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SOMMAIRE

HETEROMYSIS MURESEANUI N.SP. AND *KALLIAPSEUDES VIRIDIS*, SSP. *BRASILIENSIS* N.SSP., FROM THE BRAZILIAN LITTORAL WATERS

M. BĂCESCU

Back from Brazil, commander Petre Mureşeanu donated to our Museum a rich material of planctonic fauna from the middle of the Atlantic Ocean and 2 benthonic samples from the Brazilian waters for which we thank him. In the benthic samples, I found — besides many specimens of *Mesanthura brasiliensis* (identified by Ileana Ronai), of *Callozodion multispinosum* and *Kalliapseudes viridis* ssp. *brasiliensis* n. ssp., as well as of many Amphipods and Polychaetes — a new Mysid species that I describe below.

HETEROMYSIS (HETEROMYSIS MURESEANUI n. sp.)

(Fig. 1 A–F)

Diagnosis. Small-sized *Heteromysis* (3—4 mm), with large eyes, strong ocular peduncles ending antero-innerly in an edge that exceeds the little corneal part.

Telson short provided with a sinus adorned with 20 laminae, with sides armed with only 4 spines; apices with 2 spines, the inner one being shorter than the laminae. Uropodal endopod without any spine. On the antero-inner corner of antennule, 2 naked accuminate spinules and 2 doubly feathered setae.

DESCRIPTION OF THE ADULT MALE

Tegument soft, ivory; carapace with frontal part a little prominent between the eyes describing a large obtuse triangle.

Eyes large: peduncle massive ending in an antero-inner edge that clearly exceeds the corneal part which is very small as compared to peduncle (Fig. 1 A).

Antennule strong (Fig. 1 B), antero-innerly provided with a group of 4 characteristic phanera: a strong naked accuminate spine seta directed innerwards (a) and another one, of the same type but finer (b) directed outwards as well as 2 doubly feathered setae anteriorly directed (c and d).

The ♂ lobe, hemisphere-like, is prominent on the lower part, being armed with a long and rich bunch of fine hairs (Fig. 1 B).

Antenna distinctly shorter than antennule, with basis as long as scale (Fig. 1 A). Buccal parts common to the genus s.s.

Cheliped with dactylo-propodus not too swollen, armed as shown in fig. 1 D. The range of phanera on the lower anterior crest of carpoprodus — over which the dactyloclaw is recurved in order to form the pseudo-

chela — is relatively poor : only 2 dentiform flagellated spines (a and b Fig. D) and four feathered seetae.

The rest of peraeopods with 7-articulated tarsus which is equal in length with carpoprodus and with ischios and 9-articulated for exopod :

Penis, cylindrical, easily distinguishable between the bases of the last peraeopods by the silver brilliance of the fascicle of spermatozoa

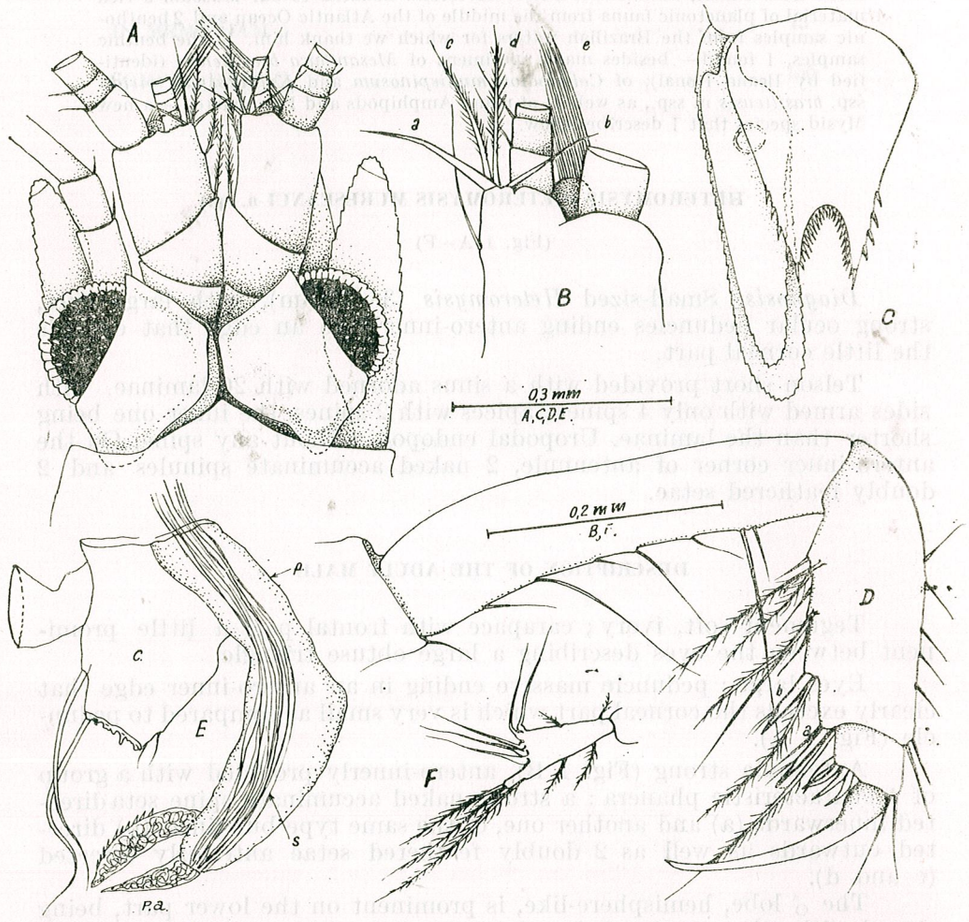

Fig. 1. — A—F, *Heteromysis (Heteromysis) mureşeanui* n. sp. ♂ = 3.5 mm. A, anterior side of cephalothorax ; B, characteristic phanera on the antero-inner side of antennule : a, inner spine abruptly thinner from its middle ; b, id., extr., c and d, feathered setae ; e, bunch of fine hairs of appendix mascula ; C, telson and uropods ; D, cheliped ; E, penis ; F, pleopod IV.

(s Fig. 1 E). It is recurved towards the ventral side and ends in a tongs-like process formed of 2 pointed parts with a special structure (p. a, Fig. 1 E). This characteristic is not met in any other *Heteromysis*.

Pleopods somewhat of *Heteromysis formosa* Sars type, i.e. simple elongate plates with long feathered setae equally on apex (Fig. 1 F) and with reduced branchial part.

Telson short, with sinus provided with about 20 lamines all along ; its sides are armed, only on the distal third, with 4 lateral spinules (3 in juveniles) and 2 apical ones, the inner one bein extremely little, distinguishing itself from the laminae only by its articulation. Even the outer apical spine is not much larger than the lateral ones.

Uropods much exceed the telson (Fig.1 C) their endopod being shorter than exopod and having no spine.

The female, 4 mm long, very crushed, made the description impossible.

Material : 1 ♂ — 3.5 mm, 1 ♀ = 4 mm and 1 juv. ♀ = 2.2 mm : locality : Brazil, on the roads of Tubarao port, 29 m depth (20°15. 5′S ; 40° 5.3′W), 12 km off the coast. Collected in calcareous concretions with foliaceous algae *Grinnellia*, on 5. V. 1985 and 10. IV. 1986.

In this coenosis : an Anthuridean, *Mesanthura brasiliensis* Koening 1980 (det. Ileana Ronai), the Monokonophoran Tanaidacea, *Kalliapseudes viridis brasiliensis* n. ssp. and *Calozodion multispinosus* Guţu 1984.

Derivatio nominis. The species is dedicated to commander Petre Mureşeanu, who collected it and brought it to the "Grigore Antipa" Museum.

♂ Holotype No. 712 in the Crustaceum Collection of the "Grigore Antipa" Museum.

Remarks

Among all the species of *Heteromysis* s.s., the new species is somewhat close only to *H. tethysiana* Băc. from Australia (2). Is differs from it however, especially in the shape of the eyes, in the lack of some special phanera on the apex of pleopod IV ♂ and also in the structure of penis (Fig. 1 E).

H. mureşeanui is the first representative of this genus collected in the Brazilian waters a strange thing if we take into consideration the fact that 23 species are known from the littoral waters of Central America (2).This might be also explained by the cooler waters of southern Brazil but also by the lack of mysidological research in the Brazilian waters, from where about 10 species have been discovered so far, however, most of which belonging to the tribe Leptomysini (3).

The occurrence of the above-mentioned Monokonophora in the Brazilian waters is also important :

Kalliapseudes viridis Menzies 1953 was also described from the Pacific waters (Lower California, Mexico) after 2 specimens. It was then found again by Sieg in a large number in the Atlantic Ocean (Bahamas) and described as *K. bahamensis* in comparison with the female of Menzies. But the differences between these two species are so insignificant that they cannot even characterize a subspecies.

I found 2 specimens (1 ♀, 1 juv.) of *K. viridis* east of Port Tubaroro, in the biocoenosis of *Heteromysis mureşeanui*, but they clearly differ from the type and from Sieg's descriptions. Consequently, I consider it as a local subspecies, *KALLIAPSEUDES V. BRASILIENSIS N. SSP.* (Fig. 2, A—D).

K. viridis brasiliensis differs both from the Pacific and Atlantic specimens in the following features : the pleonites do not show lateral setae so typical of those figured by Sieg (Figs 1 and 5) but each of them shows one simple long seta and the pair of feathered setae from the lower

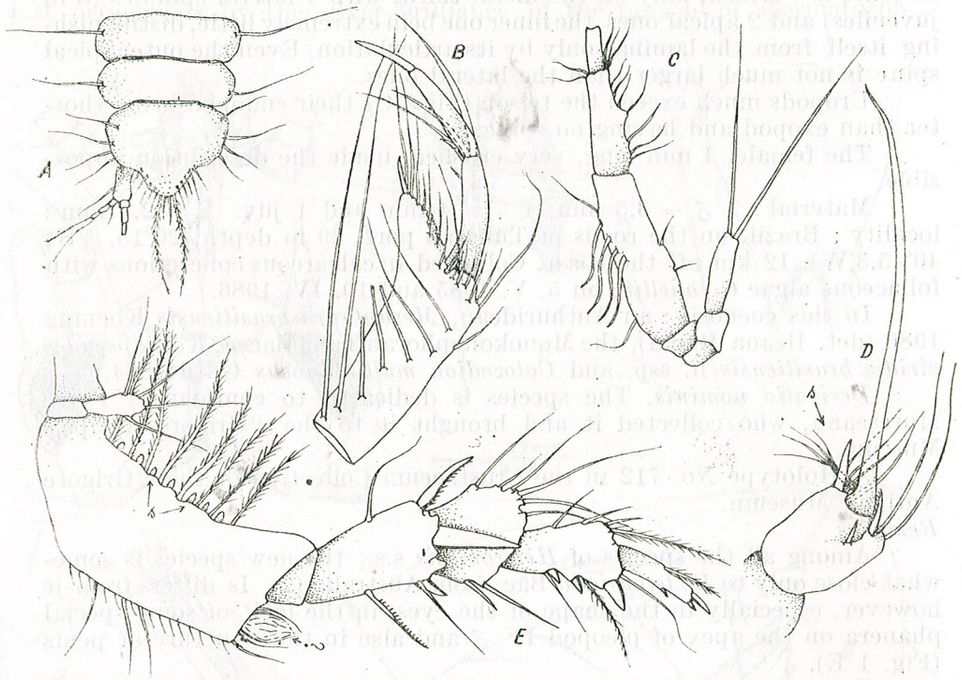

Fig. 2. — A — D, *Kalliapseudes viridis* subspecies *brasiliensis* n.sp. ♀ j. A, telson and last pleonites ; B, dactylus and extremity of propod of cheliped ; C, basis and scale of antenna ; D, dactylus and propodus of peraeopod III ; s = sensory organ ; E, *Calozodion multispinosum* Guţu peraeopod II with the series of spinula from the basipodite, characteristic of the species (arrow), spines alternating with setae, a feature characteristic of the species?

side of telsonal apex is longer than telson (Fig. 2 A).The scale of antenna is provided only with simple long setae and the basal articles are of a special shape (Fig. 2 B) ; the dactyli of peraeopods III (Fig. 2 C) IV and VII are much longer in comparison with their propodus. Only propodus I and chela are alike (Fig. 2 D).

Kalliapseudes viridis counts among the few species common to the Pacific and Atlantic waters of America. The subspecies type (1 j ♀) in the collection of the "Gr. Antipa" Museum No. 695.

Calozodion multispinosum has been just described from the Cuban waters. It forms a rich population in the biotope of calcareous concretions on which red algae of genus Grinnellia are fixed (♂♂, ovigerous ♀♀ bearing 8—10 hyaline eggs, slightly oval). From a morphological point of view it does not practically differ from the Caribbean type of the species (see Fig. 2 E, F).

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NOUVELLES ESPÈCES D'ICHNEUMONIDES *(HYM. ICHNEUM.)* PARASITES DES PUPES DE *MUSCA DOMESTICA* L. *(DIPT. MUSCIDAE)*

CONSTANTIN PISICĂ et KLAUS FABRITIUS

Six species of Ichneumonidae are presented, obtained from puparia of *Musca domestica* L. The species *Phygadeuon inflatus* Thoms. and *Phygadeuon tenuiscapus* Thoms. were obtained from puparia, collected from natural centres of development and the species *Gelis ambulans* Först. *G. nigricornis* Först. and *G. muscae* n. sp. from sentinel puparia. The species *Gelis melanophorus* Först. and *Phygadeuon tenuiscapus* Thoms. are new for the fauna of Romania.

Suite à de complexes recherches d'écologie concernant les diptères sinanthropes, initiées et développées à l'Institut d'hygiène et de santé publique de Bucarest, on a obtenu pour la première fois, des pupes de *Musca domestica* L., de nombreux espèces parasites parmi lesquelles 8 espèces d'ichneumonides. Deux espèces *(Atractodes gravidus* Grav. et *Phygadeuon subtilis* Grav.) ont été mentionnées dans des travaux antérieurs (5 et 8). Dans le présent ouvrage nous présentons six autres espèces dont deux sont nouvelles pour la faune de la Roumanie *(Gelis melanophorus* Först. et *Phygadeuon tenuiscapus* Thoms.) et une autre — nouvelle pour la science *(Gelis muscae* n. sp.).

MATÉRIEL ET MÉTHODE

Le matériel présenté dans cet ouvrage a été obtenu par deux voies :

1. On a collectionné des pupes de *Musca domestica* de foyers naturels de développement et notamment des résidus animaliers de la ferme zootechnique. Les pupes collectionnées ont été réparties dans de petits tubes de verre, qu'on a introduit dans un thermostat jusqu'à l'éclosion de l'adulte de *Musca domestica* ou du parasite respectif :

2. Les pupes obtenues dans des conditions de laboratoire ayant 24 heures d'âge au maximum, ont été exposées, quelques jours durant, dans des biotopes avec une densité élevée de mouches sinanthropes, aux alentours de places de parcage ou des campings. Avec les pupes récupérées on a procédé de la même manière qu'avec celles collectionnées dans des foyers naturels.

RÉSULTATS

La famille ICHNEUMONIDAE

La sous-famille GELINAE

1. *Gelis ambulans* Förster, 1851.

Un ♂ obtenu d'une pupe exposée de *Musca domestica* L., 15—18 VI.1983, Năvodari, département de Constanţa (leg. K. Fabritius).

REV. ROUM. BIOL. — BIOL. ANIM., TOME 31, Nº 2, P. 99—102, BUCAREST, 1986

Distribution: R. F. d'Allemagne. Espèce signalée en Roumanie (4), provenant des cocons d'*Apamteles glomeratus* L., Horlești, département de Iași. *Musca domestica* L. est un hôte nouveau.

2. *Gelis ambulans* Förster, 1851.

Un ♂ obtenu d'un pupe exposée de *Musca domestica* L., 15—18. VI.1983, Giurgeni, département de Ialomița (leg. K. Fabritius)

Distribution: le centre et l'ouest de l'Europe. *Hôtes: Macrocentrus* sp. (Braconidae) (7). *Musca domestica* L. est un hôte nouveau.

Espèce nouvelle pour la faune de la Roumanie.

3. *Gelis nigricornis* Förster, 1851.

Un ♂ obtenu d'une pupe exposée de *Musca domestica* L., 15—18 VI.1983, Giurgeni, département de Ialomița, éclosionné pendant le mois d'avril, 1984 (leg. K. Fabritius).

Distribution: l'ouest de l'Europe. En Roumanie il a été signalé à Cluj-Napoca (3). *Hôtes: Solenobia triquetrella* F.v.R. (6) *Musca domestica* est un hôte nouveau.

4. *Gelis muscae* n.sp.

♀. Le corps mesure 2,75—3,25 mm de long. La tête est plus large que le thorax, ponctué, rétréci dans la partie postérieure (fig. 1). Le visage (l'épistome) est convexe. Le clypeus est séparé du visage par un fossé évident et arrondi dans la partie antérieure. L'espace molaire est aussi grand que la largeur de la base des mandibules. Le fossé de la joue est complet et profond. Les tempes sont arrondies. La carène occipitale est complète. Les ocelles sont petits, placés au milieu du vertex. Les antennes sont relativement grosses avec le flagellum formé de 16 articles. Le premier article est plus long que le second, le huitième article est tout asussi long que large (fig. 2). Les mandibules ont des dents égales et sont très élargies à la base (fig. 3). Le thorax est lisse, finement ponctué et couvert de poils courts, rares et blanchâtres. L'écusson et les notaules manquent (fig. 1). Les ailes manquent, mais à la base de fixation des ailes antérieures il y a des excroissances, probablement le reste des tégules. Le propodeum présente des stigmes petites et rondes ; la région apicale mesure un quart de sa longueur, étant presque verticale ; les carènes latéro-longitudinales sont présentes ; elles forment dans la partie postérieure un appendice court. Les pattes sont relativement robustes, couvertes de poils courts. Les griffes sont simples. L'arolium est plus court que les griffes. L'abdomen est court, ovale-arrondi, plus large que la tête, couvert de poils courts blanchâtres. Les segments abdominaux sont beaucoup plus larges que longs excepté le premier segment, qui est plus long que large (fig. 1). Le tarière est un peu plus court que les deux premiers segments abdominaux.

La couleur du corps : la tête est noire, les mandibules, excepté la pointe des dents et la base des antennes, sont brunes-rougeâtres ; le reste des antennes est noirâtre. Thorax rouge, avec des pleures à côté de la base des pattes de couleur foncée. Les pattes ont les coxes et les trochanterelles rouges, le reste à la couleur brune-noirâtre. Le dernier article tarsal et les griffes de toutes les pattes sont noires. L'abdomen est noir ; le premier

et le second segment sont rouges, le second segment est, chez certains exemplaires, taché de noir. Les valves de la tarière sont noires.

♂ Le corps a 3 mm de long. Le mâle ressemble à la femelle, tout en se différenciant par : la présence du scutellum, l'abdomen plus étroit et

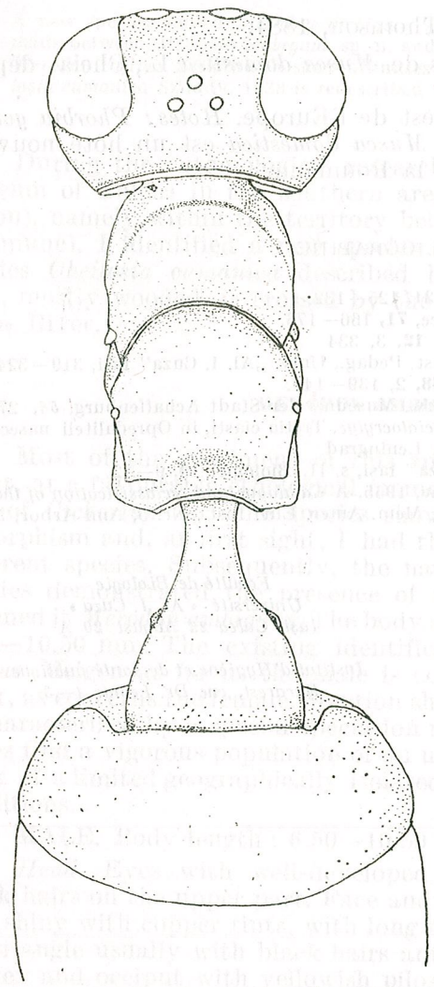

Fig. 1. — *Gelis muscae* n.sp. ♀, la tête, le thorax et les premiers segments abdominaux.

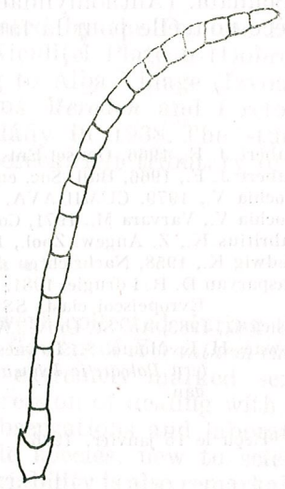

Fig 2. — *Gelis muscae* n.sp. antenne femelle.

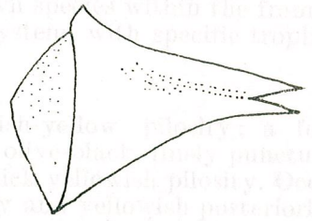

Fig. 3. — *Gelis muscae* n.sp. ♀, la mandibule.

la couleur noire du second segment abdominal, dont les gastrocoeles et le bord postérieur sont rouges. Le flagellum des antennes a 18 articles.

Matériel: Un ♀ (type), 1.VIII.1983 ; 2 ♀♀ (paratype), 2, 3.VIII. 1983 ; 1 ♂ (allotype), 1.VIII.1983, camping Năvodari, dép. de Constanța, obtenu des pupes de *Musca domestica* L. (leg. K. Fabritius).

5. *Phygadeuon inflatus* Thomson, 1884.

Trois ♀♀ obtenues des pupes de *Musca domestica* L., Versuchshof, Jensen, 2251/Eckstock, 1984, R. F. d'Allemagne (leg. S. Neuberg).

Distribution: le centre et l'ouest de l'Europe. Espèce obtenue pour la première fois des cultures.

6. *Phygadeuon tenuiscapus* Thomson, 1884.

Deux ♂♂ obtenus des pupes de *Musca domestica* L., Cheia, dép. de Vîlcea, 7.VII.1983.

Distribution: Le sud et l'ouest de l'Europe. *Hotes: Phorbia genitalis* Schnabl. (Anthomyiidae) (7). *Musca domestica* est un hôte nouveau. Espèce nouvelle pour la faune de la Roumanie.

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MERODON AMBIGUUS SP.N. AND A REDESCRIPTION OF *CHEILOSIA CUMANICA* SZILÁDY, 1938 *(DIPTERA, SYRPHIDAE)*

VLADIMIR BRĂDESCU

A new species named *Merodon ambiguus* sp. n. is described. A comparison is made between *Merodon ambiguus* sp. n. and species *Merodon aeneus* Meigen and *Merodon rufipes* Sack which show affinities with the new species. Species *Cheilosia cumanica* Szilády, 1938 is redescribed.

During the entomological research initiated by the Danube Delta Museum of Tulcea in the southern area of Niculiţel Plateau (Dobrogea region), namely within the territory belonging to Alba Village (Izvoarele Commune), I identified a new species of genus *Merodon* and I refound species *Cheilosia cumanica* described by Szilády in 1938. The studied area, mostly wooded, is crossed by the Alba creek, left tributary of the Taiţa River.

MERODON AMBIGUUS sp. n.

Most of the specimens of this species were collected during their flight, at a favourable ethological moment, on flowers of *Xeranthemum* sp. and of *Cichorium* sp. The species shows an extremely marked sexual dimorphism and, at first sight, I had the impression of dealing with two different species. Subsequently, the nature observations and laboratory studies demonstrated the presence of a single species, new to science. I named it *Merodon ambiguus*. The body size variability is also remarkable : 5.75—10.50 mm. The existing identification keys lead me to *Merodon aeneus* Meigen as far as the male is concerned and to *Merodon rufipes* Sack, as concerns the female. Mention should be made that genus *Merodon* is characterized by a special speciation malleability, when one may sometimes find a vigorous population of an unknown species within the framework of a limited geographically isolated ecosystem, with specific trophic conditions.

MALE. Body length : 6.50—10.50 mm.

Head. Eyes with well-developed whitish-yellow pilosity; a few black hairs on the upper part. Face and frons olive-black, finely punctuated, shiny with copper tints, with long and thick yellowish pilosity. Ocellar triangle usually with black hairs anteriorly and yellowish posteriorly. Vertex and occiput with yellowish pilosity. Antennae brown; 3rd article frequently paler, especially proximally, elongate, slightly recurverd upwards, about 1 1/2 longer than the 2nd article and excavated all along, on the upper part.

Thorax. Finely punctated, greenish-copper, slightly mat anteriorly and shiny posteriorly, sometimes with purple tints. Pilosity yellow, relatively abundant, upright and, rarely, with reddish tints on the discal

mesonotal area; indistinct or absent grey longitudinal stripes. Two characteristic black spots on both sides of mesonotum formed by tufts of black hairs and by the black proximal wing ends. Wings pale greyish. Squamae whitish, with black fringes. Legs: femora black, with yellow apical extremities; tibiae and tarsi rust-yellow; tarsus 4 and sometimes tarsi 3 and 5 (partially) brown. Trochanteres 3 armed with a apophysis, apically asymmetrical, almost equal in length with the 2nd tarsus of femur 3.

Abdomen. Oval; colour varying from copper-brown to bluish-black; punctuation more evident; pilosity relatively long, from pale yellow to rust-yellow. Tergite II with whitish-grey transversal band, interrupted in the middle, hardly visible, sometimes absent; tergite III with a similar band, always distinct; tergite IV with two lateral grooves, slightly elongate, devoid of pruinosity. Genital capsule shiny black, with white hairs.

FEMALE. Body length: 5.75—10.50 mm. Much differing from the male. Mesonotum greenish-black on the anterior half, with yellowish pilosity; bluish-black on the posterior half and almost entirely covered by a black pilosity; tergites II — IV with grey well-marked, interrupted, transversal band. Frons, at the level of antennae, a little narrower than 1/3 of head width, finely puctuated, shiny. The 3rd antennal article non-accuminate, gently dilated, rounded at the apical end and slightly recurved upwards. Colour of legs like in the male. Abdomen bluish-black, more intensely punctuated, with black shorter pilosity that prevails in comparison with the whitish-yellow one.

In order to point out the specific differences, two comparative tables are inserted including data on the two known species that have been mentioned previously:

MALE.

Merodon ambiguus sp.n.	*Merodon aeneus* Meigen
Mesotonum with black spots located near each postalar callus.	Mesonotum without black spots.
Trochanters 3 armed with a scoop-like apophysis, apically asymmetrical (Fig. 1).	Trochanters 3 armed with an acuminate dentiform apophysis (Fig. 2).
Tibiae and tarsi, tarsus 4 excepted, rust-yellow.	Tibiae 1 and 2 with brown median rings; tarsi yellow.
Abdomen with transversal bands, covered by an evident grey pruinosity.	Abdomen with evanescent or absent transversal bands.
Genitalia; Fig. 3, surstylus (lateral view); Fig. 4, apex aedeagi (dorsal view).	Genitalia: Fig. 5, surstylus (lateral view); Fig. 6, apex aedeagi (dorsal view).
Habitat: South-eastern region of Romania (Dobrudja); average altitude 200—350 m.	Habitat: Central and Southern Europe, Asia Minor, North-Western Africa; in the mountains up to the subalpine regions.

FEMALE.

Merodon ambiguus sp. n.	*Merodon rufipes* Sack
Antennae brown; 3rd article almost 1 1/2 longer than the 2nd.	Antennae reddisch-yellow; 3rd article subequal to the 2nd.
Tibiae and tarsi, tarsus 4 excepted, rust-yellow.	Tibiae reddish-yellow; distal tarsi brownish.
Abdomen bluish-black, with black (prevailing) and whitish-yellow pilosity.	Abdomen black, with yellowish-white pilosity.
Genitalia: Fig. 7.	

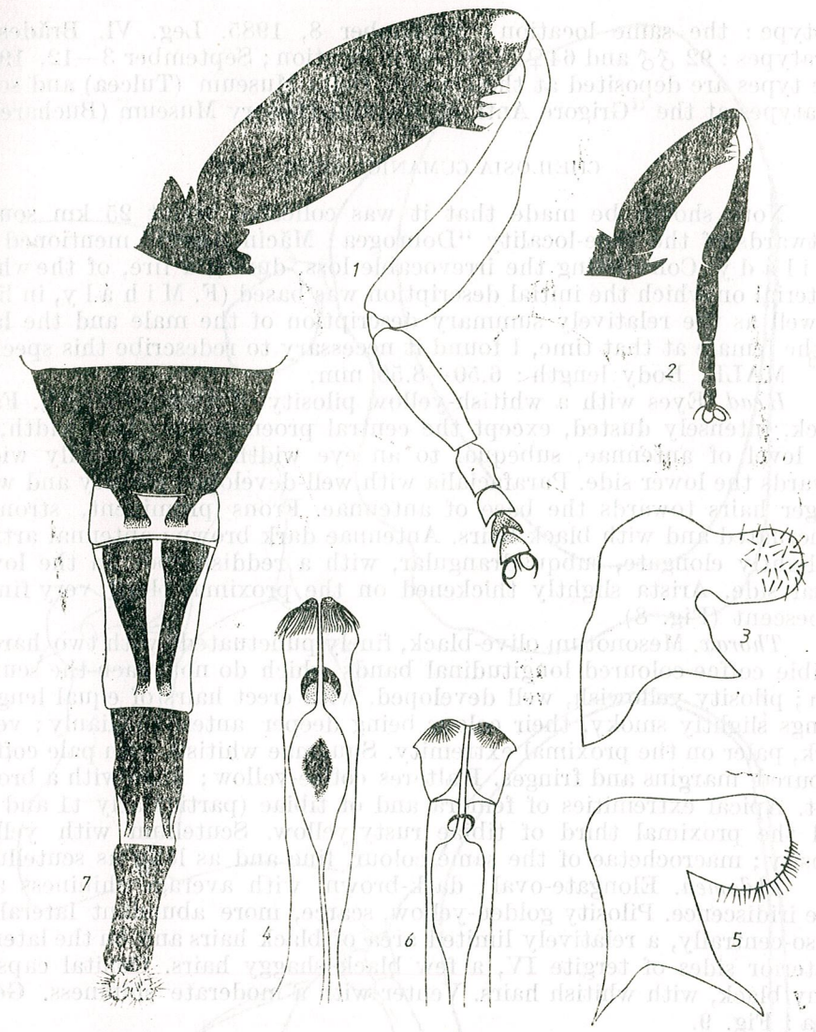

Fig. 1. — *Merodon ambiguus* sp. n. ♂, leg 3.
Fig. 2. — *Merodon aeneus* Meigen, ♂, leg 3.
Fig. 3. — *Merodon ambiguus* sp. n., ♂, surstylus (lateral view).
Fig. 4. — *Merodon ambiguus* sp. n., ♂, apex aedeagi (lateral view).
Fig. 5. — *Merodon aeneus* Meigen, ♂, surstylus (lateral view).
Fig. 6. — *Merodon aeneus* Meigen, ♂, apex aedeagi (lateral view).
Fig. 7. — *Merodon ambiguus* sp. n., ♀, genitalia (dorsal view).

MATERIAL

I had at my disposal 94 ♂♂ and 65 ♀♀ (88 ♂♂ and 53 ♀♀ leg. Vl. Brădescu; 6 ♂♂ and 12 ♀♀ leg V. Gheorghiu). Male holotype: Romania (Dobrudja), Niculițel Plateau, Alba Village — Izvoarele Commune (average altitude 200—350 m,) Tulcea Country; September 8, 1985. Female

allotype : the same location ; September 8, 1985. Leg. Vl. Brădescu. Paratypes : 92 ♂♂ and 64 ♀♀ ; the same location ; September 3—12, 1985. The types are deposited at the Danube Delta Museum (Tulcea) and some paratypes at the "Grigore Antipa" Natural History Museum (Bucharest).

CHEILOSIA CUMANICA Szilády, 1938

Note should be made that it was collected about 25 km south-eastwards of the type-locality "Dobrogea : Măcin, Greci" mentioned by Szilády. Considering the irrevocable loss, during a fire, of the whole material on which the initial description was based (F. Mihály, in lit.), as well as the relatively summary description of the male and the lack of the female at that time, I found it necessary to redescribe this species.

MALE. Body length : 6.50—8.50 mm.

Head. Eyes with a whitish-yellow pilosity of average length. Face black, intensely dusted, except the central proeminence ; face width, at the level of antennae, subequal to an eye width and gradually wider towards the lower side. Parafacialia with well-developed pilosity and with longer hairs towards the base of antennae. Frons prominent, strongly punctuated and with black hairs. Antennae dark brown ; antennal article 3 slightly elongate, subquadrangular, with a reddish spot on the lower basal side. Arista slightly thickened on the proximal third, very finely pubescent (Fig. 8).

Thorax. Mesonotum olive-black, finely punctuated, with two hardly visible coffee-coloured longitudinal bands which do not reach the scutellum ; pilosity yellowish, well developed, with erect hairs of equal length. Wings slightly smoky, their colour being deeper antero-medianly ; veins dark, paler on the proximal extremity. Squamae whitish, with pale coffee-coloured margins and fringes. Halteres coffee-yellow ; knob with a brown spot. Apical extremities of femora and of tibiae (particularly t1 and t2) and the proximal third of tibiae rusty-yellow. Scutellum with yellow pilosity ; macrochetae of the same colour, fine and as long as scutellum.

Abdomen. Elongate-oval ; dark-brown, with average shininess and blue iridiscence. Pilosity golden-yellow, scarce, more abundant laterally ; dorso-centrally, a relatively limited area of black hairs and on the lateral-posterior sides of tergite IV, a few black shaggy hairs. Genital capsule shiny black, with whitish hairs. Venter with a moderate shininess. Genitalia : Fig. 9.

FEMALE. Body length : 6.50—8.00 mm. A series of its main characters reveal a definite resemblance with the male : the aspect and ornamental characteristics of the face, the pilosity of parafacialia, the arista, the body pilosity and coloration, the rusty-yellow macrochetae of scutellum, the legs construction and coloration, the wings, the moderate shininess of the ventral side of abdomen. At the same time, several obvious characters indicate a strong enough sexual dimorphism. Accordingly, antennal article 3 with a surface at least twice larger than that of the same antennal article of the male, subroand and prevailingly reddish-yellow and with a relatively narrow black band on the upper margin (Fig. 10) ; eyes with a whitish, relatively dense, extremely short pilosity ; mesonotum strongly punctuated.

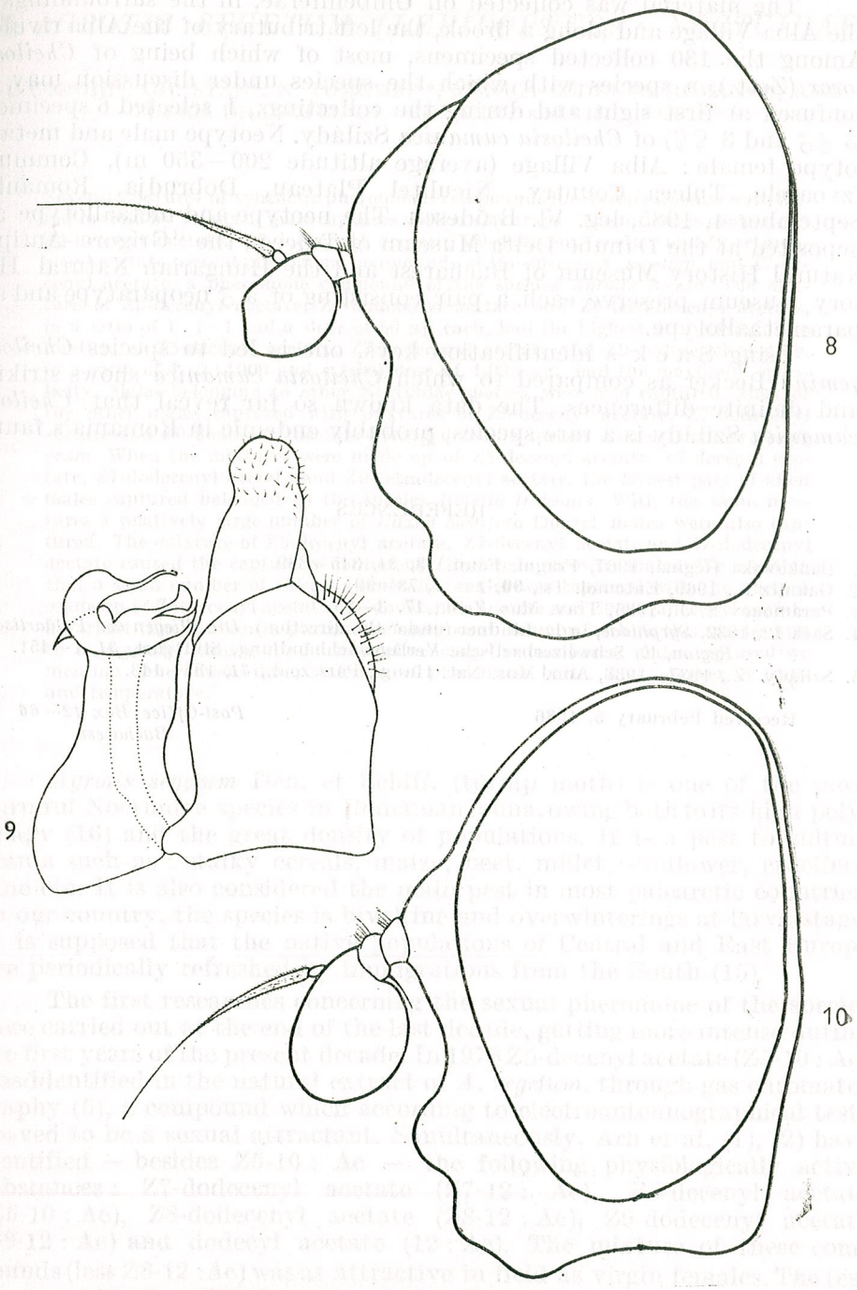

Fig. 8. — *Cheilosia cumanica* Szilády, ♂, head (profile).
Fig. 9. — *Cheilosia cumanica* Szilády, ♂, genitalia (lateral view).
Fig. 10. — *Cheilosia cumanica* Szilády, ♀, head (profile).

The material was collected on Umbelliferae, in the surroundings of the Alba Village and along a brook, the left tributary of the Alba rivulet. Among the 130 collected specimens, most of which being of *Cheilosia soror* (Zett.), a species with which the species under discussion may be confused at first sight and during the collectings, I selected 6 specimens (3 ♂♂ and 3 ♀♀) of *Cheilosia cumanica* Szilády. Neotype male and metaallotype female : Alba Village (average altitude 200—350 m), Commune Izvoarele, Tulcea Country, Niculițel Plateau, Dobrudja, Romania ; September 4, 1985, leg. Vl. Brădescu. The neotype and metaallotype are deposited at the Danube Delta Museum of Tulcea ; the "Grigore Antipa" Natural History Museum of Bucharest and the Hungarian Natural History Museum preserve each a pair consisting of a ♂ neoparatype and a ♀ parametaallotype.

Using S a c k 's identification keys, one is led to species *Cheilosia gemina* Becker as compared to which *Cheilosia cumanica* shows striking and definite differences. The data known so far reveal that *Cheilosia cumanica* Szilády is a rare species, probably endemic in Romania's fauna.

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ATTRACTIVITY AND SPECIFICITY OF SOME PHEROMONAL SYNTHETIC COMPOUNDS IN *AGROTIS SEGETUM (LEPIDOPTERA : NOCTUIDAE)*

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Several mixtures of synthetic pheromonal compounds for *Agrotis segetum* were tested in field by means of male captures with sticky traps. The mixtures contained : Z5-decenyl acetate, Z7-dodecenyl acetate, Z9-dodecenyl acetate and Z9-tetradecenyl acetate, sexual pheromone compounds of the species *A. segetum*, and Z7-decenyl acetate, a pheromone compound of the species *Agrotis fucosa*. The mixture of Z5-decenyl acetate, Z7-dodecenyl acetate and Z9-tetradecenyl acetate, in a ratio of 1 : 1 : 1 and a dose of 50 μg each, had the highest attractivity. The mixture of Z5-decenyl acetate, Z7-dodecenyl acetate and Z9-dodecenyl acetate, in a ratic of 1 : 2 : 1000 and a total dose of 1,025 μg, had the maximum specificity. Males belonging to other Noctuidae species were also captured. Most of the other males captured with mixtures of Z5-decenyl acetate, Z7-dodecenyl acetate and Z9-dodecenyl acetate belonged to the species *Xestia (Amathes) c-nigrum*. When the mixtures were made up of Z5-decenyl acetate, Z7-decenyl acetate, Z7-dodecenyl acetate and Z9-tetradecenyl acetate, the largest part of alien males captured belonged to the species *Emelia trabealis*. With the same mixtures a relatively large number of *Euxoa hastifera* Donzel males were also captured. The mixture of Z5-decenyl acetate, Z7-decenyl acetate and Z7-dodecenyl acetate caused the capture of numerous *Diachrysia chrysitis* males. We mention that a small number of *Cucullia umbratica* males was captured with the variant made up of Z5-decenyl acetate, Z7-dodecenyl acetate and Z9-tetradecenyl acetate a variant that contains a pheromone compound common both to this species and to *A. segetum*. Among the factors influencing the number of males captured we mention ; pheromone dose, staleness of pheromone baits, sticky power of traps and temperature.

Agrotis segetum Den. et Schiff. (turnip moth) is one of the most harmful Noctuidae species in Romanian fauna, owing both to its high polyphagy (16) and the great density of populations. It is a pest to culture plants such as : stalky cereals, maize, beet, millet, sunflower, crucifers, vine etc. It is also considered the main pest in most palearctic countries. In our country, the species is bivoltine and overwinterings at larva stage. It is supposed that the native populations of Central and East Europe are periodically refreshed by immigrations from the South (15).

The first researches concerning the sexual pheromone of the species were carried out at the end of the last decade, getting more intense during the first years of the present decade. In 1978 Z5-decenyl acetate (Z5-10 : Ac) was identified in the natural extract of *A. segetum*, through gas chromatography (5), a compound which according to electroantennographical tests proved to be a sexual attractant. Simultaneously, Arn et al. (1), (2) have identified — besides Z5-10 : Ac — the following physiologically active substances : Z7-dodecenyl acetate (Z7-12 : Ac), E5-decenyl acetate (E5-10 : Ac), Z8-dodecenyl acetate (Z8-12 : Ac), Z9-dodecenyl acetate (Z9-12 : Ac) and dodecyl acetate (12 : Ac). The mixture of these compounds (less Z8-12 :Ac) was as attractive in field as virgin females. The tes-

tings in the wind tunnel and in field showed that Z5-10 : Ac, Z7—12 : Ac and, probably, Z9-12 : Ac are essential for attracting. Z8-12 :Ac was highly inhibitory (2).

Tóth et al. (19) have also isolated two compounds from the raw extract of sexual pheromone : Z7-12 : Ac and Z9-tetradecenyl acetate (Z9-14 : Ac), in a ratio of 4 : 1. Their experiments, carried out in two areas of Hungary, show that *A. segetum* populations have differently responded to the two compounds. In one area, Z7-12 : Ac was more attractive alone than in the mixture of 4 : 1 with Z9-14 : Ac, while in the other area Z9-14 : Ac had a synergic effect.

Subsequently, co-operation experiments were done by researchers of four European countries : Switzerland, Denmark, France and Hungary. Z5-10 : Ac, Z7-12 : Ac and Z9-14 :Ac, alone and in mixtures, were used as active substances (3), (4). The results were different. In France, all the three compounds were attractive alone and Z7-12 : Ac has an inhibitory effect upon Z5-10 :Ac when the proportion was larger than 1 : 1 in its favour. These results were not confirmed in the other countries, where neither the compounds alone nor the mixture of Z5-10 : Ac and Z9-14 : Ac were attractive. In all the four countries, the highest attractivity for *A. segetum* males was achieved by the mixture of the three compounds in equal parts (for total dose of 25 μg). The heterogeneous results obtained with *A. segetum*, concerning the attractivity of different pheromone mixtures, suggest that the species has local populations which differ genetically and behaviourally and respond differently to the compounds of their specific sexual pheromone. Some authors also suggest a possible mistaken determination of the moths or an incomplete identification of the compounds (4).

Parallel tests done in Israel and West Germany (9) showed that besides Z5-10 : Ac, 7Z-12 : Ac and Z9-14 : Ac, 12 :Ac also plays an important part in the attractivity of the sexual pheromone in *A. segetum*.

In addition to the researches concerning the attractivity of the sexual pheromone and its compounds, pheromone traps have also been used in researches into the dynamics of *A. segetum* populations (8), at the same time investigating some factors influencing the capture of males with such traps (10).

Literature data show that there are common compounds in the sexual pheromone structure of several species, their specificity being sometimes determined by the different ratio of the compounds. A similar situation can be found in *A. segetum* and *A. fucosa* : they both have Z5-10 : Ac in the structure of their sexual pheromone. In *A. fucosa*, in addition to this compound, Z7-decenyl acetate (Z7-10 : Ac) was also identified (20). But Dunkelblum et al. (9) showed that the two species are phylogenetically very close or that they could be geographical populations of the same species.

The present paper deals with the attractivity and specificity of some synthetical pheromone compounds of *A. segetum* species, the dynamics the of the adults' flight, the influence of some factors on the capture, and density of its populations on the Transylvanian Plateau.

MATERIAL AND METHODS

The experiments were done in 1985 on an agricultural ground near the town of Cluj-Napoca. Table 1 shows the mixtures with pheromone compounds and the doses. They were supplied by the Chemistry Institute of Cluj-Napoca. Montedison traps with glue, having an increased active surface (900 cm²) (18) were used for all tests and the pheromone was conditioned on rubber septa. The traps were placed in cultures of : cabbage,

Table 1

Variants and doses of *A. segetum* synthetic sexual pheromone in 1985

Compounds Variants	Z5—10 : Ac	Z7—10 : Ac	Z7—12 : Ac	Z9—12 : Ac	Z9—14 : Ac	Proportion of compounds	Dose (μg)
A	2	—	4	199	—	1 : 2 : 100	205
A_1	10	—	20	995	—	,,	1025
B	16	—	8	—	—	2 : 1	25
C	10	100	5	—	—	2 : 20 : 1	115
D	50	—	50	—	50	1 : 1 : 1	150

sugar beet, onion, carrot and tomato. The height of each trap was of 100—120 cm above the ground, regardless of culture, and the distance between them was of 80—100 m. In the field, the traps were distributed in a randomized block, so that the distance between two traps with the same variant was not smaller than 160 m.

The tests were done during both flights, from May until September. The collecting of the captured adults was done every other day or even at 3—5 days, depending on the abundance of captures, i.e. depending on population density at a given moment and meteorological factors. After each collection, the captured individuals were taxonomically determined. The baits were changed about once a month and the sticky parts of the traps were changed whenever necessary, in order to avoid the covering of the adhesive glue with different organs of the captured moths, as well as the impregnation of the traps with corpse smell, which has a repellent effect on living males. The values of climatic factors such as : temperature, relative humidity, wind speed and rainfall have been recorded. The results have been statistically interpreted.

RESULTS AND DISCUSSION

The total number of *A. segetum* males captured during our experiments was of 11,337.

Table 1 shows the mixtures of pheromone compounds tested in fields. Besides some compounds identified in the sexual pheromone of *A. segetum* (Z5-10 : Ac, Z7-12 : Ac and Z9-14 : Ac), we also tested the second compound identified in *A. fucosa* (Z7-10 : Ac), the first one being common to both species (Z5-10 : Ac). The first two variants (A an A_1) had the same compounds but the doses were different ; the greatest difference lay in the amount of Z9-12 : Ac. The B and C mixtures differed,

first of all, in their structure as the quantitative differences between the two common compounds (Z5-10 : Ac and Z7-12 :Ac) were relatively small. The D mixture had the largest dose of the two above-mentioned compounds and also contained Z9-14 :Ac.

Furter on, attractivity and specificity will be presented as against the number of captured males and the species they belong to.

The highest attractivity was shown by D variant, where the average number/trap/night was of 12 males. With the same variant, the total number of captured males/period and the maximum number of males /trap/night in both flights were the highest (Table 2). The D variant could be compared to the A_1 variant, which had Z5-10 : Ac and Z7-12 : Ac in

Table 2

Attractivity and specificity of *A. segetum* synthetic pheromone mixtures in 1985

Variants	1st generation*			2nd generation **			Specificity %
	Total Nr. males	$\bar{x}$ (1)	Maximum Nr./trap/ night	Total Nr. males	$\bar{x}$(2)	Maximum Nr./trap/ night	
A	461	4.921a***	18	1,124	4.298a	12	94.4
A_1	824	9.050b	27	1,751	6.406b	21	96.6
B	675	8.089ab	25	1,599	6.055bc	16	84.9
C	776	10.092b	26	1,277	4.806acd	20	67.04
D	1,124	12.316b	34	1,726	6.243bd	24	89.05
Total	3,860			7,477			

* — four traps/variant; 20.5—17.7.
** — seven traps/variant; 17.7—25.9.
*** — means followed by the same letter are not significantly different at the 5% level according to "t" test.
$\bar{x}$ (1): 20.5—10.6.
$\bar{x}$ (2): 29.7—30.8.

a relatively large quantity in its composition, but the quantity in D was still larger and it also differed by its third compound. Thus, it is possible that the higher attractivity of D variant might be determined by the larger quantity of the first two compounds or by the presence of Z9-14 :Ac in the mixture. This aspects remain to be cleared up by future investigations.

The B and C variants contain relatively similar amounts of *A. segetum* compounds, but C variant also contains Z7-10 : Ac. This is a pheromone compound identified in *A. fucosa*. Although in C variant the *A. segetum* pheromone compounds were present in smaller amounts than in B, the average number of captures with C variant was larger, in spite of the fact that the statistical processing of data does not show significant differences ("t" test ; p = 0.05) (Table 2). Nevertheless, it is possible that the presence of Z7-10 : Ac in the mixture of the two compounds of *A. segetum* species might have had a slight synergical effect.

Table 2 shows in percentages the specificity of the pheromone mixtures used in the 5 variants tested. The best specificity (over 90%) was recorded in A and A_1 variants. The D variant also had a good specificity

(89%); this variant also had the highest attractivity for *A. segetum* males The poorest specificity was recorded in the C variant, which had in its mixture the pheromone compound of *A. fucosa* species. The conclusion is that the presence of an alien pheromone compound, existing in the pheromone composition of another species, brings about a decrease in specificity, even if it increases attractivity more or less. Among the captured males belonging to other species, most of them are Noctuidae species and very few are from the Lymantriidae, Pyralidae and Arctiidae families. From the Noctuidae, a relatively large number of *Xestia (Amathes) c-nigrum* L. males (Fig. 1) were captured in all the 5 variants. On the other hand, in traps with Z7-tetradecenyl acetate (Z7-14 : Ac) for *X. c-nigrum* (6), (17), a relatively large number of *A. segetum* males were also captured, but when simultaneously tested in the same area, both pheromones were specific (18). An explanation of this phenomenon will be attempted only after checking up once more the specificity of these variants in field.

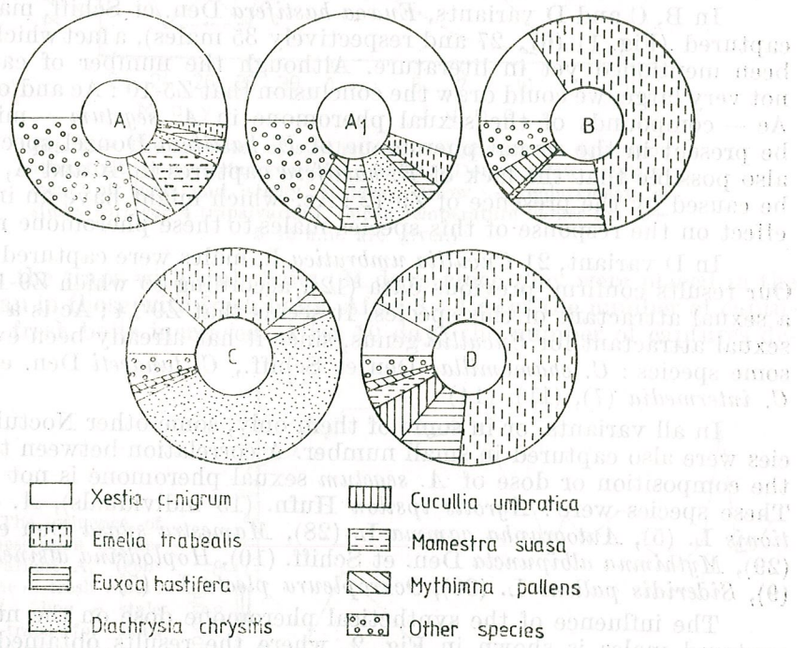

Fig. 1. — Noctuidae males captured in traps with the 5 variants of *A. segetum* synthetic sexual pheromone.

In B, C and D variants *Emelia trabealis* Scop. individuals were captured (Fig. 1) (246, 154 and 244, respectively). These results could be explained by the fact that, by screening test, it was established that Z5-10 : Ac is a sexual attractant of this species (13), and at the same time it is the main compound of the sexual pheromone in *A. segetum*. The capture of an extremely small number in A and A_1 variants (2 and respectively 5 individuals) demonstrates that Z9-12 : Ac, present only in these variants,

inhibits the response of *E. trabealis* males to Z5-10 : Ac. An interesting aspect noticed after changing the sticky part of traps, was that a great number of *E. trabealis* males were immediately captured, and that after some days their number sensibly decreased ; it increased again after changing the sticky part of traps. It seems that the decrease in captures after a certain period is caused by the corpse smell produced by the body remnants of the previously captured males, a smell having a strogly repellent effect on the males of this species.

In C variant a great number of *Diachrysia chrysitis* L. males were captured (Fig. 1) (282 males). In this case, too, there is an explanation : in *D. chrysitis*, the sexual pheromone contains Z5-10 : Ac and Z7-12 : OH [13]. The absence of response in the males of this species to the other variants could be due either to the presence of Z7-10 : Ac in C variant only, or to the absence of Z9-12 : Ac and Z9-14 :Ac in this variant. Subsequent researches will confirm one of these hypotheses.

In B, C and D variants, *Euxoa hastifera* Den. et Schiff. males were captured (Fig. 1) (67, 27 and respectively 35 males), a fact which has not been mentioned yet in literature. Although the number of captures is not very large, we could draw the conclusion that Z5-10 : Ac and/or Z7-12 : Ac — compounds of the sexual pheromone in *A. segetum* — might also be present in the sexual pheromone of *E. hastifera* Donzel species. It is also possible that the lack of *E. hastifera* captures in A and A_1 variants be caused by the presence of Z9-12 : Ac, which might have an inhibitory effect on the response of this species males to these pheromone mixtures.

In D variant, 21 *Cucullia umbratica* L. males were captured (Fig. 1). Our results confirm previous data (12), according to which Z9-14 : Ac is a sexual attractant of this species. It seems that Z9-14 ; Ac is a common sexual attractant for *Cucullia* genus, since it has already been evinced in some species : *C. chamomillae* Den et Schiff., *C. tanaceti* Den. et Schiff, *C. intermedia* (7), (11), (14).

In all variants, or in some of them only, some other Noctuidae species were also captured in small number. A correlation between them and the composition or dose of *A. segetum* sexual pheromone is not possible. These species were : *Agrotis ipsilon* Hufn. (15 individuals), *A. exclamationis* L. (5), *Autographa gamma* L. (28), *Mamestra suasa* Den et Schiff. (29), *Mythimna albipuncta* Den. et Schiff. (10), *Hoplodrina alsines* Brahm (9), *Sideridis pallens* L. (36), *Ochropleura plecta* L. (5).

The influence of the synthetical pheromone dose on the number of captured males is shown in Fig. 2, where the results obtained with A and A_1 variants are comparatively presented. During the observation period we found that in the nights when temperature was within optimum limits (namely above 10°C), the number of captured males was larger in A_1 variant, which was in higher dose. At lower temperatures (10°), the differences in captures diminished, being few in both variants. It seems that the temperatures of/and below 10°C are suboptimum for the flight of this species. Towards the end of the observation period, the number of captured males decreased in both variants, because the first flight was in a final stage, as it may be seem in Fig. 2.

The results concerning the duration of bait effectiveness in optimum dose (A_1 variant) are shown in Fig. 3. One can see that at the beginning of the observations, the average number of captured males was a little

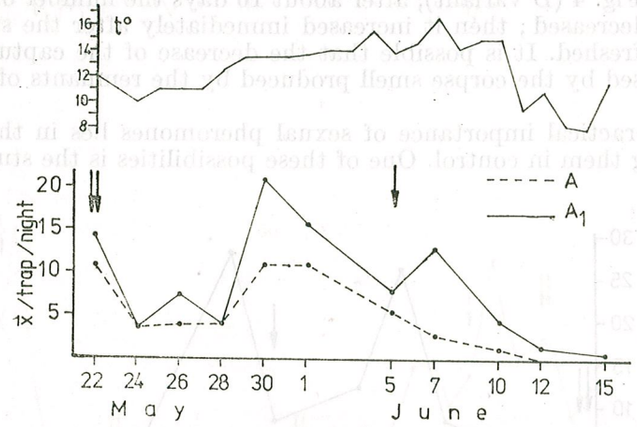

Fig. 2. — The influence of synthetic sexual pheromone dose upon attractivity in *A. segetum* (variants A and A_1). Double arrow—replacement of baits and sticky surface ; arrow -- replacement of sticky surface ; 4 traps/variant. (Mean temperature values for 00—5.15 a.m. are given).

larger in the traps with stale baits (34 days since they were placed in the field) than in those with fresh baits. After 3—4 days the number of captures with fresh baits increased. After 10 days the number of captures de-

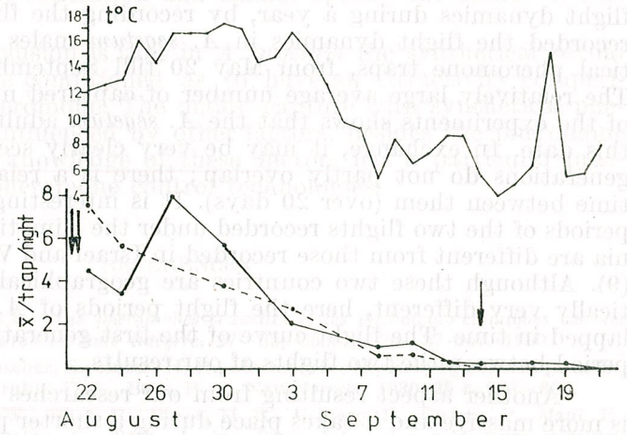

Fig. 3. — The influence of bait staleness upon attractivity in variant A_1 (continuous line — fresh baits ; interrupted line — stale baits). (7 traps for both types of baits).

creased in both bait types, due to the reduction of the intensity of adults' flight.

Another factor influencing the capture of *A. segetum* males was the covering of the sticky surface with the scales and organ remnants

of captured males. In this respect, we made observations on the dynamics of the captured moths number on the same sticky surface, during a certain number of days, in comparison with a new sticky surface. At is may be seem in Fig. 4 (D variant), after about 10 days the number of captures suddenly decreased; then it increased immediately after the sticky surface was refreshed. It is possible that the decrease of the captures might be also caused by the corpse smell produced by the remnants of captured males.

The practical importance of sexual pheromones lies in the possibility of using them in control. One of these possibilities is the study of the

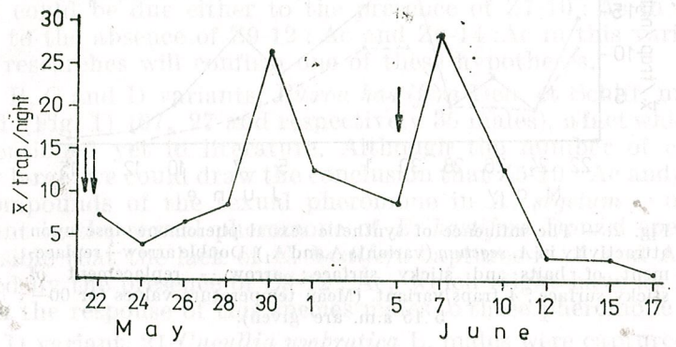

Fig. 4. — The influence of sticky surface replacement upon attractivity in *A. segetum*. (D variant — 4 traps) (the arrow shows sticky surface replacement).

flight dynamics during a year, by recording the flight curves. We have recorded the flight dynamics in *A. segetum* males by means of synthetical pheromone traps, from May 20 tilll September 25, 1985 (Fig. 5). The relatively large average number of captured males at the beginning of the experiments shows that the *A. segetum* adults' flight began before this date. In exchange, it may be very clearly seen that the two adult generations do not partly overlap; there is a relatively long period of time between them (over 20 days). It is interesting to mention that the periods of the two flights recorded under the climatic conditions of Romania are different from those recorded in Israel and West Germany in 1984 (9). Although these two countries are geographically distant and climatically very different, here the flight periods of *A. segetum* adults overlapped in time. The flight curve of the first generation coincides with the period between the two flights of our results.

Another aspect resulting from our researches is that the first flight is more massive and it takes place during a shorter period of time, as compared to the second flight.

We also studied the possible influence of the culture plant on the number of males captured in traps with identical baits. Our results show that in traps with identical baits placed in carrot and tomato cultures, more males were captured than in the other cultures. In this direction

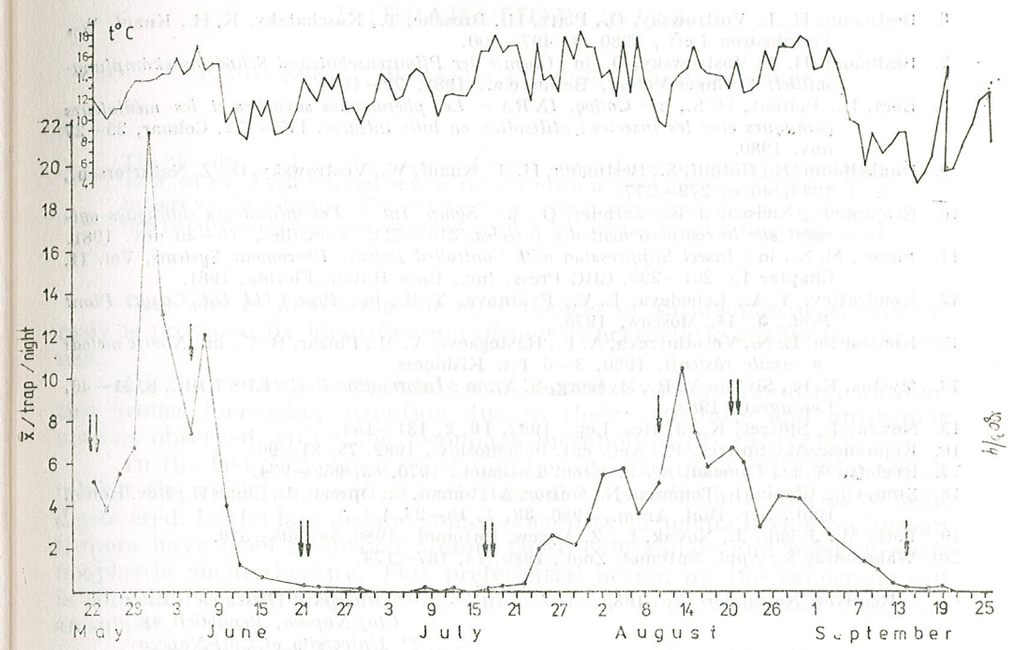

Fig. 5. — *A. segetum* flight curve in 1985, obtained by summing up the captures in all 5 variants with synthetic sexual pheromone. (Double arrow — fresh baits and sticky surface; arrow — fresh sticky surface). (Mean temperature values for 00—5.15 a.m. are given).

we cannot formulate a conclusion and cannot specify the causes of this phenomenon.

The general conclusion is that in *A. segetum* the synthetical sexual pheromone can be thus conditioned as to have an attractivity competitive to the natural pheromone. The males' capture with synthetical pheromone traps is also influenced by other factors, besides the pheromone properties. The exact knowledge of these factors has a particular practical importance for pheromone control technologies.

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CYTOSTATIC AND ANTITUMOR ACTIVITY OF THE BIOSYNTHETIC ANTIBIOTIC PREPARATION A 12.3

P. ROTINBERG, SMARANDA KELEMEN and AL. SAUCIUC*

The "in vitro" and "in vivo" action of a new biosynthetic antibiotic preparation A 12 .3, on HeLa cell cultures and on the evolution of solid and ascitic Guerin T—8 tumors was investigated. The results of the present investigation allow to consider this isolated secondary metabolite as a potential cytotoxic and antitumoral agent.

One way of increasing the effectiveness of antineoplastic chemotherapy is provided by identification of new cancerostatic agents for clinical use.

Of the natural compounds synthesized by microorganism, antibiotics arouse increasing attention due to their cytostatic and antitumor actions observed with some secondary metabolites (8), (15), (17).

In the last 25 years a great number of antitumor antibiotics which inhibit the vital functions of both normal and malignant cells have been discovered. In the last decade some cancerostatic agents acting on human tumors have been identified among these compounds and used in antineoplastic chemotherapy. This preferential action on the cancerous cell is due to its morphofunctional, biochemical, biophysical and antigenic anaplasia.

Chemotherapeutic screening programs aimed to identify new cancerostatic agents require a multistage investigation, both in "vitro" and "in vivo", into the action of drugs with supposed antitumor effects on various biological testing systems characterized by different reactivities (2), (3), (4), (6), (12), (13), (14), (16).

This paper presents the results obtained by testing the "in vitro" cytotoxic action on HeLa cells of antibiotic preparation A 12. 3 isolated at the Center for Antibiotic Research Iaşi, and its "in vivo" cancerostatic effect on rats bearing Guérin T-8 solid and ascitic tumors.

MATERIAL AND METHODS

Assessment of cytotoxic action of HeLa cell cultures

The cytotoxic action was assessed by comparative follow-up of total protein dynamics during evolution of cell cultures incubated with A 12.3 and of controls.

The test tubes were inoculated with 1×10^5 cells and after 24 hours the culture medium was replaced with a medium containing 1.5 mg/ml of the antibiotic preparation A 12.3.

At 24, 48 and 72 hours of cultures development the medium was discarded from the test tubes and the cell layer was washed with TFS and subjected to total protein determination (7), (9).

Five culture tubes were used for each type of culture and time interval, and the statistical analysis was performed using Student's "t" test.

Evaluation of antitumor activity

White Wistar female rats weighing 150 g and bearing lymphotropic epithelioma Guérin T-8 of solid or ascitic type (10) were used.

The treatment started 24 hours after the tumor transplant and lasted for 16 days in the case of solid tumor or until the death of the last control animal for the ascitic tumor.

The i.p. treatment was applied by administration of the drug A 12.3, at two day intervals, at a dose of 0.1 mg/kg. b.w.

The estimation of the antitumor activity was based on the follow-up of the mean tumor weight at sacrifice in the case of solid tumor or of the mean survival time in the case of ascitic tumor, in the treated groups comparatively to the controls.

The evaluation of antitumor action was made by the determination of mean tumor regression for solid tumor, by the increase of the mean survival time in the case of ascitic tumor and by the calculation of the statistic significance and the T/C value (where T = mean tumor weight or mean survival time for the treated groups and C = mean tumor weight or mean survival time for the controls).

RESULTS

The experimental results registered during the testing of the action of the antibiotic preparation A 12.3 on the development of HeLa cell cultures are presented in Fig. 1.

It is observed that the values of proteins which characterize the development of treated cultures are significantly decreased ($p < 0.001$)

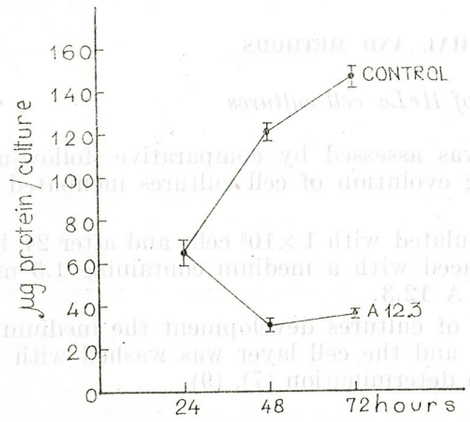

Fig. 1. — Protein content of HeLa cell cultures incubated with the antibiotic preparation A 12.3 (1.5 mg/ml).

as compared to the controls. The dynamics of total proteins in treated cultures shows that the A 12.3 isolate induces an inhibition of culture development by 75%.

The same experimental results were registered when the antibiotic preparation was retested (Fig. 2).

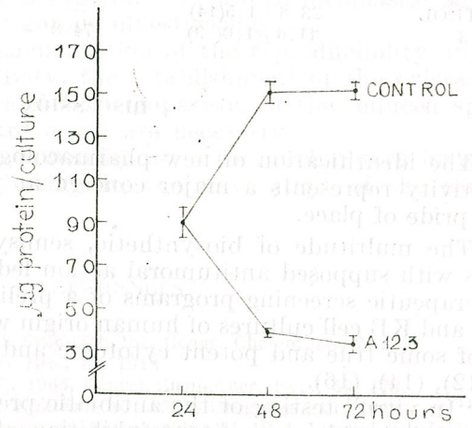

Fig. 2 — Retesting of the cytotoxic activity of the A 12.3 isolate (1.5 mg/ml) on HeLa cell cultures.

The results of the "in vivo" experiments testing the antitumor activity of the A 12.3 antibiotic isolate on rats bearing solid Guérin T-8 tumor are presented in Table 1.

Table 1

Antitumor activity of the antibiotic preparation A 12.3 (0.1 mg/kg b.w./2 days/i.p.) on solid Guérin T-8 tumor (Figures in brackets indicate the number of animals)

Group/Treatment	Mean tumor weight (g)	% tumor regression	Statistical significance	C/T value
CONTROL	10.0±1.1(14)	—	—	—
A 12.3	5.4±1.3(10)	46.0	$p<0.02$	0.54

In comparison with the control group it is observed that the treatment with A 12.3 induced a significant decrease ($p < 0.02$) of the mean tumor weight, which allows an estimate of a mean tumor regression of 46% and a T/C value of 0.54.

The investigation of the antitumor activity of the antibiotic preparation A 12.3 was extended to the ascitic type tumor (Table 2).

Once again, comparatively with the control group, it can be seen that the drug administration induced a significant ($p < 0.001$) increase of the mean survival time by 74.8% correlated with a T/C value of 1.75. Moreover, seven cases of tumor undevelopment have been registered.

Table 2

Antitumor activity of the antibiotic preparation A 12.3 (0.1 mg/kgb.w./2 days/i.p.) on ascitic Guérin T-8 tumor (Figures in brackets indicate the number of animals)

Group/Treatment	Mean survival time (days)	% increase mean survival time	Statistical significance	T/C val.	% tumor undevelopment
CONTROL	23.8±1.5(14)	—	—	—	—
A 12.3	41.6±1.0(3)	74.8	$p<0.001$	1.75	70.0

DISCUSSION

The identification of new pharmacological agents with antineoplastic activity represents a major concern at present, when chemotherapy holds pride of place.

The multitude of biosynthetic, semisynthetic and synthetic substances with supposed antitumoral action led to the inclusion in the chemotherapeutic screening programs of a preliminary "in vitro" testing on HeLa and KB cell cultures of human origin which should permit the selection of some true and potent cytotoxic and/or cytostatic agents (2), (3), (6,) (12), (14), (16).

"In vitro" testing of the antibiotic preparation A 12.3 showed that the drug induced a significant inhibition of HeLa cell culture development, illustrated by the profound alteration of the dynamics of cell total proteins.

The reproductibility, stability, statistic significance of the cell culture inhibition as well as the high intensity of action and its rapidity allow the characterization of the antibiotic preparation A 12.3 as a true and potent cytostatic and/or cytotoxic agent.

Although for some compounds positive correlations have been established between direct "in vitro" cytotoxic activity and the "in vivo" antitumor action (1), (5), (12), (14), (18), for the characterization of a drug as a cancerostatic agent a screening test on tumor bearing animals is required.

Therefore, we performed "in vivo" test of the antitumoral action of the biosynthetic secondary metabolite A 12.3 on rats bearing Guérin T-8 tumors.

"In vivo" testing of this biologically active compound revealed a significant tumor regression for solid type tumor, and a significant increase of the mean survival time, for ascitic type tumor. Antitumoral therapeutic efficiency of the antibiotic preparation A 12. 3 appears to be higher in rats bearing ascitic Guérin T-8 tumor. The seven registered cases of tumor undevelopment support this appreciation.

The comparison of the evaluation indexes of antitumor activity of the drug A 12.3 (mean tumor regression of 46% and T/C value of 0.54 registered in rats with solid Guérin T-8 tumor and mean survival time increase of 74.8% and T/C value of 1.75 registered with ascitic Guérin T-8 tumor) with those imposed by the multistage screening programs of the Institute of Microbiology and Experimental Therapy in G.D.R. and

of the Cancer Chemotherapy National Service Center in U.S.A. (4), (6), for selection of cancerostatically active agents in a first step (a mean tumor regression of at least 35% and a T/C value of 0.54 for solid tumors or an increase of the mean survival time of 30—50% and a T/C value of 1.25 for ascitic tumors), allows us to consider the isolate A 12.3 as a potential cancerostatic agent.

The characterization of a substance as an antineoplastic agent is the result of a complex and rigorous investigation.

To this purpose, the demonstration of the reproducibility and stability of the antitumoral activity, the establishment of the existence of a dose-response relationship and the comparison of the induced specific action with that of a standard agent are necesarry.

Therefore, the results obtained by us in this preliminary investigation with A 12.3 require its testing in the next steps of the complex screening program.

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HISTOLOGICAL MODIFICATION IN *MUSCA DOMESTICA* L. AND *TENEBRIO MOLITOR* L. AFTER THE APPLICATION OF SOME INSECTICIDES AND GROWTH REGULATORS (JHA)

ELENA TRACIUC and A. A. EL. FISHAWI

Insecticides and analogous juvenile hormone (JHA) were used on two species of insects for knowing the action of substance about their development.
The toxicity of Cypermethrin topically applied on chrysalis of *Musca domestica* L. induced external malformation on legs and wings with serious histological modifications.
The larvae of *Tenebrio molitor* L. in the third stage treated with analogous substances of the juvenile hormone underwent the process of gigantism. Some of these larvae started form the stage of giant chrysalis and after a few months became giant adults.

During their development, insects undergo complete or incomplete metamorphosis cycles, carried out in time according to certain living conditions and interspecific relations.

The intraspecific evolution is regulated by hormonal factors with inplications in sexuality and gametogenesis on the one side, diseases and control on the other side.

The control of some insects is natural ecosystems, mainly the anthropised ones, is very important from an economic point and raises a series of extremely difficult problems for the scientific research. Therefore the investigations initiated in the whole world have developed in two main directions; control with chemical substances and biological control (1), (4), (5), (7), (13, (14).

Insecticides have aroused a growing interest, being used more and more, in keeping with the development of the respective producing industries. Since insect control was spontaneous, or lethargical, the scientific interest coupled with the necessity of knowing the action of the substances on the animal growth, on the histological and cytological modifications at the level of some important organs, as well as the elucidation of biochemical mechanisms at the molecular level.

As regards the studies on the juvenile hormone (JHA) a series of investigations were initiated in order to use it in the control, to delay or block altogether the development of essential processes in the metamorphosis of some insects (2), (3,) (6,) (8,) (9,) (10), (11), (12).

The paper presents the evolution of organs and tissues in insects with external malformations induced by the topic application of some substances in certain moments of metamorphosis.

MATERIAL AND TECHNIQUES

Two types of insects obtained in laboratory were used. The toxicity of some insecticides in the chrysalis stages of the domestic fly *Musca domestica*, and the determination of the activity of some growth regula-

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tors of insects similar so the juvenile hormone on the larvae of coleoptera *Tenebrio molitor* were investigated.

The two species of insects were grown in laboratory at 27±2°C and relative humidity of 60%. The treatments with Cypermethrin and Sumicidin were applied topically : 1 µl of each test dilution on each chrysalis 2 and 4 days old of *M. domestica*.

Larvae of *T. molitor* in the third stage were treated topically with doses between 0.1 and 500 µg/larva. The substances were not toxic even when they were used in high doses. After the treatment, giant larvae appeared while the chrysalides of *M. domestica* produced adults with malformations on legs and wings.

The control group was treated only with acetone, the diluent of substances used in treatments in the same quantities as in the treated individuals.

Adults of malformated *M. domestica* and giant larvae of *T. molitor* were treated, the current techniques of histology being used. Tissue samples and whole animals were fixed in either Bouin-Hollande or Duboscq-Brasil fixative. The pieces were further dehydrated according to the clastical method : alcohol baths of increased concentration, then toluen baths followed by paraffin ones at 60°C and finally the pieces were embedded in paraffin. The pieces were cut at 7 µ, and the series sections treated with albumin Mayer were displayed on glass lamellas. They were stained with hemalaun Mayer-eosine and trichromic colorant according to Mallory. Micrographs representing the most evident modifications in various organs and tissues were made.

RESULTS

Musca domestica

The structural modifications denoted by us refer to parts of the tegument, wing structure, digestive apparatus, modifications of the Malpighi tubes, abundance and display of adipose cells, structure of compound eyes, as well as structure of tracheia of one day old adults of *M. domestica* originating from chrysalis treated with Cypermethrin.

In *tegument* the modifications appear in the area of the head, especially under the aspect of epicuticule thickening in certain regions accompanied by partial and rebel exfoliages.

As regards the hypoderm, which is normally formed of prismatic cells, among which the glandular ones of various types are present and which in case of treatments become pavimentous, of small size, reducing the cytoplasmic content while the nuclei become picnotic and flattened. It is evident that such a hypoderm shall generate a randomly developed cuticule since the glandular cells secreting it diminish their functionality as a result of the structural modifications undergone.

Wing structure. Macroscopically a degeneration of the wings is noticed to a larger or lesser extent. Such wings in histological sections on a lamella, present a degeneration of nervures so that the median ones can not by distinguished from the cubital ones. The spaces between

parallel nervures in some parts dilate so much as to break the separating walls. The cuticule layer covering the wing is even, crossed by fine and short pores which preserved their natural pigmentation. The generative dermic layer is structurally modified with pavimentous cells alternating with the prismatic ones, with lacunary spaces among them. The modified cells hipoderm have a finely granular cytoplasm, enlarged and chromatic nuclei.

Modifications of the compound eyes start from the outside namely in the cornean area, with the tendency to detach so that covered parts and open ones result. The cornean cells are stuck to the pigment so that in sections the area with these cells seems to be a dark strip. The crystallin of each omatid is loaded with pigment so that as a whole these formations can not be distinguished.

The pigmentary cells are grouped in the external area of the eye and present chromatic nuclei. The distribution of the pigmentary layer is different along the omatidia ; a marked distal agglomeration that covers the external part of the omatidia, which thickens in the proximal area making a bandage which covers the lower part of the omatidia.

The nuclei of pigmentary cells are visibly enlarged, while the retinian cells have slightly degenerated nuclei. Because of this bandage interfering with the basal membrane, the latter is not uniform but compressed here an there.

The layers of conjunctive and nevrogical cells under the basal membrane do not undergo any modification after the treatment applied.

The digestive system and Malpighi tubes. The strongest modifications affect mainly the Malpighi tubes, as the cells no longer have their brush-shaped margin, they become flat, in some parts of the tubes the cells detach, migrate in the lumen, and the basal membrane becomes nude on wide areas. This demonstrated a good functional activity, but as the animals were not abundantly fed, the explanation of the cellular modifications lies in the effect of the substance remanence.

As the Malpighi tubes are absorption organs of excretion production, the passage through the cells of the tube walls of some toxic molecules reduces the normal metabolism without any possibility of compensation. We mention that as there are external malformations and internal modifications in the wings which become non functional, the animal can not move and feed. It is evident that the cellular remaking of Malpighian tubes is impossible to achieve.

The same can be mentioned about some portions of the medium intestine. On histological sections the intestine is cross or longitudinally sectioned, with areas where the epithelium is well delimited, with normal structure, and close areas with portions of wide basal membrane.

The tracheal system presents the strongest morphological modification, the walls being expanded and the internal spaces widened. A process of widening is accompanied by a folding of walls with an aspect of branched tubules, which lost the elasticity characteristic of the trachea.

The trachea exocuticle lost its elasticity, becomes soft membranous, with swellings.

This evident structural modification of the trachea demonstrates in fact a clear intoxication of the organism. This leads to the enlargement

of the tracheae space with sinuous openings, alternating with narrow overcrowed parts. The cells of the trachean epithelium are retracted with large nuclei, with much chromatine. The general aspect is that of an organ which increases its volume to perform the gas exchange of the organism.

The adipose cells, which in the control adults are displayed round the digestive tube and distally under the tegument with an aspect of a subtegumentary cover, in individuals treated fill the abdominal cavity.

Anal gland. In treated individuals, the cells undergo some modification. The cell chains are close together, of visibly larger sizes and loaded with chromatic cells. The cell cytoplasm is strongly basophile and finely granular. Inside the cells there are numerous vacuoles found also in the control group.

Sexual glands. Although they do not undergo evident histological modifications as in the other described organs, mention should be made of the gonads, as they are directly implied in the reproduction of individuals. Gonads, although not affected by obvious cytological modifications are implied because certain animals having malformations, such as those in the wings and legs leading to immobilisation are therefore incapable of coupling. It is clear that the animal does not have physiological resources for coupling and the sexual glands, although under normal development could not reach maturity, the animals having not the physiological resources for coupling.

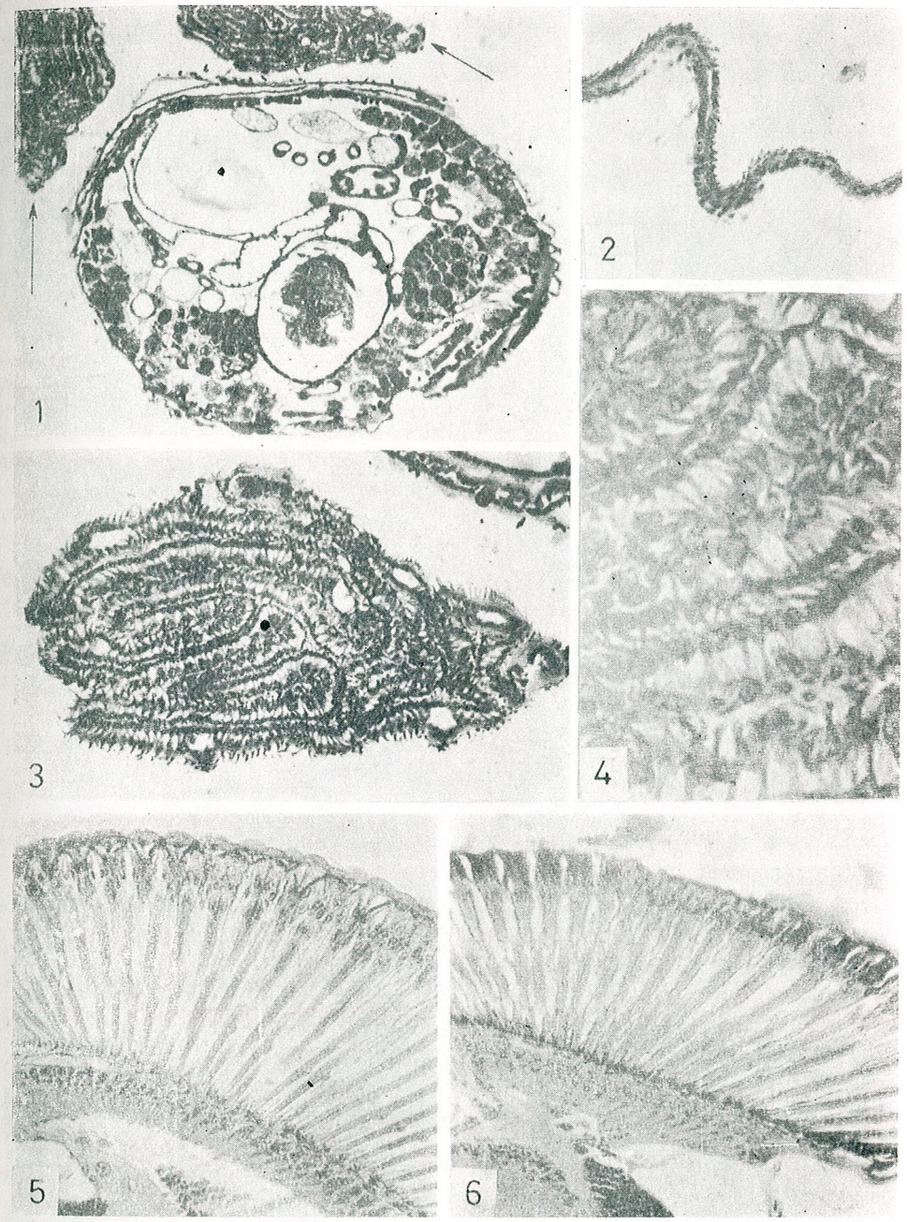
As regards the treatment with Sumicidin, no histological modifications were noticed, as well as in the treatment with (JHA)ZR-512 and RO-06-9550 applied to chrysalides of *Musca domestica* where no histological modifications were mentioned.

Tenebrio molitor. After the treatment with insecticides and juvenile hormones, by application on larvas in stage 3 we investigated the effect on the further development of the larvas treated in comparison with the control.

A growth in the size of the larvas treated in comparison with the control, with the development of giant larvas, was noticed the main interest attaching to the internal organs, mainly the histological lamellas. Besides a good development of the muscles and a large volume of the digestive system, an intense development of the adipose tissue, both of proximal strips and the distal ones was noted. The fat distal tissue is placed under the tegument and together with the muscles they form a strong subtegumentary bandage, accounting for the large size of the larva in comparison with the control. By dissection we could compare the organs of the giant larva with the control larva.

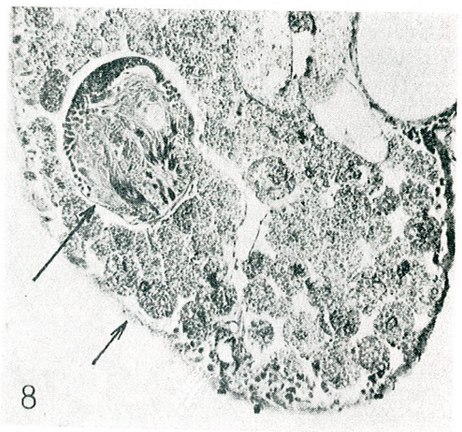
In this way we noticed large organs such as the digestive system, the adipose tissue as they do have the possibility of enlarging their size in comparison with the nervous or genital system with a strict development, because the cells of these systems do not undergo additional divisions, that is regenerative multiplication of growth. As it is known these systems do not undergo cellular multiplications during the postembrionary development.

Histological observations in giant larvas and adults in comparison with the control ones revealed neither a growth in cell volume nor modifi-



Musca domestica.

Fig. 1. — Histological section presenting the abdomen and wings of adult in the first day of life. The intestine and the Malpighi tubes are degenerated.
Fig. 2. — The tegument area.
Fig. 3. — Microscopical degeneration of the wing.
Fig. 4 — Disturbance of nervures. The generative dermic layer is structurally modified in the wings.
Fig. 5. — The eye of the control animal.
Fig. 6. — The modification of compound eyes.

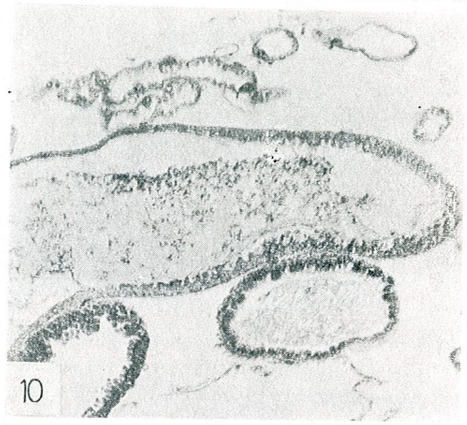
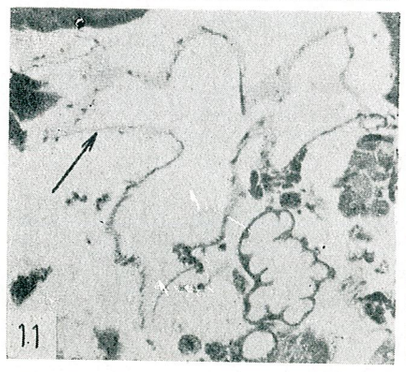
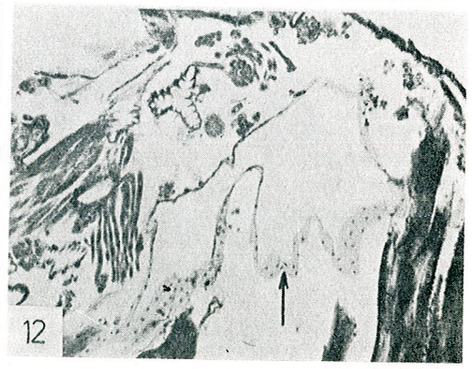




Fig. 7. — Section through abdomen. Intestin epithelum with normal structure and close areas with parts of wide basal membrane.
Fig.8. — The adipose cells fill the abdominal cavity. The sexual glands are normally developed. The tegument reveals histological modifications.
Fig. 9. — The Malpighi tubes are affected by the strongest modification.
Fig. 10. — Sections through medium intestine and Malpighi tubes.
Fig. 11. — The tracheal system is accompanied by a folding of the walls.
Fig. 12. — Aspect of branched tubules of trachea.

cations in their structure. The growth in volume of the digestive system and the fat tissue is made at the expense of the number of cells in giant individuals and not by increasing their dimensions. In some cases the giant larvas changed into giant adults; this occurred when the larva was maintained in the last stage for a longer period than the control larva in the same stage.

CONCLUSIONS

The attempt to treat chemically insect larvas and chrysalides was made to find new possibilities to prevent the development of pest insects or insects carrying infectious agents. Of course during the laboratory experiments we worked with insects that have repeated cycles in shorter periods or animals that do not require special care. In our case we worked on individuals belonging to species *Musca domestica* order Diptera and *Tenebrio molitor* order Coleoptera.

The skin treatments were made with chemical substances Cypermethrin and Sumicidin of which only Cypermethrin induced serious histological modifications, the ones mentioned above. The wing malformations were induced by both substances, but in various degrees. In this case we consider that the nature of Cypermethrin induces the disturbances at tissular level.

The epithelial cells with intense metabolism such as the tracheal cells, glandular cells and adipose cells underwent modifications.

The larvas of *T. molitor* of a third stage treated with analogous substances of the juvenile hormone, namely ZR-512 and RO-06-9550, continued their evolution to the last stage, but under the form of giant larvas maintained in the last stage much over their due time.

Some of these larvas passed to the stage of giant chrysalides and after a few months they became giant adult. Gigantism manifested by increasing the number of cells in the tissues that permit it, such as the cells of the hypoderm, the adipose cells and the cells of the digestive tractus.

After observations on the treatment with analogous juvenile hormone, we may conclude that in the treated larvas, the hormone acts on the skin as well by an increased process of cellular growth and multiplication, this being in fact one of the basic functions of these hormones. In this experiment we tested its action on the skin, its neuro-sanguine action being already known.

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HISTOLOGICAL CHANGES OF STRIATE MUSCLES OF *CYPRINUS CARPIO* UNDER THE ACTION OF MERCURY

VIORICA MANOLACHE and SIMONA MARCOCI

At different intervals of time there were pursued the effects of mercury-intoxication at doses of 1, 5 and 10 μg upon the structure of striate fibres in *Cyprinus carpio*.
Numerous aspects of degenerative modifications were noticed, especially the hypertrophy of striate fibres, the adherence of myofibrils to the sarcolemma, their reduction, pycnosis of nuclei, destruction of the vasculo-conjuctive tissue etc. All aspects affect the morpho-functionality of the striate fibre.

Aquatic organisms accumulate in their tissues and organs various pesticides and toxic metals, which do not only affect the behaviour of the animals, but cause also numerous morphological and functional changes.

There has been undertaken a great deal of research-work on different aspects of tissular degradation provoked by various toxic metals (Zn, Pb, Hg, Cd, As etc.) (1, 2, 3, 5, 6 etc.).

The present paper presents in the case of *Cyprinus carpio* structural modifications of the striate muscle-fibres as a consequence of small doses of mercury, at various intervals of time. It is well known that mercury residues influence the cellular metabolism and block the proliferation of the cells.

MATERIAL AND METHOD

The toxic action of mercury was studied for doses of 1, 5 and 10 μg. The analysis of histologic modifications in the striate muscles from the dorso-cervical region of *Cyprinus carpio* was carried out at the following time lapses since intoxication : 2 weeks, 1, 2, 3, 4, 5 and 8 months for the doses of 1 μg; at the same intervals and one year after for 5 μg; 1 1/2 month, 6 months, 1 year and 1 1/2 year for 10 μg.

RESULTS

Influence of the 1 μg Hg dose

After two weeks of treatment. Along with zones of normal aspect there are regions with rarefied muscle-fibres, partially fragmented. The conjunctive tissue wanders into the spaces freed by the destruction of the muscle fibres.

After one month of treatment. One observes the hypertrophy of muscle-fibres, the diminution of their number, the flattening and compression of nuclei. The sarcolemma becomes brittle and the normal outlines of the muscle-fibres become hazy.

After two months of treatment among some striate fibres of unmodified structure there are found some compressed in a transverse direction,

determining thus the disruption of normal topographic relations between the fibres and the appearance of some widened spaces, containing conjunctive tissue, which has become hyaline (Plate I, Fig. 1).

After four months of treatment the muscle-fibres show a trend for hypertrophy, and in some cases the fragmentation and dislocation of contractile elements occurs. The interfibrillar conjunctive tissue becomes lax (Pl. I, Fig. 2).

After 5 and 8 months of treatment, respectively, in certain areas a reduction in the number of striate fibres is noticed. The remaining fibres are hypertrophic, especially towards the insertion points. The sarcolemmae become sometimes fragile and disappear. The adhesion to the sarcolemma of the contractile elements reduces the sarcolemmal layer of cytoplasm and decreases the number of nuclei. The remaining nuclei are compressed and much flattened. Owing to the hypertrophy of the fibres the conjunctive tissue diminished, leading to the obstruction of the interfibrillar bloodvessels (Plate I, Fig. 3).

Influence of the 5 μg Hg dose

After two weeks of treatment. Besides regions of normal aspect there are regions wherein fibres appear partially or totally broken up. When fibres are fragmenting, the interfibrillar autolysis begins by the appearance of some spaces between the contractile elements and the decay of nuclei. In the places with broken up fibres the conjunctive spaces are occupied by decaying conjunctive tissue or by heaps of nuclei, originating from fragmented fibres (Plate I, Fig. 4).

After one month of treatment we noticed the condensation of fibres and interfibrillar spaces. In fibres undergoing destruction, the central contractile elements are degenerate, while only the peripheral ones remain active.

After 2 months of treatment the myofibrils diminish numerically. The nuclei of the fibres appear hypertrophic and hypochromatic, and in some cases flattened and compact. Sometimes the nuclei are displaced, protrude and push on the plasmalemma, while the cytoplasm around them is vacuolised. In the conjunctive tissue there are intensely eosinophile zones, comprising the hypochromatic nuclei of the decayed fibres. In some sites the conjunctive tissue is compacted, with blood-capillaries retracted, staying lax in other regions and being infiltrated with lymphocytes.

After 3 months of treatment the compression of striate fibres is observed; the contractile elements adhere abnormally to the sarcolemma and the nuclei are rarefied and flattened (Plate II, Fig. 5).

At the onset of the fibre decay their nuclei are hypertrophic. The spaces between fibres widen due to the reduction of the number of disorganised fibres, and the conjunctive tissue is displaced.

After 4 months of treatment we noticed the same phenomenon of muscle-fibre condensation, the degeneration being similar to that occurring after 3 months of treatment.

After 5 months of treatment. Besides fibres of normal structure we found in some regions fibres with the following signs of decay: disorga-

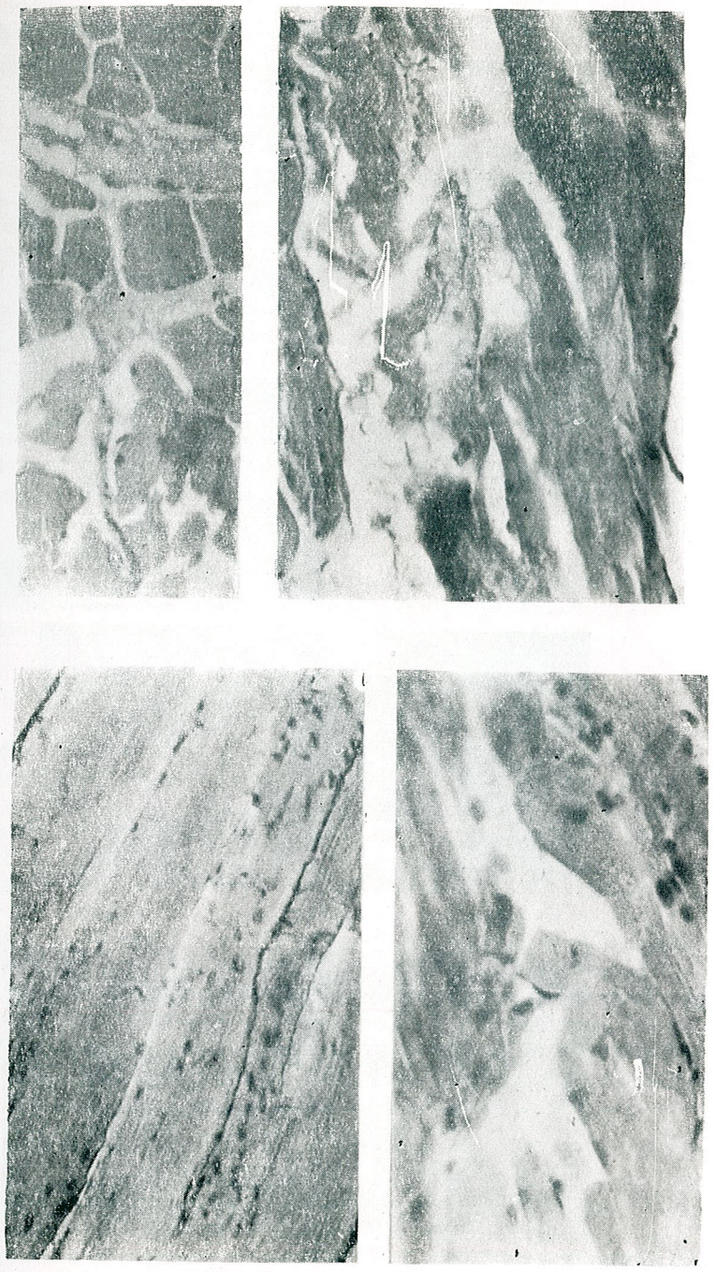

PLATE I — The striate muscle of *Cyprinus Carpio* under the action of the dose of 1 μg Hg/l

Fig. 1. — 2 months after treatment.
Fig. 2. — 4 months after treatment.
Fig. 3. — 8 months after treatment.
Fig. 4. — The striate muscle of *Cyprinus carpio* under the action of the dose of 5 μg Hg/l 2 weeks after treatment.

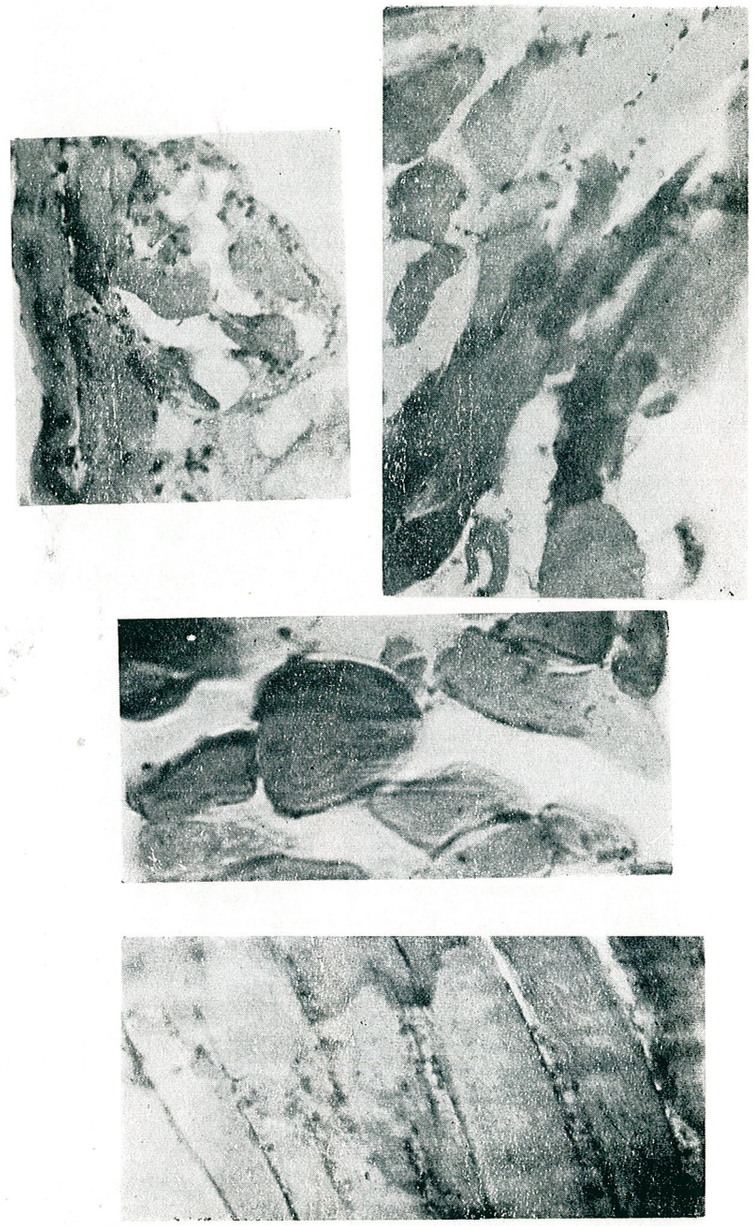

PLATE II
Fig. 5. — 3 months after treatment.
Fig. 6 — 8 months after treatment.
Fig. 7. — 1 year after treatment.
Fig. 8. — The striate muscle of *Cyprinus carpio* under the action of the dose of 10 µg Hg/l 18 months after treatment.

nised contractile elements, rarefied and autolysed. Within these fibres there appear large spots of subplasmalemmal vacuolisation, containing heavily compressed and pycnotic nuclei.

In other sites fibres are hypertrophic, the sarcolemmae become adherent to myofibrils, nuclei are flattened and pycnosis begins. Interfibrillar spaces are widened, the conjunctive tissue appears rarefied. Sometimes the interfibrillar spaces disappear and — due to the rupture of the plasmalemmae — in these regions fibres will fuse and degrade.

After 8 months of treatment one notices the obliteration of the striate fibres outlines, the rupture of sarcolemmae and the merging of fibres. The spaces between fibres become smaller, thus leading to the condensation of the conjunctivo-vascular tissue and the diminution of the normal trophicity of the muscular tissue (Plate II, Fig. 6).

After 1 year of treatment a large area exhibits the hypertrophy of striate fibres, suspension of interfibrillar spaces, rupture of sarcolemmae and merging fibres. The reduction of the conjunctive tissue and obstruction of the vessels lead to difficulties in maintaining the normal trophicity of the muscular tissue and to disturbances of interfibrillar structures (Plate II, Fig. 7).

The long-time action of the poison makes the hypertrophy and decay of striate fibres, the merging and the reduction of the vasculo-conjunctive tissue be permanent aspects.

Influence of the 10 µg Hg dose

After one and a half month of treatment. The muscular tissue shows many destroyed fibres in the degraded areas. Those which were not destroyed exhibit hypertrophy, adherence to sarcolemmae, merging fibres vacuolised subplasmalemmal cytoplasm, agglomeration of nuclei and degeneration of the latter. The fragmented fibres are autolysed. The sarcolemmae and the spoiled nuclei of the destroyed fibres remain. The vasculo-conjunctive tissue is condensed or rarefied.

After 6, 12 and 18 months of treatment we observed for each duration the same tissular type of decay, resembling that produced by the dose of 5 µg after one year of treatment. The decay consists of the spoiling of the contractile element, the destruction of sarcolemmae, the merging of fibres, the disappearance of interfibrillar spaces and the disappearance over large areas of the vasculo-conjunctive tissue (Plate II, Fig. 8).

DISCUSSIONS

The action of mercury residues at tissular level has been investigated by numerous research-workers (1, 2, 8, 9, 10 etc.).

Pursuing the effects of doses of 1, 5 and 10 µg mercury at different intervals, we noticed a series of degenerative modifications at the level of the muscular tissue, resembling those observed by (10) at the level of the hepatic tissue. Thus, in both types of tissues hypertrophy and its immediate consequences were noticed : irregular nucleo-cytoplasmic ratios, the disturbance of the metabolism and of the specific functionality

of both tissues. In the case of the striate muscular tissue of *Cyprinus carpio* we noticed the destruction of the structure of myofibrils; the same feature is reported by some authors (4) who worked on the effects of Lindan upon young specimens of *Salmo irideus*.

The reduction and spoiling of the vasculo-conjunctive tissue observed by (10) also with the hepatic tissue, leads to disturbances of the normal trophicity of the analysed tissues.

Although histologically there was noticed a reduction of the mass of muscular tissue by the loss of specific components, death of the organisms did not occur, because sufficient reserves still remained in the interval of time established as duration of the experiment.

CONCLUSIONS

— The dose of 1 μg maintains, with few exceptions, the normal structural aspect of striate muscles. The most frequent modifications, noticed at different intervals of action of the poison are: hypertrophy of the fibres, adherence of the contractile elements to the sarcolemma, reduction of the quantity of cytoplasm and even of the number of nuclei. In the interfibrillar spaces the consequence of hypertrophy is the disturbance of the morpho-functionality of the vasculo-conjunctive tissue. After 3 months the phenomenon of fibre fragmentation also occurs.

— In doses of 5 μg, mercury induces the following degenerative modifications of the striate fibres: condensation over the whole length of the fibres and hypertrophy, especially towards the insertion points. The interfibrillar contractile element suffers a reduction towards the centre resisting only at the periphery, where it adheres to the sarcolemma, spoiling also the nuclei.

— From the beginning of the experiment mercury in doses of 10 μg appears to determine the fragmentation of striate fibres. Further on, until the completion of 18 months of experimentation we remarked the hypertrophy and the merging of fibres, while extrafibrillarly there occurs the compression and reduction of the vasculo-conjunctive tissue.

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ON THE INFLUENCE OF THYROID HORMONES UPON THE INTESTINAL ABSORPTION IN CHICKENS

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Plymouth-Rock chickens were fed on fodders with 20, 18, and 16% proteins in the usual ratio of animal and vegetable ones, as well as with 18% exclusively soybean proteins. Thyroid was reversibly inhibited by thiourea treatment during the posteclosional weeks 1 and 2, and thyroid hormones were administered during weeks 7 and 8. Intestinal absorption capacity of glucose and leucine was determined *in vitro*, at the age of 8 weeks. Treatment turned out to be harmful in normally fed chickens, but stimulatory when the fodder was deficient in animal proteins.

There are many investigations on glucose or aminoacid absorption in mammals, and on the dependence of this process on various factors, like nutrients concentrations in fodder, nature of fed proteins etc. (2), (3), (6), (8). Only a small number of such studies are available with birds (5), (9). In a previous paper, we followed the *in vitro* capacity of the small intestine to absorb glucose and leucine in chickens and hens which had received fodders with quantitatively and qualitatively varying protein contents (1): we showed that differences in protein feeding do affect the absorptive capacity. Therefore, the present investigation was also conducted on the ground of various protein feedings.

MATERIAL AND METHODS

Plymouth-Rock hybrid chickens were divided in the first day after hatching into 8 groups. Four groups, constituting the control series, received fodders containing various amounts of proteins: 20% (C-20), 18% (C-18) and 16% (C-16), observing the usual ratio of animal and vegetable proteins, and the fourth group was fed on 18% exclusively vegetable one (as soybean meal; C-S). The schedule for fodders was the same for the other 4 groups (treated series, T) which received, during the first two weeks, 300 mg thiourea per kg body weight and per day, and during the 7th and 8th weeks 20 mg dry thyroid gland per kg and per day. Thus, it was intended to moderate the initial intense thyroid function, then to allow it to recover (weeks 3—6), and then to achieve a better finalization by supplementing the organism with exogeneous thyroid hormones.

Chickens were sacrificed by exsanguination at 8 weeks, after a 18—20 hour starvation. A jejunal segment of about 4 cm was immediately sampled and introduced in a physiological saline. Absorption was studied by the method of the "everted sac" (10), as modified by Petcu (7), and adapted by us to a radiobiochemical procedure. Phosphate-buffered Krebs-Henseleit saline, containing 10 mM glucose, was introduced into the sac, which was subsequently incubated for 1 hour, at 40°C, in the same saline, oxygen saturated and containing 15 mM glucose and radioactive additives. The latter were: (U-^{14}C)-D-glucose, about 4.10^5 DPM

(desintegrations per minute) per cm³, or (1-^{14}C)-DL-leucine, about 7.10^5 DPM, both products of the Institute for Physics and Nuclear Engineering, Bucharest. After incubation, the intestinal sac was rinsed in distilled water, blotted on filter paper, and opened. The liquid contained in the sac was sampled, measured with a micropipette, and processed for radioactivity determination. This was done in a Betaszint BF-5003 liquid scintillation spectrometer by using Bray's solution. Results were expressed either as DPM per mg jejunum (wet weight), or as partition coefficient, i.e. specific radioactivity of the internal fluid expressed as percentage of that of the external one.

Eight animals were processed for each experimental variant. Mean values per variant were checked for homogeneity by Chauvenet's criterion, aberrant values were eliminated, and the means were compared using Student's "t" test; $p = 0.05$ was accepted as the limit for statistical significance of differences.

RESULTS AND DISCUSSIONS

It is well known that thiourea induces a reversible blockage of the thyroid function, without morphological modifications (Makensie, 1947, and Borell and Holmgren, 1949, cited after (4)). Thus, in our experimental schedule, the function of the gland could be normalized during the 4-week interval until hormonal treatment. The latter was mild : 20 mg dry gland (as pulvered dragees of pharmaceutical Tiroida fortissimum, I.M.B. Bucharest) containing about 20 µg hormones. This did not lead to an increase in the hormonal concentration of the blood (unpublished data), the modification remaining within the limits of the homeostatic regulation. Thus, we cannot speak of a hyperthyroid state, but a sustained euthyroid one.

The treament had clearly different effects, depending on the protein content of the fodder. Both glucose and leucine absorption were strongly depressed in T-20 and T-18, as compared to C-20 and C-18; glucose absorption was clearly stimulated in T-16 and T-S, leucine absorption in T-S only (Tables 1 and 2). In the control series, a significant modification versus C-20 was found only for glucose : an increase in C-S.

In vivo and *in vitro* studies have shown that intestinal absorption of glucose and amino acids is dependent on the concentration of these nutrients in the diet (8). A protein-rich diet enhances the methionine transport in rats (3). Within the limits of our experimental schedule (20—16%), differences in the protein content of the fodder did not elicit any significant modification in intestinal absorption.

We cannot give a detailed explanation of the hormonal effects on intestinal absorption. The positive effect noticed in T-16 and T-S may be facilitated by an intensification of the ATPase activity in the intestinal wall (5).

In conclusion, a depression of thyroid function in young chickens, followed by recovery and exogeneous hormone supply seems to be harmful when associated with a normal protein feeding, but useful in chickens fed on a fodder deficient in animal proteins.

Table 1

Glucose absorption (DPM) and partition coefficient (%) in the small intestine of the chickens

	20%	18%	16%	v.p.
		DPM		
C	2.62±0.26	2.83±0.52	2.77±0.22	3.13±2.02
D_{20}		—8	+5.7	+19.5
T	0.85±0.21	0.45±0.10	5.46±0.56	4.27±0.40
D_{20}		—47*	+542.4**	+402.4**
D_C	—67.7***	—84.1***	+97.1**	+36.4*
		%		
C	11.5±0.88	11.7±1.82	9.73±0.86	25.6±4.5
D_{20}		+1.7	—15.5	+122.5**
T	1.96±0.31	1.25±0.31	13.61±1.4	12.93±0.90
D_{20}		—36.2	+594.4**	+559.3**
D_C	—83***	—89.3***	+39 9*	—49.5**

Mean values ± standard errors. Percentage differences versus C-20 and T-20 respectively,(D_{20}), and of T versus C (D_C). Statistically significant differences : *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$. Other explanations see in text.

Table 2

Leucine absorption (DPM) and partition coefficient (%) in the small intestine of the chickens

	20%	18%	16%	v.p
		DPM		
C	4.31±0.70	6.72±2.89	6.14±1.74	4.75±0.64
D_{20}		+55.9	+42.5	+10.2
T	0.63±0.07	0.15±0.06	3.63±0.48	6.08±0.84
D_{20}		—76.5**	+476.2**	+865.1***
D_C	—85.4***	—97.8*	—40.9	+28.0*
		%		
C	30.52±6.00	43.71±17.6	38.70±14.4	29.90±3.2
D_{20}		43.3	+26.8	—5.4
T	5.12±0.76	0.69±0.17	26.7±4.0	45.1±7.6
D_{20}		—86.5**	+421.9**	+780.3***
D_C	—83.2**	—98.4**	—31.0	+56.0

Explanations as in table 1.

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THE STUDY OF NORMAL HUMAN AND VARIOUS ANIMAL SPECIES SERUM AND PLASMA FIBRONECTIN

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Normal serum and plasma fibronectin concentrations from healthy blood donors and various laboratory animal species (rats, mice, rabbits) have been studied, by hemagglutination assay.
In human, the fibronectin level is significantly higher in plasma than in serum. Fibronection concentration is increased in 41—60 years age group in connection wiht age vascular changes. Higher fibronectin values in A II group, than in other blood groups, can be noticed.
In rats the fibronectin level is significantly higher than in other animal species studied.

Fibronectin (Fn) is a large molecular weight glycoprotein found in human body in insoluble form on cell surfaces, in association with basement membranes, and connective tissues, and in soluble form in plasma and other extracellular fluids (13, 15, 17, 18, 22).

Fn interacts with various macromolecules like collagen (especially gelatin), heparin (13), glycosaminoglycans, fibrinogen and fibrin (10, 13), actin (17), various complement functions (12), IgG (11), interfering in cell adhesion and in cell attachment on surfaces, in synthesis of extracellular matrix (8, 16), in opsonisation (19).

The biological importance of Fn is underlined by the fact that Fn was found in different genetic types of animals (fishes, birds, mammals) which proves its stability on phylogenetic scale, regarding the fundamental functions (6, 17).

Plasma Fn has an important role in reticuloendothelial system function, with a direct implication in phagocytic activity of debris and other bacteria and particles, as a nonspecific opsonin (14, 15, 17, 19, 20). Plasma level of Fn is changed in many diseases : septicemia (15), politrauma (7, 8), cancer (16, 23) and others, expressing its physiological function (1, 9, 15, 18, 23).

Many immunoreactive and functional techniques for plasma and serum Fn quantification have been developed (2, 4, 5, 10). In a prior study of our laboratory a hemagglutination assay for human serum Fn quantification has been described (3).

In this paper we analyse normal plasma and serum Fn levels, by hemagglutination assay, in human and various animal species, to use them as control values, the test being useful in clinical laboratory, as an indicator for disease gravity state.

MATERIAL AND METHOD

For plasma and serum Fn quantification a simple hemagglutination assay is used (3). The assay is based on the Fn stability to bind specifically to gelatin-coated red cells. Sheep red blood cells (SRBC) coated with gelatin (Serva, Heidelberg) by chromic chloride and purified human

plasma Fn, by chromatographic affinity on gelatin-Sepharose 4B (Pharmacia, Uppsala), were used (3, 6).

Sera to be tested were incubated at 56°C for 30 minutes, then adsorbed on SRBC at 37°C for 1 hour; 0.025 ml of serum were serially diluted in PBS in microtiter plates and incubated overnight at 4°C with equal volumes of 1% G-SRBC. In parallel, standard purified human plasma Fn 1 mg/ml in PBS was tested. The lowest agglutination concentration of serum and of purified Fn were compared to determine the concentration in serum (3).

Blood samples were collected, sterile by venopuncture, from 271 normal healthy blood donors (Bucharest Hematology Center); 206 for serum and 65 for plasma (on sodium citrate). Blood samples from three laboratory groups of 20 animals each: rats, mice, rabbits, were collected.

RESULTS AND DISCUSSION

The level of Fn is higher in plasma (mean ± ES) 348.0 ± 21.24 ($P < 0.0001$) as compared with serum 202.4 ± 6.6, both in men and in women (Fig. 1). Our results are in concordance with other published experimental data (21). The higher Fn level in plasma expresses its consumption in clotting blood process (21, 22).

There can not be inferred a significant differentiation in plasma and serum Fn between sexes. However, Fn level is higher in men than in

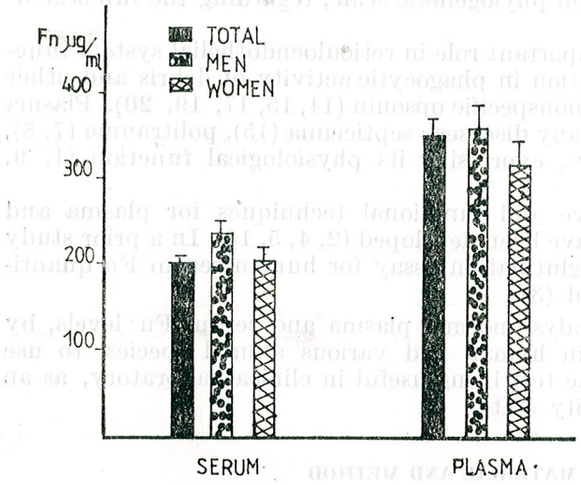

Fig. 1. — Serum and plasma Fn in men and women. Fn level is significantly ($P < 0.0001$) higher in plasma, both in men and in women. The values of Fn for all population and for two sexes are indicated as a mean ± SE.

women (Fig. 1). The same results were reported in other studies (21). The concentration of Fn is changed in various physiological periods of women's life. Stathakis N. E. et al. indicated that Fn level is significantly lower in preclimax women (21). In our study Fn level is higher in women aged between 51—60 years 250.0±34.6 ($P < 0.05$) than in yonger ones 179.7 ± 12.5, (Fig. 2).

The analysis of mean values of Fn, of all population ranged on age groups, indicates an increased Fn level in 51—60 years age group both in plasma and serum (Fig. 2 and 3). It is suggested that this increase is related to age vascular changes (18, 21, 22).

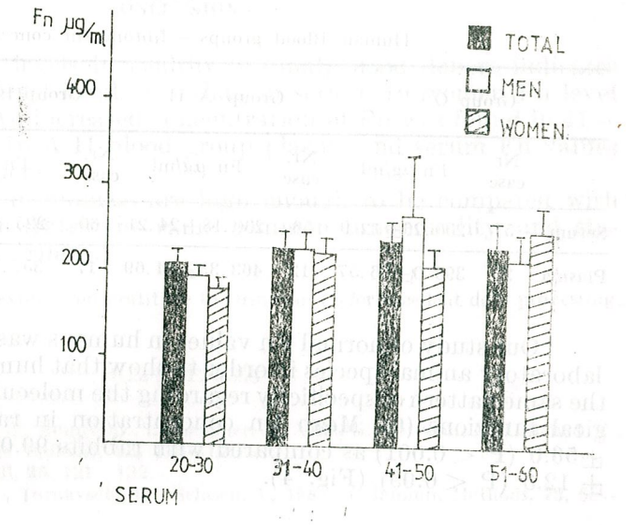

Fig. 2. — Fn concentrations in age and sex groups in serum. Mean value ± SE for total blood donors, men and women, ranged on age groups are indicated. Significant ($P < 0.05$) increased level of Fn, in women aged between 51—60 years as compared to yonger ones was observed.

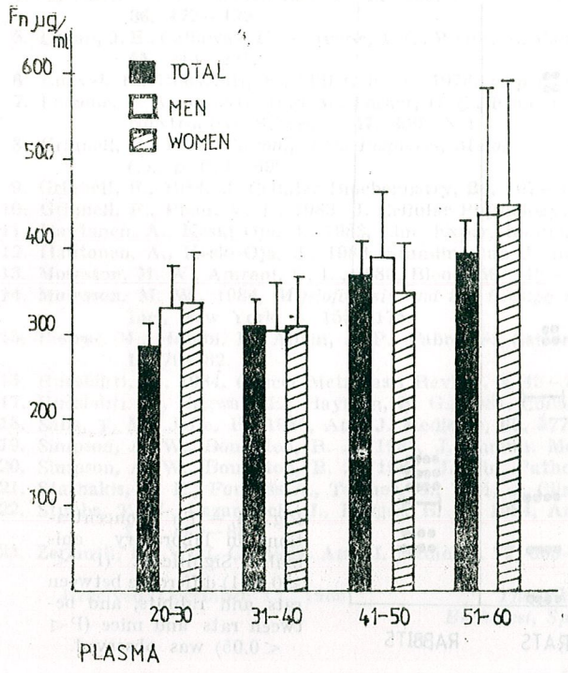

Fig. 3. — Fn concentrations in age and sex groups in plasma. Mean value ± SE for total population men and women ranged on age groups are presented, Fn level is significantly ($P < 0.05$) higher in population aged between 51—60 years.

Table I shows mean Fn concentrations in blood groups, in plasma and serum. Higher Fn levels can be seen in A II group, both in serum and plasma (230.2 ± 24.2 and 463.3 ± 64.7), than in other blood groups.

Table 1

Human Blood groups — fibronectin concentrations

	Group O I		Group A II		Group B III		Group AB IV	
	Nr. case	Fn μg/ml	Nr. case	Fn μg/ml	Nr. case	Fn μg/ml	Nr. case	Fn μg/ml
Serum	57	200.26±12.9	56	230.18±24.21	60	225.48±17.22	33	185.3±16.0
Plasma	25	395.0±36.57	12	463.33±64.69	17	357.73±52.14	10	375.0±41.7

Our study of normal Fn values in humans was extended to various the laboratory animal species in order to show that human and animal Fn have the same pattern of specificity regarding the molecular properties and biological functions (6). Mean Fn concentration in rats is increased 187.0±±56.0 ($P < 0.001$) as compared with rabbits 99.0 ± 10.0 and mice 39.5±± 12.3 ($P < 0.05$) (Fig. 4).

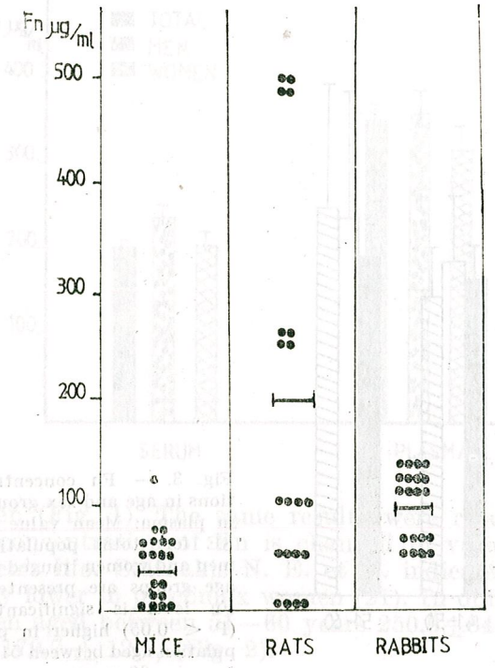

Fig. 4. — Fn concentrations in laboratory animals. Significant ($P < 0.001$) difference between rats and rabbits, and between rats and mice ($P < 0.05$) was observed.

Serum animals Fn was found in a high enough concentration to compete efficiently with human Fn, which proves an identical functionality in opsonisation, clotting and other major functions.

CONCLUSIONS

The study of Fn levels in healthy, normal, blood donors indicates a significantly higher level in plasma than in serum. In women Fn level is lower than in men. An increased concentration of Fn was found in 41—60 years age group. In A II blood group plasma and serum Fn values were found to be higher.

Serum Fn levels in animals are high enough to be compared with humans', which demonstrates the same common functionality and stability on phylogenetic scale.

Acknowledgement. We express our gratitude to Drinovan D. for excellent data processing.

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DIE STRUKTURDYNAMIK EINER POPULATION VON *DREISSENA POLYMORPHA* PALLAS UNTER DEN BEDINGUNGEN GESTEUERTEN WACHSTUMS IM STAUSEE „EISERNES TOR"

GH. BREZEANU, L. GRUIA und A. PETCU

The paper presents data on the dynamics in the structure of a population of *Dreissena polymorpha*, obtained after an experiment carried out in the dam lake Porţile de Fier, where the respective species developed abundantly after the Danube damming.

Der Bau des Wasserkraftwerks und der schiffahrtstechnischen Bauten an der unteren Donau im Gebiet des „Eisernen Tors" (Stromkilometer 944) durch Rumänien und Jugoslawien, so wie die Entstehung des Stausees hat zu schwerwiegenden Veränderungen in der Struktur der Biozönosen und der Populationen von in der Donau lebenden Organismen geführt, als auch anderer Gewässer-Ökosysteme (Flüsse, Seen, Quellen), die jetzt überflutet sind.

Als eine der Veränderungen, stellt sich die für das ganze Ökosystem äusserst wichtige massenhafte Vermehrung der Molluske *Dreissena polymorpha* Pallas dar. Während vor der Aufstauung diese Art ziemlich selten angetroffen wurde (2), ist sie in den benthonischen und epibiontischen Biozönosen dominant geworden (4). Auf verschiedenen Gegenständen im Wasser (Steine, Gehölze, Wasserbauten usw.) wurden bis zu 30 000 Ex./m² bzw. 2500 g/m² verzeichnet, was einer grösseren zahlenmässigen Dichte und Biomasse als allen anderen Organismengruppen gleichkommt.

Die ökologische Bedeutung der *Dreissena* ist um so grösser, als diese ein wichtiges Glied in der Nahrungskette und in der Stoff- und Energieüberführung der Wasser-Ökosysteme darstellt. Da *Dreissena* vornehmlich filtrierend wirksam ist, spielt sie bei der natürlichen Reinigung der Gewässer eine hervorragende Rolle. Diese beiden Aspekte genügen, um die bedeutsame Stellung der Muschel im biologischen Haushalt der Wasser-Ökosysteme zu unterstreichen.

Da *Dreissena* sich in verschiedenen Wasser-Ökosystem-Typen stark entwickelt, aber ganz besonders in solchen vom Typus des Eisernes Tor- - Stausees, haben wir daran gedacht diese Art auch auf andere Weise nutzbar zu machen. Aufgrund ihrer biochemischen* Merkmale schlossen wir, dass *Dreissena polymorpha* eine zur Fütterung von Tieren verwendbare Biomasse abgeben kann, was durch schon ausgeführte oder laufende Versuche auch bestätigt wurde. Es stellte sich uns also die Aufgabe eine Technologie zur gesteuerten Aufzucht und zur Nutzbarmachung der Molluske auszuarbeiten, was uns gelungen ist.

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* Hurghişiu Ileana, L. Gruia, Gh. Brezeanu, Date asupra compoziţiei chimice a moluştei *Dreissena polymorpha* Pallas, din lacul Porţile de Fier (im Druck).

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Unter den Versuchsbedingungen erhielten wir grosse Mengen von *Dreissena*, deren Produktion auf 30—40 Tonnen Biomasse/ha/Jahr zu schätzen ist. Im Zeitraum von 5 Monaten (Mai — September) entsteht eine überdichte Population (27.000—30.000 Ex./m²), die aus Tieren verschiedener Grösse und Gewichts bestehen (Tab. 1).

Zur Dynamik der Struktur dieser Population ist folgendes festzustellen: unter unseren Versuchsbedingungen wurden die ersten *Dreissena*-Exemplare, weniger als 1 mm gross, gegen Ende Mai beobachtet, wenn die Wassertemperatur 21—22°C erreichte. Ihre Zahl nahm schnell zu, so dass am 20. Juni 1.050—12.000 Ex./m² gezählt wurden (Tab. 1). Die Vermehrung führt zur Verdoppelung im Juli und fast zur Verdreifachung der Zahl im August. Die Kurve der Biomasse-Dichte hat einen ähnlichen Verlauf wie diejenige der numerischen Dichte, mit Werten, die von Juni bis August ansteigen (s. Tab. 1). Besonders interessant ist die Verteilung auf den Vertikalen. Die Muschelzahl ist bei 0—0,5 m Tiefe gering, erreicht ihren Höchstwert bei 1—3 m und sinkt dann wieder (Tab. 1) bis zu 16—18 m (grösste erkundete Tiefe).

Die Variation der Körpergrösse (Tab. 2) bringt die Entstehung, Entwicklung und Struktur der Population klar zum Ausdruck. Dieser Parameter wird hauptsächlich und insbesondere bei der durch gesteuerte Aufzucht erhaltenen Population bedingt durch die Fortpflazung und die Intensität der Laich-Ablage. Aus unseren Beobachtungen, wie auch anderer Autoren (1, 3) geht hervor, dass *Dreissena polymorpha* sich über eine lange Zeitspanne fortpflanzt; die Intensität der Laich-Ablage schwankt jedoch von einer Etappe zur anderen, in Abhängigkeit von zweierlei Faktoren: den hydrologischen und den chemisch-physikalischen (vor allem von der Wassertemperatur) einerseits und den biologisch-physiologischen (im Zusammenhang mit dem Vorhandensein, der Aufnahme- und Assimilierungsfähigkeit der Nahrung) andererseits. Die Faktoren erster Art bestimmen vorrangig, aber nicht ausschliesslich, die Auslösung, diejenigen zweiter Art die Intensität der Fortpflanzung. Aufgrund dieses Tatbestandes können wir grossenteils zum Verständnis der Struktur der Population in verschiedenen Etappen gelangen. Im Juni ist die Population einheitlicher und besteht aus sehr kleinen Individuen (um 1 mm Länge). Im Laufe der Zeit wird die Struktur mannigfaltiger, denn die Grössenunterschiede zwischen den Individuen treten immer klarer hervor. So zählte man z.B. im Juni 7 Grössenklassen in der Population, während in September und Oktober die Klassenzahl auf 17—18 anstieg (Tab. 2).

Von Wichtigkeit ist auch das Verhältnis der Grössengruppen in verschiedenen Etappen zueinander. Diese Tatsache erklärt die Strukturdynamik der Population, so wie die Dynamik der Laich-Ablage. Im Juli besteht die Population vorwiegend aus kleinen, etwa gleichgrossen Exemplaren; es handelt sich demnach um eine junge, nicht differenzierte Population, die aus einer reichlichen Laich-Ablage hervorging. Im August herrschen in der Population die grösseren Exemplare vor (9—11 mm), während der Anteil der kleineren viel geringer ist und diejenigen unter 1 mm völlig fehlen; dies bedeutet, dass in dieser Periode die Intensität der Fortpflanzung bzw. der Laich-Ablage viel schwächer ist. Im September und Oktober war die Zahl der kleinen Exemplare noch niedriger. Im

Oktober z.B. lag der Prozentsatz der Exemplare von bis zu 5 mm Länge unter 1 und die bis zu 2 mm Länge fehlten ganz, was bedeutet, dass vor diesem Datum die Fortpflanzung praktisch aufgehört hat.

Betrachtet man die Daten aus Tab. 2, so fällt es auf, dass die durch gesteuerte Aufzucht erhaltene Population in den anfänglichen Etappen (Mai, Juni, Juli) homogen ist, in den folgenden aber heterogener wird (Juli, August, September), was auf das differenzierte Wachstum der

Tabelle 1

Die Dynamik der zahlenmässigen Dichte und der Biomasse bei verschiedenen Tiefen, im Zeitraum Juni-August, auf 635 cm² Oberfläche

Tiefe m	numerische Dichte in Tausenden Ex/m²			Dichte der Biomasse g/m²		
	20.VI.	20.VII	20.VIII	20.VI.	20.VII	20.VIII.
0—0,5	1,05	1,75	2,25	19,95	244	598,5
0,5—1	1,75	8,50	14,00	23,75	826	3292
1— 3	12	23	27	228	3380	9850
3— 4	7	14	18	133	1640	3800
4— 5	2	3,5	5,5	38	388	700

Tabelle 2

Die Variation der Individuenzahl, auf Grösseklassen verteilt, zu verschiedenen Daten (in Prozent)

Grössenklasse mm	20.VII.	20.VIII.	20.IX.	20.X.
1	50,4	—	—	—
1,1— 2	23,5	6,4	4,5	—
2,1— 3	10,9	5,1	6,0	0,1
3,1— 4	4,2	3,1	3,9	0,15
4,1— 5	5,0	4,6	5,8	0,2
5,1— 6	4,2	5,5	7,4	2,2
6,1— 7	1,7	10,1	10,3	2,2
7,1— 8	—	11,6	10,5	2,7
8,1— 9	—	11,8	10,9	6,1
9,1—10	—	15,7	11,5	7,7
10,1—11	—	15,4	10,9	9,1
11,1—12	—	7,1	6,6	9,4
12,1—13	—	2,4	4,4	13,7
13,1—14	—	0,5	4,1	13,5
14,1—15	—	0,4	1,9	11,1
15,1—16	—	—	0,6	9,3

Individuen zurückgeht (langsamer zu Anfang und schneller im weiteren Verlauf der Beobachtungsperiode), als auch auf die unterschiedliche Fortpflazungsintensität (massiv in den Monaten Juli-August, um dann gegen den Herbst abzuklingen).

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HYDROCHEMISCHE UNTERSUCHUNGEN IM STAUSEE „EISERNES TOR" (IM GOLF DER CERNA) IM JAHRE 1983

ILEANA HURGHIŞIU

The chemical composition of water in bay Cerna is presented for 1983, in order to know the evolution of this ecosystem. The experimental data indicate a tendency of accumulation of biogenic components, namely nitrogen and organic phosphorus.

Der rumänische Donau-Abschnitt „Eisernes Tor" zeichnetesich vor der Aufstauung in physikalisch- chemischer Hinsicht durch einen Gleichgewichtszustand aus, da das Donau-wasser nicht durch Abwässer industriellen und haushaltlichen Ursprungs aus dem Oberlauf belastet war, und somit ein Musterbeispiel für den natürlichen Zustand des Stromwassers darstellte (1), (2,) (3).

Die von Ileana Hurghişiu und Dorina Nicolescu unternommenen Untersuchungen haben die Veränderung des physikalisch-chemischen Zustandes des Donauwassers in verschiedenen Abschnitten aufgezeigt im Zeitraum 1973—1974, also 4 Jahre nach der Aufstauung. (4).

Vorliegende Arbeit befasst sich im Rahmen einer Überwachung der hydrochemischen Lage mit den chemischen Kennwerten des Wassers im Cerna-Golf, nachdem 10 Jahre seit den letzten Untersuchungen und 13 Jahre seit der Aufstauung der Donau verstrichen sind.

MATERIAL UND METHODIK

Der Gesamtstickstoff wurde nach Kjeldahlisierung titrimetrisch bestimmt (1), (5), wahrend der Gesamtphosphor kolorimetrisch bestimmt wurde (7). Der anorganische Stickstoff und Phosphor wurde kolorimetrisch bestimmt. Die Differenz zwischen den gefundenen werten fur Stickstoff und Phosphor ergibt den organischen Anteil dieser Elemente.

ERGEBNISSE UND DISKUSSION

Von den *chemischen* Kenngrössen war das pH schwach alkalisch, mit mittleren Werten von 8,3 bis 8,8. Die Wasserstoffionen-Konzentration hängt ab von der Gesamtkonzentration des CO_2, in Gestalt des freien CO_2, von H_2CO_3, HCO_3^- oder CO_3^{--}.

Von den wesentlichen bigenen Komponenten weist der *Stickstoff*- als Gesamtstickstoff, organischer und mineralischer Stickstoff-kennzeichnende jahreszeitliche Schwankungen auf; der Gesamtstikstoff zeigt im Frühjahr einen Überschuss an, von Mai bis August sind die Werte hoch mit einer maximalen Konzentration von 8,2 mg/l; im August dagegen finden wir niedrige Werte von nur 3,8 mg/l. Die Kurve des *organis-*

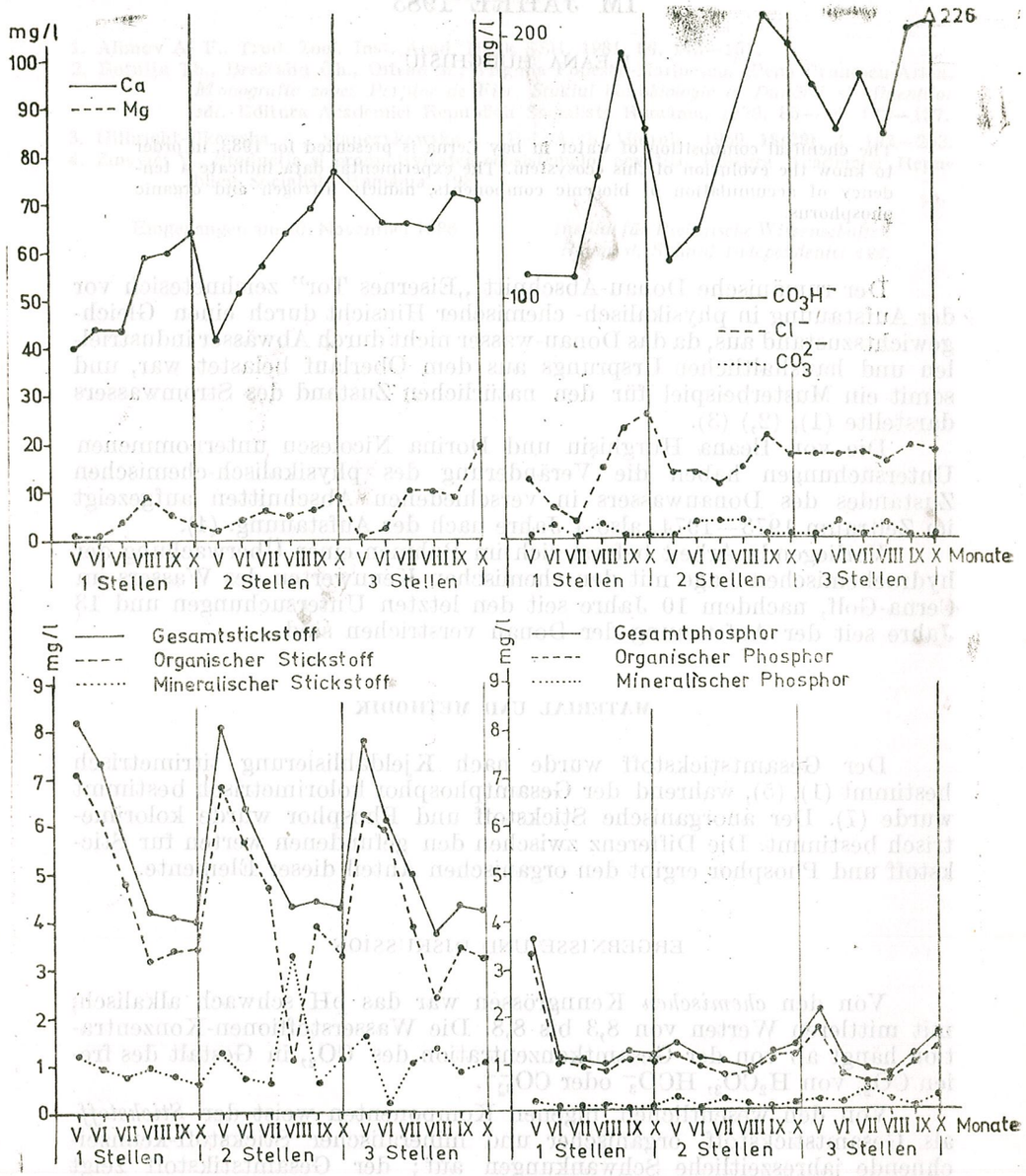

Abb. 1. — Die Dynamik des Stickstoffs, Phosphors, Calziums, Magnesiums, der Karbonate, Bikarbonate und Chloride aus dem Cerna-Golf aus dem Jahre 1983.
Standort 1 (des Probenentnahme) = Cerna, Einhillszone des Flusses in den see,
Standort 2 (des Probenthnahme) = Cerna, Golfende,
Standort 3 (des Probenthnahme) = Cerna, Zentralzone des Golfes.

chen Stickstoffs verläuft annähernd parallel mit der Variationskurve des Gesamtstickstoffs und besitzt Höchstwerte bis 7,0 mg/l, während im August einzelne Werte kaum 1,0 mg/l erreichen. Der *Mineralstickstoffgehalt* war im allgemeinen niedrig und wies Maxima von 1,6 mg/l auf. (Abb. 1).

Der *Phosphor*, wie auch der Stickstoff spielt im Ökosystem eine trophisch bedeutsame Rolle und ist für die Produktionsfähigkeit bezeichnend. Der Phosphorgehalt wurde als Gesamtphosphor, als organischer und als mineralischer Phosphor angegeben. Beim *Gesamtphosphor* bewegten sich im Mittel die Werte von 1,19 bis 1,54 mg/l, doch im Mai wurde ein Maximum von 3,6 mg/l verzeichnet. Es ist ein Vorherrschen des *organischen* gegenüber dem mineralischen Phosphor zu bemerken; die jahreszeitliche Variationskurve des organischen Phosphors hat einen ähnlichen Verlauf wie diejenige des Gesamtphosphors. Die Durchschnittswerte lagen bei 1,0—1,4 mg/l und das Maximum bei 3,4 mg/l im Mai. Quantitativ war der *mineralische* Phosphor mit 0,08—0,25 mg/l.

Die *Wasserhärte* betrug in Durchschnitt 7,4—11,4 Deutsche Härtegrade. Die zweiwertigen Ionen, und zwar das *Kalzium*, waren im Bereich der Cerna-Boje durch ein Maximum von 77 mg/l gekennzeichnet; an den Messtellen Cerna-Brücke und Einmündungsgebiet der Cerna waren die Werte-64-77 mg/l — vom Frühjahr bis zum Herbest hoch. Das *Magnesium* hingegen zeigte niedrige Werte und zwar 1—19 mg/l, wobei auch für dieses Element eine Anreicherung im Bereich der Cerna-Boje zu verzeichen war.

Die *Alcalinität* geht grösstenteils auf die hohen *Bikarbonatgehalte*- bis 226 mg/l im Bereich der Cerna-Boje — zurück. Die *Karbonate* ergaben niedrige Werte von höchstens 6 mg/l.

Die *Chloride* wiesen für Süsswasser charakteristische, niedrige Konzentrationen, mit maximal 46 mg/l auf; die Kurve der jahreszeitlichen Variationen hatte einen ähnlichen Verlauf vie bei den anderen Komponenten.

Bei einer kritischen Betrachtung der Ergebnisse ergibt sich, dass das pH innerhalb der für die III. Güteklasse gültigen Grenzwerte liegt; die Ammoniumkonzentration bleibt innerhalb der Werte für die I. Güteklasse. Die Kalzium- und Magnesiumgehalte waren gering und lagen unterhalb der zulässigen Grenzen der I. Güteklasse.

SCHLUSSFOLGERUNGEN

Der Stausse „Eisernes Tor" (im Golf der Cerna) wies im Jahre 1983 also 13 Jahre nach Aufstauung der Donau, eine normale jahreszeitliche Entwicklung der physikalischen und chemischen Kenngrösen auf.

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TRAIAN ORGHIDAN
1917—1985

C'est avec un profond regret que nous avons appris le décès inattendu du Professeur Tr. Orghidan, mort le 27 mai 1985 à Paris où il s'était rendu pour présenter son manuscrit sur les Phyllopodes que le Professeur Pierre P. Grassé lui avait demandé pour son monumental Traité de Zoologie.

Traian Orghidan est né le 27.II. 1917 à Onești-Plugari (dépt. de Iași) où sa famille, originaire de Brașov, avait trouvé refuge pendant la première guerre mondiale, dont la fin leur permit de réintégrer leur propre maison. C'est à Brașov, par conséquent que le jeune Orghidan commença ses études scolaires qu'il va finir à Bucarest, où il se fait inscrire en 1937 à la Faculté des Science de l'Université (section des sciences naturelles).

Pendant ses études universitaires il avait manifesté une réelle passion pour la recherche scientifique et, ensemble avec son collègue et ami, Nicolae Botnariuc (aujourd'hui professeur à l'Université de Bucarest et Membre Correspondant de l'Académie), il avait commencé l'étude des Phyllopodes. Ils en avaient découvert une nouvelle espèce (Imnadia Voitești) et établi une nouvelle famille (fam des Imnadiidae) dans un travail qui fut communiqué et publié aux éditions de l'Académie roumaine (1940) et leur valut un prix de l'Université de Bucarest.

Ayant passé sa licence en 1941, Traian Orghidan se fit engager en qualité d'Aide-Géologue (1942) à la Société Columbia-Petrole où il travailla sous la direction d'un grand géologue, le professeur Ioan Atanasiu, ce qui permit d'approfondir ses connaissances de géologie stratigraphique qui lui seront utiles quand il viendra à la Spéologie.

Après ce court « épisode géologique » dans sa carrière (1942—1943) il revient à la Zoologie, étant nommé assistant, puis chef de travaux à la station zoologique de Sinaia de l'Université de Bucarest où il a de nouveau la chance de travailler sous la direction d'un autre grand maître, le Professeur C. Motaș et d'y préparer son doctorat ès Sciences Naturelles qu'il passa à l'Université de Bucarest en 1949.

Nommé d'abord maître de Conférences de Zoologie à l'Université de Iassy (1950—1952), ensuite d'Hydrobiologie à l'Université de Bucarest (1951—1956), il travaille aussi au Collectif de Spéologie du Comité géologique de Bucarest. A la suite de la réorganisation du premier Institut de Spéologie fondé par Emile Racovitza, il en devient Directeur adjoint en 1956, inaugure les cours de Spéologie à la Faculté de Biologie de l'Université de Bucarest en 1957 et, enfin, est nommé Directeur de l'Institut de Spéologie de Bucarest en 1964 ainsi que Rédacteur en chef des Travaux de l'Institut de Spéologie « Emile G. Racovitza ». Dès lors l'une de ses préoccupations principales sera de le réorganiser en le modernisant et d'en dynamiser les recherches.

A cet effet il établit de nombreux rapports scientifiques avec des instituts de spéologie de l'étranger, il établit des conventions concernant des recherches en commun et organise des expéditions et des missions scientifiques en Bulgarie (1966), en Yougoslavie (1967), à Cuba (1970, 1973, 1975 dont il fait publier les résultats obtenus en 4 volumes) au Venezuela (1975, 1982) ainsi que la mission biospéologique Constantin Drăgan à Majorque (1970, 1971).

Ses propres recherches (souvent en collaboration) ont toujours donné des résultats importants et se réfèrent à de nombreux problèmes. Ceux qui concernent la taxonomie et d'autres aspects biologiques de certains invertébrés, dont les Phyllopodes, les Cladocères, les Hydracariens, les Palpigrades, les Pseudoscorpions occupent une place importante, tant par la

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découverte de nouveaux taxons que par d'autres aspects biologiques qu'il met en évidence (cycles de développement de certains arachnides, par exemple). Particulièrement interessante est aussi sa note préliminaire sur la première découverte des Pogonophores à Cuba dont il décrit un nouveau genre. Pour ce qui concerne les vértébrés, il s'est occupé (en collaboration avec Margareta Dumitrescu) de quelques Chiropteres découverts dans les grottes visitées.

L'étude des grottes représente un sujet qui l'attire par le caractère complexe des recherches et il participe à l'étude monographique de quelques-unes d'entre elles (celles de Limanu, Gura Dobrogei, de la Vallée de Fundata, de la région de Hunedoara, par exemple).

Enfin, on doit à Tr. Orghidan des contributions particulièrement importantes d'ordre écologique, concernant certains biotopes, dont il précise les caractéristiques différencielles, téls les biotopes hyporhéique, hygropétrique, celui des espaces lithoclasiques.

Il ne faut pas oublier de rappeler aussi les travaux publiés par Orghidan concernant l'évolution de la Spéologie, et le rôle des grands biologues à qui cette évolution est redevable, en commençant par le fondateur de cette science dans le monde et dans notre pays, Emile Racovitza, et en continuant par son successeur indirect, Constantin Motaş. Il revient aux biologues et spéologues qui les continuent aujourd'hui de situer sur la même lignée Traian Orghidan, à qui la situation actuelle de la Spéologie roumaine doit beaucoup.

D'ailleurs, les résultats importants de ses recherches, les collaborations internationales qu'il a réussi d'établir, les publications qu'il a dirigé, ses participations actives à de nombreuses manifestations scientifiques internationales, lui ont valu une large notoriété et de nombreuses distinctions nationales et internationales.

— Nombreux Ordres et médailles roumains et Prix « Emile Racovitza » de l'Académie de la R. S. de Roumanie (1979) ;
— Membre de l'Union internationale de Spéologie (1961) ;
— Membre Correspondant du Muséum d'Histoire Naturelle de Paris (1968) ;
— Membre du Comité de direction du Laboratoire souterrain de Moulis (1970) ;
— Membre Correspondant de la Société de Spéologie du Venezuela (1976) ;
— Membre de la Société de Biospéologie de France (1981).

Mais Traian Orghidan ne fut pas seulement un grand homme de science ; il fut aussi un homme de grande et large culture, aimant l'art et grand admirateur et ami de Georges Enesco, et aussi du sculpteur Gheorghe Anghel, fondateur de l'Association « Les amis d'Enesco » (Paris, 1970). Mais par dessus tout cela, il fut un homme de bien, un ami et collègue loyal, un maître bienveillant et juste.

Olga Necrasov

Ch. J. HUMPHRIES, L. R. PARENTI, *Cladistic Biogeography*, Oxford, Monographs on Biogeography, No. 2. Clarendon Press, Oxford, 1986 ; XII + 98 p., 67 figs.

A new biogeographical school, usually designed as "Vicariance Biogeography" emerged during the last 12 years. Its principles were exposed in less than ten short papers, published in "Systematic Zoology", 1974—1978 by Rosen, G. Nelson, Platnick and Nelson, Croizat, Nelson and Rosen, Ball. The only book containing an exposure of these principles is, according to the reviewer's knowledge, "Systematics and Biogeography : Cladistics and Vicariance" by Nelson and Platnick (Columbia University Press, 1981 : chapters 7 and 8) ; "Vicariance Biogeography : a Critique" (Nelson and Rosen eds., Columbia Univ. Press, 1981) is a sequence of articles by 12 authors, some proponents, others opponents to the school, the principles of which are not clearly exposed.

The book by Humphries and Parenti is the second one including a comprehensive exposure of the principles and methods of vicariance or cladistic biogeography and the first one devoted exclusively to the subject.

The book contains a short introduction and four main chapters. The first one (pp 1—20) reviews the development of historical or genetic biogeography, pointing out the difference between the centre-of-origin/dispersalist approach, that considers each lineage to have its own and distinct history, and Croizat's panbiogeographic approach, that groups similar distributions in "generalized tracks", giving the possibility to reconstruct the dispersal history not of single lineages, but of whole biota. A few older authors are mentioned, the ideas of whom are similar to those of Croizat, without however citing more important ones : von Ihering and Jeannel.

The second chapter (p. 21—52) deals with methodology, presenting Hennig's method of cladistic analysis and its application to biogeography (the "progression rule" and Brundin's work on the southern hemisphere chironomid midges), Rosen's 1975 (not 1976) "vicariance biogeography" and Platnick and Nelson's "cladistic" method. The authors make a distinction between these two methods, because of the titles of the two contributions and because Rosen's examples differ from Croizat's only in being based on rigorous monophyly, while the two co-operating authors use cladograms establishing the status of each subordinate taxon and consider that it is necessary to analyze at least three taxa/areas. The authors of the book present in the remaining part of the chapter their own methodology, actually a development of that worked out by Platnick and Nelson. By substituting areas to taxa in various cladograms, the authors try to determine the succession of "vicariant events" which resulted in the fragmentation of whole biota, not of single lineages. Being based mainly on Platnick and Nelson's method, Humphries and Parenti prefer the term "cladistic" to "vicariance" biogeography. The reviewer consider's that Rosen's 1975 contribution lies at the crossroads between Croizat's panbiogeography (that neglects analysis of relationships, monophyly etc.) and the rigorous vicariance biogeography of the very recent authors, including Humphries and Parenti. This school is commonly mentioned as vicariance biogeography, its proponents as vicarianists, while Hennig' and Brindin' cladistics is dispersalist. Dispersalist cladistics is not dead, has still numerous adherents above all among German zoologists (Zwick, Schmincke, Meier-Brook among those working on freshwater animals).

The two last chapters of the book ("The real world" and "A new view of the world") discuss various problems, some of them principal (significance of fossil record, of geological hypotheses, relative value of taxa, etc), others special (amphitropical distributions, biogeography of the southern end of the world, composite origin of South America and of Zealand, the expanding earth theory etc.).

Like most vicarianists, Humphries and Parenti do not deny the reality of dispersal : they simply consider that dispersal hypotheses can not be tested.

The reviewer does not accept many assumptions of vicariance biogeography ; he considers that dispersal played an important role, above all dispersal before the appearance or after the disappearance of barriers (most bariiers being short lasting). that dispersal hypotheses are compatible with continental drift etc. He welcomes, however, the publication of this book, an excellent and clear presentation of the principles of vicariance biogeography. A similar book presenting in a modern manner the principles of dispersalist biogeography (by accepting continental drift, using cladograms, giving a new interpretation to centres of origin etc.) is most desirable.

Petru Bănărescu

AVIS AUX AUTEURS

La « Revue roumaine de biologie — Série de biologie animale » publie des articles originaux d'un haut niveau scientifique de tous les domaines de la biologie animale : taxonomie, morphologie, physiologie, génétique, écologie etc. Les sommaires des revues sont complétés par d'autres rubriques, comme : 1. *La vie scientifique*, qui traite des manifestations scientifiques du domaine de la biologie : symposiums, conférences, etc. ; 2. *Comptes rendus* des livre de spécialité·

Les auteurs sont priés d'envoyer les articles, notes et comptes rendus dactylographié à double interligne (31 lignes par page) en deux exemplaires.

La bibliographie, les tableaux et l'explication des figures seront dactylographiés sur pages séparées et les diagrammes exécutés à l'encre de Chine noir sur papier calque.

Les tableaux et les illustrations seront numérotés avec des chiffres arabes.

La répétition des mêmes données dans le texte, les tableaux et dans les graphiques sera évitée. Les références bibliographiques citées par ordre alphabétique, comporteront le nom de l'auteur, l'initiale de prénom, l'année, le titre de la revue, abrégé conformément aux usances internationales, le tome, le numéro, la page.

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La responsabilité concernant le contenu des articles revient exclusivement aux auteurs.

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