glucose penetration from the incubation medium into the brain slices, depending on their chemical composition and molecular shape. It is concluded that the investigated drugs exert specific stimulatory actions upon the transmembranal glucose transport system at the level of brain hemispheres.

It is well established that under *in vivo* conditions glucose is a major energetic and metabolic substrate of the nervous system (9), (11), (12), (14), (15), (16), (22), (25), (32), (35), and that the brain is a target organ for procaine, Gerovital H₃, Aslavital and other procaine-related drugs and metabolites (1), (2), (3), (5), (6), (7).

Taking into consideration the above establishments and the fact that procaine-related substances, drugs and metabolites exert biotrophic and energizing effects upon the central nervous system (5), (6), (7), (29), depending on the rate of their hydrolysis at tissular level (1), in the present study we followed comparatively the direct influences of procaine-HCl, Gerovital H₃ and of some anesthetic tertiary amines upon the glucose uptake *in vitro* by the rat brain hemisphere sections. It is known that the metabolizing rate of these substances is reduced at the level of the brain of white rats, in comparison with that found in other tissues (1).

MATERIALS AND METHODS

For experiments 60–70-day old male albino Wistar rats, weighing 160–170 g were used as tissue donors. The animals were kept under standard laboratory conditions with free access to food and water. Before sacrifice by decapitation, the animals were fasted for 18 hours, and the brains were quickly isolated and immersed for 5 minutes in ice-cold Krebs-Henseleit bicarbonate solution, without glucose (pH = 7.4; 4°C). From each brain, at the level of right and left parietal lobes, slices of 0.8–1.0 mm were sectioned, the left ones being used for testing the basal glucose uptake (controls) and the right ones for evaluating the *in vitro* effects of the drugs upon this phenomenon. The incubation medium for each brain slice was 1.00 ml Krebs-Henseleit bicarbonate solution (pH = 7.4), containing glucose (16.7 mM) and 2 mg gelatine (p.a. "Merck") per ml.

Procaine-HCl, tetracaine, dibucaine, lidocaine, Gerovital H₃, Aslavital, 2-diethylamino-ethanol and 4-paraamino-benzoic-acid were used in a concentration of 10 micrograms/ml incubation medium, a concentration corresponding with the therapeutic dose of procaine-HCl (5), (29).

Excepting commercially available Gerovital H₃ (GH₃) and Aslavital (ASL), furnished by the Drug Enterprises Bucharest, all chemicals used were analytical grade. Procaine-HCl (PRC) p.a. cryst. was purchased from "Hoechst", GmbH, Germany; 4-paraamino-benzoic-acid (PABA) from "Merck" A. G. Germany; 2-diethylamino-ethanol (DEAE) from British Drug House; tetracaine-HCl (TC) from "Sigma" Chem. Co. U.S.A.; dibucaine-HCl (DBC) from "Sigma" Chem. Co.; lidocaine-HCl (LDC) from "Sigma" Chem. Co. U.S.A.

Incubations were performed under aerobic conditions (gas phase = 95% O₂ + 5% CO₂; 90 oscillations per minute), for 60 minutes at 37.6°C, using an original device (18) and our procedures applied for testing the *in vitro* effects of procaine-related drugs on other tissues and organs upon glucose consumption (3), (19), (20), (21).

The initial - and postincubation-contents of glucose in the medium were estimated enzymatically, using GOD-Perid Kit ("Boehringer", GmbH, Mannheim, Germany) and the method of Werner *et al.* (34).

The rate of glucose uptake by the brain slices was expressed as micromoles/100 mg fresh tissue for 60 minutes.

The results were expressed as mean values \pm S.E., and analyzed by Student's *t* test, the differences between the means being considered statistically significant when $P < 0.05$.

RESULTS AND DISCUSSIONS

From the data summarized in Table 1 it is obvious that under the direct influence of procaine-HCl (PRC) the *in vitro* glucose uptake from the incubation medium by rat brain hemisphere slices was intensified by 105.60% ($P < 0.001$) vs. the corresponding basal values. Tetracaine-HCl (TETR) have not a significant influence (14.86%), ($P > 0.25$), dibucaine (DBC) enhanced this phenomenon with 33.61% ($P < 0.01$), lidocaine (LDC) with 44.09% ($P < 0.01$), while the procaine-containing drugs, Aslavital (ASL) and Gerovital H₃ (GH₃), had a stimulatory effect of 148.07% ($P < 0.001$) and 48.03% ($P < 0.01$), respectively, upon the glucose uptake by the brain slices. At the same time, the procaine metabolite 4-paraamino-benzoic-acid (PABA) stimulated the brain glucose uptake with 61.82% ($P < 0.001$), while 2-diethyl-amino-ethanol (DEAE) did not exert any appreciable effect (+1.48%; $P > 0.50$) upon glucose uptake as compared to the basal values.

Considering arbitrarily equal to 100% the net effect of procaine-HCl upon the glucose penetration ($\Delta S-C$) from the incubation medium into the brain slices, the direct stimulatory effects of the investigated substances (Table 1 and Fig. 1) are the following: TETR = 16.37%; DBC = 39.23%; LDC = 40.47%; GH₃ = 65.48%; ASL = 67.66%; PABA = 74.10%; and DEAE = 1.87%. These modifications suggest the conclusion that the tested procaine-related substances, drugs and

Table 1

'In vitro' glucose uptake (micromoles/100 mg tissue/60 min) by rat brain slices under basal (control) conditions as well as in the presence of procaine-HCl (PRC), tetracaine (TETR), dibucaine (DBC), lidocaine (LDC), Gerovital H₃ (GH₃), Aslavital (ASL), diethylaminoethanol (DEAE) and of paraamino-benzoic-acid (PABA)

SUBSTANCE	M \pm S.E.	D %	P ₁	$\Delta(S-C)$	P ₂
CONTROL	2.989 \pm 0.46	—	—	—	—
PRC	6.147 \pm 0.48	+105.60	< 0.001	+3.158 \pm 0.45	—
CONTROL	3.479 \pm 0.30	—	—	—	—
TETR	3.996 \pm 0.27	+14.86	> 0.250	+0.517 \pm 0.12	< 0.001
CONTROL	3.686 \pm 0.19	—	—	—	—
DBC	4.925 \pm 0.24	+33.61	< 0.01	+1.239 \pm 0.16	< 0.001
CONTROL	3.302 \pm 0.27	—	—	—	—
LDC	4.758 \pm 0.17	+44.09	< 0.01	+1.278 \pm 0.15	< 0.01
CONTROL	4.306 \pm 0.34	—	—	—	—
GH ₃	6.374	+48.02	< 0.01	+2.068 \pm 0.29	\approx 0.05
CONTROL	1.635 \pm 0.17	—	—	—	—
ASL	4.056 \pm 0.32	+148.07	< 0.001	+2.421 \pm 0.14	> 0.100
CONTROL	3.988 \pm 0.08	—	—	—	—
DEAE	4.047 \pm 0.22	+1.48	> 0.500	+0.058 \pm 0.02	< 0.001
CONTROL	3.759 \pm 0.19	—	—	—	—
PABA	6.083 \pm 0.21	+61.82	< 0.001	+2.324 \pm 0.24	> 0.250

Values are expressed as means \pm S.E. Each mean represents 8 experiments. D% = percent modification of glucose uptake vs. the corresponding basal (control = C) value; P₁ = statistical significant modifications vs. the controls; $\Delta(S-C)$ = net glucose uptake in the presence of substances (S) without C; P₂ = statistical significance vs. the net glucose uptake in the presence of PRC.

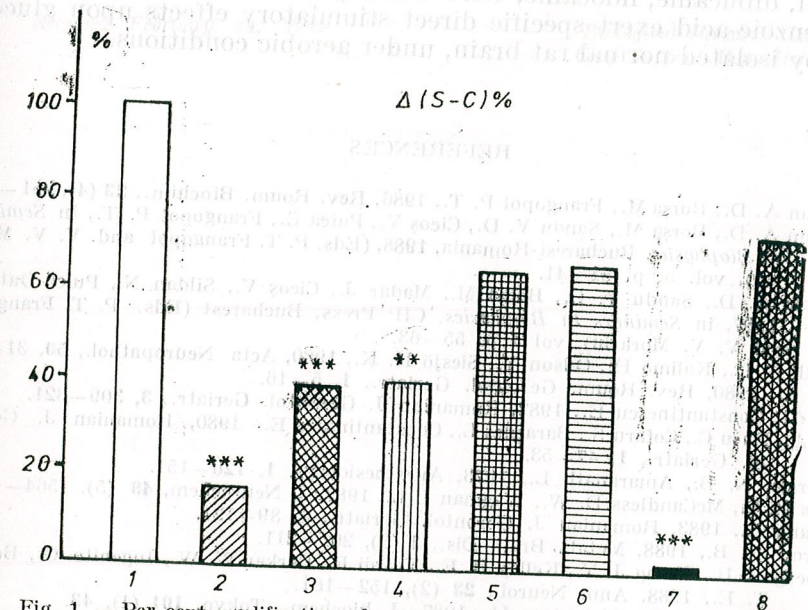


Fig. 1. — Per cent modification of net glucose uptake ($\Delta(S-C)$ %) by rat brain slices in the presence of procaine-HCl, and of various procaine-related substances, drugs and procaine metabolites. The effect of procaine-HCl is considered arbitrarily equal to 100%.

1 = procaine-HCl; 2 = tetracaine; 3 = dibucaine; 4 = lidocaine; 5 = Gerovital H₃; 6 = Aslavital; 7 = diethylamino-ethanol; 8 = paraamino-benzoic-acid. The asterisks indicate the statistical significance of modifications vs. the stimulatory effect of procaine-HCl.

(* $P < 0.05$ or = 0.05; ** $P < 0.01$; *** $P < 0.001$)

metabolites, depending on their chemical composition and molecular shape exert quantitatively different specific effects upon the transmembranal glucose transport system of brain. At the same time, the above results underline the conclusion that procaine-HCl and other procaine-containing anesthetic drugs have a reinforcing effect by activating some regions of the central nervous system (5), (6), (7), (29), probably, through the enhancement of the activity of transmembranal glucose-transporter system (8), (13), (23), (30) and by stimulating the activity of enzyme systems involved in glucose catabolism of neurocytes (4), (33), (36). In fact, it has been established that the main component of many geriatric preparations (e.g. Gerovital H₃, Aslavital, Proneuril) belongs to the group of cationic anesthetics, which act at the level of cell membrane (5), (17), (26), (27), (28), and that procaine-HCl increases the cell membrane area and changes the membrane permeability and cellular metabolic activities (1), (2), (3), (10), (24), (31). On the other hand, recent investigations in our laboratory demonstrated that procaine-related drugs (Aslavital and Gerovital H₃) and metabolites (PABA and DEAE) stimulate the rat brain mitochondrial respiratory activity (oxygen consumption, cytochrome oxidase and succinate-dehydrogenase activities) and that DEAE exert its action mainly on cell-membrane bound enzymes, while procaine-HCl caused changes in the activities of both mitochondrial and cell membrane bound enzymes at the level of rat brain hemispheres (2).

In conclusion, our experiments suggest that procaine-HCl, tetracaine-HCl, dibucaine, lidocaine, Gerovital H₃, Aslavital, as well as 4-para-amino-benzoic-acid exert specific direct stimulatory effects upon glucose uptake by isolated normal rat brain, under aerobic conditions.

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THE BIOSYNTHESIS ANTIBIOTIC PREPARATION A 37.4 — A NEW ACTIVE CANCEROSTATIC AGENT

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The comparative analysis of the antitumoral activity induced by A 37.4 with those of the Antipholan, Levopholan and Cyclophosphamid — cytostatics chosen as reference agents — revealed the significant therapeutic effectiveness of the new antibiotic preparation.

The data of the present paper complete the preclinical experimental evidences which recommend the A 37.4 biosynthesis antibiotic as an antineoplastic agent.

In previous papers the antitumoral therapeutic action of the A 37.4 antibiotic, the reproducibility of this effect, the existence of dose-response relationship and the significant cancerostatic efficiency on tumors with different degrees of development were revealed (4), (5), (6), (7).

The preclinical characterization of a substance as antineoplastic agent requires also the comparative analysis of its antitumoral effectiveness with that of a standard cancerostatic (2), (3), (10).

In the present work the results obtained by "in vivo" antitumoral activity testing of the A 37.4 antibiotic preparation and of some reference agents of clinical use (Antipholan, Levopholan and Cyclophosphamid) on rats bearing of various experimental tumoral systems are exposed.

MATERIAL AND METHODS

White Wistar female rats, weighing 150 g, bearing either of Guérin T-8 lymphotropic epithelioma in solid form and its ascitic variant, or of Walker 256 carcinosarcoma were used as experimental animals.

24 hours after the tumoral transplant the treatment started and lasted for 16-19 days, in the case of solid subcutaneous tumors, or until the death of the last animal for the ascitic tumor.

In our experimental protocol the A 37.4 antibiotic, used in a combined therapy with NsMC 2 semisynthesis polyene (chemically modified nystatin) and three clinical cytostatics (Antipholan, Levopholan and Cyclophosphamid) were tested.

The treatment was applied by intraperitoneal (ip) administration of the drugs in different doses and at various intervals, which are included in the tables containing the experimental results.

The estimation of the antitumoral activity was based on the follow up of the mean tumor weight (MTW) at sacrifice in the case of solid tumors or of the mean survival time (MST) in the case of ascitic tumor in the treated groups as compared with the controls.

The evaluation of the cancerostatic effect was made by the percentage determination of the mean tumor regression (%MTR) for solid tumors, of the mean survival time increase (%MSTI) for ascitic tumor and by the calculation of the statistical significance and of the T/C value (where T = MTW or MST for the treated group and C = MTW or MST for the control).

RESULTS

The antitumoral activity induced by A 37.4 antibiotic preparation, Antipholan, Levopholan and Cyclophosphamid on rats with Guérin T-8 solid tumor is presented in Table 1.

It can be seen that — in comparison with the control group — the A 37.4 administration (0.075 mg/kg.b.w.) at 3-days interval, concomitantly with a daily injection of NsMC 2 (50 mg/kg.b.w.), was correlated with a significant decrease ($p/0.01$) of the MTW, which allows the estimate of a MTR of 50.4% and a T/C value of 0.49.

Table 1

Antitumoral activity of the combined therapy with A 37.4 (0.0075 mg/kg.b.w./at 3 days/ip) and NsMC 2 (50 mg/kg. b.w./daily/ip), of Antipholan (0.15 mg/kg.b.w./daily/ip), Levopholan (0.30 mg/kg. b.w./daily/ip), Cyclophosphamid (1.6 mg/kg.b.w./daily/ip) treatments on solid Guérin T-8 tumor. Figures in brackets indicate the number of animals

Group/Treatment	M.T.W. (g)	% M.T.R.	T/C value	Statistical significance
Control	13.5±1.7(13)	—	—	—
A 37.4 + NsMC 2	6.7±1.1(9)	50.4	0.49	$p/0.01$
Antipholan	7.1±2.2(9)	47.4	0.53	$p/0.05$
Levopholan	6.7±2.2(9)	50.4	0.49	$p/0.05$
Cyclophosphamid	5.8±2.7(9)	57.1	0.42	$p/0.05$

It is observed that significant antitumoral effects were also induced by Antipholan, Levopholan and Cyclophosphamid daily treatments, as compared with the control. Thus, the evaluation indices values of their corresponding activities are:

- a MTR of 47.4%, 50.4% and 57.1%, respectively;
- a T/C report of 0.53, 0.49 and 0.42, respectively.

The comparison of the cancerostatic effect of the biosynthesis secondary metabolite A 37.4 with those of the Antipholan, Levopholan and Cyclophosphamid — cytostatics chosen as standard agents — was also performed on Walker 256 tumoral line (Table 2).

The values of evaluation indices revealed significant antitumoral activities ($p/0.01$ and $p/0.001$, respectively) in the case of A 37.4 and Antipholan experimental therapies (a MTR of 58.2% and 76.8%, respec-

Table 2

A 37.4±NsMC 2, Antipholan, Levopholan and Cyclophosphamid antitumoral activities on the rats bearing of Walker 256 tumor treated with the same doses as in previous experiment.

Group/Treatment	M.T.W. (g)	% M.T.R.	T/C value	Statistical significance
Control	19.4±2.1(15)	—	—	—
A 37.4 + NsMC 2	8.1±1.7(9)	58.2	0.42	$p<0.01$
Antipholan	4.5±0.8(8)	76.8	0.23	$p<0.001$
Levopholan	15.7±3.0(9)	19.1	0.81	N.S.
Cyclophosphamid	15.4±3.0(9)	20.7	0.79	N.S.

tively; a T/C value of 0.42 and 0.23, respectively) and a nonsignificant effect in the case of Levopholan and Cyclophosphamid treatments (a MTR of 19.1% and 20.7%, respectively; a T/C value of 0.81 and 0.78, respectively).

The results of the antitumoral effect investigation of the A 37.4 antibiotic preparation and of the standard cancerostatics on rats bearing ascitic tumor are exposed in Table 3.

Table 3

Cancerostatic effect of the same doses of A 37.4 antibiotic preparation, administered in association with NsMC 2 and of the reference agents on the ascitic rats

Group/ Treatment	M.S.T. (days)	% MSTI	T/C value	Statistical signific.	% tumor un- development
Control	20.1±1.6(13)	—	—	—	—
A 37.4 + NsMC.2	41.1±2.2(6)	104.4	2.04	$p<0.001$	40.0
Antipholan	44.1±4.7(6)	119.4	2.19	$p<0.001$	40.0
Levopholan	51.0±4.2(6)	153.7	2.53	$p<0.001$	40.0
Cyclophosphamid	16.2±1.2(10)	21.0	0.79	$p<0.01$	—

Comparatively with the control group it is ascertained that the A 37.4—NsMC 2 combined therapy as well as Antipholan and Levopholan treatments were associated with significant increases ($p<0.001$) of MST. The MSTI (104.4%, 119.4% and 153.7%, respectively) and T/C (2.04, 2.19 and 2.53, respectively) values suggest a strong cancerostatic effect of these substances, reflected also by the percentage of tumoral undevelopments (40%), registered in these cases.

On the contrary, as compared with the control group and also with those treated with A 37.4, Antipholan and Levopholan in the case of Cyclophosphamid treatment a significant decrease ($p<0.01$) of MST was observed.

DISCUSSION

The preclinical characterization of a substance as antineoplastic agent is the result of a complex qualitative and quantitative evaluation of its specific activity. To this purpose the evidence of the cancerostatic action, the demonstration of the reproducibility and stability of this therapeutic effect — required by the qualitative evaluation —, the appreciation of therapeutic effectiveness, by the establishment of the existence of dose-response relationship, by the recording of a significant effect on tumors with different degrees of development and by the comparison of the new drug antitumoral activity with that of a standard agent — imposed by the quantitative evaluation — are necessary (1), (2), (3), (8), (9), (10).

The comparative analysis of the antitumoral therapeutic efficiency of A 37.4 antibiotic preparation with those of Antipholan, Levopholan and Cyclophosphamid, used as reference cancerostatics, was the purpose of the present paper.

Antitumoral treatments performed with all these preparations were correlated with close inhibitory effects on the development of solid Guérin T-8 lymphotropic epithelioma.

The values of evaluation indices of the anticancerous activity registered on rats bearing Walker 256 carcinosarcoma revealed a significant therapeutic effectiveness of the A 37.4 antibiotic preparation. It is greater than those of Levopholan and Cyclophosphamid and somewhat smaller than that of Antipholan.

The A 37.4-NsMC 2 combined therapy as well as Antipholan and Levopholan treatments induced strong and close cancerostatic effects on the ascitic system, illustrated by the evaluation indices and by the tumoral undevelopment percentage.

The comparative following of the antitumoral activity induced by A 37.4 and by Antipholan, Levopholan, Cyclophosphamid, respectively, revealed that the antitumoral effectiveness of the antibiotic treatment is similar or near those of the reference agents and even higher in some cases.

The enhanced therapeutic efficiency of the A 37.4 secondary metabolite provides a positive answer to this question of the screening programs. The present data complete the experimental evidences which allows us to appreciate the A 37.4 antibiotic preparation as an active cancerostatic agent.

However, the final preclinical characterization of the A 37.4 as a new antineoplastic antibiotic requires further investigations in order to establish its antitumoral spectrum and its action mechanism.

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CHARACTERIZATION OF THE MAIN BIOGENOTIC COMPONENTS OF SOME NATURAL AND PLANTED FORESTRY ECOSYSTEMS FROM THE LETEA BANK (DANUBE DELTA)

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The paper presents elements of cenotic structure of primary producers, consumers on the soil surface and canopy and decomposers in soil of a two natural forests (Hasmacul Mare and Hasmacul Mic) and in a poplar and alder plantation, placed in Letea sand bank as well as some pedo-climatic characteristics of those biotopes. The main ecophysiological indexes: the content of chlorophyll and carotenoid pigments, osmotic pressure, sugar concentration and pH of cell sap and the accumulated supraterranean biomass presented specific values for the analysed species.

Invertebrate fauna from the herbaceous layer and canopy is dominated by phytophagous: that from edaphon, by acarina and collembola whose weak decomposing activity is compensated by the soil microbiocenoses.

The Danube Delta with its characteristic flora and fauna has for long been the object of many research-works. But extensive studies based on the structure and functioning of the ecosystems interdependence were rare.

Our research have been undertaken in two natural forestry ecosystems in the climax phase, compared with the young plantations. The directed intervention of man for the modification of some ecological factors bears on the evaluation of Delta biocenoses. That is the reason which demonstrates the necessity to preserve all the area of the Danube Delta.

Our research has been conducted in Roșca-Letea forest with Hasmacul Mare and Hasmacul Mic areas, some protected objective under the rules of biosphere reservations. Also on the Cardon bank, poplar and alder plantations have been studied in order to establish the antropic modifications.

INVESTIGATION METHODS

The research methods and working techniques in the field and in the laboratory were established depending on the biotopes, MAB standards and the type of studied animals. For the determination of the structure of the main phytocenoses, a number of surveys was taken by cutting the herbaceous layer, with three repetitions. The area was 50/50 sq. cm. The biomass of herbaceous layer was established in the fresh and dry state (at 80°C); from an ecophysiological stand point, the content in sugar was analysed by refractometry, the assimilatory pigments by the methods of Comar and Zscheille for chlorophyll and Holm for carotenoids and the pH was studied potentiometrically.

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The canopy and the herbaceous layer fauna was collected in a sweep net with a 60 cm 30 cm diameter respectively (each sample = 50 shakings).

The acariens, nematods, enchytraeids and collembola were collected at 10 cm deep in soil with a MacFadyen soil core-borer; sample extraction was made by automatic extractors. The lumbricids were collected from the first 40 cm deep, along 4 equal layers.

The dehydrogenase activity was determined according to Cassida (1963) and Ștefanic (1970).

In Hasmacul Mare area a meteorological station was installed.

The forest researched belongs to the geobotanical association of *Quercetum robori-pedunculiflorae* Simon 60, located in the higher areas of the studied surfaces and *Fraxino (pallisiae-angustifoliae)-Quercetum roboris* Popescu, Sanda, Doltu 79 subass. *fraxinetosum pallisiae* (Krausch 65) Sanda et al. 87 which occupied the lower areas of the same surfaces.

The alder (*Alnus glutinosa*) and poplar (*Populus alba* and *Populus canescens*) are rather extended at Cardon site.

RESULTS AND DISCUSSION

ELEMENTS OF BIOTOP (STATIONS)

The forestry ecosystems taken into study are to be found in the maritime delta of the Letea bank at 5 metres altitude, in low depressions with small unevennesses. The climate is dry continental of Danubian type, characterized by high thermal amplitude, by active wind conditions and little rainfall. The medium annual temperature oscillates around 11°C and the annual medium amplitude is over 22°C. The insolation has a power of 1.5 cal/cm²/minute. The rainfall quantity is the lowest in the country, below 450 mm annually.

The phreatic water works under most fluctuating conditions, going from spring marshes to below 1 m in depth, depending on the level of the Danube waters.

The soil is generally of psamosoil type, formed on sandy material with high contents of loams, most rich in calcium carbonate having in its basis a C horizon, which consists largely in lacustrine chalky stone.

The reaction of the soil is weakly alkaline, and it becomes stronger with the depth; the relation C/N is 11–12, the humus horizon is situated among the first 20 cm (Table 1).

Table 1
Some edaphic factors in soil active area

Soil depth	Acidity	Humus	CaCO ₃	N total	P mobile ppm	K assimilated ppm
2–12	7.65	3.6	14.7	0.19	50.0	60
12–22	7.9	2.1	14.7	0.08	24.0	30
40–50	8.8	0.1	7.8	0.02	36.0	20
70–80	9.0	0.07	9.5	0.01	16.0	40

ELEMENTS OF BIOCENOSIS

Primary producers

The primary producers are stratified on three layers: the tree layer which is uniform, bushes and the herbaceous layer.

Hasmacul Mare — a natural forestry ecosystem situated in low depression, with small unevennesses, 5 m altitude. The composition of the tree layer is given by *Quercus robur* and *Quercus pedunculiflora* codominant, to which *Fraxinus pallisiae*, *Populus alba*, *Populus canescens* can be sporadically added. The relative plurien total layer tree is 70–155 years in age and has a medium height of 23–25 m. The undertree layer can reach 3 to 6 metres and a medium cover of 25–30%. It consists mainly of *Crataegus monogyna*, *Pyrus pyraister*, *Cornus sanguinea*, *Malus sylvestris*, etc. The herbaceous layer is dominated by *Rubus caesius*, with the participation of several species characteristic to the alliance *Alno-Ulmion* and to *Querco-Fagetea* class (Table 2).

Table 2

Fraxino (pallisiae-angustifoliae)-Quercetum roboris Popescu, Sanda, Doltu 79 *fraxinetosum pallisiae* (Krausch 65) comb. nova (surveys 1–7) and *Quercetum robori-pedunculiflorae* Simon 60 (surveys 8–11)

Biological form	Floristic element	Survey number										
		Area (sq.m)										
		1	2	3	4	5	6	7	8	9	10	11
Vegetation high	Trees(m)	25	30	32	15	13	14	15	25	35	14	20
	Bushes(m)	6	6	4	5	4	5	4	4	6	3	6
	Herbs (cm)	70	65	25	75	100	70	70	30	25	40	50
Cover area (%)	Trees	85	80	85	80	70	70	60	85	90	80	80
	Bushes	30	25	15	15	10	25	15	30	25	25	10
	Herbs	60	60	30	60	65	60	80	40	40	50	60

Association characteristics

MM Pt-Blc	<i>Fraxinus pallisiae</i>	4	4	3–4	4	4	4	4	+	1	1
MM Pt-Blc	<i>Quercus pedunculiflora</i>	.	.	.	+1	+	+	+	4	4	4
MM Ec(Md)	<i>Quercus robur</i>	+	+	1	1	+

Local differentiation species

M–E Pt-Md	<i>Vitis sylvestris</i>	.	.	.	+	+	.	.	+	+	+
E Md	<i>Periploca graeca</i>	+	+	+	+	+	+	+	+	+	+

Alno-Ulmion

M Eua(Md)	<i>Salix cinerea</i>	1–2	+	.	+	1–2	+	+	+	.	+
M Eur	<i>Malus sylvestris</i>
H(N) Eua	<i>Rubus caesius</i>	+	2–3	+1	2–3	3	1–2	2	+	+	1–2
HH-H Ct(Md)	<i>Symphytum officinae</i>	+	+	.	+	+	+	+	.	+	1
MM Eua	<i>Populus alba</i>	+	+
MM Eua	<i>Populus canescens</i>	.	+	.	+	+	.	.	.	+	.
M Eua(Md)	<i>Salix fragilis</i>	+	.	+	.
Ch–H Eua	<i>Glechoma hederacea</i>	+	.	.	.	+	.
M Cp	<i>Viburnum opulus</i>	+
M Eua	<i>Frangula alnus</i>	+	+	+
H Eua(Md)	<i>Brachypodium sylvaticum</i>	1–2	+	+	1	.

Table 2 (continued)

Survey number	1	2	3	4	5	6	7	8	9	10	11
<i>Alnion + Alnetea</i>											
HH Eua(Md)	<i>Carex acutiformis</i>	+	1-2	+	+	+	+	+	+	+	+
G E-Md	<i>Iris pseudacorus</i>	+	+	+	+	+	+	+	+	+	+
HH Eua	<i>Lycopus europaeus</i>	+	+	+	+	+	+	+	+	+	+
HH Cs	<i>Phragmites australis</i>	+1	+	+	+	+	+	+	+	+	+
H Eua(Md)	<i>Eupatorium cannabinum</i>	+	+	+	+	+	+	+	+	+	+
H Eua	<i>Lysimachia vulgaris</i>	+	+	+	+	+	+	+	+	+	+
<i>Salicion</i>											
H Pt-Pn	<i>Galium rubioides</i>	+	+	+	+	+	+	+	+	+	+
<i>Quercio-Fagelea</i>											
G Eur	<i>Convallaria majalis</i>	+	+	+	+	+	+	+	+	+	+
Ch E(Md)	<i>Lysimachia nummularia</i>	+	+	+	+	+	+	+	+	+	+
M E(Md)	<i>Ligustrum vulgare</i>	+	+	+	+	+	+	+	+	+	+
G Eua	<i>Epipactis helleborine</i>	+	+	+	+	+	+	+	+	+	+
H Pt-Md	<i>Asparagus tenuifolius</i>	+	+	+	+	+	+	+	+	+	+
M E	<i>Crataegus monogyna</i>	+1	1-2	+	+	+	+	+	+	+	+
M Euc	<i>Cornus sanguinea</i>	+	+	+1	+	+	+	+	+	+	+
M E(Md)	<i>Pyrus pyraster</i>	+	+	+	+	+	+	+	+	+	+
N-E Euc	<i>Clematis vitalba</i>	+	+	+	+	+	+	+	+	+	+
<i>Agrostion stoloniferae</i>											
H-HH E	<i>Euphorbia palustris</i>	+	+	+	+	+	+	+	+	+	+
H-HH Eua	<i>Lythrum virgatum</i>	+	+	+	+	+	+	+	+	+	+
<i>Molinio-Arrhenatheretea + Molinietales</i>											
G E(Md)	<i>Carex hirta</i>	+	+	+	+	+	3	+	+	+	1-2
H Eua	<i>Poa silnicola</i>	+	+	+	+	+	+	+	+	+	+
H Cs	<i>Potentilla reptans</i>	+	+	+	+	+	+	+	+	+	+
H Cp	<i>Mentha arvensis</i>	+	+	+	+	+	+	+	+	+	+
H Cp	<i>Poa pratensis</i>	+	+	+	+	+	+	+	+	+	+
<i>Accompanying species</i>											
H Eua	<i>Plantago major</i>	+	+	+	+	+	+	+	+	+	+
H Cp(Md)	<i>Galium palustre</i>	+	+	+	+	+	+	+	+	+	+
H Eua	<i>Galium mollugo</i>	+	+	+	+	+	+	+	+	+	+
H Eua	<i>Calystegia sepium</i>	+	+	+	+	+	+	+	+	+	+
H Eua(Md)	<i>Stachys officinalis</i>	+	+	+	+	+	+	+	+	+	+
H Eua(Md)	<i>Coronilla varia</i>	+	+	+	+	+	+	+	+	+	+

Present species in a survey: *Asparagus tenuifolius* (2), *Alnus glutinosa* (2), *Astragalus glycyphyllos* (7), *Anthericum ramosum* (8), *Alliaria petiolata* (9), *Agropyron repens* (10:2), *Berula erecta* (7), *Berberis vulgaris* (9), *Asparagus pseudoscaber* (10), *Carex vulpina* (5), *C. contigua* (8:2), *Corylus ovellana* (9), *Galium oparine* (3), *Festuca valesiaca* (8), *Hedera helix* (1), *Lythrum salicaria* (3), *Linaria vulgaris* (8), *Lepidium latifolium* (10), *Heracleum sphondylium* (9), *Polygonatum officinale* (3), *P. latifolium* (9:1), *Prunus spinosa* (3), *Physalis alkekengi* (7), *Pulicaria dysenterica* (8), *Ranunculus repens* (7), *Rhamnus cathartica* (9), *Rosa canina* (5), *Scutellaria galericulata* (5), *Stachys palustris* (7), *Typha angustifolia* (7), *Verbascum phoeniceum* (9), *Vicia cracca* (11), *Vincetoxicum hirundinaria* (8), *Viburnum lantana* (1).
The place of surveys: 1, 2, 8-Hasmacul Mic: 3, 4, 5, 6, 7, 9, 10, 11-Hasmacul Mare.

Hasmacul Mic, a natural forestry ecosystem, the medium height of the trees is 14 to 16 metres. The composition of the tree layer is *Fraxinus angustifolia*, *Fraxinus pallisae*, *Quercus robur*, *Q. pedunculiflora*. The undertree layer consists of *Berberis vulgaris*, *Crataegus monogyna*, *Cornus sanguinea*, *Pyrus pyraster*, etc., the medium height being 6-7 m.

The species *Alnion* and *Alnetea*: *Carex acutiformis*, *Iris pseudacorus*, *Lycopus europaeus*, *Phragmites australis* are dominant in the herbaceous layer. They indicate higher humidity conditions for this biotope. 16% of the trees are covered by *Perliploca graeca* and *Vitis sylvestris* liana.

Biomass accumulation in the primary producers

Studied vegetable populations have a characteristic biomass because of the alternation of subhygrophilous and mesophilous associations. In the frame of subhygrophilous vegetation, biomass was determined to the herbaceous species belonging to the following associations: *Fraxino (pallisae-angustifoliae)-Quercetum roboris* Popescu, Sanda, Doltu 79, *Alnetum glutinosae* Mejer-Drees 36 (ass. cult.), *Populetum albae* Br.-Bl. 31 (ass. cult.).

Herbaceous species belonging to the association *Fraxino (pallisae-angustifoliae)-Quercetum roboris*, identified in both biotopes Hasmacul Mare and Hasmacul Mic, are characterized by large values of the total biomass determined by environmental factors (air and soil temperature and humidity, recorded rainfall). Fresh biomass per sq.m. assumed the largest values: 650.22 g/sq.m; dry biomass showed smaller values (120.22 g/sq.m) because of the fact that herbaceous plants have a rather large content of water (77.9%) (Table 3).

Table 3

The variation of vegetation biomass and water content of the plants from Letea bank

Nº	The association	Type	Station	Fresh biomass g/m ²	Dry biomass g/m ²	The mean of water content(%)
1	<i>Fraxinetum pallisae</i>	subhygrophilous	Hasmacul Mare	657.92	120.93	78.23
2	<i>Fraxinetum pallisae</i>	"	Hasmacul Mic	642.53	119.54	77.69
3	<i>Alnetum glutinosae</i>	"	Cardon	322.11	97.77	61.7
4	<i>Populetum albae</i>	"	Cardon	318.02	100.39	69.44
5	<i>Quercetum robori-pedunculiflorae</i>	mesophilous	Hasmacul Mare	267.86	64.64	73.88

Herbaceous populations from poplar and alder populations at Cardon are characterized by a smaller total biomass, because of the smaller water content in soil and of higher temperature in air and soil. (The variation amplitude of fresh biomass was 318.1-322.1 g/sq. m, 97.7-100.4 g/sq.m of dry biomass and 61.7-69.4% of the average water content.

Herbaceous populations of *Quercetum robori-pedunculiflorae* Simon 60 association is characterized by a small biomass because of the fact that quercines species, as dominant species with high densities, produced a pronounced shade of the herbaceous layer disturbing the development of plants.

Fresh biomass was 267.8 g/sq m, dry biomass was 64.6 g/sq m and the average water content of plants was 73.8%.

Indicator of plant metabolic activity

The link between sun energy and the capacity of biomass producer of autotrophic plants consists in the chlorophyllian and carotene pigments. The contents of assimilatory pigments from the edifying leaves of the plants is small with each of the analyzed populations in the natural forests (Letea), especially in the plants from Cardon (Table 4). In comparison with the wooden populations belonging to the same species in the hill and field forests, the populations in the Danube Delta act as "light plants" even under more shadowy conditions which denotes that there are optimum possibilities for ecological evaluation of these stations.

Table 4

The contents of assimilatory pigments in the forests populations

The species	The station	Total chlorophyll	Relation chlorophyll a/b	Carotenoid pigments	Relation chlorophyll/carotenoid	Osmotic pressure	Sacchars (%)	pH
<i>Quercus pedunculiflora</i>	Hasmacul Mare	88	2.61	16	5.67	25.11	13.0	7.8
<i>Quercus robur</i>	"	89	2.72	14	6.33	25.83	15.0	3.7
<i>Alnus glutinosa</i>	"	143	3.27	36	4.00	21.04	8.0	4.7
<i>Populus canescens</i>	"	75	3.00	18	4.25	22.0	15.0	5.0
<i>Quercus pedunculiflora</i>	Hasmacul Mic	105	2.69	16	6.54	26.16	13.0	8.0
<i>Quercus robur</i>	"	109	2.64	16	6.69	25.35	13.0	4.3
<i>Populus canescens</i>	Cardon	68	3.25	21	3.23	21.76	10.0	5.4
<i>Alnus glutinosa</i>	"	100	3.06	24	4.05	25.83	13.0	4.3

The osmotic potential of the vegetal species varies with the species, with the development degree, the pedoclimatic conditions. As part of the research we made on the analyzed phytocoenosis, we chose the dominant forestry species which characterize best the respective biotope.

For the great majority of the species we took into study, remarks must be made on the lower value of the osmotic pressure in Hasmacul Mare as well as in Hasmacul Mic, within the poplar and alder trees; thus, this ecophysiological parameter varies between 21.04 bars (*Alnus glutinosa*) and 25.83 bars (*Quercus pedunculiflora*), both species being present in Hasmacul Mare. The values of the osmotic pressure for *Quercus pedunculiflora* in Hasmacul Mare and Hasmacul Mic are rather close. As concerns the concentration of sacchars in the cellular juice, there is a differential dynamics correlated with the phenophases sequence and with the osmotic pressure (Table 4).

The cellular juice pH indicates a rhythm of evolution of the plant metabolism in direct relation with the possibilities of evaluation of the station. For the analysed species, the pH rises above the 4.0 value in most of the cases, which is in agreement with the idea of the existence of conditions favourable to a metabolic activity development; the values

deviating from those mentioned above represent the consequence of plant adaptation to stational conditions.

CONSUMERS AND DECOMPOSERS

The fauna of invertebrates in the canopy is a precise function in the trophic chain of forestry ecosystems. The forest biocoenosis contains in its structure a trophic chain made up of 3-4 links; each link produced does not surpass 10% from the consumed one. Thus a small production of consuming organisms may be reached from a large production of phytomass.

The fauna in the canopy comprises phytophagous organisms dip-terous, homopterous, etc. (Table 5). The accumulation of biomass achieved by these is most reduced (a few kg/year/ha), and so is the efficiency of using the biomass at the trophic level of phytophags, below 10%. Their biomass is used by the upper trophic chain, by secondary consumers (some insects, aranea), this aspect being stronger when leaves-eater insects interfere.

Table 5

The relative abundance of invertebrates fauna in canopy

Taxonomic group	Hasmacul Mare	Hasmacul Mic	Alder plantation	Poplar plantation
Thysanoptera	3.09	—	—	2.15
Heteroptera	8.26	4.28	—	2.15
Homoptera	30.94	6.38	5.43	39.15
Hymenoptera	2.08	2.10	32.41	—
Coleoptera	9.28	4.28	—	10.89
Lepidoptera	7.21	8.49	13.53	6.52
Diptera	19.59	55.33	10.79	10.89
Dermoptera	7.21	2.10	13.53	8.68
Mecoptera	1.02	—	—	—
Araneae	9.28	10.66	21.64	8.68
Acarina	1.02	—	—	4.37
Opilionidae	1.02	6.38	—	6.52

In the research surfaces, the Diptera are mostly represented by mosquitoes, Coleoptera of numerous curculionids, Thysanoptera by *Dendrothrips ornatus* and *Haplothrips kurdjumovi*, the lepidoptera *Lymantria dispar* and *Tortrix viridana* in the Hasmacul Mare and Hasmacul Mic and by *Melasoma populi* in poplar plantation. The lepidoptera species leaves-eater insects, most main forestry pest, do not exhibit a mass reproduction phenomenon.

The invertebrate fauna in the herbaceous layer is represented by organisms belonging to 10 taxons, primary and secondary consumers. A maximum of microarthropods were collected in Hasmacul Mare, where the heteroptera are dominated by Reduviidae, coleoptera by *Malachuis bipustulatus* * (Fam. Cantharidae), Araneas by *Thomisidae* (juvenile forms) and hymenoptera by *Lasius niger* *. In poplar plantation the invertebrate

* The coleopter species were determined by Mr. Th. Nalbant, scientific researcher, and the Formicide ones by Dr. D. Paraschivescu.

fauna is the least numerically represented. The relative abundance, which expresses the participation degree of each component to the making of zoocenosis, indicated maximum percentage values for mosquitoes (Diptera) on all the four researched surfaces.

Table 6

The relative abundance of invertebrates fauna in herbaceous layer

Taxonomic groups	Hasmacul Mare	Hasmacul Mic	Alder plantation	Poplar plantation
Collembola	2.68	—	—	—
Thysanoptera	8.00	8.33	4.46	—
Heteroptera	4.00	5.58	6.67	2.61
Homoptera	14.68	11.08	4.46	5.30
Hymenoptera	9.32	11.08	26.65	34.23
Coleoptera	5.32	2.75	28.85	23.72
Diptera	48.00	30.58	13.32	18.42
Dermaptera	—	—	4.46	2.61
Orthoptera	—	—	—	—
Araneae	5.32	22.27	11.13	10.30
Opilionidae	2.68	8.33	—	2.61

THE INVERTEBRATE IN THE SOIL

The processes of storage and using of matter and energy by the decomposers within the studied ecosystems are estimated at the main invertebrate edaphic groups, including mostly the group of detritivorous, bacteriophage but also phytophagous, fungivorous and predatory organisms.

The differentiated contribution of the invertebrate groups to the making of edaphic consumer communities is established in terms of mean density/m² parameters, and by estimating the storage of organic matter under the form of biomass, which expresses their participation degree to the process of matter transformation within the ecosystem. The mean density/m² of invertebrates fauna is maximum on the surface soil of Hasmacul Mare and Hasmacul Mic and diminishes in the planting soil of poplar and alder. Lumbricidae are sporadically present only in the limitrophe layer and by *Allolobophora dubiosa*. In all research surface, the enchytraeids are characterized by the dominant species *Fridericia ratzei* and collembols by *Folsomia quadrioculata*, *Onychiurus armatus* and *Friesea mirabilis*. The oribatids are characteristic bioindicators for the delta types of ecosystems by: *Protoribates monodactylus*, *Oppia minus*, *Oppia absoluta* in Hasmacs, *Mycrozetorcheses emeryi*, *Trichoribates trimaculatus* in poplar plantation and *Tectocephus alatus*, *Zetorcheses myronicus* in alder plantation.

The distribution of the edaphon upon the vertical is influenced by the richness of food and by the pedoclimatic characteristics of the researched surfaces. The great majority of organisms inhabit the limitrophe layer, their numerical density being in a negative connection with the depth of prelevation samples.

Table 7
Numerical density of edaphic fauna

Surfaces	Group of organism														
	Lumbricidae			Enchytraeidae			Nematoda			Collembola			Acarina-Oribatei		
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
Hasmacul Mare	4.56	9.6	—	1000	800	200	330000	266800	159200	13400	11600	9200	18200	49000	13200
Hasmacul Mic	2.28	—	—	600	600	200	282000	340000	105200	15900	12400	8400	2000	115800	19600
Alder plantation	—	—	—	400	200	0	136000	78600	44000	6200	3000	3400	7600	10600	7200
Poplar plantation	—	—	—	200	0	0	184200	90000	58600	10000	9200	6800	17000	47600	19400

1—1985; 2—1986; 3—1987.

Table 8

Biomass of edaphic fauna mg/s.u./m²

Surfaces	Group of organisms														
	Lumbricidae			Enchytraeidae			Nematoda			Collembola			Acarina		
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
Hasmacul Mare	95.76	201.6	—	32.0	25.6	6.4	16.5	13.34	7.96	36.18	31.32	24.84	96.46	259.70	69.96
Hasmacul Mic	47.88	—	—	19.2	19.2	6.4	14.1	17.0	5.26	42.93	33.48	22.68	10.60	613.74	103.88
Alder plantation	—	—	—	12.8	6.4	—	6.8	3.93	2.2	16.74	8.1	9.18	40.28	56.18	38.16
Poplar plantation	—	—	—	6.4	—	—	9.21	4.5	2.93	27.0	24.84	18.36	90.10	252.28	101.52

1—1985; 2—1986; 3—1987.

The estimation of the organic matter storage as biomass completes the information on the soil decomposing fauna. The quantity of organic matter stored as biomass by invertebrates is generally low, owing to the specific conditions on Letea bank (Table 8). In Hasmacul Mare the achieved biomass is 4.57 times higher than that from alder plantation and 1.71 times higher than that in poplar plantation. The values of the populational ecological indexes of soil fauna assume a descending curve, from 1985 up to 1987. This complex causal phenomenon is correlated with station and general pedoclimatic factors unfavourable to edaphic coenosis development.

DEHYDROGENASE ACTIVITY

An index of the total activity of soil microbiocenosis, the present and potential dehydrogenase activity exhibited the highest values on Hasmacul Mic surface (Table 9). In poplar and alder plantation, the present and potential dehydrogenase activity is lower than that in the natural forestry ecosystems under study. The potential dehydrogenase activity is constantly higher than the existing one, on all surfaces under examination. An outstanding increase of the dehydrogenase activity could be noticed from 1985 up to 1987; this happened in conditions in which the main components of the edaphon strongly diminished while the microorganisms equilibrated. Thus, the soil decomposing activity and, in this way, the process of pedogenesis accomplishes under normal conditions.

Table 9

Level of dehydrogenase activity (expressed in formazan mg/100 g dry soil)

Surface	Present Potential		Present Potential		Present Potential	
	1	2	3	4	5	6
Hasmacul Mare	4.23	8.26	3.55	5.98	7.78	15.69
Hasmacul Mic	9.55	5.75	3.84	7.23	15.13	34.79
Alder plantation	0.07	0.13	1.30	2.60	4.75	10.98
Poplar plantation	0.35	0.50	0.75	2.08	6.24	12.33

CONCLUSIONS

The primary producers, the primary and secondary consumers and decomposers studied in the present paper differ according to the station and to the type of natural or planted forestry ecosystem. In Letea forest, a balanced representation of the main trophic levels is revealed, ensuring thus a normal ratio of the matter circulation in the ecosystem. The ecological indexes of the vegetal and animal populations show higher values in the natural forestry ecosystems if referred to the planted ones.

Hence, the extreme importance of the preservation of the entire nature in the Danube Delta.

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L'ÉVOLUTION MULTIANNUELLE DU PLANCTON BACTÉRIEN DANS LES ÉCOSYSTÈMES AQUATIQUES DU DELTA DU DANUBE ET DU SECTEUR PRÉDELTAÏQUE

DORINA NICOLESCU

Les recherches sur la densité numérique des bactéries planctoniques dans les écosystèmes aquatiques du Delta du Danube et du secteur prédeltaïque pendant 14 années ont mis en évidence le fait que le développement des producteurs primaires régit l'évolution de la microflore bactérienne planctonique, le développement de celle-ci suggérant un caractère cyclique, à des maxima et minima entre 4 et 6 ans. On relève en même temps que les écosystèmes aquatiques appartenant au biome deltaïque évoluent d'une manière unitaire, tout en étant marqués par le spécifique local.

A la suite de recherches complexes sur les biocénoses des écosystèmes aquatiques de Delta du Danube et du secteur prédeltaïque, dans la période 1974—1987, on a mis en évidence la place du plancton bactérien dans la hiérarchie trophique et son rôle dans le fonctionnement de ces écosystèmes (1; 2).

Les recherches détaillées sur les paramètres structurels et fonctionnels du bactérioplancton (avec référence à la densité numérique), pendant 14 années, ont surpris la dynamique de cette composante biocénotique, dans le contexte général de l'évolution des écosystèmes deltaïques et prédeltaïques.

Dans ce travail, nous allons porter notre attention sur les déterminations numériques des bactéries chimio-organotrophes du type hétérotrophe, dominantes dans les écosystèmes aquatiques deltaïques du type autotrophe. Les recherches se sont déroulées, par étapes, dans les suivantes catégories d'écosystèmes :

- a) lacustres : Roşu (1974—1979; 1987); Puiu (1977—1979); Porcu (1976—1979); Iacob (1975); Răducu (1975; 1977); Matiţa et Merheiu (1980—1982); Roşuleţ (1987);
- b) fluviaux : les bras du Danube à l'embouchure (1974—1975), notamment Sulina et Sf. Gheorghe;
- c) lagunaires : le secteur prédeltaïque, notamment le golfe Musura, la lagune Sacalin.

Un tableau synoptique de la densité numérique bactérienne du plancton dans les écosystèmes lacustres deltaïques, en fonction de la période pendant laquelle les recherches ont eu lieu (Tableau 1), met en évidence de grandes variations, de 0 à 8 par ordre de grandeur, durant la même année. Une vue d'ensemble (Tableau 1, Fig. 1) nous permet d'apprécier que :

— une comparaison de la microflore bactérienne planctonique ne peut se réaliser qu'entre des écosystèmes étudiés pendant la même période de temps, étant donné l'évolution d'ensemble des écosystèmes deltaïques. Ainsi, pendant la période 1976—1979, les écosystèmes lacustres

Tableau 1

La microflore bactérienne planctonique — densité numérique (limites de variation) — n° 1

Lac	Année					
	1974	1975	1976	1977	1978	1979
Roșu	10^8-10^9	10^8-10^9	10^7-10^9	10^7-10^{13}	$10^{11}-10^{17}$	$10^{12}-10^{13}$
Puiu				10^8-10^{13}	$10^{11}-10^{16}$	$10^{12}-10^{13}$
Porcu			10^7-10^{10}	$10^{11}-10^{13}$	$10^{12}-10^{16}$	10^{12}
Iacob	10^8-10^9	10^8-10^{10}				
Răducu			10^8	10^9-10^{14}		
Matia		1980	1981	1982		
Merheiu		$10^{10}-10^{14}$	10^8-10^{14}	$10^{10}-10^{13}$		
		10^9-10^{17}	10^6-10^{13}	10^8-10^{14}		
			1987			
Roșu			$10^{10}-10^{12}$			
Roșuleț			$10^{10}-10^{12}$			

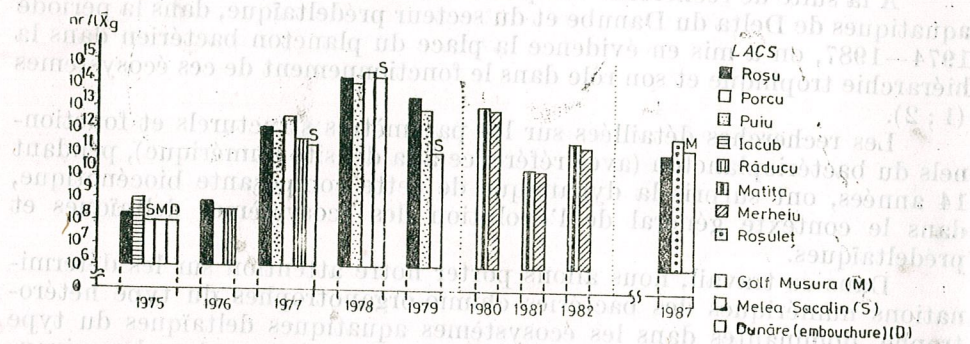


Fig. 1.— Evolution multiannuelle de la microflore bactérienne planctonique des écosystème aquatiques du Delta du Danube et du secteur prédeltaïque (densité numérique) — moyenne géométrique annuelle.

analysés peuvent être classifiés, du point de vue de la densité numérique bactérienne, dans l'ordre de priorité suivant : Porcu, Roșu, Puiu, Răducu ; les recherches effectuées pendant l'année 1974—1975 mettent en évidence des valeurs plus élevées dans le lac Iacob que dans le lac Roșu ; les lacs Matia et Merheiu, analysés pendant la période 1980—1982, présentent des valeurs similaires, avec un légère priorité du lac Matia. Il est à remarquer que, pendant la même année, les différences entre les écosystèmes (comme limites de variation surtout) ne sont pas significatives ; — l'évolution de la courbe de la dynamique multiannuelle de la densité numérique de la microflore bactérienne planctonique, pendant les deux périodes de recherches dans les écosystèmes lacustres, reflète : un bond quantitatif en 1977 (avec trois ordres de grandeur) dans les lacs Porcu, Roșu, Puiu, Răducu, avec le maximum atteint en 1978, après quoi suit un léger déclin (1979) ; la baisse brusque, en 1981 par rapport à 1980, dans les écosystèmes de Matia et Merheiu, de presque trois ordres de grandeur, suivie d'un léger redressement en 1982.

L'alignement des données sur la densité numérique bactérienne du Danube (l'embouchure), du golfe Musura et de la lagune Sacalin — dans le secteur prédeltaïque — aux données des écosystèmes lacustres deltaïques

(Fig. 1) nous justifie à supposer un développement cyclique de la microflore bactérienne planctonique, avec des maxima et minima entre 4 et 6 ans, fait signalé aussi pour la zone du lac Portile de Fier I (1 ; 2). Cette dynamique multiannuelle est engendrée par la dynamique des producteurs primaires — le phytoplancton et les macrophytes submerses, qui enregistrent des maxima de développement dans les lacs Puiu, Roșu, Porcu et la lagune Sacalin en 1978, et seulement le phytoplancton en 1983 dans les lacs Matia et Merheiu (auxquels s'ajoute le lac Portile de Fier I). D'ailleurs, faisant référence à l'évolution des niveaux de trophicité phytoplanctonique dans le Delta du Danube, pendant la période 1976—1985, M. Oltean (3) montre que cela « suggère un caractère cyclique multiannuel des processus de floraison des eaux dans le Delta du Danube ». Il est à remarquer que les valeurs minimales sont enregistrées pendant les années (1975—1976 (Tableau 1, Fig. 1) pour tous les systèmes analysés du Delta et du secteur prédeltaïque, tandis que les années 1982 et 1987 se trouvent dans l'ascendance de la courbe, pareillement à l'année 1977, vers un maximum en 1983 (mis en évidence dans le lac Portile de Fier) et un autre présomptif en 1988 (89).

Le fait que tous les écosystèmes dans lesquels on a mis en évidence ce caractère cyclique du développement des producteurs primaires et, implicitement, de la microflore bactérienne planctonique se trouvent sous l'incidence du Danube nous pousse à attribuer ce phénomène à une dynamique des nutriments ou au rapport entre certains éléments comme N:P (la hausse de la valeur de ces éléments dans le Danube et au Delta étant mise en évidence par V. Vadineanu et S. Cristofor — 4) ; cette explication apparaît comme insuffisante et ne peut justifier que des valeurs partielles maximales ou minimales et non pas le caractère cyclique de celles-ci ; si cette dynamique était caractéristique des systèmes lacustres seulement, nous pourrions la mettre au crédit de la capacité locale de remise en circulation des nutriments ; mais, dans cette situation générale, comme nous l'avons montré, le seul facteur commun dans le développement des producteurs primaires est la quantité et, surtout, la qualité de l'énergie lumineuse (cosmique) qui peut déterminer tant le développement sélectif de ceux-ci que l'intensité de la biosynthèse, associée, certes, à l'accroissement de la quantité de nutriments du Danube, à la modification du régime hydrologique de celui-ci. En même temps, la dynamique de la densité numérique de la microflore bactérienne pendant 14 années, pour les 12 écosystèmes étudiés, met en évidence le fait que les écosystèmes aquatiques qui font partie du biome deltaïque évoluent d'une manière unitaire, tout en étant marqués par le spécifique local.

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L'ÉVOLUTION DE LA STRUCTURE GRAVIMÉTRIQUE DU ZOOPLANCTON DANS LES ÉCOSYSTÈMES DE TYPE LACUSTRE DU DELTA DU DANUBE SOUS L'IMPACT DU PROCESSUS D'EUTROPHISATION (1975-1987)

VICTOR ZINEVICI et LAURA TEODORESCU

Des recherches déployées pendant 13 ans dans 11 écosystèmes de type lacustre représentatifs pour le Delta du Danube poursuivent les modifications intervenues dans la structure gravimétrique du zooplancton (biomasse, abondance relative, dominance).

La dynamique ascendante des nutriments dans les eaux du Delta du Danube détermine des modifications correspondantes dans la structure et la fonction biocénotique. Les caractéristiques de ces mutations diffèrent d'un type d'écosystème à un autre. De la sorte, à partir de 1981, dans les lacs ayant une profondeur au-dessus de 1,7-2,0 m signale-t-on de puissants phénomènes de « floraison de l'eau », la disparition du riche stock de macrophytes submerses et la prolifération massive du bactérioplancton. Dans les lacs ayant des profondeurs inférieures aux niveaux mentionnés, les macrophytes submerses représentent, encore, le principal producteur primaire. Le zooplancton de ce type d'écosystème, analysé au cours des années 1975-1987, met en évidence un large spectre taxonomique (454 éléments) et des valeurs modérées de la densité numérique ($\bar{X}_a = 232 \text{ ex/l}$). En échange, dans les écosystèmes dont les producteurs primaires de type macrophytique ont été remplacés par ceux de type algal, la structure de la zoocénose planctonique diminue de plus de 53% et la densité numérique augmente de presque 5 fois (1), (2). Tous ces changements biocénotiques influencent bien sûr, d'une manière significative, la structure de la biomasse zooplanctonique.

MATÉRIEL ET MÉTHODE

Les recherches portent sur le zooplancton de 11 écosystèmes lacustres différenciés du point de vue temporel et spatial par la nature du producteur primaire : 1) écosystèmes des producteurs primaires de type macrophytique (Iacub, année 1975; Roșu, période 1975-1978; Porcu, 1976-1978; Puiu, 1977-1978; Merhei et Matița, 1980; Bogdaproste et Băclănești, 1982-1986; Roșuleț, 1982); 2) écosystèmes des producteurs primaires de type planctonique (Isacova et Babina, 1982-1986; Merhei, 1981-1983; Matița, 1981-1986; Puiu, 1983; Roșu, 1982-1987). Les valeurs de la biomasse sont exprimées en $\mu\text{g/l}$ substance sèche.

RÉSULTATS ET DISCUSSIONS

Une large gamme typologique des écosystèmes pris en étude, ainsi que la variabilité temporelle accentuée des paramètres ambiants, déterminent des différences significatives d'ordre temporel et spatial

dans la dynamique structurelle de la biomasse zooplanctonique qui se font remarquer au niveau des moyennes annuelles minima (19,4–443 $\mu\text{g/l}$) (Băclănești, respectivement Isacova–1984) et surtout dans le cas des moyennes annuelles maxima (66,4–3138,8 $\mu\text{g/l}$) (Porcu–1976, respectivement Isacova–1983) (tableau 1). Naturellement, les plus amples variations sont propres à la dynamique saisonnière (2,7–8603,2 $\mu\text{g/l}$ Băclănești, décembre 1984, respectivement Isacova, septembre 1983). Les valeurs saisonnières maxima de la biomasse présentent un décalage d'un mois par rapport à celles de la densité numérique (l'intervalle septembre-octobre); en échange, les valeurs minima des deux paramètres structuraux coïncident, étant enregistrées aux extrémités de la période végétative (mars-novembre) (tableau 2).

L'analyse de la biomasse aux niveaux trophiques relève la dominance des consommateurs primaires (63,81–100%) mais cela d'une manière moins nette que pour la densité numérique (2). L'étude de l'abondance gravimétrique par groupes systématiques permet de mettre en évidence le rôle déterminant des cladocères pour le zooplancton c_1 , ainsi que l'apport accru des copépodes ou (rarement) des rotifères pour le zooplancton c_2 (tableau 1).

La dynamique de la biomasse zooplanctonique au cours des années 1975–1987 est caractérisée par un sens évidemment ascendant pendant les premières 8–9 années, avec des maxima remarquables en 1981–1983 et un autre, descendant, d'amplitude plus réduite, au cours des 4–5 derniers ans. Par conséquent, dans les limites de la période mentionnée, on peut apprécier l'existence d'une tendance générale d'accroissement en temps, des valeurs qui caractérisent ce paramètre structural.

De l'ensemble des 498 éléments qui se trouvent dans la structure taxonomique du zooplancton lacustre du Delta du Danube (1), 84 sont dominants sous rapport gravimétrique (54 c_1 + 31 c_2) (tableau 3). Le spectre taxonomique de ces éléments qui ont un rôle important dans l'équilibre dynamique de la zoocénose est déterminé surtout par les cladocères (40,75%) et rotifères (27,78%) dans le cas des consommateurs primaires, ou par les copépodes (77,42%) au niveau des consommateurs secondaires.

Il est à remarquer que la plupart des taxa c_2 (94,12%) exercent d'une manière périodique le rôle d'éléments dominants, ce qui suggère l'existence de certaines mutations fréquentes dans les éléments ayant une importance écologique. La labilité structurale des éléments dominants du point de vue gravimétrique se reflète négativement dans le mécanisme de l'équilibre écologique du zooplancton.

La moyenne multiannuelle de la biomasse du zooplancton des écosystèmes lacustres aux producteurs primaires de type macrophytique est de 114,2 $\mu\text{g/l}$ (100,3 $\mu\text{g/l}$ c_1 + 13,9 $\mu\text{g/l}$ c_2) valeur qui illustre l'existence d'un niveau d'eutrophie modéré.

Dans le type mentionné, les limites de variation des moyennes annuelles maxima, calculées par écosystème, sont 63,9–931,6 $\mu\text{g/l}$ (Porcu–1977, respectivement Bogdaproste–1983), et celles des moyennes annuelles minima–19,4–31,5 $\mu\text{g/l}$ (Băclănești–1984, respectivement Bogdaproste–1985) (tableau 1). La moyenne annuelle qui représente le

maximum absolu des écosystèmes aux producteurs primaires de type macrophytique dépasse nettement les moyennes maxima des autres écosystèmes aux producteurs similaires, étant, au fond, comparable, pour ce qui est de l'ordre de mesure, aux valeurs qui caractérisent les écosystèmes aux producteurs de type planctonique. C'est le résultat d'une évolution saisonnière exceptionnelle, dans laquelle la moyenne du mois de septembre (1828,2 $\mu\text{g/l}$) représente le maximum absolu pour la catégorie mentionnée des écosystèmes (tableau 2).

La dynamique de l'abondance de la biomasse zooplanctonique de ce type d'écosystèmes, évaluée par groupes taxonomiques, relève l'apport supérieur des cladocères et copépodes parmi les consommateurs primaires, des copépodes et quelquefois des rotifères, même des cladocères, pour les consommateurs secondaires (tableau 1).

Le zooplancton des écosystèmes aux producteurs de type macrophytique présente 76 éléments dominants de point de vue gravimétrique (46 c_1 + 30 c_2). Leur structure taxonomique reflète le rôle déterminant des cladocères (43,49%) au niveau des consommateurs primaires et surtout des copépodes (76,66%) dans le cas des consommateurs secondaires (tableau 3).

L'analyse de la fréquence des éléments dominants relève le rôle tout particulier réalisé dans la structure trophique de la biocénose planctonique par les formes naupliales de copépodes, copépodites I–III, *Bosmina longirostris*, *Diaphanosoma orghidani*, *Chydorus sphaericus*, *Daphnia cucullata* et *Synchaeta oblonga* (pour les consommateurs primaires), copépodites IV–V de cyclopides, *Leptodora kindti* et *Asplanchna priodonta* (pour les consommateurs secondaires).

La moyenne multiannuelle de la biomasse zooplanctonique dans les écosystèmes aux producteurs primaires de type planctonique (789,8 $\mu\text{g/l}$) (701,4 $\mu\text{g/l}$ c_1 + 88,4 $\mu\text{g/l}$ c_2), ce qui reflète, même dans ce plan, l'évolution de l'état trophique des écosystèmes du niveau d'eutrophie modérée vers ceux d'eutrophie avancée, polytrophie au même hypertrophie. L'accroissement des valeurs de la biomasse s'accorde avec la dynamique ascendante de la densité numérique de la zoocénose, par ensemble, mais elle reflète en même temps la tendance vers l'accroissement de l'abondance numérique de certaines espèces ayant des individus de taille plus grande.

Les variations des moyennes annuelles, calculées par écosystème à l'intérieur du type mentionné, sont comprises entre 825,2–3138,8 $\mu\text{g/l}$ pour celles maxima (Matița–1986, respectivement Isacova–1983), et entre 209,9–751,6 $\mu\text{g/l}$, pour celles minima (Roșu–1985, Merhei–1981) (tableau 1). La moyenne annuelle représentant le maximum absolu de la période 1975–1987, propre à l'écosystème d'Isacova, se situe aux côtes particulièrement élevées. C'est la conséquence d'une dynamique particulière en tant qu'amplitude, enregistrée au cours de 1983. C'est ici que la moyenne du mois de septembre (8603,2 $\mu\text{g/l}$) représente la plus grande valeur mensuelle de la biomasse rencontrée dans les écosystèmes de type lacustre du biôme, pendant les 13 années de recherche.

La dynamique de l'abondance gravimétrique, par groupes taxonomiques, du zooplancton des écosystèmes aux producteurs primaires de type planctonique relève, d'une manière unitaire, la domination nette des cladocères (pour c_1) et des copépodes (pour c_2) (tableau 1).

Tableau 1
La dynamique de la biomasse ($\mu\text{g/l}$ substance sèche) et l'abondance (%) de la biomasse du zooplancton (\bar{X}_a annuelles)

L'écosystème	L'années	Total Zoopl. $\mu\text{g/l}$	Consommateurs primaires							Consommateurs secondaires						
			Zoopl. c_1 $\mu\text{g/l}$	Test.	Cil.	Rot.	Lam.	Ostr.	Cop.	Clad.	Zoopl. c_2 $\mu\text{g/l}$	Rot.	Cop.	Clad.		
			%	%	%	%	%	%	%	%	%	%	%	%		
ROȘU	1975	20,1	19,9	0,50	0,10	5,52	3,01	12,05	57,74	21,08	0,2	50,00	50,00	—		
	1976	205,3	163,0	0,31	0,98	8,89	0,06	2,58	28,22	58,96	42,3	65,48	14,18	20,34		
	1977	82,0	81,8	0,11	0,23	4,33	0,35	1,17	89,48	4,33	0,23	86,96	13,04	—		
	1978	33,2	33,0	—	0,03	1,51	0,91	20,60	63,92	13,03	0,2	100,00	—	—		
	1983	1222,9	1148,7	0,03	0,05	1,37	0,04	—	6,80	91,71	74,2	0,81	82,61	16,58		
	1984	339,4	314,4	0,06	0,16	1,49	0,03	—	15,37	82,89	25,0	12,00	28,40	59,60		
	1985	209,9	171,7	0,17	0,64	6,82	0,35	—	22,13	69,89	38,2	27,23	56,81	15,96		
1986	634,5	552,0	0,09	0,43	2,95	0,09	—	7,50	88,94	82,5	18,42	61,33	20,25			
1987	283,7	240,2	0,50	0,37	9,78	0,17	—	14,28	74,90	43,5	18,62	52,18	29,20			
ROȘULET	1987	130,5	111,0	1,26	—	9,73	0,45	—	15,58	72,98	19,5	22,05	74,36	3,59		
PORCU	1976	66,4	64,6	0,77	0,15	7,89	0,03	1,55	39,32	50,29	1,8	94,44	5,56	—		
	1977	27,9	27,7	0,72	0,36	3,97	0,36	0,72	68,24	25,63	0,2	100,00	—	—		
	1978	46,6	46,6	0,21	0,09	0,64	0,04	0,21	50,79	48,02	—	—	—	—		
PIIU	1977	63,9	57,8	0,17	0,35	6,40	1,21	4,84	36,16	50,87	6,1	22,95	63,94	13,11		
	1978	27,17	27,07	0,07	0,37	14,41	7,02	3,14	59,11	15,88	0,1	—	50,00	50,00		
	1983	1038,7	892,9	0,01	0,10	2,70	0,13	—	10,80	86,26	145,8	0,75	67,70	31,55		
IACUB	1975	31,3	29,3	1,71	12,97	1,71	2,39	3,07	37,88	40,27	2,0	—	95,00	5,00		
ISACOVA	1983	3138,8	2986,6	0,003	0,01	0,97	0,003	0,06	6,56	92,39	152,2	11,43	70,70	17,82		
	1984	443,0	370,8	0,05	0,92	5,74	0,03	0,19	20,31	72,76	72,2	16,34	72,03	11,63		
	1985	578,6	470,4	0,09	0,13	3,56	0,02	—	13,46	82,74	108,2	24,58	40,57	34,85		
	1986	1006,2	844,4	0,02	0,07	0,93	0,01	—	9,85	89,12	161,8	1,30	86,96	11,74		
	1975	—	—	—	—	—	—	—	—	—	—	—	—	—		

MATIȚA

1980

1981

1982

1983

1984

1985

1986

149,0

808,6

721,2

612,3

220,6

310,9

825,2

95,1

733,8

673,1

553,8

178,8

261,1

754,5

0,11

0,04

0,18

0,04

0,17

0,04

0,01

0,63

1,47

0,41

0,11

0,38

0,38

0,28

19,24

4,80

4,44

3,32

7,10

1,99

1,30

0,16

0,38

0,68

0,33

0,11

0,23

0,11

0,32

0,05

13,42

24,90

28,37

20,91

12,27

35,12

15,66

81,27

71,30

63,71

20,91

86,03

43,42

77,60

48,1

21,4

76,44

70,7

—

53,9

74,8

48,1

58,5

41,8

49,8

3,96

31,54

12,43

6,65

14,53

18,42

5,62

3,96

32,10

61,37

52,39

77,61

88,42

94,48

—

26,20

40,90

7,86

67,70

42,37

1,56

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Abbreviations : Zoopl. = Zooplankton ; Test. = Testacea ; Cil. = Ciliata ; Rot. = Rotifera ; Lam = Lamellibranchia ; Ostr. = Ostracoda ; Cop. = Copepoda ; Clad. = Cladocera.

5 EFFET DE L'EUTROPHISATION SUR LA STRUCTURE DU ZOOPLANCTON

159

Tableau 2
La dynamique de la biomasse ($\mu\text{g/l}$ substance sèche) et l'abondance (%) de la biomasse de zooplancton (\bar{N}_a par mois)

L'écosystème	Mois Année	III	IV	V	VI	VII	VIII	IX	X	XI	XII
ROȘU	1975	32,4	422,3	20,0	9,4	23,1	7,8	35,9	24,4	41,0	24,4
	1976	9,7	13,8	68,3	90,7	708,9	105,5	245,4	132,9	479,7	132,9
	1977		7,2	35,6	11,6	24,9	24,0	70,4			
	1978		322,1	155,0	2,0	2451,2	2354,5	1711,5			
	1983	31,9	17,6	222,8	19,0	351,4	293,6	1619,8			43,3
	1984		16,8	31,8	22,9	317,7	215,7	638,0			455,3
	1985		26,4	31,8	470,0	188,2	334,1	2040,0			230,5
1986				81,4	52,6		974,5				95,3
1987			10,7	25,9	26,5		458,9				
ROȘULEȚ	1987										
PORCU	1976	11,9	19,9	3,3	20,2	171,0	114,3	231,8	15,8	9,4	
	1977	0,7	2,1		22,9	47,0	31,0			63,8	
	1978		2,0				47,6			46,6	
PUIU	1977	15,7	43,1	81,4	31,7	48,5	115,3			111,3	
	1978		25,9	35,2			13,2			34,7	
	1983		209,3	180,5	495,7	758,1	1990,0	2598,7			
IACUB	1975			25,1	35,0	7,2	8,5	104,2	7,6		
ISACOVA	1983	167,4	1199,2	3068,3	946,8	3776,8	1238,7	8603,2		278,4	64,8
	1984		327,1	1288,5	127,9	47,2	403,8	1281,7		226,3	
	1985		61,0	68,5	381,5	758,9	1091,6	975,4	555,3	540,5	
	1986		117,3		597,2	667,7		2108,6	2943,5		

MĂIȚA	1980	157,5	27,1	135,2	18,7	263,4	232,1	160,6	304,0	43,4	31,5
	1981		16,8	55,0	979,3	1041,8	1238,5	1775,2	1331,9		
	1982		66,3	145,1	115,7	605,9	1276,9	1762,9	1076,2		
	1983	32,2	263,3	863,3	779,8	1468,8	231,9	573,0	106,2		
	1984		12,7	53,4	97,4	678,5	429,9	502,8		108,4	70,4
	1985		20,3		81,2	501,2	629,4	508,6	312,2	1557,3	123,6
1986		21,4	54,7	224,6	1126,8		1755,7	1036,0			
MERHEI	1980	129,8	65,4	72,4	39,3	57,2	36,1	104,3	137,4	51,9	
	1981		21,1	107,8	103,8	269,7	1297,3	1392,3	2037,8	1318,7	
	1982		171,0	360,7	272,4	782,7	2375,8	888,0	1489,4		
	1983		574,2	707,4	691,0	2207,5	583,2	1113,3	333,7		216,0
BABINA	1982			506,6	132,3			2107,0			
	1983		384,5	25,0	831,5	1339,1	287,6	680,2	85,4		61,6
	1984	18,6	22,2		242,5	615,2	505,9	295,5		229,6	32,5
	1985		2,8		8,4	433,3	591,4	752,9	1381,9		
1986		15,0		347,0	965,7		4915,0	3930,8	1566,7		
BOGDAPROSTE	1982				59,8			1636,2			
	1983		203,4	82,5	579,0	1017,5	2019,5	1828,2	791,6	31,2	13,0
	1984	20,4	19,5	15,7	103,7	13,5	23,5	262,2	68,6	267,8	12,3
	1985		12,9		20,0	20,1	86,6				
1986		27,8	24,3	85,3	53,3		600,6	307,6			
BĂCLĂNEȘTI	1982				3,0			4,6			
	1983		79,6	55,5	77,7	36,5	35,9	189,5		38,1	2,7
	1984	16,5	49,8	19,7	5,1	5,2	25,8	11,8			
	1985		10,1		5,7	17,8	54,3		215,0		
	1986		25,5	136,4	2,7	17,8		13,5	128,4		

Tab-

Les éléments dominants du zooplancton

Eléments dominants	L'écosystème L'année	Consommateurs																
		I										II	III			IV		V
		a	b	c	d	h	i	j	k	l	l	b	c	d	c	d	h	a
TESTACEA																		
<i>Arcella arenaria</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
ROTIFERA																		
<i>Brachionus calyciflorus amphyceros</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Br. calyciflorus anuraeiformis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Br. calyciflorus doreas</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Br. calyciflorus doreas spinosa</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Br. calyciflorus</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Br. diversicornis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Br. leydigi tridentatus</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Euchlanis dilatata</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Keratella cochlearis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>K. quadrata</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Synchaeta oblonga</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>S. pectinata</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>S. vorax</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Synchaeta sp.</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Throcospaera aequatorialis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
LAMELLIBRANCHIA																		
<i>Dreissena polymorpha</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
OSTRACODA																		
<i>Ostracoda g. sp.</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Physocypria kraepelini</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
GLADOCERA																		
<i>Alona quadrangularis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>A. rectangula</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Alonella exigua</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Acerperus harpae</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Biapertura affinis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Bosmina longirostris</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Camptocercus rectirostris</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Ceriodaphnia pulchella</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>C. quadrangularis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Chydorus sphaericus</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Daphnia cucullata</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>D. galeata</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>D. longispina caudata</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Diahanosoma orghidani</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Eubosmina coregoni</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Eurycercus lamellatus</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Moina brachiata</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>M. micrura</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Pleuroxus aduncus</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>P. laevis</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Simocephalus vetulus</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Simocephalus sp.</i>		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
COPEPODA																		
Nauplii Copepoda g. sp.		+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	
Copepoditi st. I-III		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
Copepoda g. sp.		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	

teau 3

du point de vue gravimétrique

primaires	Consommateurs																											
	VI				VII				VIII				IX			X			XI									
	h	i	j	k	e	f	g	h	i	j	k	e	f	g	h	h	i	j	k	h	i	j	k	h	i	j	k	
	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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Le zooplancton de ce groupe d'écosystèmes présente un nombre plus réduit d'éléments dominants (43) par rapport à celui des écosystèmes dont la production primaire est assurée par des macrophytes submerses. La réduction massive du nombre de ces éléments (de presque 44%) reflète la baisse du degré de hétérogénéité ambiante. Du total mentionné, 25 sont c_1 , 18 c_2 . La structure taxonomique des éléments dominants est déterminée surtout par des cladocères et copépodes (les deux, à raison de 36%) dans le cas de c_1 , par des copépodes (72,22%) dans le cas de c_2 (tableau 3). De leur total, un rôle tout particulier de point de vue écologique, avec des implications dans la dynamique saisonnière et multiannuelle, dans l'équilibre biocénotique et dans les relations trophiques, présentent les copépodites I—III, *Diaphanosoma orghidani*, *Chydorus sphaericus*, *Bosmina longirostris* et les formes naupliales de copépodes pour le premier niveau de consommateurs, les copépodites IV—V de cyclopidés, *Leptodora kindti* et *Asplanchna priodonta* dans le cas du deuxième niveau.

CONCLUSIONS

— L'évolution ascendante du processus d'eutrophisation détermine des mutations amples et en même temps accélérées dans la structure de la zoocénose planctonique des écosystèmes lacustres du Delta du Danube. En corrélation avec la réduction de la structure taxonomique et la prolifération numérique des individus, la biomasse enregistre des variations significatives en temps et espace.

— La dynamique de la biomasse au cours des années 1975—1987 est caractérisée par un sens évidemment ascendant au cours des premières 8—9 années, avec des maxima remarquables pour la période 1981—1983 et un autre descendant, d'amplitude plus réduite, pendant les dernières 4—5 années, de sorte qu'on puisse apprécier que la tendance d'évolution, pour l'entière période étudiée, est ascendante.

— La moyenne multiannuelle de la biomasse du zooplancton des écosystèmes des producteurs primaires de type macrophytique (114,2 $\mu\text{g/l}$ substance sèche). (100,3 $\mu\text{g/l}$ c_1 + 13,9 $\mu\text{g/l}$ c_2) est comparable aux valeurs enregistrées même avant 1975, en s'encadrant dans les limites de variation propres aux écosystèmes lacustres de type modéré eutrophe. Celle des écosystèmes des producteurs primaires de type planctonique est 6,9 fois plus grande (789,8 $\mu\text{g/l}$) (701,4 $\mu\text{g/l}$ c_1 + 88,4 $\mu\text{g/l}$ c_2) et correspond à l'évolution de l'état de trophicité vers des niveaux d'eutrophie avancée, polytrophie ou même hypertrophie.

— La valeur maximum des moyennes annuelles, déterminées par écosystèmes, varie entre 63,9—931,6 $\mu\text{g/l}$ (Puiu—1977, respectivement Bogdaproste—1983) dans le cas des écosystèmes aux producteurs primaires de type macrophytique et entre 825,2—3138,8 $\mu\text{g/l}$ (Matia—1986, respectivement Isacova—1983) dans le cas de ceux aux producteurs primaires de type planctonique, et celle minimum entre 19,4—31,5 $\mu\text{g/l}$ (Băclănești—1984, respectivement Bogdaproste—1985) pour le premier type d'écosystèmes et entre 209,9—751,6 $\mu\text{g/l}$ (Roșu—1985, respectivement Merhei—1981) dans le cas du deuxième type.

— Pour les deux types d'écosystèmes, la dynamique de l'abondance gravimétrique par groupes taxonomiques relève la domination des cladocères, au niveau c_1 et des copépodes, dans le cas c_2 .

— Le zooplancton du premier type d'écosystèmes comprend 76 éléments dominants de point de vue gravimétrique, tandis que le deuxième type n'en comprend que 43. La réduction mentionnée exprime la tendance de baisse du degré d'hétérogénéité du milieu.

— Parmi les éléments dominants de point de vue gravimétrique du premier type d'écosystèmes, il faut mettre en évidence le rôle particulièrement important réalisé par les formes naupliales de copépodes, les copépodites I—III, *Bosmina longirostris*, *Diaphanosoma orghidani*, *Chydorus sphaericus*, *Daphnia cucullata* et *Synchaeta oblonga* (dans le cas des consommateurs primaires,) les copépodites IV—V de cyclopidé, *Leptodora kindti* et *Asplanchna priodonta* (dans le cas de consommateurs secondaires); dans le cas du deuxième type d'écosystèmes on relève l'apport particulier réalisé par les copépodites I—III, *Diaphanosoma orghidani*, *Chydorus sphaericus*, *Bosmina longirostris* et les formes naupliales de copépodes (pour c_1 , les copépodites IV—V, *Leptodora kindti* et *Asplanchna priodonta* (pour c_2)).

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Institut de Sciences Biologiques
Bucarest, Splaiul Independenței 296

BEITRÄGE ZUM KENNEN DER STRUKTUR DER JUNGFISCHPOPULATIONEN AUS SPEZIFISCHEN TROPHISCHEN ZONEN DES DONAU-DELTA

GH. BREZEANU und R. POPA

On the surface of the natural lakes in the Danube Delta there are areas where a rich vegetation, constituted by aquatic macrophytes, develops. The majority of fishes (especially *Cyprinidae*) reproduce in these areas and the fry remains among the vegetation where excellent conditions for food and protection are found.

A number of 15 species have been identified, their populations reaching a density and a biomass exceeding 60000 individuals/ha and 500 kg/ha, respectively. In the areas without vegetation, the density and biomass of the fry were of 2300 individuals/ha and 15 kg/ha, respectively.

Ein Überblick über die natürlichen Seen des Donau-Deltas lässt eine große Mannigfaltigkeit derselben erkennen, welche auf die geomorphologischen, hydrologischen und ökologischen (1, 2) Eigenschaften zurückzuführen ist. Auf Grund dieser Kriterien können die Gewässer-Ökosysteme je nach ihrem Entwicklungsstadium in eine Reihe von Typen eingeordnet werden. Innerhalb jeder Kategorie weisen die Seen Flächen-gebiete auf, die sich durch ihre ökologischen Eigentümlichkeiten unterscheiden (2).

Solche Zonen sind beispielsweise diejenigen, in welchen eine üppige Vegetation einerseits aus Sumpf-Makrophyten (*Phragmites communis*, *Typha latifolia*, *Hydrocharis morsus-ranae*, *Scirpus sp.*, *Carex sp.* u.a.) und andererseits aus eigentlichen Wasserpflanzen (*Ceratophyllum demersum*, *Myriophyllum spicatum*, *Trapa natans*, *Nymphaea alba*, *Nuphar luteum*) besteht; in anderen Zonen fehlen die Makrophyten und der Grund der Seen ist von einer dicken Schlickschicht bedeckt (3).

Die in den Makrophyten-Zonen entstehenden Biozönosen sind von höherer Komplexität und enthalten eine große Artenzahl, welche lange Nahrungsketten erzeugt, während in den vegetationslosen Zonen die Struktur der Biozönosen viel einfacher ist und eine geringere Artenzahl aufweist (4,5). Diese Zonen, die für die Seen des Donau-Deltas spezifisch sind, haben für die Entwicklung und den Aufbau der Fisch-Populationen eine große Bedeutung.

Die Vegetationszonen stellen die Fortpflanzungsstätten der meisten Fische dar, hier halten sich auch die Larven und die Jungfische auf, da hier günstige Bedingungen für Schutz und Nahrung bestehen. Die vegetationslosen Zonen, deren Angebot an Nährstoffen für die Fische viel geringer ist, dienen mehr oder weniger dem Durchgang der Fische.

Tabelle 1
Die Struktur der Fisch-Populationen (Jugendstadium) aus Zonen besonderer Trophizität

Art	Die zone mit üppiger Vegetation				Die Vegetationslose Zone			
	Numerische Dichte Ex/100 m ²	Numerische Abundanz %	Dichte der Biomasse g/100 m ²	Abundanz der Biomasse %	Numerische Dichte Ex/100 m ²	Numerische Abundanz %	Dichte der Biomasse g/100 m ²	Abundanz der Biomasse %
<i>Esox lucius</i>	2,5	0,04	330	4,96	1,5	6,56	140	92,2
<i>Abramis brama</i>	154	2,32	119	1,79	0	0	0	0
<i>Abranus alburnus</i>	40	0,6	157	2,37	2,04	8,8	4,4	2,9
<i>Blicca bjoerkna</i>	2320	35,08	1570	46,9	0	0	0	0
<i>Carassius auratus gibelio</i>	41	0,62	24,3	0,37	0	0	0	0
<i>Rhodeus sericeus amarus</i>	690	10,43	1091	10,5	0	0	0	0
<i>Rutilus rutilus</i>	2983	44,44	1375	20,78	0	0	0	0
<i>Scardinius erythrophthalmus</i>	160	2,42	342	5,17	0	0	0	0
<i>Syngnathus nigrolineatus</i>	10	0,15	8,1	0,12	1,8	7,6	1,7	1,14
<i>Percia fluviatilis</i>	5	0,17	36	0,54	0	0	0	0
<i>Acerina cernua</i>	0,5	0,07	0	0	0	0	0	0
<i>Pomatoschistus microps</i>	212	3,19	33	0,5	15,5	67	3	1,9
<i>Neogobius fluviatilis</i>	0	0	0	0	1,02	4,4	1,3	0,8
<i>Proterorhinus marmoratus</i>	0	0	0	0	1,3	5,5	1,6	1,04
<i>Cobitis taenia</i>	0	0	0	0	0,08	0,5	0,04	0,07
Gesamtzahl	6617	100	5083	100	23,2	100	152,3	100

Die Struktur der Jungfisch-Populationen, welche den Gegenstand unserer Untersuchungen darstellt, ist in den beiden Zonen grundlegend verschieden (Tabelle 1). Im Rahmen der Untersuchungen wurden in den Vegetationsgebieten 14 Fischarten gefangen und in den anderen Zonen lediglich 3 Arten. Es liegt jedoch auf der Hand, daß in den Seen, in welchen eine ausgeglichene Entwicklung aller Biozönosen durch die Anwesenheit der Makrophyten ermöglicht wird, die Fischfauna günstige Entwicklungsbedingungen vorfindet.

In den Makrophyten-Gebieten beträgt die zahlenmässige Dichte und die Biomasse 6617 Exemplare bzw. 5083 g, diese Werte sind auf 100 m² bezogen (Tabelle 1).

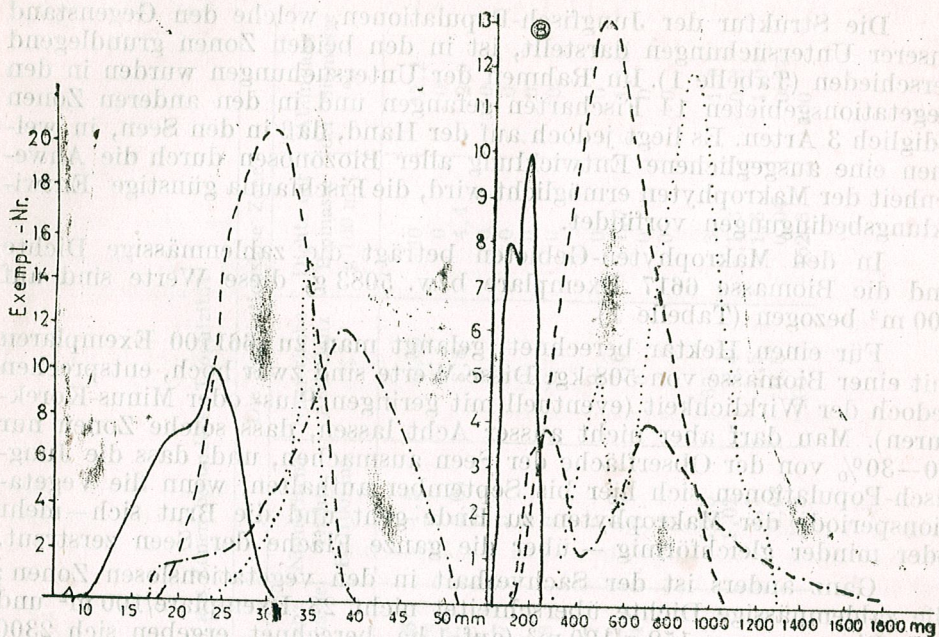
Für einen Hektar berechnet, gelangt man zu 661700 Exemplaren mit einer Biomasse von 508 kg. Diese Werte sind zwar hoch, entsprechen jedoch der Wirklichkeit (eventuell mit geringen Plus- oder Minus-Korrekturen). Man darf aber nicht ausser Acht lassen, dass solche Zonen nur 20–30% von der Oberfläche der Seen ausmachen, und, dass die Jungfisch-Populationen sich hier bis September aufhalten, wenn die Vegetationsperiode der Makrophyten zu Ende geht und die Brut sich—mehr oder minder gleichförmig—über die ganze Fläche der Seen zerstreut.

Ganz anders ist der Sachverhalt in den vegetationslosen Zonen; die zahlenmässige Dichte überschreitet nicht 23 Exemplare/100 m² und eine Biomasse von 152 g/100 m² (auf 1 ha berechnet ergeben sich 2300 Ex. bzw. 15 kg). Dies ist ein Beweis dafür, dass solche Zonen für die wirtschaftliche Produktion von Fischbrut und die Bildung der Fisch-Reserve in den natürlichen Seen des Donau-Deltas von untergeordneter Bedeutung sind.

Die Werte der zahlenmässigen Dichte und der Biomasse bringen auch die quantitativen Verhältnisse der verschiedenen Arten zum Ausdruck. So ist in den Makrophyten-Gebieten die vorherrschende Art *Rutilus rutilus*, gefolgt von *Blicca bjoerkna*, *Rhodeus sericeus*, *Gobius fluviatilis* und *Pomatoschistus microps* (Tabelle 1). Dieses Verhältnis kann sich von See zu See oder sogar auf der Fläche desselben Sees ändern, in Abhängigkeit von gewissen lokalen ökologischen Kennwerten, ohne dass sich aber die Grundstruktur der Jungfisch-Populationen in den betreffenden trophischen Zonen verändern würde.

Es soll hierbei erwähnt werden, dass speziell die Gobiden *Gobius fluviatilis*, *Pomatoschistus microps* und *Proterorhinus marmoratus* sich auf der nekto-benthonischen Ebene aufhalten und deshalb in den vegetationslosen Zonen häufig sind.

Verfolgt man die Längen- und Gewichtsschwankungen der dominierenden Arten (Abb. 1), so ergibt sich, daß die Populationen die aus Individuen der gleichen Generation bestehen, in Klassen verschiedener Grösse zusammengefaßt werden können. Diese Tatsache spiegelt die Dynamik der Fortpflanzung der betreffenden Arten (Etappen der Laich-Ablegung), welche sich über einen gewissen Zeitraum erstreckt, der spezifisch für die Biologie jeder Art ist.



Dominante Arten		Es wurden untersucht:	
1. <i>Blicca</i>	50 ex. <i>Blicca</i>	$\bar{W} = 22,1$ mm	$\bar{W} = 503,47$ mg
2. <i>Rhodeus</i>	50 ex. <i>Rhodeus</i>	$\bar{W} = 29,31$ mm	$\bar{W} = 701,75$ mg
3. <i>Rutilus</i>	50 ex. <i>Rutilus</i>	$\bar{W} = 38,06$ mm	$\bar{W} = 995,1$ mg
4. <i>Pomatoschitus</i>	50 ex. <i>Pomatoschitus</i>	$\bar{W} = 22,1$ mm	$\bar{W} = 126,9$ mg

Abb. 1. — Die Schwankung der Länge (A) und des Gewichtes (B) bei den vorherrschenden Arten.

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