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TAXONOMIC COMPOSITION, NUMERICAL DENSITY AND BIOMASS OF THE ZOOBENTHOS IN THE DAMLAKE IRON GATES II (ROMANIAN STRETCH), IN 2002

VIRGINIA POPESCU-MARINESCU

In the paper, the composition, variability and frequency of taxa, as well as the numerical density and biomass on taxonomic groups, and the spatial and seasonal variability of total numerical density and biomass of the zoobenthos organisms in the damlake Iron Gates II, in 2002 are presented. The analysis of data in the tables and figures shows that: a) *Hypania invalida*, *Limnodrilus hoffmeisteri*, *Tubifex tubifex* and *Sphaerium* sp. were the constant-dominant taxa; b) the taxonomic groups, dominant as numerical density, were polychaetes and oligochaetes, while bivalves were dominant as biomass; c) generally, the highest values of the numerical density and biomass were found during summer, usually in the middle navigable zone; d) significant spatial variations were evidenced both in summer and autumn, in 2002.

1. INTRODUCTION

The strong morphologic, hydrologic, chemical and biologic changes which occur in newly created aquatic ecosystems, such as the damlakes from Iron Gates, impose complex and detailed studies, inclusively at the level of all links of the trophic chain.

Our researches on the zoobenthos in the accumulation Iron Gates II, carried out in 2002, within a contract concluded with the "Branch of Iron Gates Hydro-power stations", cover this domain. The names and location of the profiles and stations for biological sample collection are presented in Table 1. We mention that no thorough and consistent researches on the zoobenthos organisms in the said ecosystem were carried out before 2002. Our researches were performed on material of 6 transversal profiles situated along both the accumulation and around 80 km of the Danube. Among all obtained results, we present in this paper the zoobenthos organism state in summer and autumn 2002, in damlake Iron Gates II, under various aspects.

To remark that the 2002 year, from hydrologic point of view, was an atypical one, because the high waters were present also in autumn, which influenced the benthos fauna development.

The few previous studies on the respective links of the trophic chain in the Danube, between 932 and 866 rkm, provided some reference data (4, 5, 8-10, 11).

Table 1

Profiles and stations of zoobenthos sample collection, in the damlake Iron Gates II, in 2002

Profile name	Danube (km)	Stations	Water depth (m)		Station code
			June	September	
AVANPORT ECLUZA P.F.I.	942.5	Left riparian	9.00	6.56	1
		Middle navigable zone	9.00	7.00	2
TOPOLNIȚA	928.0	Middle navigable zone	11.00	12.00	3
OSTROVU CORBULUI	911.0	Left riparian	1.45	3.50	4
		Middle navigable zone	9.80	15.46	5
CRIVINA	894.0	Left riparian	2.60	4.30	6
		Middle navigable zone	13.00	13.00	7
ȚIGĂNAȘ	878.0	Left riparian	8.30	8.00	8
		Middle navigable zone	18.00	18.00	9
OSTROVUL MARE	866.0	Left riparian	8.70	6.40	10
		Middle navigable zone	15.87	14.75	11

2. MATERIALS AND METHODS

The benthos sample collection, both on longitudinal and transversal profiles, was performed using a quantitative method. The Bodengreiffer device, with a surface of 400 cm², was dipped in different stations, on the muddy, sandy, stony or mosaic substratum. After the biologic material selection, the obtained data were related to the 1 m² surface of benthos facies.

Together with the sample collection we performed field observations and measurements regarding the facies nature, water depth, water flow rate, pH and temperature, as these environment factors had a significant role in survival and development of benthos organisms.

3. RESULTS AND DISCUSSION

Both the **spatial distribution** and **taxon frequency** in benthos fauna of the damlake Iron Gates II, in 2002 (Table 2) were dominated by a small number of genera and species, namely the pelophilic organisms *Hypania invalida*, *Limnodrilus hoffmeisteri*, *Tubifex tubifex*, *Sphaerium* sp. These forms exceeded 50% frequency, having the highest numerical density, favored by the large extension of the muddy facies. Otherwise, the said four taxa, occupied, from the same point of view, the first places in the damlake Iron Gates I, too, both in 2002 and 1995–1996 period (9). We also mention that in the 1995–1996 period, more than 10 years from the damlake Iron Gates II formation, in the zone between 931 and 898 rkm, the two mentioned oligochaetes had a wide development followed by the polychaet *Hypania invalida* (4).

Table 2

Taxonomic composition, spatial distribution and frequency of zoobenthos organisms, in the damlake Iron Gates II, in 2002

Taxonomic unit name	Collection month: J = June; S = September Station code: 1–11 (Table 1)		Frequency %
1	2		3
COELENTERATA			
<i>Cordylophora caspia</i>	J: –	S: 3; 7	9
<i>Hydra viridis</i>	J: 3; 8	S: 5	14
TURBELLARIA			
<i>Otoplana antipai</i>	J: 7; 9	S: –	9
<i>Palaeodendrocoelum romanodanubialis</i>	J: 4; 7; 9; 11	S: 5; 9; 11	32
<i>Planaria torva</i>	J: 5	S: –	5
OLIGOCHAETA^{*)}			
<i>Branchiura sowerbyi</i>	J: 11	S: 2; 10; 11	18
<i>Limnodrilus hoffmeisteri</i>	J: 1; 2; 4; 6; 7; 11	S: 1; 2; 4; 6; 9–11	55
<i>Tubifex tubifex</i>	J: 1; 2; 4; 6; 7; 11	S: 1; 2; 4; 6; 9–11	55
POLYCHAETA			
<i>Hypania invalida</i>	J: 1–5; 7–9; 11	S: 5; 7–9; 11	63
<i>Manayunkia caspica</i>	J: 3; 5; 8	S: 3; 5; 7; 11	32
LAMELLIBRANCHIA			
<i>Dreissena polymorpha</i>	J: 3; 5; 8	S: 1; 3–5; 7; 8; 10	45
<i>Sphaerium</i> sp.	J: 3; 5; 7–9; 11	S: 3–5; 7; 8; 11	55
<i>Unio tumidus</i>	J: 7–9	S: 4; 8	23
GASTROPODA			
<i>Bithynia tentaculata^{**)}</i>	J: 7–9	S: 6; 7; 10	
<i>Esperiana esperi^{**)}</i>	J: 4; 5	S: 4; 8; 10	
<i>Lithoglyphus naticoides^{**)}</i>	J: 2; 4; 5; 8; 11	S: 1; 2; 4; 6; 8; 10	
<i>Theodoxus danubialis</i>	J: 3; 5; 8; 9	S: 4–6; 8; 9; 11	45
<i>Theodoxus fluviatilis</i>	J: 4	S: 4; 5; 9; 11	23
<i>Theodoxus palasi^{**)}</i>	J: –	S: 11	
<i>Viviparus acerosus^{**)}</i>	J: –	S: 11	
AMPHIPODA			
<i>Corophium curvispinum</i>	J: 3–5; 8; 9	S: 3; 5; 7	36
<i>Chaetogammarus tenelleus behningi</i>	J: 4; 5; 7–9; 11	S: 5; 8	36
<i>Dikerogammarus haemobaphes fluviatilis</i>	J: 3–5; 8; 9	S: 9; 11	32
<i>Dikerogammarus villosus</i>	J: 5	S: –	5
<i>Dikerogammarus villosus bispinosus</i>	J: 5; 7	S: 5	14
<i>Obesogammarus obesus</i>	J: 3; 4; 8	S: –	14
<i>Pontogammarus sarsi</i>	J: 4	S: –	5
ISOPODA			
<i>Jaera istri</i>	J: 3–5; 7–9	S: 3; 5; 7; 9; 11	50

^{*)} Oligochaetes were not completely identified.

^{**)} The gastropods, denoted by ^{**)} in this table, were found only as empty shells, in the respective stations.

To remark that the lithorheophilic organisms *Jaera istri* (frequency 50%), *Dreissena polymorpha* (frequency 45%) and *Theodoxus danubialis* (frequency 45%) usually found on mosaic substratum were spread almost along the entire second accumulation of Romanian river stretch, in 2002.

Regarding the spatial distribution of zoobenthos organisms in damlake Iron Gates II, in 2002, we observed the presence of the Ponto-Caspian elements on large areas (1-3, 7, 10). Among these, the main places were occupied by the already mentioned *Hypania invalida*, *Dreissena polymorpha*, *Jaera istri*, as well as by *Corophium curvispinum*, *Manayunkia caspica* and *Palaeodendrocoelum romanodanubialis*, followed by *Chaetogammarus tenellus behningi* and *Dikergammarus haemobaphes fluviatilis*. *Cordylophora caspia*, *Otoplana antipai*, *Dikergammarus villosus*, *D. v. bispinosus*, *Obesogammarus obesus*, *Pontogammarus sarsi* were found with a low frequency.

By diversity point of view, the greatest number of taxa was found at rkm 911, rkm 894 and rkm 878 in benthos fauna of damlake Iron Gates II, in 2002 (Table 2). In that place, we remarked the agglomeration of many Ponto-Caspian elements (as was observed in the same period of time, in the upper part of the damlake Iron Gates I) (7). As regards the taxonomic groups, the amphipods occupied the first place, by form diversity point of view, within it the gamarids dominated. On the contrary, scarce gastropod taxa were recorded (the main part of them were found as empty shells) (Table 2). We emphasize that the gastropods were well represented, as species number, in fauna of local submerged vegetation (6).

A comparison between the literature taxonomic diversity of benthos fauna in damlake Iron Gates II (4, 5, 9-11) and our data in Table 2 (considering that the oligochaetes were identified to a small extent, and chironomids, not at all) supports that the diversity presented in the table should be higher in 2002. But, we must also consider that, before 2002, the researches in that zone were carried out on few profiles and stations (4, 9). The higher form diversity, in 2002, was caused also by the abundant macrophyte vegetation in the respective ecosystem, with a rich phytofauna. The last one reaches the benthos especially after the plant fall on the damlake bottom.

The numerical density of some taxa, as well as of zoobenthos organism groups (presented in Table 3), in damlake Iron Gates II, in 2002.

The polychaet *Manayunkia caspica* had the highest number of individuals/m² (maximum value of 24,125 ind/m²) on a sandy-stony facies, water depth of 11 m. The polychaet *Hypania invalida* had the maximum value (of 20,375 ind/m²) on a muddy facies, water depth of 13 m. The crustaceans *Jaera istri* (maximum value of 4,250 ind/m²) and *Corophium curvispinum* (maximum value of 3,250 ind/m²) follow, the both species were identified on sandy-stony facies at depth of 11 m. All maximum values reached by these taxa were located in the middle navigable zone, in summer.

Table 3

Numerical density (ind/m²) of zoobenthos organisms on the taxonomic groups, in the damlake Iron Gates II, in June/September, 2002

Profiles and stations	Taxonomic group					
	Coelenterata	Turbellaria	Nematoda	Polychaeta	Oligochaeta	Cocoons Olig.
En avant Lock P.F.I						
Left riparian	—	—	2,550	25	2,375	25
Middle	—	—	25	—	2,025	1,625
navigable zone	—	—	10,300	25	1,125	50
	—	—	250	—	3,500	1,925
TOPOLNIȚA						
Middle	1,425	—	50	33,375	2,125	—
navigable zone	3,612	—	—	600	75	—
OSTROVU CORBULUI						
Left riparian	—	—	—	25	7,900	50
Middle	—	—	—	—	2,250	—
navigable zone	50	25	12	2,212	350	—
			—	4,575	450	100
CRIVINA						
Left riparian	—	—	—	—	6,550	—
Middle	—	—	200	—	2,575	1,075
navigable zone	75	450	350	20,375	9,900	800
		50	—	3,600	6,625	—
ȚIGĂNAȘ						
Left riparian	488	—	38	1,237	688	12
Middle	—	—	—	275	8,175	300
navigable zone	—	250	50	14,400	9,000	625
	—	25	100	4,100	11,825	—
OSTROVUL MARE						
Left riparian	—	—	—	1,525	3,400	—
Middle	—	—	—	—	1,275	150
navigable zone	—	3,325	25	875	79,075	1,650
	—	3,800	—	1,775	9,000	1,250

Table 3 (continued)

Profiles and stations	Taxonomic group					
	Lamelli-branchia	Gastropoda	Isopoda	Gamma-ridae	Corophi-idae	Diptera
En avant Lock P.F.I						275
Left riparian	25	-	-	-	-	25
Middle navigable zone	-	-	-	-	-	175
TOPOLNIȚA						
Middle navigable zone	1,100 238	25 -	4,250 163	650 -	3,250 1,487	- -
OSTROVU CORBULUI						
Left riparian	25	200	25	150	75	7,300
Middle navigable zone	132 450	25 75	1,125 625	163 3,575	200 725	12 50
CRIVINA						
Left riparian	-	-	-	-	-	4,050 75
Middle navigable zone	175 475	-	450 375	175 -	- 550	325 825
ȚIGĂNAȘ						
Left riparian	87 50	25 25	700 -	313 125	113 -	163 275
Middle navigable zone	2,175 -	- 125	200 50	375 250	50 -	50 -
OSTROVUL MARE						
Left riparian	25 -	-	-	-	-	50
Middle navigable zone	550 2,475	- 100	- 150	850 75	-	6,650 500

As regards the numerical density of different taxonomic groups of zoobenthos organisms (Table 3) the oligochaetes occupied the first place (maximum value of 79,075 ind/m²) followed by polychaetes (max. 33,375 ind/m²) (Table 2). The groups with a density of few thousands ind/m², but with a high frequency are: dipters, isopods, lamellibranchiate and gammarids. But, some of the mentioned taxonomic groups presented also minimum values among the benthos organisms in the damlake (Table 3).

The spatial and seasonal fluctuations of total numerical density of the zoobenthos organisms on longitudinal and transversal profiles in accumulation Iron Gates II, in 2002, are illustrated by the data of figures 1–3.

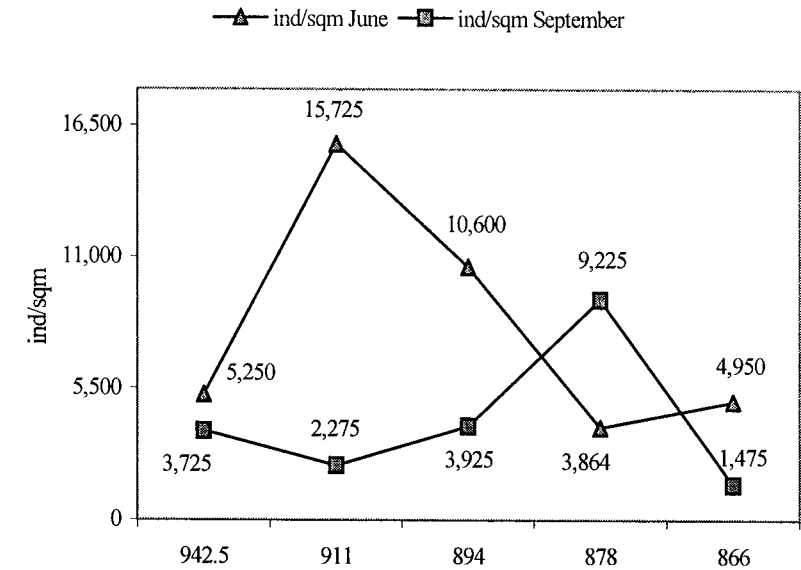


Fig. 1. – Spatial and seasonal variations of the total numerical density (ind/sqm) of the zoobenthos organisms, from longitudinal profile, in the left riparian zone of the damlake Iron Gates II, in 2002.

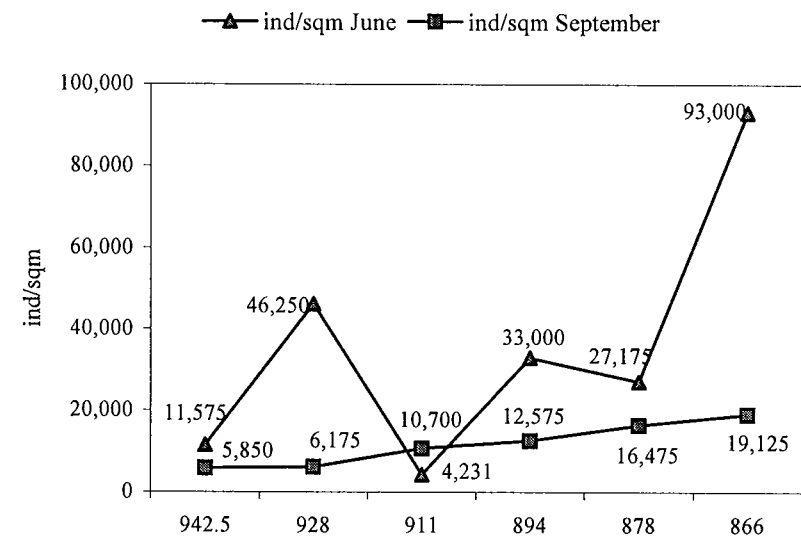


Fig. 2. – Spatial and seasonal variations of the total numerical density (ind/sqm) of the zoobenthos organisms, from longitudinal profile, in the middle navigable zone of the damlake Iron Gates II, in 2002.

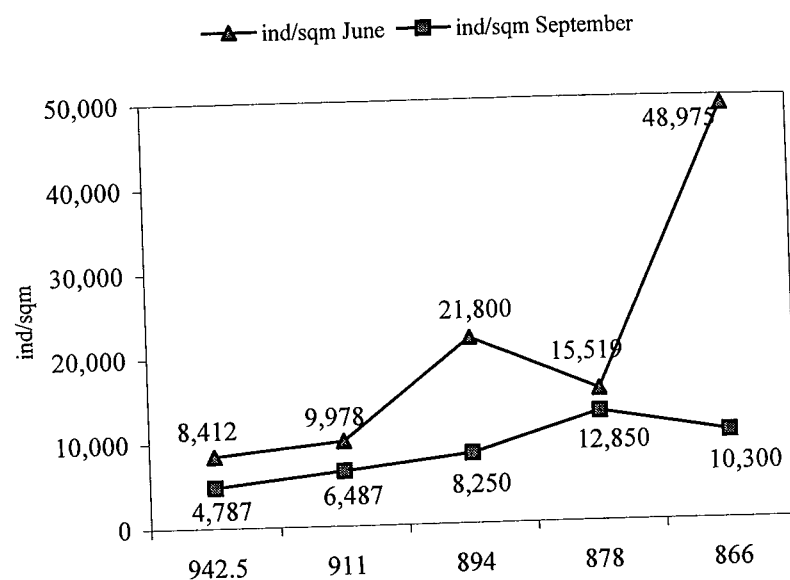


Fig. 3. – Spatial and seasonal variations of the total numerical density (ind/sqm) of the zoobenthos organisms, from transversal profiles, of the damlake Iron Gates II, in 2002.

The fluctuations, on the longitudinal profile, of total zoobenthos in the various stations of the left riparian zone (Romanian), in summer and autumn, are presented in figure 1. The two curves were divergent on their major part of direction. The maximum value in summer, at water depth of 1.45 m, on predominant muddy facies, belonged mainly to the oligochaetes and chironomids. In autumn, at a water depth of 8 m, on the sandy-muddy substratum, the oligochaetes alone amounted to 88.38% of zoocenosis components, and the values were generally lower in autumn.

The averages of the total numerical density of zoobenthos organisms on the longitudinal profile in the left riparian zone were 8,078 ind/m², in summer and 4,125 ind/m² (almost a half of the summer figure) in autumn.

The curves in figure 2 showing the total number of zoobenthos organisms, also on the longitudinal profile, but in the stations of the middle navigable zone, had different directions. The summer one, sinuous, included high values, the maximum was owed to the numerous oligochaetes dominant on sandy facies, at water depth of 16 m. The minimum value was observed on a stony-sandy facies, at water depth of 9.8 m, where the principal role belonged to the polychaetes (*Manayunkia caspica*) and isopods. The curve of the autumn values, much lower against summer ones, was ascendant upstream towards downstream.

The average of the total numerical density of zoobenthos organisms on the longitudinal profile in middle navigable zone of the damlake was much higher, 35,872 ind/m², in summer, against 11,817 ind/m², in autumn.

On the transversal profiles of Iron Gates II, the fluctuation of the total numerical density of zoobenthos organisms is presented in figure 3. The two curves had, generally an ascendant direction upstream towards downstream. The summer curve showed higher values than autumn ones, its peak being strongly marked.

Table 4

Biomass (mg/m²) of the zoobenthos organisms on the taxonomic groups, in the damlake Iron Gates II, in June/September, 2002

Profiles and stations	Taxonomic group					
	Coelenterata	Turbellaria	Nematoda	Polychaeta	Oligochaeta	Cocoons Olig.
En avant Lock P.F.I	–	–	–	–	–	–
Left riparian	–	–	3.06	25.00	733.00	12.50
Middle navigable zone	–	–	0.04	–	1,275.75	715.00
	–	–	12.37	12.50	–	55.00
	–	–	0.50	–	–	500.00
TOPOLNIȚA	–	–	–	–	–	–
Middle navigable zone	57.00	–	0.10	12,592.00	1,300.00	–
	36.13	–	–	6.25	0.75	–
OSTROVU CORBULUI	–	–	–	–	–	–
Left riparian	–	–	–	0.25	481.75	5.00
Middle navigable zone	–	–	–	–	531.00	–
	–	–	0.02	582.50	213.38	–
	2.00	0.50	–	125.00	1,250.00	30.00
CRIVINA	–	–	–	–	–	–
Left riparian	–	–	–	–	4,980.00	–
Middle navigable zone	–	–	0.34	–	2,079.75	317.60
	–	–	–	–	–	–
	–	4.50	2.45	6,920.00	155.75	60.00
	1.75	1.50	–	1,881.75	1,325.00	–
ȚIGĂNAȘ	–	–	–	–	–	–
Left riparian	19.50	–	0.06	386.25	70.00	1.25
Middle navigable zone	–	–	–	110.00	2,452.50	60.00
	–	–	–	–	–	–
	–	2.75	0.08	30,240.00	2,880.00	63.50
	–	1.50	0.25	5,387.50	1,875.00	–
OSTROVUL MARE	–	–	–	–	–	–
Left riparian	–	–	–	1,175.00	1,580.00	–
Middle navigable zone	–	–	–	–	930.75	60.00
	–	–	–	–	–	–
	–	33.25	0.04	750.00	4,325.00	230.00
	–	112.50	–	125.75	875.00	162.50

Table 4 (continued)

Profiles and stations	Taxonomic group					
	Lamelli-branchia	Gastropoda	Isopoda	Gammaridae	Corophiidae	Diptera
En avant Lock P.F.I						
Left riparian	— 37.50	— —	— —	— —	— —	82.50 12.50
Middle navigable zone	— —	— —	— —	— —	— —	51.25 325.00
TOPOLNIȚA Middle navigable zone	1,670,000.00 6,050.00	1,625.00 —	170.00 6.50	975.00 —	1,550.00 892.50	— —
OSTROVU CORBULUI Left riparian	— 460,325.00	22,590.00 —	1.00 —	105.00 —	33.00 —	3,705.00 —
Middle navigable zone	52,500.00 10,075.50	919.75 225.00	22.90 2.00	1,043.13 5,000.00	68.75 210.00	1.25 17.50
CRIVINA Left riparian	— —	— —	— —	— —	— —	5,900.00 25.00
Middle navigable zone	460,000.00 483,000.00	— —	18.00 15.00	2,625.00 —	— 825.00	3.25 100.00
ȚIGĂNAȘ Left riparian	205,062.50 262,650.00	1,875.00 1,169.75	42.00 —	360.00 37.50	33.75 —	16.25 132.50
Middle navigable zone	1,940,000.00 —	— 6,250.00	8.50 2.00	1,125.00 1,000.00	35.00 —	15.00 —
OSTROVUL MARE Left riparian	850.00 —	— —	— —	— —	— —	— 12.50
Middle navigable zone	428,500.00 3,002.50	— 2,562.50	— 6.00	1,346.00 225.00	— —	66.50 120.00

The summer average of the numerical density of the zoobenthos organisms on the transversal profiles of damlake 20,937 ind/m² was about 3 times greater than autumn average 8,535 ind/m².

The numerical density of the zoobenthos organisms had higher values in summer than those in autumn, in major part of stations. This fact was mainly due to the prolonged water flooding towards autumn. This hydrologic condition caused the washing and drifting of the mobile substratum, disturbing the development of the benthos fauna.

The biomass of zoobenthos organism groups (Table 4) and of some taxa in damlake Iron Gates II, in 2002, reached high values, comparable to those obtained in both Danube accumulations (in Romanian stretch) in 1995–1996 period (9, 10). They were much higher than 1986 data in zone between rkm 931 and 898 (damlake Iron Gates II) (4). In the whole 1986–2002 period, the mollusks (the figures include the shell weight) dominated as biomass. In 2002 the biomass maximum value, due to lamellibranchiate (1,940,000 mg/m² of which *Sphaerium* sole had 1,690,000 mg/m²) was registered in middle navigable zone, in summer. The polychaetes, with figures of about 60–70 times lower, were represented by *Hypania invalida* (maximum value of 30,240 mg/m²) also in the middle navigable zone, in summer.

The gastropods followed (maximum value of 22,590 mg/m²) with the dominance of *Theodoxus fluviatilis* (22,500 mg/m²), present on left riparian. The gammarids and oligochaetes reached close biomass values of only few thousands mg/m², in different damlake zones.

The spatial and seasonal fluctuations of total biomass of zoobenthos organisms on longitudinal and transversal profiles in damlake Iron Gates II, in 2002, are presented in figures 4–6.

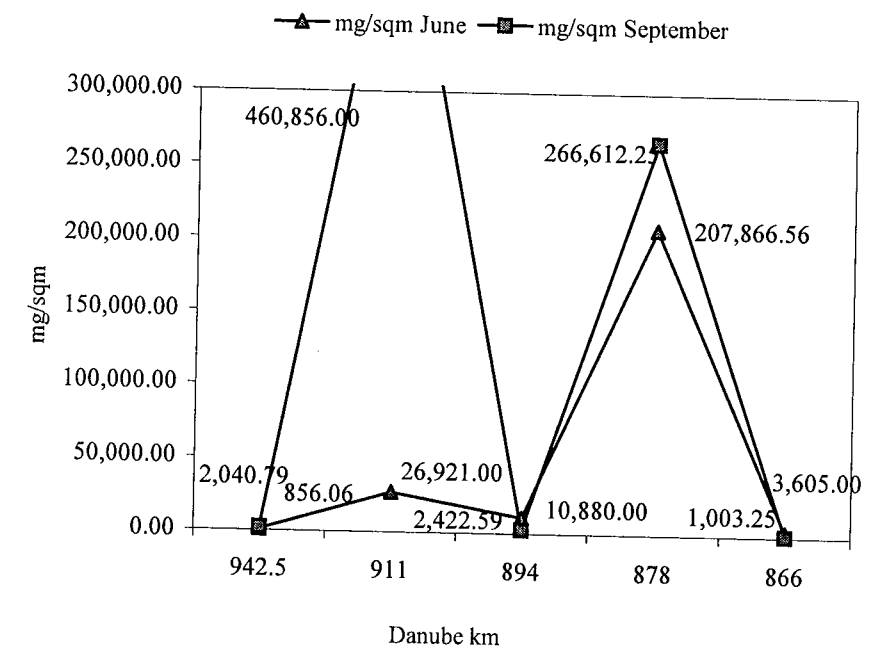


Fig. 4. – Spatial and seasonal variations of the total biomass (mg/sqm) of the zoobenthos organisms, from longitudinal profiles, in the left riparian zone of the damlake Iron Gates II, in 2002 (for the ▲ the value is placed on the right; for the ■ the value is placed on the left).

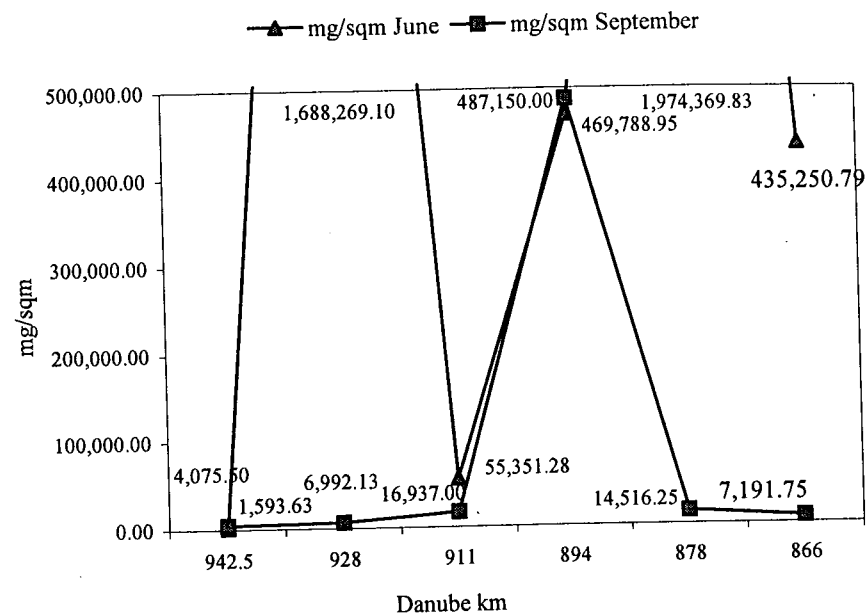


Fig. 5. – Spatial and seasonal variations of the total biomass (mg/sqm) of the zoobenthos organisms, from longitudinal profile, in the middle navigable zone of the damlake Iron Gates II, in 2002 (for the ▲ the value is placed on the right; for the ■ the value is placed on the left).

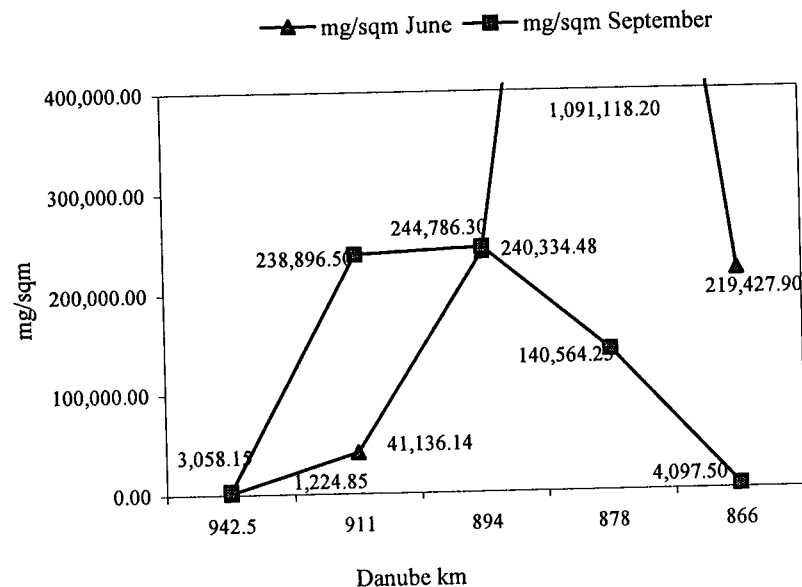


Fig. 6. – Spatial and seasonal variations of the total biomass (mg/sqm) of the zoobenthos organisms, from transversal profiles, of the damlake Iron Gates II, in 2002 (for the ▲ the value is placed on the right; for the ■ the value is placed on the left).

The curves of the fluctuations of zoobenthos organism biomass, along the damlake left riparian (fig. 4), showed one peak of development in summer and two peaks in autumn. The peaks are much far from the minimum values of both summer and autumn.

The seasonal averages of total biomass of zoobenthos organisms in stations along the damlake riparian were of 50,025.72 mg/m² in summer and of 146,586.98 mg/m² in autumn.

The biomass values of riparian zone, higher in autumn than in summer, although in atypical hydrologic conditions, may be explained, on the one side, through the development and weight increase of individuals, especially of mollusks, and, on the other side, by low water flow.

The fluctuation of zoobenthos organism biomass (fig. 5) along the middle navigable zone of damlake was opposed to that in riparian zone, namely: two development peaks in summer and one peak in autumn. The summer peaks were 3 times higher than autumn peaks. The difference between the maximum and minimum values was especially large.

The biomass averages of the 6 stations in middle navigable zone were of 770,770.60 mg/m² in summer and of 89,477.11 mg/m² in autumn.

On the transversal profiles, the fluctuation of total biomass of zoobenthos organisms in damlake Iron Gates II showed ascendant curves, both in summer and autumn (from rkm 942.5 until rkm 879 in summer and until rkm 894 in autumn). In both seasons there was a sharp discrepancy between the maximum and minimum values of the zoobenthos organism weights (fig. 6).

The averages of zoobenthos organism biomass on the damlake and on the transversal profiles were higher in summer 318,648.31 mg/m² than in autumn 126,280.54 mg/m².

As regards the biomass of benthos fauna organisms in the middle navigable zone, also on transversal profiles, the autumn lower values were determined by higher waters long prolonged in 2002. These generated the washing and drifting of the mobile substratum, together with that of zoobenthos organisms.

4. CONCLUSIONS

The results of the studies carried out in 2002, on the zoobenthos organisms in accumulation damlake Iron Gates II, allow us to conclude.

1. The taxonomic diversity in 2002 was relatively higher than that before the river blocking.
2. The constant-dominant taxa were *Hypania invalida*, *Limnodrilus hoffmeisteri*, *Tubifex tubifex*, *Sphaerium sp.*
3. The taxonomic groups dominant as numerical density were polychaetes and oligochaetes, and as biomass lamellibranchiate.

4. The highest values of the numerical density and biomass were registered in summer.
5. The spatial distribution of zoobenthos organisms changes, generally, on middle navigable zone registered the highest values of both numerical density and biomass.

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THE ZOOPLANKTON STRUCTURE IN THE IRON GATES II RESERVOIR (DANUBE km 863–942), IN 2002

VICTOR ZINEVICI, LAURA PARPALĂ

The dam Lake Iron Gates II reservoir represents a young anthropic ecosystem, having less than two decades of existence. It includes within its perimeter almost 80 km of lower Danube section. It is located far downstream of the Lake Iron Gates I, from which differ by its size and slope clearly reduced. Its shoreline is more uniform, while its affluent network is poor represented. The river, implicitly the lake, shows an asymmetric transversal profile; it has higher depths along the right bank and low deep zones, populated with submerged and emerged macrophytes along its left bank. The maximum depth is of 18 m, and the water flow rate varies around 1 m/s. In downstream extremity, just against Drobeta-Turnu Severin town there exists a significant source of domestic and industrial wastewater pollution. The analyses evidenced that the zooplankton was characterized by low values of species diversity ($\Sigma = 70$ taxa), numerical density ($X_a = 24.5$ ex/l) and biomass ($X_a = 51.2$ μg wet weight/l). The species with the most significant role in the dynamics of the zooplankton ecological equilibrium was *Dreissena polymorpha*, in the plankton larva stage, having the highest values of constancy, numerical and gravimetric dominance.

Key words: dam lakes, zooplankton, species diversity, numerical density, biomass.

1. INTRODUCTION

The dam Lake Iron Gates II represents a young anthropic ecosystem, having less than two decades of existence. It includes within its perimeter almost 80 km of lower Danube section. It is located beyond the downstream extremity of the Lake Iron Gates I, from which differ by its size and slope clearly reduced. Its shoreline is more uniform and its affluent network is poor represented. The higher right bank is cut in hard or solid rocks, while the left bank consists especially of alluvial matter. As a result, the river and implicitly the lake, have an asymmetric transversal profile, with greater depths along the right bank, while the plain flooding areas are present along the left bank (I). A part of plain flooding zones, such as those at km 911–918, initially, were littoral marshes. These were included within the lake perimeter, after the water level increase caused by the Danube blocking at Ostrovul Mare. The lower depths in the left bank zone, as well as the mud deposits, favoured the appearance of a narrow and almost continuous belt of submerged and floating vegetation, in the last years. Previously the lake making, the water flow rate varied around 1 m/s (2).

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Similar values were found quite all time after the lake appearance. Intermittently, lower values were registered depending on the maintenance conditions, hydroelectric exploitation and lock construction on both dam lakes (8). Topolnița river, the main affluent, discharges downstream the lake (at km 928) carrying the wastewaters of Drobeta-Turnu Severin town. All these natural and anthropic local conditions influence, in turn, the lake biocenosis characteristics, including the zooplankton structure.

2. MATERIALS AND METHODS

The zooplankton sample collection was carried out in 2002 summer and autumn, in 7 sectors comprising 12 stations (Table 1). These were placed in medial and littoral zones. The littoral zone stations were situated in the vicinity of the submerged and emerged macrophyte belt. Each sample resulted from filtration of 50 l water taken from the entire water column. On this purpose, a Patalas-Schindler device of 5 l volume was used. A net of 65µm mesh was used for the filtration.

Table 1

The stations of zooplankton sample collection, water depth (m), transparency (m) and temperature (°C)

No.	Station	River km	Depth		Transparency		Temperature	
			VI	IX	VI	IX	VI	IX
1	Gura Văii – medial	942.5	8.5	7.3	0.9	0.9	19.5	22.0
2	Gura Văii – littoral	942.5	8.0	7.0	0.9	0.9	19.5	22.3
3	Upstream of Topolnița river confluence – medial	928	13.7	15.0	0.8	1.3	20.0	22.4
4	Upstream of Topolnița river confluence – littoral	928	1.0	1.4	1.0	1.0	20.6	22.4
5	Upstream Hinova – medial	918	0.8	0.5	1.0	0.5	20.0	22.1
6	Ostrovul Corbului – medial	911	10.0	15.5	0.8	1.5	20.0	22.0
7	Ostrovul Corbului – littoral	911	1.4	1.4	1.0	0.9	20.0	22.0
8	Crivina – medial	894	13.4	13.0	0.7	1.6	20.2	22.2
9	Crivina – littoral	894	1.5	4.0	1.2	0.9	20.2	22.2
10	Țigănași – medial	978	18.4	17.5	1.3	1.5	20.0	21.5
11	Țigănași – littoral	978	1.0	2.0	1.3	1.0	20.0	22.2
12	Ostrovul Mare – medial	866		16.0		1.9		21.3

3. RESULTS AND DISCUSSION

Very few data exist referring to the zooplankton structure, in the river sector between 863 and 942 km, before 1984. At that time the said river sector was included within the perimeter of the dam Lake Iron Gates II, through the dam

construction at Ostrovul Mare. The results obtained by Popescu-Marinescu (4) in zone of Drobeta-Turnu Severin town, in 1972, referred to the taxonomic structure and numerical density of the zooplankton. The biomass data could not be obtained. The 1972 data analysis showed that in sector Drobeta-Turnu Severin, upstream of the future dam lake, the mentioned zooplankton ecological parameters were low, close to the minimum level, in sector Drobeta-Turnu Severin – Turnu Măgurele. According to the author opinion, the reason for this state was the discharge of domestic and industrial wastewater of Drobeta-Turnu Severin.

The species diversity of the zooplankton present in Drobeta-Turnu Severin zone, previous to the creation of the dam Lake Iron Gates II, was of 28 taxa (4), the ciliates and testacea being not determined. If these groups could be considered, the actual value of the zooplankton species diversity should be higher, in our opinion, with about 1/3 of the mentioned value.

Relatively few data of zooplankton taxonomic structure were registered in the first years of the lake existence. They are included in researches, which cover the entire Romanian Danube sector, not comprising details about the zooplankton in the dam Lake Iron Gates II (3, 6, 7).

The researches carried out in the first decade of lake existence were focused on the zooplankton structure (6, 7) or on the entire biocenosis structure (3) in Romanian sector of the Danube, not dealing with the sector between 863 and 942 km. Thus, the 2002 researches, the object of the present work, furnishes the first detailed information.

In 2002 ecological and anthropic conditions, the zooplankton of the dam Lake Iron Gates II amounted 70 taxa (Fig. 1). This figure is clearly lower than the value evidenced in the Lake Iron Gates I (125 taxa) in the same year, using the same research

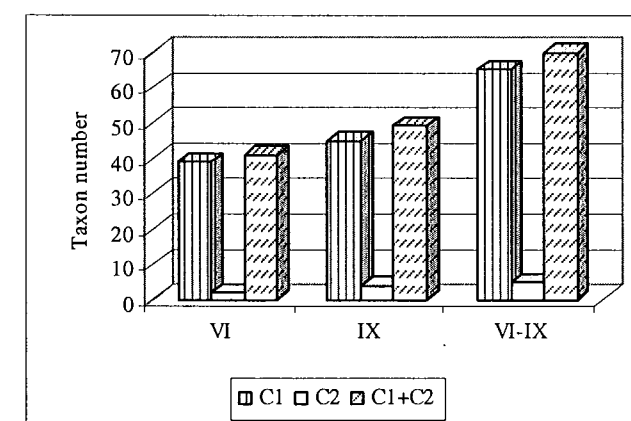


Fig. 1. – Seasonal variations of the zooplankton species diversity (no. taxa).

method (5). Among the determining factors of this state, we mention the greater size of the upstream lake, a wider range of ecological conditions, and larger water

contribution of affluent network. On the contrary, the downstream lake, of lower sizes, was more influenced by the effects of the maintenance regime, hydroelectric exploitation and lock construction. To these, the effects of hydrologic regime of the upstream lake, the large level difference between the two basins, the location far upstream the lake (in front of Drobeta-Turnu Severin) of a significant pollution source, as well as the uniformity of the lake ecological conditions are added.

92.86% of the zooplankton taxa in the Lake Iron Gates II belonged to the trophic level of plankton primary consumers, and the rest of 7.11% to the secondary consumers (Fig. 1).

The systematic groups with determinant role in zooplankton taxonomic spectrum were rotifers and testacea, amounting 50.77%, respectively 18.46% of c_1 total taxon number and cyclopids, 60% of c_2 total taxon number (Table 2). The contribution of the first two systematic groups with main role in the dynamics of ecological equilibrium, decreased, while that of cyclopids increased, from summer towards autumn.

Table 2

Seasonal variation averages of the taxonomic structure of the zooplankton in two trophic groups distributed on the systematic groups (% taxa)

Systematic group	Month		
	VI	IX	VI-IX
Primary Consumers	100.00	100.00	100.00
Ciliata	12.82	8.89	10.77
Testacea	23.08	20.00	18.46
Lamellibranchia	2.56	2.22	1.54
Rotatoria	51.29	46.67	50.77
Cladocera	7.69	17.78	13.85
Copepoda	2.56	4.44	4.61
Secondary Consumers	100.00	100.00	100.00
Ciliata	-	25.00	20.00
Rotatoria	50.00	25.00	20.00
Copepoda	50.00	50.00	60.00

The comparative analysis of the zooplankton species diversity in the 7 transversal sections of the river revealed higher values in the littoral zone against those in medial zone. As a consequence, the seasonal maximum values of this lacustrine ecosystem were located in 2 of 6 littoral stations: station 4 (km 928) and station 5 (km 918) (Fig. 2).

The constant frequency elements amounted to 13.48% of total taxon number, the accessory elements 19.10%, and accidental ones 67.42%. The value of the first category showed an increase from summer towards autumn (Table 3). Among the 16 forms with constant frequency, the plankton larva of the lamellibranchiate organism *Dreissena polymorpha* sole had a maximum coefficient, both during

summer and autumn. *Keratella cochlearis* had the maximum value only in summer, and *Bosmina longirostris* only in autumn (Tables 3 and 4).

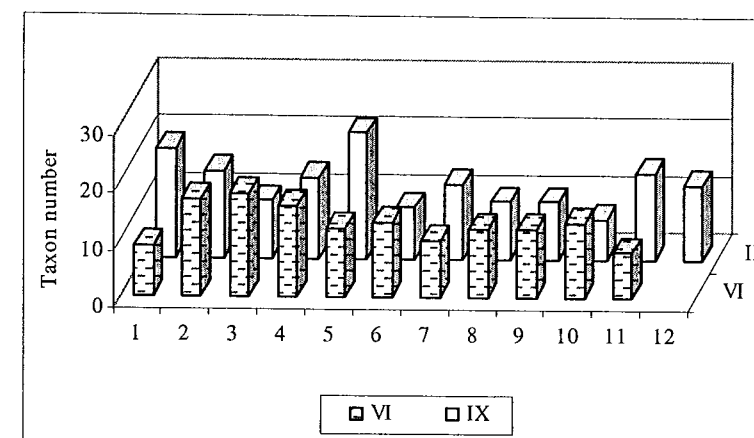


Fig. 2. – Seasonal spatial variations of the zooplankton species diversity (no. taxa).

Table 3

Contribution of constant, accessory and accidental frequency elements of the zooplankton (%)

Taxonomic elements	Month		X_a
	VI	IX	
Constant frequency elements	11.36	14.58	13.48
Accessory frequency elements	25.00	16.67	19.10
Accidental frequency elements	63.64	68.75	67.42

Table 4

Zooplankton constant elements as frequency (1), numerical dominance (2) and biomass (3)

Composition	1		2		3	
	Month					
	VI	IX	VI	IX	VI	IX
Primary consumers						
Testacea						
<i>Arcella arenaria</i> Greef		+				
<i>Centropyxis discoides</i> (Pénard) Deflandre		+				
Ciliata						
<i>Codonella cratera</i> Leidy	+					
Lamellibranchia						
<i>Dreissena polymorpha</i> Pallas	+	+	+		+	
Rotatoria						
<i>Euchlanis dilatata</i> Ehrenberg	+					
<i>Keratella cochlearis</i> Gosse	+		+			
<i>Synchaeta oblonga</i> Ehrenberg	+	+		+		

Table 4 (continued)

Cladocera						
<i>Bosmina longirostris</i> Schoedler		+		+		+
<i>Sida crystallina</i> (O.F.Müller)				+	+	
Copepoda						
<i>Eudiaptomus gracilis</i> (Sars) (naupliar stage)	+	+		+		
<i>Eurytemora velox</i> (Lill.) (naupliar stage)	+	+		+		
<i>Acanthocyclops vernalis</i> (Fischer) (I–III stage copepodids)		+				
<i>Mesocyclops crassus</i> (Fischer) (I–III stage copepodids)		+				
Secondary Consumers						
Rotatoria						
<i>Asplanchna priodonta</i> Gosse						+
Copepoda						
<i>Acanthocyclops vernalis</i> (Fischer) (IV–V stage copepodids + adult stage)		+		+		+
<i>Mesocyclops crassus</i> (Fischer) (I–III stage copepodids)		+		+		+

The zooplankton ecological structure comprised euplankton, phytophilous and nectobenthonic forms. The first category was relatively uniformly widespread in the whole lake, the other two being confined to the littoral zone.

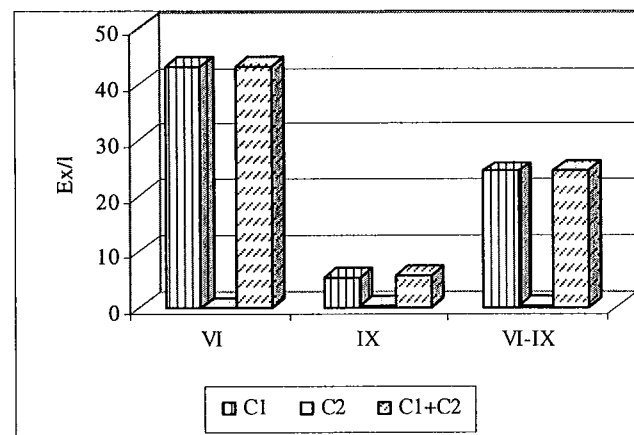


Fig. 3. – Seasonal average variations of the zooplankton numerical density distributed on the trophic levels (ex./l).

In ecological conditions of 2002 the zooplankton numerical density in Lake Iron Gates II was characterized by very low values ($X_a = 24.5$ ex/l) (Fig. 3). They were about 10 times lower than those evidenced in Drobeta-Turnu Severin river section, where the lacustrine ecosystem previously occurred (5). The low 2002 average of the mentioned parameter was a result of, to a great extent, its descendant dynamics in

summer-autumn (42.3–5.6 ex/l) (Fig. 3). In normal ecological conditions the dynamics of the zooplankton numerical abundance was ascendant from summer towards autumn in all categories of aquatic ecosystems from Romania. As a consequence, the maximum value was found in September or October. The atypical decrease in 2002 September was determined by a significant increase of the river flow during August, a seldom phenomenon for this period. To assure the dam security, the lake management authority partially emptied the basin, before the increase of the river flow. The sequence of these events strongly disturbed the normal zooplankton dynamics, in early autumn. But, even in these conditions, the numerical density average in 2002 (24.5 ex/l) was similar to that registered during 1995 (36.4 ex/l) (4).

To notice that the 2002 average of the zooplankton numerical density, in the Lake Iron Gates II was almost 3 times lower than that in the Lake Iron Gates I (70 ex/l) (6). Gomoiu et al. evidenced a difference of similar amplitude, in 1995 (3).

The data comparative analysis revealed that, generally, the Lake Iron Gates I assures better conditions for the zooplankton development. Surely, we refer especially to small depth zones, flooded after the lake occurrence, characterized by a slow water flow, higher temperature and abundant development of aquatic macrophytes. These are located both on upstream extremity of the basin, and in gulfs formed in affluent mouths (Mraconia, Eșelnița, Cerna, Bahna) situated mainly downstream the lacustrine basin.

The spatial dynamics of the numerical density evidenced, both in summer and autumn, higher values in littoral zone against medial zone. The annual maximum value (126.1 ex/l) was registered in littoral zone at km 918 (upstream Hinova locality), on the place where few marshes previously occurred. The lowest value was in the medial zone located close to the basin at Ostrovul Mare (1.8 ex/l) (Fig. 4) and represented, probably, the effect of the actions performed for energetic scopes and navigation.

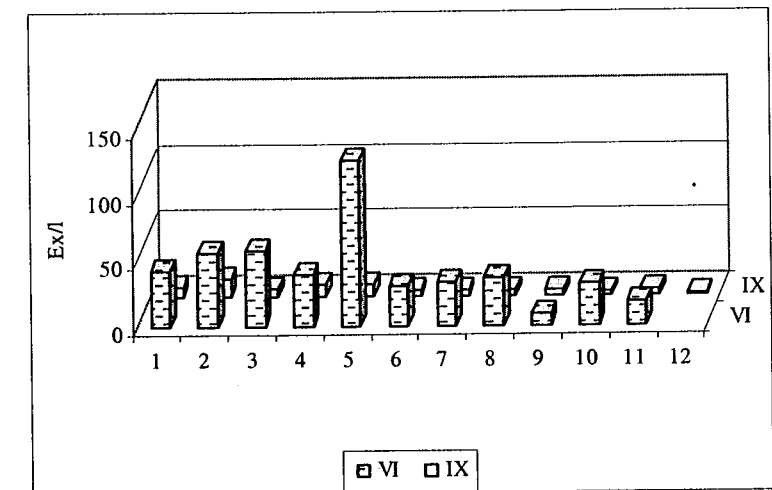


Fig. 4. – Seasonal spatial variations of the zooplankton numerical density (ex./l).

The detailed taxonomic structure, on systematic groups, showed the clear dominance of Lamellibranchia larvae (60.79%) followed by the Rotatoria (25.33%), within the primary consumers, and of Cyclopidae followed by the Rotatoria, in the case of secondary consumers. To remark that the first group had a descendant seasonal dynamics (66.8–11.35%), the second group a constant one (25.33–25.24%), while the dynamics of the third group was ascendant (0–96.46%) (Table 5).

Table 5

Seasonal variations of the relative numerical abundances of the two trophic groups of the zooplankton distributed on systematic groups (%)

Systematic groups	Month		X _a
	VI	IX	
Primary Consumers	100.00	100.00	100.00
Ciliata	2.17	1.11	2.06
Testacea	1.71	9.05	2.52
Lamellibranchia	66.81	11.35	60.79
Rotatoria	25.33	25.24	25.34
Cladocera	0.23	19.26	2.27
Copepoda	3.75	33.99	7.02
Secondary Consumers	100.00	100.00	100.00
Ciliata	-	3.54	3.33
Rotatoria	100.00	-	1.11
Copepoda	-	96.46	95.56

Similar changes in the seasonal dynamics were also observed at taxon level. Thus, in the case of primary consumers, the distinct numerical dominance of *Dreissena polymorpha* larva, in early summer (66.87%), was replaced with that of *Eudiaptomus gracilis* and *Eurytemora velox* naupliar stages, early autumn, amounting to 29.55%. In the case of secondary consumers, the numerical dominance of *Asplanchna priodonta* (100% in June) was replaced with that of *Mesocyclops crassus* and *Acanthocyclops vernalis* (amounting to 49.75% in September) (Table 4).

The zooplankton biomass in the Lake Iron Gates II was characterized by low values, correlated with its numerical abundance. To remark that the 2002 biomass average (51.2 µg wet weight /l) (Fig. 5) was 5 times greater than that in 1995 (10.1 µg/l) (3).

As the numerical density in the Lake Iron Gates I was remarkably higher than that in the Lake Iron Gates II, similarly was the biomass difference (3, 5). This parameter had a descendant summer–autumn dynamics, too (38.4–12.8 µg/l) (Table 5).

The biomass spatial dynamics showed the highest values in littoral zones situated in the middle part of the lake. Thus, the summer highest value (220 µg/l) was recorded at km 928, and at km 918 (149.2 µg/l) the early autumn one (Fig. 6).

The minimum value of littoral zone was at km 894 (10.6 µg/l), in summer, and that of medial zone at km 978, close to the dam (9.3 µg/l), in autumn.

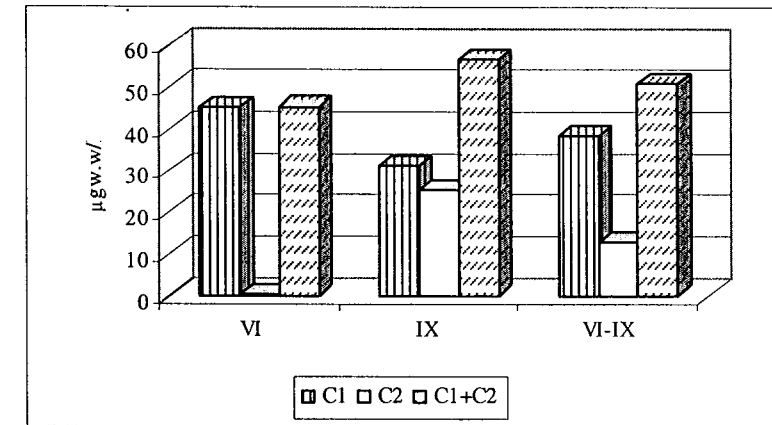


Fig. 5. – Seasonal average variations of the biomass zooplankton distributed on the trophic levels (µg wet weight/l).

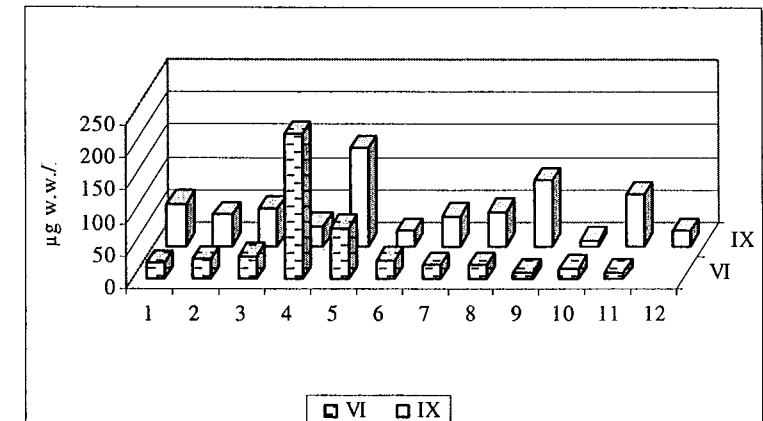


Fig. 6. – Seasonal spatial variations of the biomass zooplankton (µg wet weight/l).

The analysis of the gravimetric contribution of the systematic groups evidenced the dominance of Cladocera and Lamellibranchia larva stage (49.95%, respectively 23.39%) for primary consumers, and of Cyclopidae (99.41%) for secondary ones, in 2002 (Table 6).

The seasonal dynamics of primary consumers revealed the determinant role of Lamellibranchia (38.75%) and Cladocera (35.73%), in summer, and of Copepoda (24.02%) and Cladocera (70.45%) in autumn. In the case of secondary consumers, the Rotatoria dominated (100%) in summer and the Cyclopidae (99.96%) in autumn (Table 6).

Table 6

Seasonal variations of the relative biomass abundances of the two trophic groups of the zooplankton, distributed on systematic groups (%)

Systematic groups	Month		X _a
	VI	IX	
Primary Consumers	100.00	100.00	100.00
Ciliata	0.57	0.03	0.35
Testacea	0.92	0.92	0.91
Lamellibranchia	38.75	1.24	23.39
Rotatoria	10.31	3.34	7.45
Cladocera	35.73	70.45	49.95
Copepoda	13.74	24.02	17.95
Secondary Consumers	-	100.00	100.00
Ciliata	-	0.04	0.04
Rotatoria	100.00	-	0.55
Copepoda	-	99.96	99.41

The analysis of the gravimetric contribution of c₁ taxa, revealed the major role of *Dreissena polymorpha* (larva stage) (41.42%), followed by *Sida crystalina* (18.59%), in early summer. The gravimetrical dominance of the cladocer *Bosmina longirostris* (55.07%), followed by that of *Eudiaptomus gracilis* and *Eurytemora velox* (larva stages) (12.92%) was remarked during autumn. In the case of secondary consumers the sole contribution of *Asplanchna priodonta* (100%) during summer, and of *Mesocyclops crassus* and *Acanthocyclops vernalis* (IV–V copepodids and adults) (amounting 94.10%) during autumn, was observed (Table 4).

3. CONCLUSIONS

- The zooplankton in the dam Lake Iron Gates II was characterized by relatively low values of species diversity ($\Sigma = 70$ taxa), numerical density ($X_a = 24.5$ ex/l) and biomass ($X_a = 51.2$ μ g wet weight/l).
- Comparatively to the zooplankton in the dam Lake Iron Gates I, the zooplankton in the dam Lake Iron Gates II had a species diversity 1.8 times lower, and a numerical density and biomass of almost 3, respectively 2.2 times lower.
- The species with the most important role in the dynamics of the ecological equilibrium of the zooplankton, with the highest values of constancy, numerical and gravimetrical dominance was *Dreissena polymorpha* in the plankton larva stage.

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FRESHWATER OLIGOCHAETA (ANNELIDA) OF THE SOMEȘUL MIC RIVER DRAINAGE IN ROMANIA

CLAUDIA PAVELESCU*, CLAUDIU TUDORANCEA**

This study is a contribution to the knowledge of the aquatic Oligochaeta (Annelida) fauna of Romania. Freshwater oligochaetes were sampled quantitatively at eight sites on the Someșul Mic River and on headwater streams of one of its main tributaries, the Someșul Cald, in 2000. Thirty-eight species of oligochaetes belonging to seven families and twenty-two genera were recorded during this study. The family Naididae had the highest species richness (18 species) followed in decreasing order by Enchytraeidae (8 species), Tubificidae (5 species), Lumbricidae and Lumbriculidae (2 species each), and Propappidae and Haplotaxidae (one species each). In general, species richness and density increased on the longitudinal gradient of the river, although the highest number of species occurred in the middle stretches of the river. Several species of *Nais* appeared to be tolerant to organic enrichment of the river. *Nais alpina* along with *Pristina foreli*, *Pristina* sp. and *Specaria josinae* occurred in the headwaters and in the middle transitional zone of the river from clean towards more organically polluted stretches. The tubificid *Rhyacodrilus coccineus* proved to be an indicator for pristine waters of the headwaters zone.

INTRODUCTION

Oligochaeta assemblages are important components of the freshwater benthic fauna. They are encountered in both lentic and lotic ecosystems inhabiting a diversity of habitats ranging from silty and sandy substrates with organic matter to submerged vegetation and even stony or rocky bottoms.

Except for the Danube Delta, very little information on the taxonomy and ecology of freshwater Oligochaeta of Romania is available (3, 13, 14). Information on river and stream oligochaetes is virtually non-existent in Romania. This paper summarizes the results of one study associated with a multidisciplinary and long-term project on the fauna of the Someșul Mic River, a trans-boundary river in Romania. This river is part of the Someș River catchment area, the largest in the north-west of Romania. The catchment area of the Someșul Mic River is comprised of a variety of ecosystems, from the pristine headwater streams to the very organically polluted stretches downstream. The oligochaete assemblages identified from the river during this study will contribute significantly to our knowledge of the oligochaete diversity in this insufficiently studied European region. In addition, the habitat information associated with our collections will allow us to establish substrate preferences for oligochaete species, and allow us to define more clearly their tolerances to organically polluted water.

STUDY AREA

The Someșul Mic River is formed by the upstream confluence of two major tributaries, the Someșul Cald and Someșul Rece rivers, about 5 km upstream the Gilau (Fig. 1). The river has a length of 153 km, measured from the headwater springs of Someșul Cald (the longer of these two major tributaries). The Someșul Mic River drains the eastern side of the Apuseni Mountains and has a catchment area of about 775 km² and an annual mean discharge of 24.4 m³/s at Dej (both measurements measured at a point just upstream of its confluence with the Someșul Mare River) (6). The Someșul Cald, the longest tributary of Someșul Mic, originates at an altitude of 1385m a.s.l. on the southeastern side of Vladeasa Mountains, a karstic region. The Someșul Cald has a length of 64 km and a catchment area of about 534 km² (7). For most of its length, the Someșul Cald is a mountainous river, relatively shallow, torrential, and shaded by forest. The river system is located in a moderate continental climate characteristic for the western and north-western part of Romania.

MATERIAL AND METHODS

Quantitative samples were collected from eight sites located along a longitudinal gradient on the main river and on the Someșul Cald on 25 May, 26 July, and 22 November 2000. Four of the stations located on the headwater tributaries of Someșul Cald were considered pristine water ecosystems protected from the human impact (Fig. 1). A fifth sampling station was established on the Someșul Cald upstream of Lake Tarnita, a man-made lake. Three more sampling stations were selected on the Someșul Mic River, near major organic pollution sources. One station was located at Gilau, about 15 km upstream of the city of Cluj-Napoca. At this station there are no major sources of pollution except that of domestic discharge from the surrounding villages, with no wastewater treatment. The other two sites, Apahida and Gherla, are situated 15 and 50 km, respectively, downstream of the city of Cluj-Napoca. The Someșul Mic River, in particular the lower course downstream of Cluj-Napoca, is exposed to various sources of organic pollution, including animal husbandry, agricultural, and industrial activities (17).

Three benthic samples were taken across the river bed at each site during each sampling period using a Surber type sampler (surface area = 1060 cm²). Samples were washed through a fine mesh net (250μ). The samples were preserved in the field in 4% formaldehyde. Several physical and chemical parameters were measured at each site during the collection of biological samples. The oxygen and temperature were measured with a portable YSI Model 52 oxygen meter – one that allows adjustment of oxygen saturation to altitude. The hydrogen ion concentration (as pH) was measured with a CONSORT Model P 902 pH meter; conductivity and salinity

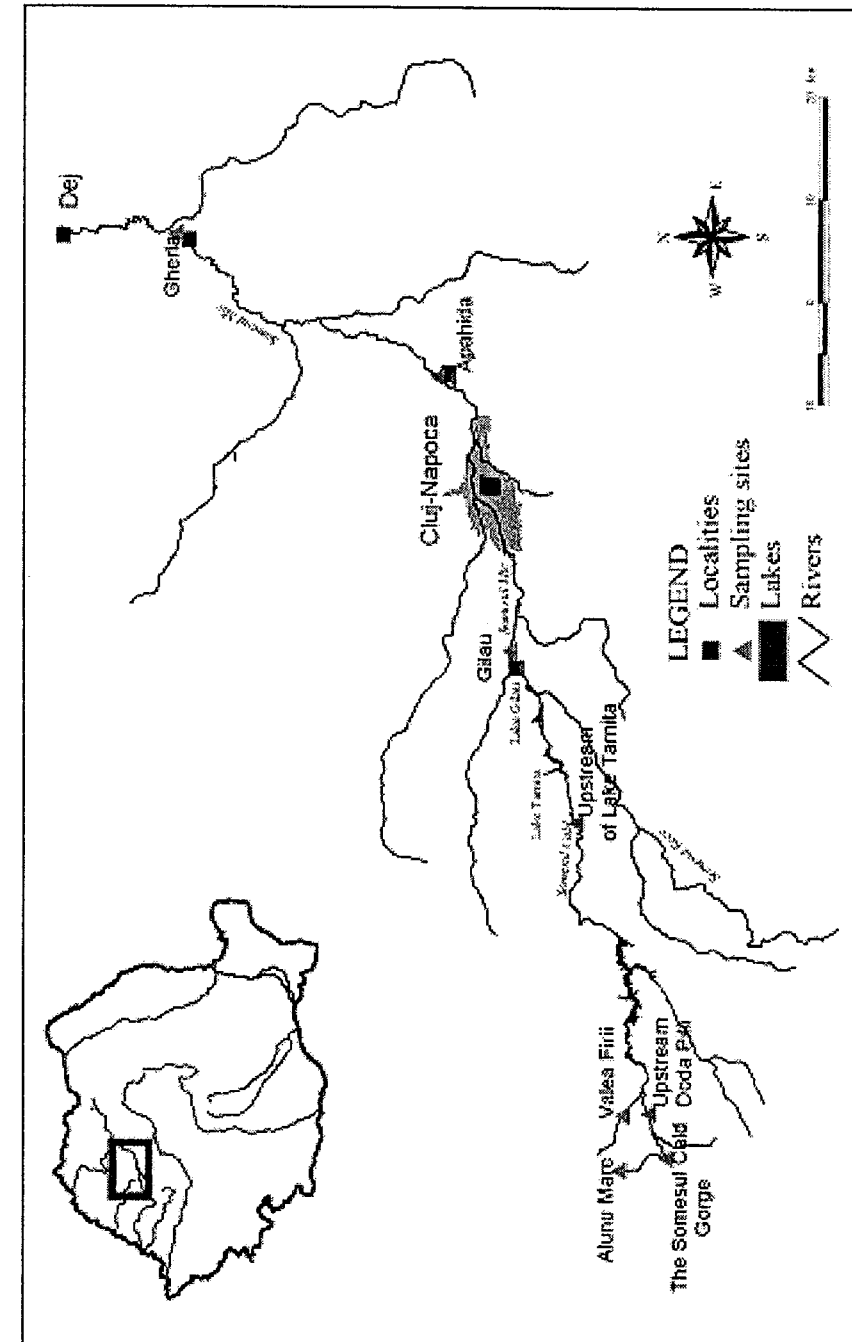


Fig. 1. – The sketch map of the study area, the Someșul Mic River and its tributary Someșul Cald, and its general location in Romania, where surveys for aquatic oligochaetes at eight stations were conducted during 2000.

were measured with a CONSORT Model K 119 conductivity meter. The medium depth and the river bed width were also measured. Total organic content in river sediment was determined by the standard method of loss on ignition at 550°C after drying at 105°. Biological samples were sorted on major invertebrate groups in the laboratory under a dissecting microscope. Prior to identification, oligochaete specimens were allowed to clear for a day in Amman's lactophenol (glycerol, lactic acid, carbonic acid and water at a ratio of 2:1:1:1). We are aware of some recent molecular and morphological studies conducted on family Naididae which place this group as a subfamily (Naidinae) within the larger family Tubificidae (4, 5). However, our identifications were done by using Sperber (15), Brinkhurst (2) and Timm (16).

RESULTS AND DISCUSSION

The values for physical and chemical parameters measured were recorded along a longitudinal gradient of the stream (Table 1). Temperatures recorded during 2000 at the eight sampling stations did not vary much between the stations. The lowest temperatures, 1.5°C and 2°C, were recorded in November 2000 at station Doda Pili and the station located immediately upstream of Lake Tarnita, respectively. The highest temperatures were recorded in July 2000 at Gherla (20.8°C) and at the site located immediately upstream of L. Tarnita (22.3°C). No major differences were noticed with respect to other physicochemical factors among the eight sampling sites except for conductivity and salinity – higher values for these two parameters were recorded at the stations near Apahida and Gherla, those exposed to human impact. Lower values for oxygen saturation were also recorded at these two stations in comparison with values recorded at upstream stations. During a previous study (17) measured several other physical and chemical factors (e.g. suspended solids, total dissolved solids [TDS], nitrates, ammonia, and chemical oxygen demand [COD]) along the same sector of the Someşul Mic River, recording higher values at the stations near Apahida and Gherla than at the station near Gilau (upstream of the city of Cluj-Napoca). Sediment analyses showed rocky and stony substrates mixed with cobbles and pebbles at the most headwater stations on small tributaries and gravel and sparse cobbles at Apahida, a substrate similar to that present at Gilau. At Gherla, the substrate had a high proportion of silts and sands to gravel and pebbles (17). The amount of organic matter in the sediment reflects not only the nature of the substrate but also its location along the longitudinal gradient. The highest values of the organic matter were found at Alunul Mare (in the headwaters zone), and at Apahida and Gherla, the two stations exposed to anthropogenic impacts. All this evidence is indicative of the exposure of the Someşul Mic River to organic enrichment along its low reaches.

The high values of organic matter in the sediment at station Alunul Mare most likely results from the sawdust and other forest industry byproducts abandoned along the river shorelines. The primitive practices of the forest product industry contribute to the ever-increasing and detrimental anthropogenic effects on the water quality and on the aquatic communities in the Someşul Mic River.

Table 1

Physical and chemical characteristics of water and sediment at the sampling stations on the Someşul Mic River and its tributary Someşul Cald, Romania, during 2000

Location	Altitude (m)	Month	Water temperature (°C)	Riverbed width (m)	Medium depth (cm)	Conductivity (µS/cm)	Salinity (mg/l)	Dissolved oxygen (mg/l)	Dissolved oxygen (%)	Hydrogen ion concentration (as pH)	Organic matter in sediment (% by weight)
The Someşul Cald Gorge	1145	May	9.7	4-5	20	63	31.2	9.2	80.5	8.3	1.99
		July	13.6	4-5	30	82	43	8.6	87.0	7.9	1.13
		Nov	-	-	-	-	-	-	-	-	-
Alunul Mare	1170	May	8.2	1-2	15	34	17.8	10.5	89.4	8.1	2.26
		July	9.1	1-2	20	145	76.8	11.2	98.0	7.9	1.28
		Nov	6.4	1-2	18	165	87.5	6.2	50.0	8.1	1.79
Doda Pili	1000	May	15.2	14	45	94	49.8	7.3	72.4	8.5	0.66
		July	16.6	14	20	121	64.1	8.2	82.0	8.1	0.71
		Nov	1.5	8-10	15	130	69.1	12.7	90.6	8.1	0.73
Valca Firii	1100	May	14.9	3-4	15	110	58.2	7.7	76.7	8.1	1.18
		July	15.6	3-4	25	103	55	7.1	71.0	7.9	1.53
		Nov	3.0	2	5	149	78.9	11.6	86.4	9.2	1.63
Upstream of Lake Tarnita	490	May	16.8	4-5	10	107	56.4	7.4	76.0	8.2	0.65
		July	22.3	4-5	15	114	60.3	7.7	91.0	7.7	0.73
		Nov	2.0	7	10	131	69.7	9.8	71.3	5.3	1.03
Gilau	350	May	21.8	18	30	394	208	10.0	84.2	8.2	0.48
		July	17.0	17	20	263	141	9.2	82.7	7.9	0.52
		Nov	8.4	22	30	95	50.4	9.1	77.4	7.4	0.25
Apahida	300	May	21.7	50	40	384	204	11.3	97.6	7.8	0.69
		July	18.4	55	40	265	140	6.8	73.0	7.4	0.35
		Nov	7.7	70	30	561	296	9.0	76.9	7.4	1.21
Gherla	300	May	-	-	-	-	-	-	-	-	-
		July	20.8	35	45	696	372	6.0	66.6	7.3	2.20
		Nov	6.5	30	30	693	371	7.5	62.3	7.6	1.09

A total of 38 species of oligochaetes were identified at the eight stations during the sampling period. These species belong to 24 genera and 7 families (Table 2). The Naididae (considered by Erséus and Gustavsson to be subfamily Naidinae) (4, 5) represented by 10 genera and 18 species, appeared to be the most diverse. The family Enchytraeidae was represented by eight species, while the family Tubificidae was represented by 5 species. Other families such as Lumbricidae, Propappidae, Haplotaxidae, and Lumbricidae were represented by fewer species. Other invertebrate groups occurring with oligochaetes in samples included Ephemeroptera, Plecoptera, Trichoptera, and Diptera larvae. In general, oligochaetes numerically constituted less than 10 % of the total benthic fauna at the headwater stations, including that documented for the station at Gilau, located upstream city of Cluj-Napoca. Conversely, oligochaetes were the dominant group, numerically, at the two polluted stations Apahida and Gherla on the low part of the main river, reaching (as high values) 98 % of the total benthic fauna (Fig. 2).

Most of the identified species occurred at each sampling station (Table 2). As a general pattern of the distribution of Oligochaeta in the Someşul Mic River, both species richness (Table 1) and numerical abundance (Table 3) increased along the longitudinal gradient from headwaters towards middle and low reaches of the river.

At headwaters stations of Someşul Cald Gorge (elevation: 1145 m) and Alunul Mare (elevation: 1170 m), oligochaetes were represented by 12 species (3 Naididae, 1 Tubificidae, 3 Lumbricidae, and 5 Enchytraeidae). Twenty species were identified at the station located upstream of Lake Tarnita, while 12 and 14 species, respectively, were recorded at the lowland stations Apahida and Gherla. We believe that the stony substrate and torrential regime of the river in the headwaters zone rather than the altitude prevent more species of oligochaetes from establishing themselves. Similar species richness was recorded at the stations Apahida and Gherla, most likely because of the change in substrate nature as well as the higher levels of organic pollution at these sites.

Most Naididae species occurred at the majority of the sites sampled along the longitudinal gradient. *Nais elinguis* and *Nais communis* were the most widespread species, each occurring at six sampling stations, but in varying densities. The former had densities ranging between 2 ind./m² (station Doda Pili) and 13,976 ind./m² at the polluted station Gherla, downstream of the Someşul Mic River (Table 3). *Nais communis*, although widely distributed, occurred in very low densities at all stations. These two species occurred at both clean and polluted stations, suggesting that they have wide tolerances to organic pollution and thus may not be indicators of water quality. *Nais elinguis* has previously been recorded as a pollution tolerant species in rivers (10). Four naidids occurred at stations that were devoid of, or subject to but little human impact, such as the station Gilau – *Pristina foreli*, *Pristina* sp., *Specaria josinae*, and *Nais alpina*. These four taxa seem to be characteristic of both clean and the transitional zone, from pristine to more organically polluted waters. Three species, *Vejdovskyella comata*, *Amphichaeta leydigi*, and *Stylaria lacustris* occurred only at the station Gilau, located in a transitional stretch of the river with little human impact. One naidid

Table 2

List of Oligochaeta species collected from eight sampling sites on the Someșul Mic River and its tributary Someșul Cald, Romania, during 2000

	The Someșul Cald Gorge	Alunu Mare	Doda Pili	Valea Firii	Upstream of Lake Tarnita	Gilau	Apahida	Gherla
Family Naididae								
<i>Amphichaeta leydigi</i> Tauber, 1879						+		
<i>Dero (Aulophorus) furcata</i> (Müller, 1773)								+
<i>Chaetogaster diaphanus</i> (Gruithuisen, 1828)			+		+		+	+
<i>Chaetogaster diastrophus</i> (Gruithuisen, 1828)					+		+	
<i>Dero (Dero) obtusa</i> d'Udekem, 1855								+
<i>Dero</i> sp. Oken, 1815								+
<i>Nais alpina</i> Sperber, 1948	+		+		+	+		
<i>Nais barbata</i> Müller, 1773							+	+
<i>Nais bretscheri</i> Michaelsen, 1923			+		+	+		+
<i>Nais communis</i> Piguet, 1906	+		+	+	+	+	+	
<i>Nais elinguis</i> Müller, 1773			+	+	+	+	+	+
<i>Nais pardalis</i> Piguet, 1906			+	+	+	+		+
<i>Nais stolci</i> Hrabe, 1981						+		
<i>Paranais frici</i> Hrabe, 1941								+
<i>Pristina foreli</i> (Piguet, 1906)			+		+	+		
<i>Pristina</i> sp. Ehrenberg, 1828 ; emended Brinkhurst, 1985			+	+	+	+		
<i>Specaria josinae</i> (Vejdovsky, 1883)		+	+		+	+		
<i>Stylaria lacustris</i> (Linnaeus, 1767)						+		
<i>Vejdovskyaella comata</i> (Vejdovsky, 1883)						+		
Family Tubificidae								
<i>Limnodrilus hoffmeisteri</i> Claparède, 1862					+	+	+	+
<i>Limnodrilus udekemianus</i> Claparède, 1862							+	+
<i>Rhyacodrilus coccineus</i> (Vejdovsky, 1875)	+		+	+	+			
<i>Tubifex newaensis</i> (Michaelsen, 1902)				+	+		+	
<i>Tubifex tubifex</i> (Müller, 1774)					+		+	+
Family Lumbriculidae								
<i>Stylodrilus herringianus</i> Claparède, 1862			+	+	+			
<i>Trichodrilus</i> sp. Claparède, 1862		+	+	+	+		+	
Family Propappidae								
<i>Propappus volki</i> Michaelsen, 1916			+	+	+			
Family Haplotaxidae								
<i>Haplotaxis cf. gordioides</i> (Hartmann, 1821)			+	+				
Family Lumbricidae								
<i>Dendrobaena octaedra</i> (Savigny, 1826)	+							
<i>Eiseniella tetraedra</i> (Savigny, 1826)		+		+	+		+	+

	The Someșul Cald Gorge	Alunu Mare	Doda Pili	Valea Firii	Upstream of Lake Tarnita	Gilau	Apahida	Gherla
Family Enchytraeidae								
<i>Cernosvitoviella</i> sp. Nielsen & Christensen, 1959			+	+				
<i>Cognettia glandulosa</i> (Michaelsen, 1888)	+	+	+		+			
<i>Cognettia sphagnetorum</i> (Vejdovsky, 1878)		+	+					
<i>Fridericia</i> sp. Michaelsen, 1889	+	+						
<i>Henlea perpusilla</i> Friend, 1911				+				
<i>Marionina</i> sp. Michaelsen, 1889		+						
<i>Mesenchytraeus</i> sp. Eisen, 1878		+		+				
<i>Mesenchytraeus armatus</i> (Levinsen, 1884)	+	+						
Other Enchytraeidae	+	+		+	+	+	+	+

species, *Nais barbata*, occurred only at the most impacted stations, Apahida and Gherla, reaching a mean density of 29,249 ind./m² (Station Apahida) (Table 3). It is well known that species of the Family Tubificidae are usually associated with low amounts of oxygen and high organic enrichment, particularly in lakes. According to Brinkhurst (1, 2) and Milbrink (11), *Limnodrilus hoffmeisteri* and *Tubifex tubifex*, two species present in our samples, are characteristic of highly eutrophic waters. Out of the 5 species of Tubificidae identified during this study, three – *Limnodrilus hoffmeisteri*, *Tubifex tubifex*, and *Rhyacodrilus coccineus* – were the most common, occurring at four stations each. Both *L. hoffmeisteri* and *T. tubifex* occurred at all four stations located on the main river, a sector exposed to human impact. *Tubifex tubifex* had densities which varied from as low as 3 ind./m² (at the station located upstream Lake Tarnita and 22505 ind./m² at organically polluted station Gherla (Table 3). *Limnodrilus hoffmeisteri* occurred in densities ranging from 3 ind./m² at the station upstream of Lake Tarnita, to 95,711 ind./m² at Gherla. *Rhyacodrilus coccineus* occurred in much lower abundances at all upstream stations located in the headwater zone, including the station at Someșul Cald Gorge (Table 3). The densities of this species ranged from 2 ind./m² (the station upstream of L. Tarnita) to 21 ind./m² at the station near Valea Firii. *Rhyacodrilus coccineus* is one of several species considered to be restricted to oligotrophic conditions by Howmiller and Scott (8). Milbrink (12) considers this species characteristic for transitional zones, those ranging from eutrophic to oligotrophic conditions. Our data indicate that *R. coccineus* seems to be a species sensitive to organic enrichment and appears to be rather a stable indicator of good quality water. It is obvious that the dissolved oxygen is not the limiting factor for the oligochaetes at these organically enriched stations because the saturation in oxygen was over 62% (Table 1) at Apahida and Gherla. We believe that it was the nature of the sediment

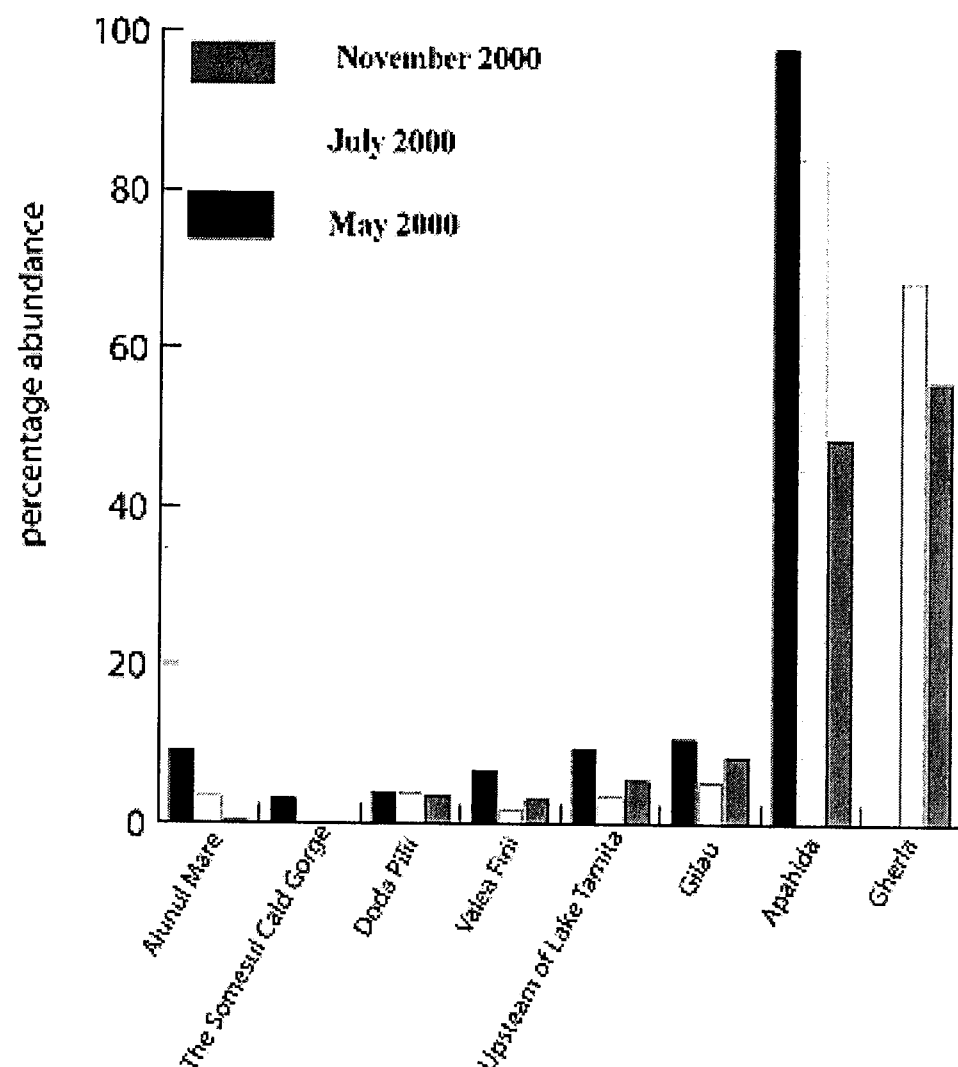


Fig. 2. – The oligochaetes as a percentage by densities of the total benthic macroinvertebrates collected from eight sampling stations on the Someșul Mic River and its tributary Someșul Cald, in Romania, during 2000.

and the amount of organic matter that control the distribution of oligochaetes at the stations along the low sector of Someșul Mic River. Only three oligochaete species appear to be restricted in distribution to the most polluted stations, Apahida and Gherla – the naidids *Dero (Aulophorus) furcatus*, *Dero (Dero) obtuse*, and *Paranais frici*. Other species of oligochaetes were present in this river but occurred in low numerical abundance.

Table 3

Densities (nr. ind./m²) of the commonest oligochaete species occurring at eight sampling stations on the Someșul Mic River and its tributary Someșul Cald, Romania, during 2000

Species	Sampling site							
	Someșul Cald Gorge	Alunul Mare	Doda Pili	Valea Firii	Upstream of Lake Tarnita	Gilau	Apahida	Gherla
<i>Nais alpina</i>	2	1	84	1	473	422	0	0
<i>Nais barbata</i>	0	0	0	0	0	0	29249	1444
<i>Nais bretscheri</i>	0	0	3	0	14	361	0	0
<i>Nais elinguis</i>	0	0	2	9	5	585	13976	85
<i>Pristinella rosea</i>	0	0	3	1	32	307	0	0
<i>Limnodrilus hoffmeisteri</i>	0	0	0	0	3	58	2560	95711
<i>Rhyacodrilus coccineus</i>	3	0	8	20	2	0	0	0
<i>Tubifex tubifex</i>	0	0	0	0	2	26	1373	22505
<i>Stylogrilus heringianus</i>	0	5	7	22	30	0	0	0
<i>Propappus volki</i>	0	0	65	169	11	0	0	0
<i>Haplotaxis cf. gordioides</i>	0	0	2	3	0	0	0	0
Enchytraeidae	16	80	20	50	129	43	74	85

Because of the relatively low number of samples, no advanced statistical analyses could be performed. However, a similarity of Oligochaeta assemblages at the eight sampling locations during 2000, based on Morisita index (9, 18), is displayed in the dendrogram of Fig. 3. There seem to be three different assemblages: one group is composed of the assemblages from the highest altitudes (Someșul Cald Gorge and Alunul Mare); a second group is formed by the assemblages recorded at four stations between 1000 m and 350 m (Doda Pili, upstream of Lake Tarnita, Valea Firii, and Gilau); a third group is represented by the assemblages at the polluted Apahida and Gherla sites.

In conclusion, longitudinal zonation and factors associated with human activities both influenced the Oligochaeta distribution. Although additional data are needed in order to determine the importance of Oligochaeta assemblages, particularly their value as biological indicators, the data presented in this paper indicate that this major invertebrate group has an important potential of being used efficiently in management and monitoring practices during water quality assessments.

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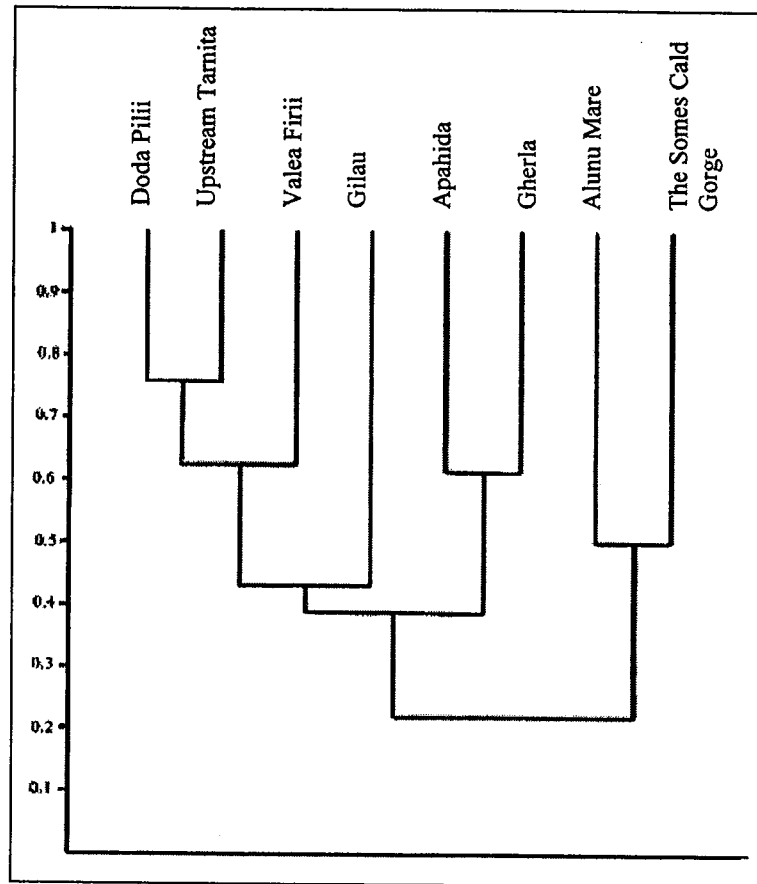


Fig. 3. – Dendrogram of the similarity of the Oligochaeta species composition, based on Morisita index, between eight sampling sites located on the Someșul Mic River and its tributary Someșul Cald in Romania, surveyed during 2000.

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CONTRIBUTIONS TO THE COGNITION OF GROWTH OF CERTAIN POPULATIONS OF AQUATIC GASTROPODS ACCORDING TO AGE AND SEASON

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By analysing a number of 4,000 individuals we succeeded in emphasizing the specific features of growth of the aquatic gastropods populations according to body dimensions and age categories. These parameters vary also according to seasons. Thus, it resulted that there is a close correlation among the body dimensions, age categories and growth rate, fact that allows to estimate the ecological characteristics of the aquatic gastropods populations.

Key words: gastropods, age categories, growth rate.

1. INTRODUCTION

Gastropods represent one of the most important groups as concerns the biological production of the lacustrine ecosystems built.

By knowing their growth rate during different life stages it can be estimated their contribution to the production of aquatic ecosystems (2).

2. MATERIAL AND METHOD

In order to determine the growth rate, there have been seasonally drawn samples containing variable quantities of gastropods. There have been made measurements to determine the height (the value rendered in mm of the distance between the apex and the top of the siphonal channel) and the length (the distance between the external side of the last anfract and the external side of the aperture).

The structure on dimensions and age has been established by analysing a number of 4,000 individuals. There have been analysed 17 species.

3. RESULTS AND DISCUSSIONS

Establishing the dimensions of the individuals plays an important role in determining the physiological ages. When certain populations do not clearly have the mark of the physiological ages, the age is estimated according to the body dimension (Table 1).

Table 1

Dimensional values for different age categories at gastropods populations

SPECIES	BODY DIMENSIONS CHARACTERISTIC FOR DIFFERENT AGE CATEGORIES (h, l=mm)		
	Pre-reproductive	Reproductive	Post-reproductive
<i>Viviparus acerosus</i>	h=10 – 20 l=7 – 15	h=20,1 – 40 l=15,1 – 30	h=over 40 l=over 30
<i>Viviparus viviparus</i>	h=5 – 15 l=3 – 10	h=15.1 – 35 l=10.1 – 25	h=over 35.1 l=over 25.1
<i>Physa acuta</i>	h=0,1 – 5 l=0.1 – 4	h=5.1 – 11 l=4.1 – 6	h=over 11.1 l=over 6.1
<i>Radix ovata</i>	h=0.1 – 5 l=0.1 – 4	h=5.1 – 11 l=4.1 – 6	h=over 11.1 l=over 6.1
<i>Aplexa hypnorum</i>	h=0.1 – 6 l=0.1 – 2	h=6.1 – 8.9 l=2.1 – 3.9	h=over 9 l=over 4
<i>Radix auricularia</i>	h=0.1 – 15 l=0.1 – 10	h=15.1 – 21.1 l=10.1 – 15	h= over 21 l= over 15
<i>Valvata piscinalis</i>	h=1.5 – 5 l=1 – 3	h=5.1 – 8 l=3.1 – 5	h=over 8 l=over 5
<i>Radix peregra</i>	h=0.1 – 15 l=0.1 – 10	h=15.1 – 20.9 l=10.1 – 11.5	h=over 20 l=over 12
<i>Planorbis planorbis</i>	h=0.1 – 1.5 l=0.1 – 9	h=1.6 – 3.5 l=9.1 – 17	h=over 3.5 l=over 17
<i>Physa fontinalis</i>	h=0.1 – 5 l=0.1 – 3	h=5.1 – 9 l=3.1 – 7.5	h=over 9 l=over 7.5
<i>Segmentina nitida</i>	h=0.1 – 1.2 l=0.1 – 4	h=1.3 – 1.8 l=4.1 – 7	h=over 1.8 l=over 7
<i>Stagnicola palustris</i>	h=0.1 – 15 l=0.1 – 8	h=15.1 – 28 l=8.1 – 15	h=over 28 l=over 15
<i>Fagotia acicularis</i>	h=0.1 – 10 l=0.1 – 3	h=10.1 – 17 l=3.1 – 6	h=over 17 l=over 6
<i>Stagnicola corvus</i>	h=0.1 – 20 l=0.1 – 10	h=20.1 – 35 l=10.1 – 18	h=over 35 l=over 18
<i>Fagotia esperi</i>	h=0.1 – 10 l=0.1 – 5	h=10.1 – 18 l=5.1 – 8	h=over 18 l=over 8
<i>Anisus spirorbis</i>	h=0.1 – 0.6 l=0.1 – 2.5	h=0.7 – 1.4 l=2.6 – 5.5	h=over 1.4 l=over 5.5
<i>Succinea elegans</i>	h=8 – 14 l=3 – 5	h=14.1 – 20 l=5.1 – 8	h=over 20 l=over 8

The measurements emphasize that the dimensions of juvenile individuals (pre-reproductive) of the species of *Physa acuta*, *Radix ovata*, *Aplexa hypnorum*,

Physa fontinalis, *Segmentina nitida*, *Fagotia acicularis*, *Fagotia esperi*, *Anisus spirorbis*, *Succinea elegans* and *Valvata piscinalis* oscillate between h = 0.1 and 5 – 6 mm and l = 0.1 and 2 – 5 mm; the dimensions of reproductive individuals (mature from the sexual point of view) oscillate between h = 4.1 – 18 mm, l = 1.9 – 20 mm, while the dimensions of post-reproductive individuals (senescent) oscillate between h = 1.8 – 11 mm, l = 3 – 18 mm.

The dimensions of the other species (Table 1) oscillate between h = 0.1–15 mm, l = 0.1–10mm for pre-reproductive individuals, h = 10–40 mm, l = 15–40mm for reproductive individuals and h = 18–40 mm for post-reproductive individuals.

By analysing Tables 1 and 2, it can be noticed that in the case of 2 species, namely *Fagotia acicularis* and *Stagnicola corvus*, there predominates the age category of pre-reproductive individuals with a rate of 71.43 per cent respectively, 64.43 per cent. The age category of pre-reproductive individuals predominates at most of the species (15 species) and its values oscillate between 44.08 per cent and 75 per cent. In the case of only one species, *Fagotia esperi*, the individuals belonging to both pre-reproductive and reproductive categories present similar values 45.45 per cent (1, 3).

Table 2

Ages distribution at gastropods populations according to their body dimensions

SPECIES	AGES					
	Pre-reproductive		Reproductive		Post-reproductive	
<i>Viviparus acerosus</i>	264	21.15%	882	70.67%	102	8.17%
<i>Viviparus viviparus</i>	163	16.44%	682	72.47%	96	10.20%
<i>Physa acuta</i>	191	43.31%	198	44.19%	59	13.38%
<i>Radix ovata</i>	191	43.31%	198	44.19%	59	13.38%
<i>Aplexa hypnorum</i>	46	18.77%	153	62.45%	46	18.77%
<i>Radix auricularia</i>	47	35.08%	76	56.72%	11	8.20%
<i>Valvata piscinalis</i>	19	32.40%	59	54.62%	30	12.98%
<i>Radix peregra</i>	35	37.63%	55	59.14%	3	3.22%
<i>Planorbis planorbis</i>	27	29.03%	41	44.08%	1	1.07%
<i>Physa fontinalis</i>	16	32%	27	54%	7	14%
<i>Segmentina nitida</i>	7	17.5%	30	75%	3	7.5%
<i>Stagnicola palustris</i>	18	45%	21	52.5%	1	2.5%
<i>Fagotia acicularis</i>	15	71.43%	12	42.86%	1	3.57%
<i>Stagnicola corvus</i>	11	64.70%	5	29.41%	1	5.88%
<i>Fagotia esperi</i>	5	45.45%	5	45.45%	1	9.09%
<i>Anisus spirorbis</i>	2	40%	3	60%	"	"
<i>Succinea elegans</i>	–	–	2	66.66%	1	33.33%

At the species *Succinea elegans* there were not registered pre-reproductive individuals, while in the case of the species *Anisus spirorbis* there were not registered post-reproductive individuals. In our case, the present situation can be also induced by certain sampling errors.

The predominance of pre-reproductive and reproductive individuals at most species illustrates that their populations register a clear numerical progress. At only one species, *Fagotia esperi*, it is registered a stationary equilibrium, while at the species *Succinea elegans* and *Anisus spirorbis* the lack of some age categories illustrates a diminution of the total number of individuals.

The seasonal analysis on ages and dimensions categories (Table 3) proves that 9 species presented all the categories of age and dimension during the three seasons of the study: *Viviparus acerosus*, *Viviparus viviparus*, *Valvata piscinalis*, *Physa acuta*, *Radix ovata*, *Aplexa hypnorum*, *Radix auricularia*, *Radix peregra* and *Segmentina nitida* (4, 5). The other 8 species rarely appear only during certain seasons and they are considered accidental species.

Table 3

Gastropods populations seasonal structure on dimensions and age categories

SPECIES	AGE CATEGORIES	SEASON					
		SPRING		SUMMER		AUTUMN	
<i>Viviparus acerosus</i>	1	147	30.1%	31	8%	86	23.1%
	2	282	57.6%	323	83.5%	277	74.5%
	3	60	12.3%	33	8.5%	9	2.4%
<i>Viviparus viviparus</i>	1	83	16.2%	42	18.5%	38	19%
	2	399	77.6%	157	89.2%	126	63%
	3	32	6.2%	28	12.3%	36	18%
<i>Physa acuta</i>	1	29	31.2%	39	36.1%	123	49.8%
	2	60	64.5%	48	44.4%	90	36.4%
	3	4	4.3%	21	19.4%	34	13.8%
<i>Radix ovata</i>	1	29	31.2%	39	36.1%	123	49.8%
	2	60	64.5%	48	44.4%	90	36.4%
	3	4	4.3%	21	19.4%	34	13.8%
<i>Aplexa hypnorum</i>	1	24	36.36%	16	24.24%	6	5.3%
	2	24	36.36%	12	72.72%	81	71.68%
	3	18	27.3%	2	3.03%	26	23%
<i>Radix auricularia</i>	1	9	23.68%	13	50%	25	35.71%
	2	25	65.79%	12	46.15%	39	55.71%
	3	4	10.52%	1	3.84%	6	8.57%

<i>Valvata piscinalis</i>	1	6	16.6%	8	19.1%	5	16.6%
	2	16	44.5%	23	54.7%	20	66.8%
	3	14	38.9%	11	30.2%	5	16.6%
<i>Radix peregra</i>	1	23	54.8%	5	19.2%	7	28%
	2	18	42.9%	20	76.9%	17	68%
	3	1	2.3%	1	3.9%	1	4%
<i>Planorbis planorbis</i>	1	"	"	18	45%	9	30.5%
	2	1	2.22%	22	48.9%	19	42.2%
	3	"	"	"	"	1	100%
<i>Physa fontinalis</i>	1	8	22.2%	8	57.2%	"	"
	2	23	63.8%	4	28.6%	"	"
	3	5	14%	2	14.2%	"	"
<i>Segmentina nitida</i>	1	3	42.8%	2	8.3%	2	22.2%
	2	3	42.8%	21	87.5%	6	66.6%
	3	1	14.4%	1	4.2%	1	11.2%
<i>Stagnicola palustris</i>	1	9	53%	6	46%	3	30%
	2	8	47%	7	54%	6	60%
	3	"	"	"	"	1	10%
<i>Fagotia acicularis</i>	1	13	52%	2	66.6%	"	"
	2	11	44%	1	33.4%	"	"
	3	1	4%	"	"	"	"
<i>Stagnicola corvus</i>	1	7	70%	4	66.6%	"	"
	2	3	30%	2	33.4%	"	"
	3	"	"	"	"	"	"
<i>Fagotia esperi</i>	1	1	33.3%	4	50%	"	"
	2	2	66.7%	3	37.5%	"	"
	3	"	"	1	12.5%	"	"
<i>Anisus spirorbis</i>	1	"	"	"	"	2	40%
	2	"	"	"	"	3	60%
	3	"	"	"	"	"	"
<i>Succinea elegans</i>	1	"	"	"	"	"	"
	2	"	"	2	66.6%	"	"
	3	"	"	1	33.7%	"	"

1 – juvenile age period; 2 – mature age period; 3 – senescent age period

Thus, it can be noticed that most of the populations present an increasing tendency of the number of individuals in the conditions of the ecosystems studied

by us. The rate of young individuals (belonging to the pre-reproductive category), as well as of the mature ones is eloquent (5, 7, 8).

It is also obvious that even if the category of juvenile pre-reproductive individuals predominates, the difference as compared to the other groups (reproductive and senescent individuals) is not as big as it seems; in this case, we can conclude that there is an equilibrium among the different age categories which is an essential factor of a constant, well-balanced increase of the populations.

As a conclusion, it results that there is a close correlation among the body dimensions, age categories, growth rate, and the seasonal presence fact that allows us to estimate the ecological characteristics of the aquatic gastropods populations.

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NOTES UPON THE HERPETOFAUNA OF THE SOUTH-WEST AREA OF THE COUNTY OF CARAȘ-SEVERIN, ROMANIA

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We studied the composition and the geographical distribution of the herpetofauna of the South-West area in the County of Caraș-Severin. We identified 236 new herpetological localities in the South-West of the County of Caraș-Severin for the 26 species and a hybrid form. We identified 12 species of amphibians, hybrids between *Triturus cristatus* and *Triturus dobrogicus* and 14 species of reptiles. Five species of amphibians and four species of reptiles are identified in the investigated region for the first time. Our results indicate the fact that *Testudo hermanni* is spread on a continuous area in the zone of the slopes that are contiguous to the Danube in the County of Caraș-Severin.

1. INTRODUCTION

The herpetofauna of Romania has been little investigated so far. According to Ghira and his collaborators, the data about its composition and geographical distribution prior to the year 2000 is only 5% of the real situation [9]. The herpetofauna of the County of Caraș-Severin was little investigated. Data about the herpetofauna in this region appear in volumes of “The Fauna of The People’s Republic of Romania” [11, 14], in the monograph of the amphibians [8], in the monograph referring to the Iron Gates [13], and in some scientific papers [18, 19]. The most recent study about this region refers only to the turtle [22]. We decided to study the composition and the geographical distribution of the herpetofauna of the South-West region of the county of Caraș-Severin because of the precariousness of the prior data.

2. MATERIAL AND METHODS

The investigated area lies in the basins of the Danube, the Nera and the Caraș and it comprises a highland area represented by Locvei Mountains and a lowland area made out of the plain in the inferior area of the Caraș basin [2]. The investigated region stretches to the South and to the West up to the border with Serbia and to East it reaches the limit of the County of Mehedinți. Our study needed repeated fieldtrips during several years (2002–2004). We used the method of

transecting [7] in order to map the herpetofauna, organizing several fieldtrips in each investigated locality. The informations gathered from the local people served only as guidance. The animals were identified mainly directly and their capture was not necessary; we used the method of direct observation [6]. When their capture of some individuals was needed, this was done by hand. In the case of amphibians, during the aquatic period the capture was done using a square drag or round nets at the end of long iron rods. After identification, the captured animals were released. We also used dead animals in the inventory, killed either by the local people or by cars. For each assessed species we made up a list with the localities where it was found. The final list comprises the newly identified localities, as we met in a certain locality in the territory several species of amphibians and reptiles.

3. RESULTS AND DISCUSSIONS

We identified 25 species of the herpetofauna and a category of hybrids between two species of amphibians. Among the 25 species, 12 are of amphibians (*Salamandra salamandra*, *Triturus cristatus*, *Triturus vulgaris*, *Bombina bombina*, *Bombina variegata*, *Bufo bufo*, *Bufo viridis*, *Hyla arborea*, *Pelobates fuscus*, *Rana ridibunda*, *Rana dalmatina* and *Rana temporaria*) (Table 1). We identified a hybrid population between *Triturus cristatus* and *Triturus dobrogicus*. The reptiles are represented by 14 species (*Testudo hermanni*, *Emys orbicularis*, *Lacerta agilis*, *Lacerta viridis*, *Podarcis muralis*, *Podarcis taurica*, *Anguis fragilis*, *Ablepharuskitabelii*, *Natrix natrix*, *Natrix tessellata*, *Elaphe longissima*, *Coluber caspius*, *Coronella austriaca* and *Vipera ammodytes*) (Table 2). Five species of amphibians are signaled for the first time in the investigated region (*Salamandra salamandra*, *Triturus cristatus*, *Bombina variegata*, *Bufo bufo*, *Rana temporaria*). *Triturus cristatus* was identified for the first time in the County of Caraș-Severin. The discovery of a hybrid population between *Triturus cristatus* and *Triturus dobrogicus* is also a premier. Four species among the reptiles were identified for the first time in the investigated region: *Emys orbicularis*, *Anguis fragilis*, *Ablepharus kitabelii* and *Coronella austriaca*.

Following our study, we identified 226 localities for the 25 species and a hybrid form in the South-West of the County of Caraș-Severin. The situation of the distribution of these species is presented in Table 1, next to the mentioning of the status of their signaling (premier signaling, species first signaled and found or not found through our investigations).

Salamandra salamandra (Linnaeus, 1758) is signaled in premier in this region and was first known in the contiguous sector of the Danube in the County of Mehedinți [13]. The salamander is present in the region we investigated mainly

Table 1
The identified amphibian species in the region studied

Species	S	T	T	T	B	B	Bf	B	H	P	R	R	R
Locality	s	c	x	v	b	v	b	f	a	f	r	d	t
Bazias	-	-	-	-	X	-	X	X	X	-	X	X	-
Belobreșca	-	-	-	-	-	-	X	X	-	-	X	X	-
Berliște	-	-	-	-	-	-	-	-	-	-	X	-	-
Berzasca	-	-	-	-	-	-	X	-	-	-	X	-	-
Câmpia	-	-	-	-	-	-	-	-	-	-	-	-	-
Ciuchici	-	-	-	-	-	-	-	-	-	-	X	-	-
Ciortea	-	-	-	-	X	-	X	-	-	-	X	X	-
Cornea	-	-	-	-	-	-	-	-	-	-	-	-	-
Coronini	X	-	-	-	-	-	X	-	-	-	X	X	-
Cozla	-	-	-	-	-	-	X	-	X	-	X	-	-
Crușovița	-	-	-	-	-	-	-	-	-	-	-	-	-
Dalboșet	-	-	-	-	-	-	-	-	-	-	-	-	-
Divici	-	-	-	-	-	-	X	X	-	-	X	X	-
Greoni	-	X	X	X	X	-	X	-	X	X	X	X	-
Iam	-	-	-	-	-	-	-	-	-	-	X	-	-
Leșcovița	-	-	-	-	-	-	X	-	-	-	X	X	-
Liborajdea	X	-	-	-	-	-	X	-	-	-	X	X	-
Liubcova	-	-	-	-	-	X	X	-	-	-	X	X	-
Macoviște	-	-	-	-	-	-	-	-	-	-	-	-	-
Măcești	-	-	-	-	-	-	X	-	-	-	X	X	-
Mercina	-	-	-	-	X	-	X	-	-	-	X	X	-
Milcoveni	-	-	-	-	-	-	-	-	-	-	X	-	-
Moldova Nouă	X	-	-	-	-	X	X	-	-	-	X	X	X
Moldova Veche	-	-	-	-	-	-	X	-	-	-	X	X	-
Moldovița	X	-	-	-	-	X	X	-	-	-	-	-	X
Naidăș	X	-	-	-	-	-	X	-	-	-	X	X	-
Nicolinț	-	-	-	-	-	-	-	-	-	-	X	-	-
Pârnea	-	-	-	-	-	-	-	-	-	-	-	-	-
Pojejena	-	-	-	-	-	-	X	X	X	-	X	X	-
Radimna	X	X	-	-	-	-	X	X	-	-	X	X	-
Răcășdia	-	-	-	-	-	-	-	-	-	-	X	-	-
Rusova Nouă	-	-	-	-	-	-	-	-	-	-	X	-	-
Rusova Veche	-	-	-	-	-	-	-	-	-	-	X	-	-
Sf. Elena	-	-	-	-	-	-	-	-	-	-	-	-	-
Sichevița	-	-	-	-	-	-	X	-	-	-	-	-	-
Socol	-	-	-	-	X	-	X	X	X	-	X	X	-
Șușca	-	-	-	-	-	-	X	X	X	-	X	X	-
Ticvanu Mare	-	-	-	-	-	-	-	-	-	-	-	-	-
Vărădia	-	X	-	X	X	-	-	-	X	-	X	X	-
Vrani	-	-	-	X	X	-	X	-	-	-	X	X	-
Zlatița	-	-	-	-	-	-	-	-	-	-	X	-	-
Total no. of localities	7	3	1	3	7	3	23	7	7	1	32	20	2

(Ss=*Salamandra salamandra*, Tv=*Triturus vulgaris*, Tc=*Triturus cristatus*, Tx=*Triturus cristatus* x *Triturus dobrogicus*, Bb=*Bombina bombina*, Bv=*Bombina variegata*, Bf.b=*Bufo bufo*, Bf.v=*Bufo viridis*, Ha=*Hyla arborea*, Pf=*Pelobates fuscus*, Rr=*Rana ridibunda*, Rd=*Rana dalmatina*, Rt=*Rana temporaria*).

Table 2

The identified reptilian species in the region studied

Species	E	T	L	L	P	P	A	E	A	C	N	N	V	C
Locality	o	h	a	v	m	t	f	l	k	a	n	t	a	c
Bazias	-	X	-	X	X	X	-	-	-	-	X	X	-	X
Belobreșca	-	X	-	X	X	X	-	-	-	-	X	X	-	-
Berliște	-	-	-	-	-	-	-	-	-	-	X	-	-	-
Berzasca	-	-	-	X	-	-	-	-	-	-	X	X	-	X
Câmpia	-	-	-	X	-	-	-	-	-	-	-	-	-	-
Ciuchici	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Ciortea	-	-	X	-	-	-	-	-	-	-	-	-	-	-
Cornea	-	X	-	X	-	-	-	-	-	-	X	X	-	-
Coronini	-	-	-	X	X	-	X	-	-	-	X	X	X	-
Cozla	-	X	-	X	X	-	X	X	X	-	-	X	X	X
Crușovița	-	X	-	X	-	-	-	-	-	-	X	X	-	-
Dalboșet	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Divici	-	X	-	X	X	X	X	-	-	-	X	X	-	X
Greoni	-	-	-	-	-	-	-	-	-	-	X	-	-	-
Iam	-	-	X	-	-	-	-	-	-	-	-	-	-	-
Leșcovița	-	-	-	X	X	-	-	X	-	-	-	-	-	-
Liborajdea	-	-	-	X	X	-	X	X	-	-	X	X	X	-
Liubcova	-	-	-	X	-	-	-	-	-	-	X	X	-	-
Macoviște	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Măcești	-	-	-	-	-	-	-	-	-	-	X	X	-	-
Mercina	-	-	X	-	-	-	-	-	-	-	-	-	-	-
Milcoveni	-	-	-	-	-	-	-	-	-	-	X	-	-	-
Moldova Nouă	-	-	-	X	X	-	X	X	-	-	X	-	X	-
Moldova Veche	-	-	-	-	-	-	-	-	-	-	X	X	-	-
Moldovița	-	-	-	X	X	-	X	X	-	-	-	-	-	-
Naidăș	-	-	X	X	X	-	-	X	-	X	X	-	-	-
Nicolinț	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Pârneaurea	-	-	-	X	-	-	-	-	-	-	-	-	-	-
Pojejena	-	-	-	X	-	-	-	-	-	-	X	X	-	-
Radimna	-	-	-	X	X	-	X	X	-	X	X	X	-	-
Răcășdia	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Rusova Nouă	-	-	X	-	-	-	-	-	-	-	-	-	-	-
Rusova Veche	-	-	X	-	-	-	-	-	-	-	X	-	-	-
Sf. Elena	-	-	-	X	X	-	-	-	-	-	-	-	X	-
Sichevița	-	X	-	X	-	-	-	-	-	-	-	X	-	-
Socol	-	-	-	X	-	-	-	-	-	-	X	-	-	-
Șușca	-	-	-	X	X	-	-	-	-	-	X	X	-	-
Ticvaniu Mare	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Vărădia	-	-	-	-	-	-	-	-	-	-	X	-	-	-
Vrani	-	-	X	-	-	-	-	-	-	-	-	-	-	-
Zlatița	X	-	-	X	X	-	-	X	-	-	-	-	-	-
Total no. of localities	1	7	7	23	14	3	7	8	1	2	23	16	5	4

(Eo=*Emys orbicularis*, Th=*Testudo hermannii*, La=*Lacerta agilis*, Lv=*Lacerta viridis*, Pm=*Podarcis muralis*, Pt=*Podarcis taurica*, Af=*Anguis fragilis*, Ak=*Ablepharus kitaibelii*, Nn=*Natrix natrix*, Nt=*Natrix tessellata*, El=*Elaphe longissima*, Ca=*Coronella austriaca*, Va=*Vipera ammodytes*, Cc=*Coluber caspius*).

in the forested areas of the Locvei Mountains. Although it is more frequently met in the beech forests [13], place where we identified as well most of the individuals, the species descends a lot, reaching the close proximity of the Danube following the valleys that drain in it.

Triturus cristatus (Laurenti 1768) is as well signaled in premier in the South-West of the County of Caraș-Severin where it is rare. Although it is a species that is attached to high areas [10], it is extremely rare in the Locvei Mountains as we met it only in the area of the locality of Radimna. It is better represented in the basin of the Caraș River. According to the data in the literature [8], *Triturus cristatus* was identified by us in premier in the County of Caraș-Severin.

Triturus cristatus X *Triturus dobrogicus*. We met hybrids between the two species of crested newts in only one locality, at Greoni. The hybrids populate a system of swamps and deep puddle areas with vegetation. The hybrids between *Triturus cristatus* and *Triturus dobrogicus* are signaled in premier in the investigated region and in the County of Caraș-Severin on the whole.

Triturus vulgaris (Linnaeus, 1758) is a rare species in the investigated region, as we met it in just three localities situated in the basin of the Caraș River.

Bombina bombina (Linnaeus, 1761) was first signaled in the Bazias region [8, 11]. We identified it in that area, but we also signal it in premier in the basin of the Caraș River. *Bombina bombina* is characteristic to plain areas in the East and Center of Europe [1], and yet, at Bazias comes to populate under-mountainous regions with rocks. It populates large permanent puddles in the plain areas [20]; we met it in this type of habitats in the basin of the Caraș River. It is also present in the area in the canals on the edges of the roads. But, at Bazias, appears in different habitats, as we met it in the divagations of the Danube in close proximity of the abrupt and rocky walls that edge the river to the North. These habitats with rocky under-layer, usually, do not have any vegetation and are flooded by the Danube.

Bombina variegata (Linnaeus, 1758) is a species identified in premier in the investigated area, where it is relatively rare. Numerous exemplars show up in the streams in the high areas of the Locvei Mountains. The species descends along the arms of the Danube down to the close proximity of the river. *Bombina variegata* is present at some tens of meters of the Danube, a situation first signaled in the Mehedinți sector of the defile [13]. It does not show up in the low areas in the neighborhood of the Danube next to *Bombina bombina*.

Bufo bufo (Linnaeus, 1758) is signaled in premier in the investigated area where it is a common species. The number of individuals we met is high, but the largest part of them is the dead bodies we identified on the roads, victims of the traffic. They die by traffic because of their affinity for the biotope of reproduction [16], as they cross the roads in the spring while they move towards their spawning

habitats. The situation applies especially in the close proximity of the Danube where the reproduction habitats are close to the river and after the reproduction the animals live in the forested areas of the Locvei Mountains. A road where during spring thousands of bodies can be found intercedes the two habitats.

Bufo viridis (Laurenti, 1768) was first signaled in the region and we met it only in the contiguous area of the Danube. It is more rare than the precedent species although it is more thermophile [24] and it should be thriving in the investigated area because it has a higher thermic regime [23].

Hyla arborea (Linnaeus, 1758) was first signaled in the region. We met it in both the close proximity to the Danube and the Caraș basin, but the number of localities is relatively reduced, although there are plenty favorable habitats. This fact is probably due to the life style of the species that is arboreal most of their activity period and present on the soil only during the spring, in the mating season and in the fall [17].

Pelobates fuscus (Laurenti, 1768) is a rare species in the investigated region and we identified it in only one locality. This fact is probably due to their nocturnal life style [12].

Rana ridibunda Pallas, 1771 is the most common amphibian species in the investigated region. It is present both along the Danube and in the basins of the Nera and Caraș Rivers. It is spread in both the plain area in the alluvial plain of the Caraș River and in the contiguous high areas of the Danube. It is present on both the shores of the Danube and on its arms going upstream a lot. None of the investigated exemplars showed any of the characteristics of *Rana lessonae* species, signaled upstream, in the Dubova area [13], or of its hybrid, *Rana Kl. esculenta* [3, 4, 5]. Our results show the fact that *Rana ridibunda* is the only species of the group of green frogs in the investigated region, the other two preferring colder regions [25].

Rana dalmatina Bonaparte, 1839 is a common species in the investigated region.

Rana temporaria (Linnaeus, 1758) is a species we signaled in premier in the area. We met the species in only two localities, as it is spread in the high area of the Locvei Mountains

Emys orbicularis (Linnaeus, 1758) is a new species in the investigated region. The water turtle is rare in the South-West of the County of Caraș-Severin and we identified it in only one locality.

Testudo hermanni (Gmelin, 1798) is a species known before only from the sector of the Danube in the County of Mehedinți [14] and it was recently signaled in the Divici-Baziaș area [22]. Compared to this data, we noticed a wider distribution of the species in the region. What is important is the discovery of the turtle in the

Șichevița region. According to prior data the species seemed to be spread in the proximity of the Danube on a disjunctive area, on one side at Divici-Baziaș and at the Șichevița region on the other side. The populations at Șichevița fill this gap, suggesting a continuous distribution of the species on the slopes that edge the Danube both in the County of Caraș-Severin and in the County of Mehedinți. *Testudo hermanni* is a widely spread species in the bordering region of the Danube and the number of exemplars we investigated is large, especially in the month of May. The habitats where we met turtles correspond to the one indicated in the literature [22], the species is present in grassy areas, but it also populates forested areas.

Ablepharus kitaibelii (Bibron & Bory, 1833) was first identified in the forests nearby Cozla.

Lacerta agilis (Linnaeus, 1758) is a relatively rare species in the investigated region and is exclusively present in the hydrographic basins of the Nera and Cerna rivers. Although it was first signaled at Baziaș [14], we did not meet it nearby the Danube. This fact is probably due to the ecological demands of the species that prefers regions with a high level of humidity.

Lacerta viridis (Laurenti, 1768) is common in the investigated region.

Podarcis muralis (Laurenti, 1768) is also a common species in the investigated region, although we identified it in a more reduced number of localities than the precedent. We met it only in the abutting zone of the Danube and of the Nera River. It mainly populates abrupt, rocky mountain slopes, but it appears in the sand areas in the close proximity of the Danube. It is met together with *Podarcis taurica* in this kind of habitat. We met even a melanistic individual at Divici.

Podarcis taurica (Pallas, 1814) is a rare species in the region and it was first signaled in only one locality, at Baziaș [14, 21]. We re-identified it at Baziaș but we also met it in premier in other two localities: at Divici and at Belobreșca. We did not identify it more eastwards than Belobreșca. It populates mainly sandy habitats, covered exclusively with grassy vegetation like in other areas [9]. It also appears in regions where the under-layer is not sandy, but it has only grassy vegetation. The number of identified exemplars is reduced.

Anguis fragilis (Linnaeus, 1758) is a species identified in premier in the investigated region and is relatively well represented in the forested areas of the Locvei Mountains.

Coluber caspius (Gmelin 1789) was first identified at Baziaș [14], but we met it in other three localities. *Coluber caspius* is spread in the uncovered areas on the slopes in close proximity to the Danube, probably on a whole sector of the Danube in Caraș-Severin.

Elaphe longissima (Laurenti, 1768) was first signaled in the Baziaş region [14]. We met it in the forested area of the Locvei Mountains where it is common, being spread in numerous localities. Unlike the precedent species, we did not meet at all in the dry areas close to the Danube, as it is present at some distance from it, in forests with a higher level of humidity.

Coronella austriaca (Laurenti 1768) is a species that was signaled in premier in the investigated region. *Coronella austriaca* is rare in the area and we met it in only two localities in forested regions belonging to the Locvei Mountains.

Natrix natrix (Linnaeus, 1758) was first signaled in the Baziaş region [14]. We identified it in numerous other localities in the investigated region (Table 1). It appears both in the close proximity of the Danube, where the most of exemplars can be met and in the hydrographic basins of Nera and Caraş rivers. It populates a large variety of habitats being present along the Danube, in the rocky areas on its shore and on the arms of the Danube in forested areas. In the hydrographic basin of the Caraş River it appears in swampy areas or in the neighborhood of the canals on the side of the roads.

Natrix tessellata (Laurenti, 1768) was, like the precedent species, first signaled in the Baziaş region [14]. Following our studies we identified the water snake in other 15 localities, as it is a well-represented species in the area. It appears only in the close proximity of the Danube. Most of the individuals can be met in the Danube but they also go upstream along its arms.

Vipera ammodytes (Linnaeus, 1758) was first signaled in the region at Moldova Nouă and at Baziaş. We re-identified it at Moldova Nouă and added two new localities. It is a well-represented species in the area of the abrupt landscape in the proximity of the course of the Danube.

4. CONCLUSIONS

Salamandra salamandra reaches the immediacy of the Danube, as it is present in the streams that flow in the river. *Triturus cristatus* is signaled in premier for this region, as it is rare. *Bombina variegata* is 20 meters close to the Danube, populating the streams and the gills. *Bufo bufo*, *Rana ridibunda* and *Rana dalmatina* are widely distributed in the area. *Lacerta agilis* is rare in the region and we met it only in the wet areas close to the Nera River. *Lacerta viridis* and *Podarcis muralis* are common in the investigated region. Our results indicate the fact that *Testudo hermanni* is distributed on a continuous area in the zone of the slopes that are contiguous to the Danube in the County of Caraş-Severin.

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BIOGEOGRAPHICAL DISTRIBUTION OF GAMASINA MITES FROM ROMANIA (ACARI-MESOSTIGMATA)

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The paper presents a list of the gamasina mites (Acari; Mesostigmata) from Romania with species of two suborders: Epicriina and Gamasina. The order Epicriina, family Epicriidae, has 6 species. The suborder Gamasina has the following families: Parasitidae, Veigaiidae, Ameroseiidae, Aceoseiidae, Phytoseiidae, Antennoseiidae, Podocinidae, Rhodacaridae, Ologamasidae, Macrochelidae, Pachylaelaptidae, Laelaptidae, Eviphididae, Haemogamasidae, Zerconidae; with 58 genera and 273 species. A number of 34 species are identified only for Romania.

These species are found in the Eastern area of Romania in 26 regions, in the South area in 14 regions, in the West area in 24 regions, in the North area in 6 regions and in the central area in 20 regions.

Key words: Mesostigmata; Gamasina; mites; microarthropod; habitat.

INTRODUCTION

The Mesostigmata (Arachnida; Acari) is a large and successful group of acarines which occupies a variety of habitats. While the majority of mesostigmatid species are predators (Parasitidae), many are external or internal parasites of mammals, birds, reptiles or invertebrates (Haemogamasidae; Laelapidae). The order Mesostigmata is considered to comprise the following 12 suborders: Antennophorina, Arctacarina, Cercomegistina, Dermanyssina, Diarthrophallina, Epicriina, Heterozerconina, Microgyniina, Gamasina, Sejina, Uropodina and Zerconina with 60 families.

METHODS

The paper presents an inventory of gamasina mites from areas of Romania, (pointed by Roman signs such as: I=North area of Romania, II=East area, III=South area, IV=West area, V=Central area) and every taxa was classified and synonymized, according to the taxonomical references (1, 6, 11, 12, 25). The species are followed by a number corresponding to the bibliography (1-24). Every species pointed by order number of the localities and geographic regions of Romania are followed by the bio-geographical distribution:

- I. 2=Oașului Mountains; 3=Gufii Mountains; 4=Maramureș Region; 5=Maramureș Mountains; 6=Tibleș Mountains; 7=Rodnei Mountains.
- II. 1=Oriental Carpathians; 8=Moldavia; 9=Obcina Feredeului; 10=Obcina Mare; 11=Moldavia Plateau; 12=Dealul Mare; 13=Dealul Bour; 14=Iași Regions; 15=Suceava Regions; 18=Stânișoarei Mountains; 19=Rarău and Giumalău Peaks; 20=Iași Meadow; 21=Central Plateau of Moldavia; 22=Cîlnăului Meadow; 23=Ceahlău Mountains; 25=Ciuc Mountains; 27=Nemira Mountains; 28=Vrancea Mountains; 29=Penteleu Peak; 74=Dobrogea Region; 75=Măcin Mountains; 76=Babadag Plateau; 77=Galați Regions; 79=the Danube Delta; 80=Miercurea Ciuc Mountain; 82=Poiana Stampei; 86=Gura Humorului Region.
- III. 39=Sinaia Region; 40=Bucegi Mountains; 41=Baiului Mountains; 42=Ploiești Region; 43=Dîmbovița Region; 44=Argeș Region; 45=Lotrului Mountains; 51=Gorj Valley; 78=Bărăgan Region; 83=Râmnicu-Vâlcea Valley; 84=Oltului Valley; 85=Prahova Valley; 87=Gârbova Mountains.
- IV. 24= Tarcului Mountains; 49=Rețezat Mountains; 50=Vîlcanului Mountains; 52=Cerna Mountains; 53=Hateg Valley; 54=Banat Region; 55=Mehedinți Mountain; 56=Mehedinți Plateau; 57=Caraș-Severin Valley; 58=Herculane; 59=Porțile de Fier; 60=Anina Mountains; 61=Almașului Mountains; 62=Semenic Mountains; 63=Sebeș Mountains; 64=Apuseni Mountains; 65=Trascăului Mountains; 66=Bihor Mountains; 67=Vlădeasa Peak; 68=Muntele Mare; 69=Ciceului Hills; 70=Timiș Region; 71=Timiș Meadow.
- V. 16=Călimani Mountains; 17= Bistriței Mountains; 26=Harghita Mountains; 30=Siriului Mountains; 31=Meridionali Carpathians; 32=Ciucăș Mountains; 33=Roșu Mountains; 34=Brașov Region; 35=Postăvaru Mountain; 36=Făgăraș Mountain; 37=Perșani Mountains; 38=Piatra Craiului Mountains; 46=Cibinului Mountains; 47=Căpățânei Mountains; 48=Parîng Mountains; 72=Transilvania Plateau; 73=Cluj Region; 81=Porțile de Fier ale Transilvaniei; 88=Harghita Region.

RESULTS AND DISCUSSION

The specific diversity shows the presence of two suborders: Epicriina and Gamasina. The suborder Epicriina has only 1 family: Epicriidae with 1 genus and 6 species. The suborder Gamasina has the following families: Parasitidae, Veigaiidae, Ameroseiidae, Aceoseiidae, Phytoseiidae, Antennoseiidae, Podocinidae, Rhodacaridae, Ologamasidae, Macrochelidae, Pachylaelaptidae, Pseudolaelaptidae, Laelaptidae, Eviphididae, Haemogamasidae, Zerconidae; with 67 genera and 273 species, known till now in Romania. They represent 42.66% of the total number of gamasina mites in the world.

These species show the following biogeographical distribution: 51.12% Europe; 21.40% Central Europe; 8.98% Palearctic region; 4.49% East Europe;

3.93% Holarctic region; 2.24% East and Central Europe; 2.24% West Europe; 2.24% South Europe; 1.68% South and Central Europe; 1.12% North and Central Europe; 0.56% North Europe; 0.56% Europe and Australia; 0.56% Central and West Europe; 1.12% California, North Africa, South America, Japan. A number of 34 species are identified by Dr. Ilinca Juvara-Balș, for Romania only.

Ten species are present in the highest number of regions: *Pergamasus kelemeneus* in 27 regions, *Pergamasus dimitrescui* in 16 regions, *Amblygamasus mirabilis*, *Ernogamasus simplex*, *Leptogamasus suecicus*, *Leptogamasus tectegynellus*, *Pergamasus primorellus*, *Lasioseius berlesei*, *Podocinum pacificum*, *Zercon schweizeri*, each species being present in 15 regions, as listed below.

PARASITIFORMES REUTER, 1909

MESOSTIGMATA CANEASTRINI, 1891

Suborder Epicriina Berlese, 1885

Family Epicriidae, 1885

Genus Epicrius G. Caneastrini and Fanzago, 1877

Epicrius kargi Solomon, 1978 (33): 11; 14; 66; – South Europe

Epicrius resinae Karg, 1871 (9; 33): 5; 14; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe

Epicrius stellatus Balogh, 1959 (9; 10): 49; 65; 66; – South Europe

Epicrius mollis Kramer, 1876 (9; 10): 5; 14; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Epicrius bureschi Balogh, 1959 (9; 33): 5; 14; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Epicrius magnus Solomon, 1984 (34): 11; – Romania.

Suborder Gamasina Leach, 1815

Family Parasitidae Oudemans, 1901

Subfamily Pergamasinae Juvara-Bals, 1972

Genus Amblygamasus Berlese, 1903

Amblygamasus mirabilis Willmann, 1951 (9; 19): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Central Europe.

Amblygamasus hamatus C.L.Koch, 1839 (19): 73; – Europe.

Amblygamasus romanicus Juvara-Bals, 1976 (19): 54; 60; – Europe.

Genus Ernogamasus Athias-Henriot, 1971

Ernogamasus leruthi Cooreman, 1951(9; 24): 16; 49; 65; – Europe.

Ernogamasus decui Juvara-Bals, 1974 (17): 55; – Romania.

Ernogamasus simplex Juvara-Bals, 1974 (17): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Romania.

Genus Holoparasitus Oudemans, 1936

- Holoparasitus calcaratus* C.L.Koch, 1839 = *Holoparasitus excipuliger* s. Karg, 1971(18): 49; 66; – Europe.
- Holoparasitus ampullaris* Witalinski, 1994 (18): 36; 88; – Europe.
- Holoparasitus fortunatus* Juvara-Bals, 1975 (18): 36; 38; 48; 56; – East Europe.
- Holoparasitus tuberculatus* Juvara-Bals, 1975 (18): 31; 48; 68; – East Europe.
- Holoparasitus caesus* Micherzinski, 1969 (9): 16; – Central Europe.
- Holoparasitus rotulifer* Willmann, 1940 = *Ologamasus rotulifer*; Willmann 1941 (9): 5; 53; 65; 66; 67; – South Europe.
- Holoparasitus minimus* Holzmann, 1969 (9; 10): 16; 65; 66; – Central Europe.
- Holoparasitus excisus* Berlese, 1905 (18): 1; 31; 36; – Europe.
- Holoparasitus cornutus* Juvara-Bals & Witalinski, 2000 (22): 32; 63; 85; 87; – Central Europe.
- Genus *Leptogamasus* Tragardh, 1936
- Leptogamasus suecicus* Tragardh, 1936 (9; 22): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Central Europe.
- Leptogamasus tectegynellus* Athias-Henriot, 1967 = *Pergamasus tatrensis* Micherzinski, 1969 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Central Europe.
- Leptogamasus alstoni* Bhattacharyya, 1963 (22): 16; – Europe.
- Leptogamasus (Holoperigamasus) tabacarui* Juvara-Bals, 2003 (24): 83; – Romania.
- Leptogamasus doinae* Juvara-Bals, 1981 (22): 33; 51; 55; 56; 66; – Romania.
- Leptogamasus orghidani* Juvara-Bals, 1981 (22): 33; 51; 55; 56; 66; – Romania.
- Leptogamasus margaretae* Juvara-Bals, 1981 (22): 33; 51; 55; 56; 66; – Romania.
- Leptogamasus motrensis* Juvara-Bals, 1981 (22): 33; 51; 55; 56; 66; – Romania.
- Leptogamasus paracarpaticus* Juvara-Bals, 1981 (22): 33; 51; 55; 56; 66; – Romania.
- Leptogamasus variabilis* Juvara-Bals, 1981 (22): 33; 51; 55; 56; 66; – Romania.
- Genus *Mixogamasus* Juvara-Bals, 1972
- Mixogamasus intermedius* Juvara-Bals, 1972 (8): 51; 66; – Romania.
- Genus *Ologamasiphis* Athias-Henriot, 1971
- Ologamasiphis (Holzmannia) aff. disfistulatus* Athias-Henriot, 1967 (23): 79; – Europe.
- Genus *Paragamasus* Hull, 1918
- Paragamasus (Paragamasus) diversus* Halbert, 1915 (8): 16; 65; – Europe.
- Paragamasus (Paragamasus) robustus* Oudemans, 1902 = *Parasitus longulus* var. *robustus* Oudemans, 1902 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
- Paragamasus (Aclerogamasus) alpeastris* Berlese, 1904 (9; 20): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.

- Paragamasus (Aclerogamasus) bicalliger* Athias-Henriot, 1967 (20): 51; – Europe.
- Paragamasus (Aclerogamasus) motasi* Juvara-Bals, 1977 (20): 35; 36; – Romania.
- Paragamasus (Anidogamasus) misellus* Berlese, 1904 = *Pergamasus brevipes* Micherzinski, 1969 (4, 9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
- Paragamasus (Anidogamasus) armatus* Halbert, 1915 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
- Paragamasus (Anidogamasus) schweizeri* Bhattacharyya, 1963 (8): 16; 65; – Europe.
- Paragamasus (Anidogamasus) wasmanni* Oudemans, 1902 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
- Paragamasus (Anidogamasus) vagabundus* Karg, 1968 = *Pergamasus mniophilus* Micherzinski, 1969 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
- Paragamasus (Dyogamasus) puerilis* Karg, 1963 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
- Paragamasus crassipilis* Holzmann, 1969 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe. – Incertae sedis.
- Genus *Pergamasus* Berlese, 1904
- Subgenus *Triadogamasus* Athias-Henriot, 1971
- Pergamasus caneastrinii* Berlese, 1905 (19): 84; 85; 86; – Europe.
- Pergamasus dacicus* Juvara-Bals, 1976 (19): 51; 55; – Romania.
- Subgenus *Thenargamasus* Athias-Henriot, 1971
- Pergamasus theseus* Berlese, 1903 (19): 16; 83; – Europe.
- Pergamasus quisquiliarum* R. and G. Caneastrini, 1882 (19): 5; – Europe; South America.
- Pergamasus barbarus* Berlese, 1904 = *Pergamasus viator* Halaskova 1959; s. Micherzinski, 1969 (19): 37; 52; 57; – Central Europe.
- Pergamasus septentrionalis* Oudemans, 1902 = *Pergamasus germanicus* Berlese, 1906 (9; 10): 65; 73; – Central Europe.
- Pergamasus inexpectatus* Juvara-Bals, 1976 (19): 19; 26; – Romania.
- Subgenus *Pergamasus* Berlese, 1904
- Pergamasus athiasae* Juvara-Bals, 1970 (14): 36; 38; 40; – Romania.
- Pergamasus dimitrescui* Juvara-Bals, 1970 (14): 3; 18; 19; 21; 27; 28; 32; 36; 38; 40; 45; 46; 50; 55; 60; 62; – Romania.
- Pergamasus laetus* Juvara-Bals, 1970 (14; 16; 19): 26; 32; 36; 40; 41; 45; 47; – Romania.
- Pergamasus buerebistai* Juvara-Bals, 1973 (10; 19): 40; 52; 55; 57; 60; 61; 63; 69; 71; – Romania.
- Pergamasus biharicus* Juvara-Bals, 1973 (10; 19): 65; 66; 67; – Romania.
- Pergamasus decebali* Juvara-Bals, 1973 (19): 47; 48; 50; 65; 66; 67; – Romania.

Pergamasus scorilai Juvara-Bals, 1973 (19): 24; 49; 50; 52; 55; 57; 60; 62; 63; 66; 67; – Romania.

Pergamasus primorellus Athias-Henriot, 1967 (19): 3; 10; 12; 13; 20; 21; 22; 24; 45; 46; 49; 71; 75; 76; 79; – Hungary.

Pergamasus crassipes Linne, 1758 (9): 3; 16; 40; 49; 65; 66; 73; 74; – Holarctic Region.

Pergamasus adinae Juvara-Bals, 1970 (16; 19): 7; 9; 10; 16; – Romania.

Pergamasus brevicornis Berlese, 1903 (19): 2; 5; 9; 10; 12; 18; 19; 25; 26; 36; 66; – Europe.

Pergamasus beklemischevi Sellnick, 1929 (19): 9; 16; 17; 18; 23; 26; 27; 28; 29; 30; 32; 40; 41; – East and Central Europe.

Pergamasus kelemeneus Athias-Henriot, 1967 (19): 5; 6; 7; 9; 10; 16; 17; 18; 20; 21; 23; 26; 27; 36; 37; 38; 45; 46; 48; 49; 52; 53; 54; 62; 65; 66; 67; – Europe.

Pergamasus tuberopalpus Juvara-Bals, 1976 (19): 2; – Romania.

Subfamily Parasitinae Oudemans, 1901

Genus Neogamasus Tichomirov, 1969

Neogamasus islandicus Sellnick, 1940 = *Parasitus islandicus* Sellnick, 1940 (9): 49; – North and Central Europe.

Genus Eugamasus Berlese, 1892

Eugamasus magnus Kramer, 1876 = *Gamasus magnus* s. Berlese, 1892 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Eugamasus berlesei Willmann, 1935 = *Eugamasus magnus* Berlese; 1892 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Eugamasus monticolus Berlese, 1905 = *Eugamasus kobyi* Schweizer; 1961 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Genus Parasitellus Willmann, 1939

Parasitellus fucorum De Geer, 1778 = *Parasitus fucorum* Tragardh, 1910 (5; 9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Europe

Genus Trachygamasus Berlese, 1906

Trachygamasus gracilis Karg, 1965 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Trachygamasus ambulacralis Willmann, 1949 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Genus Poecilochirus G and R. Caneastrini, 1882

Poecilochirus carabi Willmann, 1951 = *Gamasoides eurasiaticus* Tragardh, 1937 (9): 39; 79; – Europe; Asia.

Genus Gamasodes Oudemans, 1939

Gamasodes spiniger Tragardh, 1910 = *Parasitus lunarisimilis* Schweizer, 1961 (4): 73; – Europe.

Genus Parasitus Latreille, 1795

Parasitus furcatus G. and R. Caneastrini, 1882 (4; 9): 16; 49; 66; – Europe.

Parasitus fimentorium Berlese, 1903 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Parasitus numerus Karg, 1965 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe

Parasitus nollii Karg, 1965 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Germany.

Genus Vulgarogamasus Tichomirov, 1969

Vulgarogamasus kraepelini Berlese, 1905 (32): 16; – Europe.

Vulgarogamasus remberti Oudemans, 1912 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.

Vulgarogamasus zschokkei Schweizer, 1922 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Vulgarogamasus oudemansi Berlese, 1903 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Genus Porrhostaspis Müller, 1859

Porrhostaspis lunulata Müller, 1859 = *Eugamasus epsilon* Oudemans et Voigts, 1904 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe

Family Veigaidae Oudemans, 1939

Genus Veigaia Oudemans, 1905

Veigaia nemorensis C.L.Koch, 1839 (9; 10; 32): 16; 49; 65; 66; 73; – Europe; Asia.

Veigaia cerva Kramer, 1876 (9; 10; 32): 16; 49; 65; 66; – Europe; Asia; North America.

Veigaia planicola Berlese, 1892 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Asia.

Veigaia kochi Tragardh, 1901 (9; 10): 16; 49; 65; 66; – Europe; Asia.

Veigaia exigua Berlese, 1917 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.

Veigaia propinqua Willmann, 1936 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Asia.

Veigaia transisalae Oudemans, 1902 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.

Veigaia paradoxa Willmann, 1951 (32): 16; – Central Europe.

Genus Gamasolaelaps Berlese, 1904

Gamasolaelaps multidentatus Karg, 1965 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Germany.

Gamasolaelaps excisus C.L.Koch, 1879 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Asia.

Family Ameroseiidae Berlese, 1919

Genus Ameroseius Berlese, 1903

- Ameroseius plumigerus* Oudemans, 1930 (33): 11; 16; – Europe.
Ameroseius corbiculus Sowerby, 1806 (33): 11; – Europe.
Ameroseius eumorphus Bregetova, 1977 (33): 11; – Russia and Romania.
Ameroseius lanatus Solomon, 1969 (33): 11; – Russia and Romania.
Ameroseius delicatus Berlese, 1918 (33): 11; – Central and East Europe.
Ameroseius plumosus Oudemans, 1902 (33): 11; – Europe; Australia.

Family Aceosejidae Baker and Wharton, 1952

Genus Cheroseius Berlese, 1916

Subgen Cheroseius Berlese, 1816

- Cheroseius viduus* C.L.Koch, 1839 (4): 73; – Central Europe
Cheroseius aequalis Schweizer, 1949 (4): 73; – Central Europe.
Cheroseius feideri Solomon, 1984 (34): 11; – Romania.

Subgen Plesiosejus Evans and Hyatt, 1960

- Plesiosejus major* Halbert, 1923 (4): 73; Central Europe.

Genus Iphidozercon Berlese, 1903

- Iphidozercon gibbus* Berlese, 1903 (31): 14; – Central Europe.

Genus Zerconopsis Hull, 1918

- Zerconopsis remiger* Kramer, 1876 (31): 14; – Central Europe.

Genus Aceoseius Sellnick, 1941

- Aceoseius muricatus* C.L.Koch, 1839 (4): 73; – Central Europe.

Genus Lasioseius Berlese, 1916

- Lasioseius berlesei* Oudemans, 1938 (9): 5; 14; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Europe; Asia; North America.
Lasioseius lawrencei Evans, 1958 (4): 14; 73; – Europe.
Lasioseius youcefi Athias-Henriot, 1959 (31): 14; – Europe.

Genus Leioseius Berlese, 1916

- Leioseius bicolor* Berlese, 1948 (4): 14; 73; – Europe.
Leioseius minusculus Berlese, 1905 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Leioseius magnanalis Evans, 1958 (31): 14; – Central Europe.

Genus Arctoseius Thor, 1930

- Arctoseius brevicheles* Karg, 1969 (31): 14; – Central Europe
Arctoseius venustus Berlese, 1917 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Arctoseius magnanalis Evans, 1958 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Arctoseius sessiluncus Karg, 1965 (27): 65; – Central Europe.
Arctoseius eremitus Berlese, 1918 (27): 14; 73; – South and Central Europe.

- Arctoseius semiscissus* Berlese, 1892 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.

- Arctoseius cetratus* Sellnick, 1940 (31): 14; 16; – Europe.

- Arctoseius dendrophilus* Karg, 1969 (31): 14; – Central Europe.

- Arctoseius minutus* Halbert, 1915 (31): 14; – Central Europe.

- Arctoseius elegans* Bernhard, 1963 (31): 14; – Central Europe.

Genus Melichares Hering, 1838

- Melichares juradeus* Schweizer, 1949 (9): 40; – Russia.

Genus Proctolaelaps Berlese, 1923

- Proctolaelaps pomorum* Oudemans, 1929 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Australia.

- Proctolaelaps pygmaeus* Muller, 1860 (31): 14; 34; – Holarctic Region.

- Proctolaelaps longanalis* Westerboer, 1963 (31): 14; – Europe.

Family Phytoseiidae Berlese, 1916

Genus Amblyseius Berlese, 1904

Subgenus Amblyseius Berlese, 1904

- Amblyseius meridionalis* Berlese, 1914 (33): 11; 74; – Europe.

- Amblyseius obtusus* C.L.Koch, 1839 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.

Subgenus Typhlodromips De Leon, 1965

- Amblyseius massei* Wesbitt, 1951 (33): 11; – Europe; North America.

Subgenus Neoseiulus Hughes, 1948

- Amblyseius reticulatus* Oudemans, 1930 (33): 11; – Europe; North America; Australia.

- Amblyseius foramenis* Karg, 1970 (33): 11; – Central Europe.

Genus Anthoseius De Leon, 1959

- Anthoseius rhenanus* Oudemans, 1905 (33): 11; – Europe.

Family Antennoseiidae Karg, 1965

Genus Antennoseius Berlese, 1916

- Antennoseius bullitus* Karg, 1969 (21): 14; – Central Europe.

- Antennoseius oudemansi* S. Thor, 1930 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – North Europe.

Family Podocinidae Berlese, 1913

Genus Podocinum Berlese, 1882

- Podocinum pacificum* Berlese, 1895 (15): 5; 19; 33; 38; 48; 49; 52; 53; 58; 62; 65; 66; 67; 81; 82; – South Europe; India; Japan; California; South America; North Africa.

Family Rhodacaridae Oudemans, 1902

Genus *Hololaelaps*; Berlese and Troussart, 1889*Halolaelaps punctulatus* Leitner, 1946 (32): 16; – Central Europe.Genus *Leitneria* Evans; 1957*Leitneria granulata* Halbert, 1923 (32): 14; – Central and West Europe.Genus *Asca* Heyden, 1826*Asca bicornis* Caneastrini and Fanzago, 1887 (9; 10; 26): 14; 16; 65; – Europe.*Asca aphidoides* Linne, 1758 (9; 10): 5; 14; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.Genus *Gamasellus* Berlese, 1892*Gamasellus montanus* Willmann, 1936 (9): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.*Gamasellus falciger* G. and R. Caneastrini, 1881 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.Genus *Dendroseius* Karg, 1965*Dendroseius reticulatus* Sheals, 1956 (33): 11; – Europe and North Africa.Genus *Dendrolaelaspis* Lindquist, 1975*Dendrolaelaspis angulosus* Willmann, 1936 (33): 11; – Europe.Genus *Dendrolaelaps* Halbert, 1915*Dendrolaelaps septentrionalis* Sellnick, 1958 (33): 11; 16; – Europe.*Dendrolaelaps brevipilis* Leitner, 1949 (33): 11; – Central Europe.*Dendrolaelaps rectus* Karg, 1962 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.*Dendrolaelaps sellnicki* Hirschmann, 1960 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.Genus *Punctodendrolaelaps* Hirschmann and Wisniewski, 1982*Punctodendrolaelaps strenzkei* Hirschmann, 1960 (33): 11; – Europe.*Punctodendrolaelaps arvicolus* Leitner, 1949 (33): 11; – Central Europe.*Punctodendrolaelaps rotundus* Hirschmann, 1960 (33): 11; – Europe.*Punctodendrolaelaps latior* Leitner, 1949 (33): 11; – Central Europe.Genus *Cornodendrolaelaps* Hirschmann and Wisniewski, 1982*Cornodendrolaelaps cornutulus* Hirschmann, 1960 (33): 11; – Central Europe.Genus *Rhodacarellus* Willmann, 1935*Rhodacarellus perspicuus* Halaskova, 1958 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.*Rhodacarellus silesiacus* Willmann, 1936 (8): 14; 16; – Europe; Asia; North Africa; North America; Australia.*Rhodacarellus minimus* Karg, 1961 (9; 10): 14; 65; 66; 73; – Central and East Europe.*Rhodacarellus kreuzi* Karg, 1965 (33): 11; – Russia and Central Europe.Genus *Rhodacarus* Oudemans, 1902*Rhodacarus calcaratus* Berlese, 1921 (8): 66; – Europe.*Rhodacarus coronatus* Berlese, 1921 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.*Rhodacarus roseus* Oudemans, 1902 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Palearctic Region.*Rhodacarus denticulatus* Berlese, 1921 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – South and Central Europe.

Family Ologamasidae Ryke, 1962

Genus *Sessiluncus* G. Caneastrini, 1898*Sessiluncus hungaricus* Karg, 1964 (9; 10; 26): 5; 14; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Hungary.

Family Macrochelidae Vitzthum, 1930

Genus *Neopodocinum* Oudemans, 1902*Neopodocinum mrciaki* Sellnick, 1968 (33): 40; – Europe.*Neopodocinum ilincae* Iavorski, 1975 (33): 40; – Romania.Genus *Geholaspis* Berlese, 1918*Geholaspis longispinosus* Kramer, 1876 (10; 33): 11; 65; 66; – Europe.*Geholaspis mandibularis* Berlese, 1904 (9; 10; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Europe.*Geholaspis ponticus* Bregetova and Koroleva, 1960 (9; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – East Europe.*Geholaspis longisetosus* Balogh, 1958 (33): 11; – South Europe.Genus *Macrocheles* Latreille, 1829Subgenus *Glyptholaspis* Filipponi and Pegazzano, 1960*Macrocheles americana* Berlese, 1888 (9): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Asia.Subgenus *Macrholaspis* Oudemans, 1931*Macrocheles (M.) dentatus* Evans and Browing, 1956 (33): 11; – Europe.*Macrocheles (M.) opacus* C.L.Koch, 1839 (33): 11; – Europe.*Macrocheles (M.) thianschanicus* Bregetova, 1977 (33): 11; – Russia; Romania.Subgenus *Macrocheles* Latreille, 1829*Macrocheles montanus* Willmann, 1951 (9; 33): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.*Macrocheles matrius* Hull, 1925 (33): 11; 34; 54; 73; 74; – Europe; Asia.*Macrocheles decoloratus* C.L.Koch, 1839 (33): 11; 54; – Europe; Asia.*Macrocheles recki* Bregetova and Koroleva, 1960 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.*Macrocheles monchadskii* Bregetova, 1960 (33): 11; – Russia; Romania.

- Macrocheles merdarius* Berlese, 1889 (33): 11; – Europe; North America; Africa; Australia.
Macrocheles pisentii Berlese, 1882 (33): 11; – Europe.
Macrocheles montanus Willmann, 1951 (33): 11; – Europe.
Macrocheles glaber Muller, 1960 (33): 11; – Europe; Asia.
Macrocheles mammifer Berlese, 1918 (33): 73; – Europe.
Macrocheles banaticus Iavorski, 1977 (13): 62; – Romania.
Macrocheles biharicus Iavorski, 1977 (13): 64; 66; – Romania.
Macrocheles margaretae Iavorski, 1977 (13): 51; 55; – Romania.

Family Pachylaelaptidae Vitzthum, 1931

Genus Pachylaelaps Berlese, 1888

- Pachylaelaps imitans* Berlese, 1921 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Pachylaelaps latior Berlese, 1920 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – South Europe.
Pachylaelaps alpinus Willmann, 1953 (33): 73; – Europe.
Pachylaelaps furcifer Oudemans, 1903 (9; 10): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Pachylaelaps pectinifer G. and R. Caneastrini, 1882 (9; 10; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Pachylaelaps regularis Berlese, 1921 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Pachylaelaps tessellatus Berlese, 1920 (9; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 73; 81; 82; – Europe.
Pachylaelaps vexillifer Willmann, 1956 (33): 11; 16; 73; – Central Europe.
Pachylaelaps sculptus Berlese, 1921 (33): 11; 73; – Europe.
Pachylaelaps humusorum Schweizer, 1962 (33): 11; 73; – Central Europe.
Pachylaelaps reticulatus Berlese, 1904 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Pachylaelaps magnus Halbert, 1915 (9): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Pachylaelaps dubius Hirschmann and Krauss, 1965 (33): 11; – Europe.
Pachylaelaps fusciniger Berlese, 1921 (33): 11; – Europe.
Pachylaelaps ineptus Hirschmann and Krauss, 1965 (33): 11; – Central Europe.
Pachylaelaps laeuchlii Schweizer, 1922 (33): 11; – Central Europe.
Pachylaelaps latus Schweizer, 1961 (33): 11; – Central Europe.
Pachylaelaps undulatus Evans and Hyatt, 1956 (33): 11; – Central and North Europe.

Genus Olopachys Berlese, 1910

- Olopachys scutatus* Berlese, 1910 (9; 10; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – South Europe.

- Olopachys suecicus* Sellnick, 1950 (9; 10; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Olopachys vysotskajae Koroleva, 1976 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – East Europe.
 Genus Pachyseius Berlese, 1910
Pachyseius angustus Hyatt, 1956 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – West Europe.
Pachyseius humeralis Berlese, 1910 (9; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Pachyseius strandtmanni Solomon, 1982 (33): 11; – Romania.

Family Pseudolaelapidae Evans and Till, 1966

Genus Pseudolaelaps Berlese, 1916

- Pseudolaelaps doderoi* Berlese, 1910 (33): 11; – Europe.

Family Laelaptidae Berlese, 1892

Genus Ololaelaps Berlese, 1903

- Hypoaspis venetus* Berlese, 1903 (4): 73; – Europe.
Ololaelaps placentula Berlese, 1887 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Ololaelaps sellnicki Bregetova and Koroleva, 1964 (27): 74; – Europe.

Genus Hypoaspis Caneastrini, 1884

- Hypoaspis montana* Berlese, 1904 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Hypoaspis miles Berlese, 1892 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 74; 81; 82; – Europe; Asia.
Hypoaspis astronomica Koch, 1892 (28): 44; – Europe.
Hypoaspis austriaca Sellnick, 1935 (28): 65; 70; – Europe.
Hypoaspis nolli Karg, 1962 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Asia; North America.
Hypoaspis aculeifer Caneastrini, 1883 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Asia; North and South America.
Hypoaspis antipai Solomon, 1968 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Hypoaspis angusta Karg, 1962 (33): 11; – Europe.
Hypoaspis claviger claviger Berlese, 1883 (33): 11; – Europe.
Hypoaspis krameri G. and R. Caneastrini, 1881 (4): 73; – Europe.
Hypoaspis humerata Berlese, 1904 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.

- Hypospis oblonga* Evans, Till, 1966 (33): 11; 40; 73; – Europe.
 Genus *Androlaelaps* Berlese, 1903
Androlaelaps fahrenheitzi Berlese, 1911 (24): 59; 74; 79; – Palearctic Region; North America.
Androlaelaps casalis Berlese, 1887 (4): 73; – Palearctic Region; North America.
 Genus *Eulaelaps* Berlese, 1903
Eulaelaps stabularis C.L. Koch, 1836 (24): 39; 59; 79; – Europe; Asia; North America.
Eulaelaps novus Vitzthum, 1925 (27): 56; 59; 73; – Europe.
 Genus *Pseudoparasitus* Oudemans, 1902
 Subgenus *Pseudoparasitus* Oudemans, 1902
Pseudoparasitus germanicus Karg, 1965 (9): 11; – Central Europe.
Pseudoparasitus centralis Berlese, 1921 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
 Genus *Laelaps* C.L.Koch, 1836
Laelaps agilis Koch, 1836 (31): 36; 74; 78; 81; – Europe; Asia.
Laelaps hilaris Koch, 1836 (31): 31; 74; – Europe; Asia.
Laelaps muris Ljungh, 1799 (31): 34; 42; – Europe; Asia.
 Genus *Hyperlaelaps* Zachvatkin, 1948
Hyperlaelaps microti Ewing, 1933 (31): 59; – Europe.
Hyperlaelaps amphibijs Zachvatkin, 1948 (31): 59; – Europe; Asia.
- Family *Eviphididae* Berlese, 1913
 Genus *Iphidosoma* Berlese, 1892
Iphidosoma fimentarium Muller, 1859 (9; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Iphidosoma multiclavatum Willmann, 1953 (33): 11; – Europe.
 Genus *Eviphis* Berlese, 1903
Eviphis uropodinus Berlese, 1903 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Italy.
Eviphis ostrinus C.L.Koch, 1836 (9; 10; 29; 33): 5; 11; 14; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
 Genus *Alliphis* Halbert, 1923
Alliphis siculus Oudemans, 1905 (9; 33): 5; 11; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe; Asia.
- Family *Haemogamasidae* Oudemans, 1926
 Genus *Haemogamasus* Berlese, 1889
Haemogamasus nidi Michael, 1892 (31): 39; 49; 59; 74; 79; – Europe.
Haemogamasus hirsutosimilis Willmann, 1952 (7): 32; 36; – Europe.

Family *Zerconidae* Caneastrini, 1891Genus *Zercon* C.L.Koch, 1836

- Zercon sellnicki* Schweizer, 1948 (3): 16; 73; – Central Europe.
Zercon triangularis C.L.Koch, 1836 (9; 10): 16; 65; – Europe.
Zercon berleseii Sellnick, 1958 (30): 8; 16; – Central Europe.
Zercon arcuatus Tragardh, 1931 (30): 8; 16; – Europe.
Zercon romagniolus Sellnicki, 1944 (30): 9; 16; 19; 73; – Central and South Europe.
Zercon peltatoides Halaskova, 1970 (30): 9; 16; 19; – Europe.
Zercon hungaricus Sellnick, 1958 (30): 8; 49; – Europe.
Zercon montanus Willmann, 1953 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Zercon spatulatus C.L.Koch, 1839 (30): 49; – Europe.
Zercon carpathicus Sellnick, 1958 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – East and Central Europe.
Zercon baloghi Sellnick, 1958 (10; 30): 8; 65; – Hungary.
Zercon echinatus Schweizer, 1922 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Zercon keiseri Schweizer, 1949 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Zercon foveolatus Halaskova, 1970 (30): 8; – Central Europe.
Zercon peltatus C.L.Koch, 1836 (30): 8; Central Europe.
Zercon perforatulus Berlese, 1904 (9; 30): 5; 8; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Zercon storkani Halaskova, 1970 (30): 8; – Central Europe.
Zercon schweizeri Schweizer, 1948 (9): 1; 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Zercon alpinus Willmann, 1953 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Zercon joduthae Sellnick, 1944 (9): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Zercon aniellae Solomon, 1984 (3): 8; – Romania.
Zercon fageticola Halaskova, 1969 (3): 8; – Central Europe.
Zercon moldavicus Călugăr, 1997 (2): 8; – Romania.
Zercon montigenus Blaszak, 1972 (3): 8; – Central Europe.
Zercon pinicola Halaskova, 1970 (3): 8; – Central Europe.
Zercon juvarae Călugăr, 2003 (3): 8; – Romania.
Zercon marinae Călugăr, 2003 (3): 8; – Romania.
Zercon magdae Călugăr, 2003 (3): 8; – Romania.
Zercon similifoveolatus Călugăr, 2003 (3): 8; – Romania.

Genus Prozercon Sellnick, 1943

- Prozercon traegardhi* Halbert, 1923 (3): 16; 49; – Central and North Europe.
Prozercon sellnicki Halaskova, 1963 (9; 10): 5; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Prozercon kochi Sellnick, 1943 (9; 10; 27): 5; 8; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Prozercon fimbriatus C.L.Koch, 1839 (9; 10): 5; 16; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Europe.
Prozercon carsticus Halaskova, 1963 (9, 27): 5; 8; 19; 38; 48; 49; 52; 53; 62; 65; 66; 67; 81; 82; – Central Europe.
Prozercon tragardhisimilis Solomon, 1984 (3): 8; – Romania.
Prozercon plumosus Călugăr, 2003 (3): 8; – Romania.

Genus Parazercon Tragardh, 1931

- Parazercon radiatus* Berlese, 1914 (29): 16; – Europe.

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AN OVERVIEW OF THE OLDER AND RECENT CONTRIBUTIONS TO THE KNOWLEDGE OF THE FISH FAUNA OF THE BANAT

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Two species of lampreys (both mis-identified), one sturgeon and 37 bony fishes have been recorded prior to 1918 from the sector of the Banat province, now belonging to Romania, most of them from a single or a small number of localities or rivers. Field investigations, most of them made by the authors of the present paper, carried on from 1943 till 1998 resulted in the addition of two lampreys (others than those mis-identified by older authors), three sturgeons and 11 species of bony fishes priorly not recorded from one province. For 36, out of the 37 native of bony fishes recorded earliest, numerous new localities have been signalled. The status of three species has recently been modified through molecular investigations. Nine non native species were introduced.

Key words: Freshwater fishes; Banat; Danube basin.

1. INTRODUCTION

The province Banat is delimited by the river Mureș (tributary of the Tisa in the north, the Tisa in the west, the Danube in the south and the Carpathians in the east). It is drained by five big rivers, the Bega, Timiș, Caraș, Nera and Cerna and many small ones, most of these in the south of the province, between the Nera and the Cerna. Numerous papers have been published about its fish fauna (the oldest one in 1726) and it was considered useful to make a synthesis on them.

2. MATERIALS AND METHODS

The data included in the present paper are the results of our own field investigations, between 1943 and 1999 and of a critical overview and evaluation of the results of earlier publications.

3. RESULTS AND DISCUSSION

The first author who published data on the fishes from the Banat was Marsigli. In his "Danubius Pontico Misicus" (1726) he mentions the occurrence of five

species of sturgeons and of many bony fishes in the Danube. The title of the book suggests that Marsigli's data refer to the species in the Middle and Lower Danube as well; yet he spent long time in the island Ada-Kaleh in the southern Banat, his informations refer to a large extent to the Danube in this province.

Reisinger (1830) records some sturgeons and teleosts from "Tibiscus". This was the Latin name of the Timiș, the main inner river in the Banat (probably the latinisation of older Dacian name); however, Reisinger meant the river Tisa, not the Timiș.

Few contributions to the fish fauna of the Banat were published during the XIX-th century and the first decades of the XX-th century: books on the fishes from the former Habsburg empire (e.g. Heckel & Kner, 1858) or from the pre-1919 Hungary (Herman, 1887), besides minor contributions. A special mention deserves the paper of Vütskits (1911) who records the occurrence of a goby of marine Ponto-Caspian origin, *Gobius* (now *Ponticola*) *kessleri*, in the Danube at O-Palanka, near the confluence of the Nera River.

G.kessleri was the second species of Ponto-Caspian goby recorded upstream of the Iron Gates, after *Proterorhinus marmoratus*.

All data concerning the fishes from the Banat were summarized in another contribution by Vütskits – the chapter of fishes in "Fauna Regni Hungariae" (1913). Curiously enough, he does not mention the species *G.kessleri* which he found himself earlier in the Danube.

Vütskits records from the Banat two species of lampreys (*Lampetra fluviatilis* and *L.planeri*, both misidentified), the sturgeon *Acipenser ruthenus* from the Bega and 37 species of teleosts.

A. ruthenus occurs also in the Danube and the Mureș; it is practically extinct from the Bega; isolated specimens have been fished in this river during the 30-ties of the last century.

The teleosts listed by Vütskits were recorded, with few exceptions, only from one or two localities or rivers; most have been recorded or found, after 1946, in many other rivers.

These species are:

1. *Hucho hucho* recorded by Vütskits (as *Salmo h.*) from the Cerna. This is the only river from the Banat in which the huchen was native; it is now extinct, the last specimen being fished in 1912 (information from a competent forestry engineer).
2. *Salmo trutta labrax*, *f. fario*: recorded by Vütskits (as *S. fario*) only from the Caraș; there are recent records from the upper reaches of the rivers Bega, Timiș, Bistra, Sebeș, Bârzava, Nera, Caraș and Cerna, as well as from the Sirinea, Berzasca, Mraconia and Eșelnița, in which the species seems to have been introduced.

3. *Esox lucius*, recorded only from the Bega, is actually present also in the Mureș, Danube, Timiș, Caraș, Nera and in numerous standing water bodies in drainage areas of these rivers.
4. *Abromis ballerus*, listed by Vütskits from the Danube, later recorded from the Bega and Timiș; it became quite abundant in the new Iron Gates damlake.
5. *A. brama*. Recorded by Vütskits from the Danube, later also from the Timiș and the Bega; also present in the Mureș.
6. *A. sapa*. Recorded by Vütskits from the Danube at Baja and the Caraș at Crebenăț. Recent investigations in the latter river do not confirm the presence of the species there. It is present in the Banat sector of the Danube; sporadic specimens were found in the Bega and Timiș.
7. *Alburnoides bipunctatus*. Recorded by Vütskits (as *Spirilinus b.*) only from the river Caraș; after 1946 it was also recorded from the Mureș, Bega, Timiș, Pogănici, Caraș, Nera, Miniș, Radimna Camenița, Sirinea, Berzasca, Elișeva, Plavișevița, Mraconia, Eșelnița, Cerna, Belareca, Mehadica and Behna.
8. *Alburnus alburnus*. Recorded by Vütskits (as *A.lucidus*) from the rivers Caraș and Cerna; after 1946 also from Mureș, Bega, Beregsău, Timiș, Pogănici, Bistra, Nera, Berzasca and many pools.
9. *Aspius aspius*. Recorded by Vütskits (as *A.rapax*) from the Danube and Caraș; after 1946 also recorded from the Mureș, Bega, Beregsău, Timiș, Nera, Berzasca and Cerna.
10. *Barbus barbus*, the common barbel, was recorded by Vütskits (as *B.fluviatilis*) from the Danube, being after 1946 recorded also from the Mureș, Bega, Timiș, Caraș, Nera and Cerna.
11. *Barbus petenyi* (usually listed as *B.meridionalis petenyi* or *B.peloponesius petenyi*, the Carpathian barbel was recorded by Vütskits from the Caraș and Cerna, was found and recorded in the other rivers in the southern Banat, Nera, Radimna, Camenița, Berzasca, Sirinea, Elișeva, Plavișevița, Mraconia and Eșelnița being absent from Tișovița. It was also found in the rivers Bistra and Bârzava, tributaries of the Timiș, where it appeared later. It is absent from the Bega. Recent molecular investigations (Kotlick *et al.*, 2002) demonstrated that the Danubian *B.petenyi* must be split in three species or subspecies, the populations from the rivers in southern Banat belong to *B.p.balcanicus*, also present in the south-western tributaries of the Danube. The status of the populations from the Timiș River and its tributaries is unknown.
12. *Blicca bjoerkna*, the white bream. Recorded by Vütskits only from the Danube, after 1946 also from the Mureș, Bega, Timiș, Beregsău, Caraș, Nera, Cerna; juvenils also numerous in the lowermost sectors of the small rivers in the Iron-Gates area.
13. *Carassius carassius*, the crucian-carp. Listed by Vütskits (as *C.vulgaris*) from "Iutosis Mosztonga", probably near Lugoj; it was present and abundant, until

the 50-ties in the sectors with slowly running water of the Danube and of the rivers Aranca, Bega, Behela, Beregsău, Timiș, Caraș and in all pools in the low lands; it became extinct from everywhere, it is not known if it survives or not in some places.

14. *Chondrostoma nasus*, the nose; reported by Vütskits from the Danube and the Caraș, after 1946 also from the Mureș, Bega, Nera, (where it is very abundant) and Berzasca; juveniles also from the lowermost sectors of the Danube tributaries in the Iron-Gates area.
15. *Cyprinus carpio*, the carp.; reported by Vütskits from the rivers Bega, Timiș, Caraș, also present in the Danube, Mureș, Pogăniș, Nera, Berzasca, Cerna and in many oxbows, pools, reservoirs and farms.
16. *Gobio gobio*, the common gudgeon: reported by Vütskits (as *G. fluviatilis*) only from the rivers Caraș and Cerna; since the species is now absent from the latter river, its record may be based on misidentified *G. albipinnatus*. After 1946 the species was recorded from the rivers Mureș, Aranca, Bega, Beregsău, Timiș, Pogăniș, Bârzava, Caraș, Nera, Radimna, Berzasca, Sirinea, Mraconia and Eșelnița.
17. *Gobio uranoscopus frici*, the stone-gudgeon; recorded by Vütskits from the Cerna at Mehadia, after 1946 from the Bega (Curtea to Chizătău), Timiș (Teregova to Chizătău, juvenils occasionally farther downstream to Albina), Nera (upstream Bozovici almost to Socol) its tributary Miniș, Cerna (upstream Pecinișca to Orșova) and its tributary Belareca near Mehadia.
18. *Idus idus*, the ide, recorded by Vütskits (as *I. melanotus*) from the Cerna, later also from the Mureș, Bega, Timiș, Nera and Berzasca.
19. *Pelecus cultratus*, recorded by Vütskits from the Danube; ascends occasionally the Bega and the Timiș.
20. *Phoxinus phoxinus*, the minnow, recorded by Vütskits (as *P. laevis*) from Caraș, after 1952 also from the upper Bega, Timiș, Nera, Cerna and their tributaries, but absent from the rivers in the Iron-Gates area.
21. *Rhodeus sericeus amarus*, the bitterling recorded by Vütskits from the Danube, later from the Mureș, Bega, Beregsău, Timiș, Pogăniș, Caraș, Nera, Cerna and numerous standing waters.
22. *Rutilus rutilus*, the roach; recorded by Vütskits (as *Leuciscus r.*) from the Cerna, after 1946 also from the Danube, Mureș, Bega, Beregsău, Timiș, Caraș, Nera, Berzasca and neighbouring pools.
23. *Scardinius erythrophthalmus*, the rudd; mentioned by Vütskits from the Danube and the Caraș, found later also in the Mureș, Bega, Beregsău, Timiș, Nera and especially in stagnant water bodies within the drainage areas of these rivers.
24. *Squalius cephalus*, the chub; recorded by Vütskits from the Danube and the Caraș, later reported also from the Mureș, Bega, Beregsău (and its tributaries Matca and Ier) Timiș (from Teregova downstream), Bistra, Bistra Mărului,

- Pogăniș, Bârzava, Nera, Miniș, Cerna, Belareca and in the nine Danube tributaries in the Iron Gates area.
25. *Tinca tinca*, the tench, recorded by Vütskits (as *T. vulgaris*) from three stagnant waters, "Apatin", "lacu Ludas" and "Iutosis Mosztonga"; and later from many oxbows and pools in the drainages of Bega, Timiș.
 26. *Vimba vimba*, the vimba bream, recorded by Vütskits (as *Abramis v.* and *A. melanops*), from the Danube and Caraș, by later authors from the Bega, Timiș, Nera, Berzasca and Cerna.
 27. *Cobitis danubialis*, the spined loach, recorded by Vütskits (as *C. taenta*) from the Cerna and Caraș (possibly on the base of misidentified specimens of *Sabanejewia montana*), later recorded from the Danube, Mureș, Aranca, Bega, Beregsău, Niarad, Ier, Timiș, Pogăniș, Caraș, Nera, Cerna, ponds in the drainage areas of these rivers, ponds of Satchinez, Biled, Becicherec and Biled.
 28. *Misgurnus fossilis*, the mud loach, recorded by Vütskits from the pools at Mosztonga, by later authors from the rivers Aranca, Beregsău, Bega and Timiș, from oxbows and pools in the drainage areas of these rivers and of the Danube and Caraș, from ponds at Satchinez, Bărateaz and Biled.
 29. *Orthrias barbatulus*, the stone loach, recorded by Vütskits (as *Nemachilus b.*) only from the Cerna, by later authors from the upper and partly middle sectors of the Bega, Timiș, Bistra, Bârzava, Caraș, Nera, Radimna, Berzasca, Sirinea, Mraconia, Eșelnița, Cerna and tributaries.
 30. *Silurus glanis*, the wels; recorded by Vütskits from the Bega, Timiș and Caraș, by later authors also from the Danube and Mureș; it ascends occasionally the lower sectors of the Nera and Cerna.
 31. *Lota lota*, burbot; recorded by Vütskits (as *L. vulgaris*) from the Danube, by later authors from the Bega, Timiș, Caraș, Berzasca and Cerna.
 32. *Gymnocephalus cernuus*, the ruffe. Recorded by Vütskits (as *Acerina c.*) from the Danube and the Caraș, by Bănărescu (1946, 1964) from the Bega, Beregsău, Niarad and Timiș and at the confluence of the Nera and Cerna.
 33. *Gymnocephalus schraetser*, the yellow pope; recorded by Vütskits from the Danube, by Bănărescu (1946) from the Bega and Timiș.
 34. *Perca fluviatilis*, the perch; recorded by Vütskits from the Danube and Cerna, by later authors from the Mureș, Bega, Beregsău and tributaries, Timiș, Caraș, Nera, Sirinea, in pools near Timișoara and Satchinez, at "Lacul Dracului" near the Nera River.
 35. *Stizostedion lucioperca*, the pike-perch; recorded by Vütskits from the Danube, Bega, Caraș, Cerna, by Bănărescu (1946) from the Timiș.
 36. *Zingel streber*; recorded by Vütskits (as *Aspro vulgaris*) from the Danube, by Bănărescu (1946, 1964) from the Timiș, Bega and Nera, the population from the latter river being ascribed to a distinct subspecies, *Z. streber nerensis*.
 37. *Z. zingel*; recorded by Vütskits only from the Danube, by Bănărescu (1946) from the Timiș and the Bega.

Two species of lampreys, three sturgeons and 14 of teleosts, not reported by Vütskits, have been recorded after 1946, or at least are known to be, or to have recently been present in the Banat.

- the predatory lamprey *Eudontomyzon danfordi* in the upper sector of the river Timiș, in its tributary Bistra Mărului, according to some information also in the Cerna;
- the non-predatory lamprey *E.vladykovi* in the Timiș, Bistra, Bistra-Mărului and in the Bega;
- the migratory sturgeons *Acipenser gueldenstaedti*, *A. stellatus* and *Huso huso* were present, prior to the damming of the Iron Gates area, in the Banat sector of this river; they became very rare;
- the Danube shad, *Alosa immaculata* (*A. pontica*) ascended, occasionally upstream the Iron Gates);
- the ceel *Anguilla anguilla*: specimens have occasionally been collected from the Iron Gates sector of the Danube and from the rivers Bega and Cerna;
- a large-sized species of bleak, *Chacalburnus, chalcoides mento*, was present before the damming of the Danube, in the Iron Gates sector of the river where it probably became extinct;
- the dace (*Leuciscus leuciscus*); its recordings in the rivers Beregsău (Bănărescu, 1946) and Bega (1964) are based on mis-identified specimens. A single locality is known in the Banat for this species: the lowermost sector of the rivulet Mraconia (tributary of the Danube, Iron-Gates area). This sector is now covered by the dam-lake, the local population of dace became extinct;
- *Leucaspius delineatus*. Abundant in small pools in the drainage areas of the rivers Bega, Timiș and Caraș (many of which disappeared); also in the rivers Bega and Bârzava;
- the whitefin gudgeon *Gobio albiginnatus vladykovi* recorded first by Bănărescu (1964) from the rivers Bega and Timiș; numerous also in the Danube, in the rivers Mureș, Aranca, Caraș, Nera and at the confluence of the Berzasca, Mraconia and Cerna with the Danube.
- the sand gudgeon, *G.kessleri*. Recorded first in Romania from the Timiș and Bega (Bănărescu, 1946) later from the Mureș, Caraș, Nera, absent from the Cerna, Berzasca; the populations from the Timiș, Bega and Caraș are ascribed to a distinct subspecies, *G.kessleri banaticus* Bănărescu, 1953;
- the elongate loach, *Cobitis elongata*; the range of this species in Romania is restricted to the river Nera and its tributary Miniș, the species being abundant (Bănărescu and Nalbant, 1957);
- the Danubian golden loach, *Sabanejewia montana* Vladykov, was until recently listed as *S.aurata balcanica* or *S.balcanica*. Molecular investigations

(Perdices *et al.*, 2003) demonstrated that the Danubian golden loach is distinct from the Transcaucasian *S.aurata* and from the Macedonian *S.balcanica*. Two subspecies are present in the Banat. *S.montana montana* was recorded initially from the river Timiș (Bănărescu, 1946), as *Cobitis aurata balcanica*, found later also in the Bega, Caraș, Nera, Berzasca, Sirinia, Mraconia, Eșelnița and Cerna; in the Danube, Tisa and the lowermost sectors of the Bega and Timiș lives the subspecies *S.montana bulgarica*. A gradual transition (intergradation between both subspecies) takes place in the lower Bega and Timiș;

- *Sabanejewia radnensis* formerly considered a subspecies restricted to the upper sector of the River Mureș in Transylvania, actually inhabits the entire Mureș River, as far downstreams as its confluence with the Tisa (Perdices *et al.*, 2003; Bănărescu in preparation). In the lowermost sector of the Mureș (north-western Banat) it lives sympatrically with *S.montana bulgarica* (which ascends from the Tisa) without hybridization; this means, both are reproductively isolated, i.s.g. "good" species.
- the Balon's pope, *Gymnocephalus baloni*, until recently confounded with the ruffe (*G. cernuus*), has been identified in the Danube (Iron Gates area) and in the Timiș; it is surely present at least in the Mureș, Bega and Caraș.
- *Proterorhinus marmoratus* recorded from the river Beregsău at Pischia (Bănărescu, 1946), found later in the tributary Niarad at Sânaandrei and in the river Timiș at Peciul-Nou. Another fish species may be present in the Banat: *Umbra krameri*, which has not been recorded.

Besides the 45 fish species listed above which are native in the Banat (possibly except for *Anguilla anguilla*), nine exotic species are now present in the province:

- two valuable salmonids of North American origin – *Salvelinus fontinalis* in montane waters and *Oncorhynchus mykiss*;
- three valuable East Asian carpfishes – *Ctenopharyngodon idella*, *Hypophthalmichthys molitrix* and *H. (Aristichthys) nobilis* in fishery farms, occasionally found also in lowland rivers;
- two North American species, introduced by aquarium hobbyists in the German sector of the Danube basin wherefrom they dispersed eastwards in the Banat and further: *Lepomis gibbosus* (the sunfish or pumpkinseed) and *Ictalurus nebulosus* (the catfish);
- the East Asian *Pseudorasbora parva*, introduced in southern and western Romania in 1961–1962 with fry of valuable Chinese carpfishes, now widely distributed throughout Europe;
- another species of East Asian ancestry, *Carassius auratus gibelio* (the German carp), which was present in eastern and southern Romania in the 20-th century and was involuntarily introduced in the Banat after 1948

with fry of the common carp. It is now abundant and widely distributed in the Banat, has almost totally replaced and eliminated the native congener *C. carassius*.

During the last few decades several fish species of ponto-caspian brackish water origin ascended from the lower Danube into the Iron-Gates damlake and even further upstream: *Neogobius fluviatilis* (the most abundant species, present also in tributary rivulets), *N. (Apollonia) melanostomus*, *N. (Ponticola) kessleri*, *N. (Babka) gymnotrachelus*, *Benthophilus stellatus*, *Syngnathus abaster (S.nigrolineatus)* also possibly others.

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EURYGASTER SPECIES (HETEROPTERA – SCUTELLERIDAE) ATTACK ON WHEAT PRODUCTION, IN ONE LOCALITY OF BRĂILA DISTRICT (1996–2003)

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Following the examination of the grains in the wheat stores, carried out between 1996 and 2003, we estimated the degree of wheat grains “stinging”, produced by *Eurygaster* species. The total production of wheat harvested in Brăila district during the eight years showed no or low intensity attacks. The non-attacked wheat quantity increased from 44% in 1996 to 71% in 1998, and then it decreased significantly in the 1999–2001 period and especially in 2002 (0.02%) and 2003 (0.12%). Except for 2003 (11.44%), the quantities of wheat grains with 2%–4 % attack intensity were very low (under 1%).

Key words: *Eurygaster* species attack, *Eurygaster* species damages.

1. INTRODUCTION

In Romania, the complex of *Eurygaster* species from the cereal crops (“sunn pest”, “cereal bugs”) is represented by *Eurygaster integriceps* Put., *Eurygaster maura* L., *Eurygaster austriaca* Schr. and *Eurygaster testudinaria* Geoffr., of the Scutelleridae family.

Eurygaster species are dominant in comparison to other cereal bugs, like *Aelia* species, from Pentatomidae family, and cause higher damages to the wheat crops. They represent 80% in Moldova region, 93% in Muntenia and Oltenia and 97% in Dobrogea, of whole cereal bugs populations (8).

Eurygaster integriceps, present in all areas, with more than 75% of the total *Eurygaster* and *Aelia* species weight, became on the first place, as dominance and, consequently, as importance of the produced damages. It originates from high Asia plateaus, and, at present, is the main pest of the cereal crops, despite the fact that at the beginning of the 20th century it was not known in our fauna (11). This is an alien invasive species. *Eurygaster maura* was present mainly in Oltenia Plateau, in the Center and the Northwest of Romania, in the North of Moldova, with about 15% of weight. *Eurygaster austriaca* was present especially in Banat and Crișana, with 10% of weight and *Eurygaster testudinaria* had a small weight and therefore an insignificant economic importance (4).

In Romania, *Eurygaster* species attacks were registered on a very large area, covering 41 districts. Their populations size and attack intensity are correlated with the climate conditions, the quercineae forests distribution (as place of adults hibernation) and wheat crops distribution (as food resources). The association of these three categories of factors is favorable to the *Eurygaster* species development and, in consequence, creates the premises for the quantitative and qualitative decreases of the cereals productions.

With the piercing-sucking mouthparts, both the *Eurygaster* adults and larvae perforate the plant tissues, inject their saliva and suck out the sap from the leaves, stalk, rachis, flowers and grains.

The aspect of the plants depends on the attacked organ and on the attack intensity: the conic protuberances named "salivary cones" appear on the location of the sting; the superior part of the attacked leaves dries; the central leaf and the stalks become yellow and dry; supplementary stalks appear. In the case of the attack that occurs at the ear basis, the ears remain enclosed. The attacked flowers become sterile. The ears lose their color when the rachis is stung. The awns are positioned in a characteristic way forming a 45° angle when the stinging is under the ear. The grains are emptied of their contents, when the attacks occur on the developing grains. Their contents are altered when the attacks occur on the mature grains, without modifying their shapes or weights.

The qualitative implications are very important: attacks induced by sunn pest severely change the gluten quality, which influence the panification process (dough quality, weight and yield of bread) (5, 12, 16).

In Romania, *Eurygaster* species and their oophagous Scelionidae were very much investigated, especially by Popov *et al.* 7, 8, 9, 10, 11, 12) and Teodorescu *et al.* (3, 13, 14, 15, 16).

2. MATERIAL AND METHODS

The attack intensity of the *Eurygaster* species in the vegetation period of cereal crops, on the grain production, was estimated at the wheat stores (warehouses) by the "stinging" degree of wheat grains.

From wheat grains quantities harvested in one day, of each agricultural unit, 0.5 kg grains were selected. From this, in maximum 24 hours, 25 grams were examined, by using the gravimetrical method.

Under the microscope the healthy grains were separated from the attacked ones. The attacked grains were weighed and the obtained data were extrapolated to 1000 grams.

3. RESULTS AND DISCUSSIONS

The investigated samples of wheat showed a very good grain quality: the greatest quantities of grains were without attack or with the reduced attack intensity, especially between 0.1% and 2%. Whole quantities of wheat grains registered the attack intensity below 4% (Table 1).

Table 1

Stinging degrees of the wheat grains by *Eurygaster* registered in Insurăței locality, Brăila district

Stinging percentage of wheat grains in warehouse	1996	1997	1998	1999	2000	2001	2002	2003
Without attack/Unharmed	44.46	57.18	71.12	35.69	26.44	22.35	0.03	0.12
0.0–1 % attack	16.54	42.82	28.88	39.66	34.74	48.85	62.84	53.42
1.1–2 % attack	36.95	–	–	24.64	38.50	28.75	37.01	33.91
2.1–3 % attack	0.88	–	–	0.007	0.007	0.04	0.11	12.32
3.1–4 % attack	1.07	–	–	–	0.3	0.007	–	0.23
4.1–5 % attack	–	–	–	–	–	–	–	–
5.1–50 % attack	–	–	–	–	–	–	–	–
over 50 % attack	–	–	–	–	–	–	–	–

A very low attack intensity was a consequence of a low *Eurygaster* population density in the fields. Under these conditions, in Brăila district a very good quality flour was obtained.

In 1996, more than 44% of the entire wheat production showed no *Eurygaster* attack (Fig. 1 A.). If we add to this value 54.37% with less 3% attack results that approximately 99 % of the entire wheat production was of the best quality. The state was even better (Fig. 1 B.), in 1997: the wheat whole production showed no attack (over 57%) or an attack below 1% (over 42%). In the next year the same best conditions were recorded, when less than 1% attack decreased, and in exchange the non-attacked wheat production increased (over 71%) (Fig. 1 C.).

Beginning with 1999 year, the *Eurygaster* species attack was intensified, evidenced both by the registration of 3–4% attacks degree (Fig. 1 D–H) and by the decrease of non-attacked wheat production (from 35.69% in 1999 to 22.35% in 2001) (Fig. 2 A). During 2002 and 2003, the quantity weights without attack were very small (0.03–0.12%). In 1997 and 1998 the wheat quantity up to 1% attack was missing. The wheat quantities with 0.01–4% attack increased (to 64.30%, in 1999; 73.54% in 2000; 77.64 in 2001; 99.96% in 2002; 99.88% in 2003) (Fig. 2 B–E). Small quantities of wheat grains with over 3% attack were found in the whole period of investigation (Fig. 2 E).

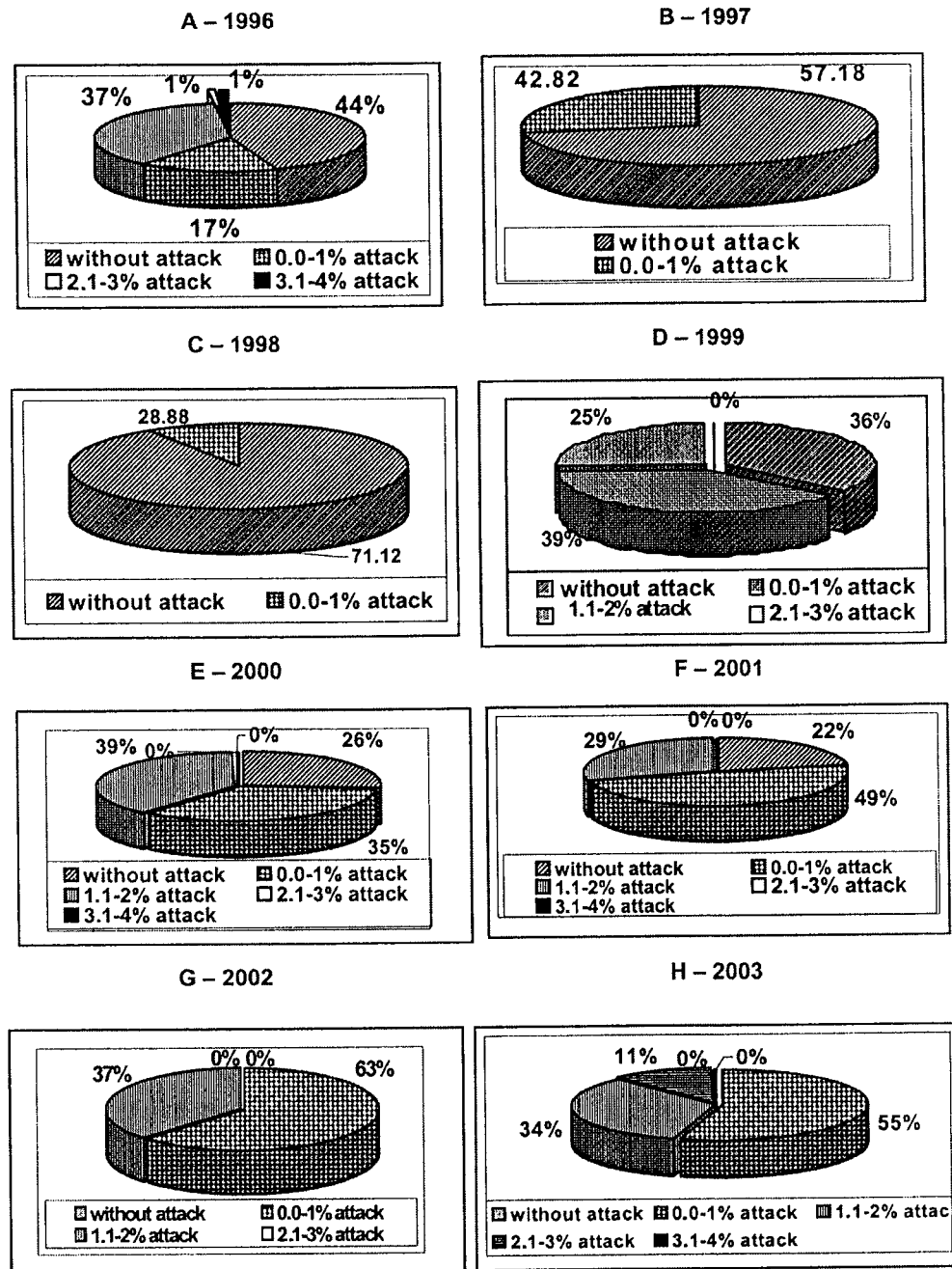


Fig. 1. – Proportions of wheat grains without *Eurygaster* attack and with various attack degrees.

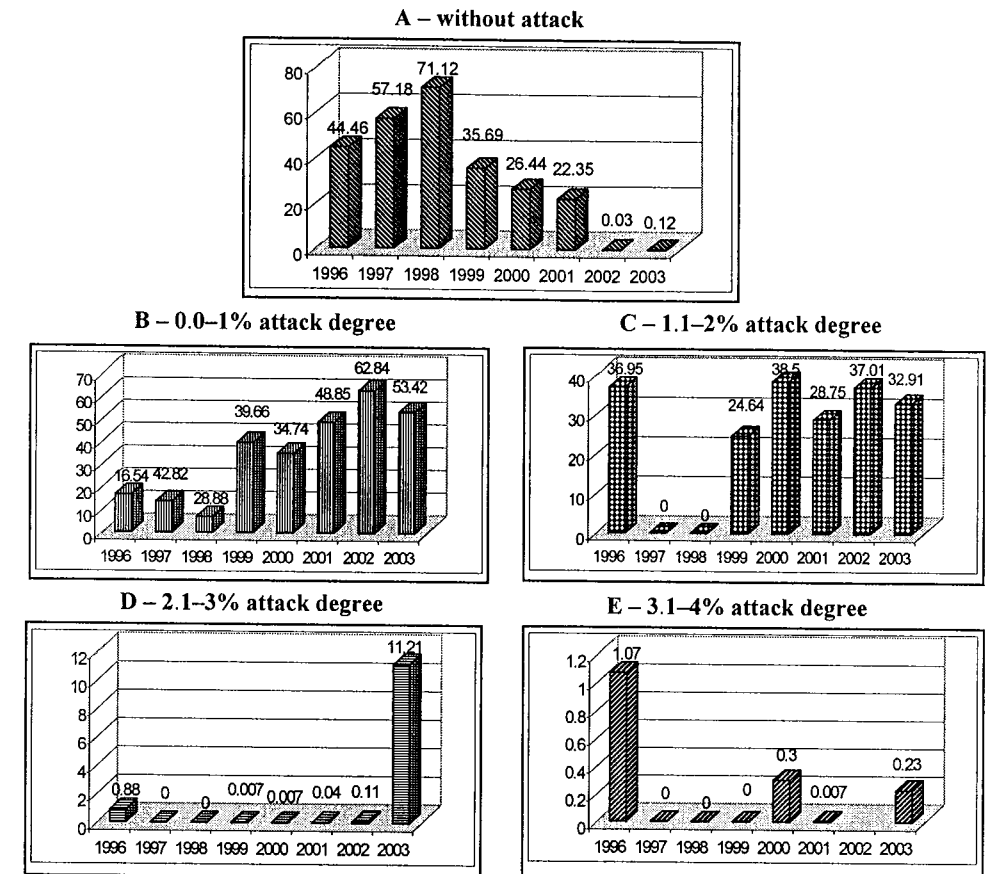


Fig. 2. – Dynamics of wheat grains (%) of total production without attack by *Eurygaster* (A) and with different attack degrees (B–E), in Insurăței, 1996–2003.

At the same time, our attention was focused not only on the correlation between the attack intensity (percentage of stung grains) with *Eurygaster* adult densities (Table 2), but also with the harvest moment. The cases with a reduced attack, at higher pest densities (above the economic threshold of damages), were due to early cereals harvesting which impeded the adults feeding. The cases with higher attack intensity, at low *Eurygaster* densities (under the economic level of damages) were due to the harvesting delay, which allowed the adults to sting more grains, intensifying the attack.

Table 2

Dynamics of the *Eurygaster* adults in wheat crops

1996	1997	1998	1999	2000	2001	2002	2003
3.4	0.8	0.6	0.3	0.6	1.3	1.4	4.4

4. CONCLUSIONS

The 1996–2003 period was not favorable for the sunn-bug populations, but very favorable from the quantitative and mainly qualitative points of view for the wheat production, in Brăila district.

The weight of the quantity of grains not attacked by the *Eurygaster* species increased in the first 3 years of the interval, later decreased, concomitantly with a gradual increase of the quantities of cereals with a higher attack degree. The quantities of wheat with attack below 2% ranged between 62.88% of total production in 2002 and 100% in 1998 and 1999. An increase of wheat quantities with attack of over 2% was recorded after 1998. An important quantity (over 11%) of wheat with attack between 2.1 and 4% was for the first time recorded in 2003.

The obtained results draw attention on the importance of the moment of cereals harvesting, considering not only the grain maturity but also avoiding the aggravation of the *Eurygaster* species attack. The choosing of the best moment for the cereals harvesting is one of the alternative, non-polluting solutions to avoid the high loss of crops.

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THE MAIN ALIEN/INVASIVE NEMATODA AND ACARINA SPECIES IN ROMANIA

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The paper is the result of the authors' observations in the orchards, vineyard plantations, field crops, and also of an elaborate study concerning the nematoda and acarina alien/invasive species introduced in Romania. The data refer to 22 species of invertebrates belonging to Nematoda (9 Tylenchida and 5 Dorylaimida) and Acarina (1 Eriophyiidae, 1 Bryobiidae, 4 Phytoptipalpidae, 1 Tarsonemidae, 1 Phytoseidae). A number of 8 species are considered invasive (*Heterodera schachtii*, *Globodera rostokiensis*, *Meloidogyne incognita*, *M. hapla*, *M. arenaria*, *Ditylenchus destructor*, *Polyphagotarsonemus latus*, *Bryobia rubrioculus*) and at present time 14 are considered non-invasive, but with a future potential of turning into invasive species.

1. INTRODUCTION

The aliens, exotics, foreign, non-indigenous species (Agenda 21) are the species which enlarged their natural areal, invading new territories, as a consequence of their natural spreading tendency or of human intervention.

Unintentional introduction is also very frequent, since all species can move between countries or continents.

The introduction of species beyond their natural range is rising sharply, due to increased transport, trade, travel and tourism and the unprecedented accessibility of goods resulting from globalisation. These activities provide vectors and pathways from live plants, animals and biological material to cross biogeographical barriers that would usually block their way (European Strategy on Invasive Alien Species).

The intentional introduction is the deliberate movement and/or release by human of an alien species outside its natural range. In biological control methods, especially in the classical period, many programs were carried out in different countries for the introduction of the natural enemies (parasitoid, predator, some phytophagous species) against alien invasive pest species.

A key emerging global environmental issue for the new century are invasive species. Endemic species, ecosystems and human communities are ravaged (McDowell, 1998).

Successfully alien species level increases explain: "in its new range it may have better resources for growth, it may be free from competition with other species, or it may be free from its natural predators or parasites" (Waage J., 1998).

The invasive species are the second greatest threat to biodiversity after habitat destruction. I would say that on islands, invasive species is the number one threat to biological diversity. Invasive species are a kind of habitat destruction. When invasive species take over a habitat, they erase the native richness and diversity of species. Of course, habitat destruction in its naked form is clearly the worst threat to species conservation worldwide (Mooney and Shiva, 1998).

To solve the problem of alien invasive species, the Convention on Biological Diversity established a three-stage hierarchical approach: prevention of introduction, eradication, and control of the population level. In general, if an alien species has been introduced, no effective methods of eradication are known.

Biological control is becoming increasingly popular in the toolbox of alien species management methods because, unlike other methods, it does not require repeated intervention with control measures and therefore can combine environmental friendliness with high cost-effectiveness (Waage, 1998).

The Convention on Biological Diversity provides the best opportunity for developing a comprehensive global approach to the introduction of alien species and their eradication or control (Glowka Lyle, 1998).

2. MATERIAL AND METHODS

The paper is the result of personal observations in the orchards, vineyard plantations, and field crops and, also of an elaborate study of the literature concerning the invasive and non-invasive alien species introduced in Romania.

3. RESULTS AND DISCUSSION

Among the species listed, 21 are pests and only one is a beneficial species, introduced for Tetranychidae pest biological control. A number of 8 species are invasive and 14 are non-invasive with a future potential of turning into invasive species.

The most numerous alien species introduced in Romania belong to Nematoda class, especially Heteroderidae family (*Globodera*, *Meloidogyne* and *Heterodera* species), many of them being a very polyphagous pest. In vineyard plantation, five *Xiphynema* polyphagous species were unintentionally introduced, which produce certain damages at present.

With one exception (the intentional introduction of predaceous *Phytoseiulus persimilis*) the others were unintentionally (accidentally) introduced along with vegetal

Table 1
Alien/invasive vineyard pests

Species, Orders, Families	Hosts trophic range	Native area, year of introduction, actual status in Romania
<i>Xiphynema americanum</i> Dorylaimida Longidoridae	Polyphagous: <i>Vitis vinifera</i> , <i>Medicago sativa</i> , <i>Canabis sativa</i> , <i>Ribes nigrum</i> , <i>Cerasus vulgaris</i> , <i>Persica vulgaris</i>	North America. In Europe it was mentioned in 1913. In Romania it was detected in 1969, in Ialomița district; at present it occurred in many vineyards, where it caused important yield losses.
<i>Xiphynema italiae</i> Dorylaimida Longidoridae	Polyphagous: <i>Vitis vinifera</i> , <i>Medicago sativa</i> , <i>Canabis sativa</i> , <i>Ribes nigrum</i> , <i>R. grossularia</i> , <i>Prunus domestica</i> , <i>Persica vulgaris</i> , <i>Malus domestica</i>	South and Central America. In Europe it was mentioned in 1953. In Romania it was detected in 1969, and now is present in the vineyard under the sandy soils (Bechet, Șimnic), where it caused important yield losses.
<i>Xiphynema index</i> Dorylaimida Longidoridae	Polyphagous: <i>Vitis vinifera</i> , <i>Ficus carica</i> , <i>Jasminum officinale</i> , <i>Rosa canina</i> , <i>R. foetida</i> , <i>R. sempervirens</i> , <i>R. centifolia</i> , <i>Morus alba</i> , <i>Citrus limon</i> , <i>Persica vulgaris</i> , <i>Quercus borealis</i>	South and Central America. In Europe it was mentioned in 1950. In Romania it was signalled in 1972, in Buzău district; now it is present in the Pietroasele, Segarcea and Valea Călugărească vineyard, with important damages.
<i>Xiphynema vuitenezi</i> Dorylaimida Longidoridae	Polyphagous: <i>Vitis vinifera</i> , <i>Cydonia oblonga</i> , <i>Pyrus communis</i> , <i>Malus sylvestris</i> , <i>Prunus domestica</i> , <i>P. armeniaca</i> , <i>Ribes nigrum</i> , <i>Pinus sylvestris</i>	Central America. In Europe it was mentioned in 1964. In Romania it was detected in 1970, in Constanța district; now are present in Murfatlar vineyard, with important damages.
<i>Xiphynema brevicolle</i> Dorylaimida Longidoridae	In native areal, on <i>Coffea brasiliensis</i> , <i>Citrus aurantium</i> , and in Romania on <i>Vitis vinifera</i> , <i>Ribes nigrum</i> , <i>Rosa foetida</i> , <i>R. centifolia</i> , <i>Persica vulgaris</i>	South America (Brazil). In Europe it was mentioned in 1961. In Romania it was detected in 1970, in Ialomița and Constanța districts; now it is present in Murfatlar, Jidvei, Drăgășani and Ștefănești vineyard and caused important damages.
<i>Brevipalpus lewisi</i> Acar Phytotipalpidae	<i>Vitis vinifera</i> , <i>Citrus aurantium</i>	Australia. In Europe it was mentioned in 1949. In Romania it was detected in 1977, in the south districts; now it caused sporadic damages.

Table 2

Alien/invasive crop pests		
Species, Orders, Families	Hosts trophic range	Native area, year of introduction, actual status in Romania
<i>Heterodera schachtii</i> Tylenchida Heteroderidae	Polyphagous: <i>Chenopodiaceae</i> , <i>Polygonaceae</i> , <i>Cariophyllaceae</i> , <i>Cruciferae</i> , <i>Labiatae</i>	In Europe it was introduced in 1859 in Germany. In Romania it was mentioned in 1910, in Braşov district; at present it caused important yield losses in the sugar beet crops. Invasive species.
<i>Globodera rostokiensis</i> Tylenchida Heteroderidae	<i>Solanum tuberosum</i> and other <i>Solanaceae</i> species	South America. In Europe it was introduced in 1889 in Germany. In Romania it was mentioned in 1984, in Harghita district; now it is present in the centre of Romania and affects seriously the potato crops. Invasive species.
<i>Globodera pallida</i> Tylenchida Heteroderidae	<i>Solanum tuberosum</i> , <i>S. melongena</i> , <i>Lycopersicum esculentum</i>	South America. In Europe it was introduced in 1889 in Germany. In Romania it was mentioned in 1988, in Harghita district; now it is present in the centre of Romania and affects seriously the potato crops. Invasive species.
<i>Meloidogyne hapla</i> Tylenchida Heteroderidae	Polyphagous: over 550 plant species (<i>Daucus carota</i> , <i>Beta vulgaris</i> , <i>Solanum tuberosum</i> , <i>Lycopersicum esculentum</i> , <i>Sonja hispida</i> , <i>Allium cepa</i> , <i>Asparagus</i>)	Tropical regions. In Europe it was introduced in 1961. In Romania it was mentioned in 1972, in Ilfov district; caused important yield losses in the carrot crops. Invasive species.
<i>Ditylenchus destructor</i> Tylenchida Tylenchidae	<i>Solanum</i> , <i>Beta vulgaris</i> , <i>Triticum</i>	North America. In Europe it was introduced in 1888, in Germany. In Romania it was detected in 1966, in Harghita district; it caused severe yield losses. Invasive species.
<i>Deladenus durus</i> Tylenchida Neotylenchidae	<i>Solanum tuberosum</i>	North America. In Romania it was mentioned in 1968, in Cluj district; it caused some damages in the potato crops.

Table 3
Alien/invasive greenhouses pests

Species, Orders, Families	Hosts trophic range	Native area, year of introduction, actual status in Romania
<i>Meloidogyne arenaria</i> Tylenchida Heteroderidae	Polyphagous: vegetables and ornamental plants	Tropical regions. In Europe it was introduced in 1889, in England. In Romania it was mentioned in 1971; important damages registered on <i>Zantedeschia aethiopica</i> and other ornamental plants. Invasive species.
<i>Meloidogyne incognita</i> Tylenchida Heteroderidae	High polyphagous: over 2000 plant species, from 40 families (Solanaceae, Cruciferae, Poaceae, Cucurbitaceae, Umbelliferae, Malvaceae, many ornamentals).	Tropical regions. In Europe it was introduced in 1855, in England. In Romania it was mentioned in 1970; severe yield losses caused in greenhouses at the tomato, eggplant and cucumber crops. Invasive species.
<i>Heterodera cacti</i> Tylenchida Heteroderidae	Polyphagous: <i>Phyllocactus</i> , <i>Cereus</i> , <i>Opuntia</i> , <i>Echinocactus</i> , <i>Epiphyllum</i> , <i>Mammillaria</i> , <i>Aporocactus</i>	Mexic. In Europe it was introduced in 1932, in Holland. In Romania it was mentioned in 1972; now it is sporadically present in greenhouses and collection of cacti.
<i>Brevipalpus inornatus</i> Acari Phytotipalpidae	Polyphagous: <i>Azalea indica</i> , <i>Aralia</i> , <i>Cissus</i> , <i>Campanula</i> , <i>Gardenia</i> , <i>Hedera</i> , <i>Rhododendron</i>	South America. In Europe it was mentioned in 1953, in France. In Romania it was signalled in 1964, in Bucharest.
<i>Brevipalpus cactorum</i> Acari Phytotipalpidae	<i>Mammillaria zeilmanniana</i> , <i>M. oliviae</i> , <i>M. pitcayensi</i> , <i>Echinocactus</i>	Mexic. In Europe it was mentioned in 1832, in Germany (München). In Romania it was signalled in 1929, in Bucharest; important damages were registered only on cactus species.
<i>Tenuipalpus orchidarum</i> Acari Phytotipalpidae	Many <i>Orchidaceae</i> species	Asia (Indonesia). In Europe it was mentioned in 1859. In Romania it was detected in 1978, in Bucharest; reduced damages were registered on <i>Orchidaceae</i> .
<i>Polyphagotarsonemus latus</i> Acari Tarsonemidae	Polyphagous: <i>Camellia sinensis</i> , <i>Citrus aurantium</i> , <i>Gossypium herbaceum</i> , <i>Lycopersicum esculentum</i> , <i>Capsicum annum</i>	Southern Asia. In Europe it was mentioned in 1922. In Romania it was signalled in 1976, in Bucharest; important yield losses were registered in greenhouses on pepper and tomato crops. Invasive species.

Table 4
Alien/invasive orchard pests

Species, Orders, Families	Hosts trophic range	Native area, year of introduction, actual status in Romania
<i>Bryobia rubrioculus</i> Acari Bryobiidae	Polyphagous: <i>Malus domestica</i> , <i>Pyrus sativa</i> , <i>Cydonia vulgaris</i> , <i>Prunus cerasifera</i> , <i>P. domestica</i> , <i>Armeniaca vulgaris</i> , <i>Persica vulgaris</i> , <i>Cerasus vulgaris</i> , <i>C. avium</i> , <i>Vitis vinifera</i> , <i>Amygdalus communis</i>	North America. In Europe it was introduced in 1888, in Italy. In Romania it was signalled in 1958; important damages in apple and cherry tree orchards, registered in the whole country. Invasive species.
<i>Eriophyes pyri</i> Acari Eriophyidae	<i>Pyrus sativa</i> , <i>Cydonia vulgaris</i>	North and South America. In Europe it was mentioned in 1857. In Romania it was detected in 1913; important damages were registered in pear and quince tree orchards.

Table 5

Alien useful species: intentional introduction

Species, Orders, Families	Hosts	Native area, year of introduction, actual status in Romania
<i>Phytoseiulus persimilis</i> Acari Phytoseiidae	Tetranychidae	In Romania it was introduced in 1969 in greenhouses for <i>Tetranychus urticae</i> populations control.

materials, other merchandises, imported ornamental plants or vegetative organs for seeding and breeding; dissemination through natural factors (wind, water). The species of Acarina with piercing-sucking mouthparts are vectors from viral, bacterial and fungal plant diseases.

4. CONCLUSIONS

In Romania between 1910 and 1988, 22 species belonging to Nematoda class (Longidoridae, Heteroderidae, Tylenchidae, Neotylenchidae families) and Acari order (Tarsonemidae, Bryobiidae, Eriophyidae, Phytoptalpidae families) were introduced. Most of these alien species are pests in vineyard, orchard and field crops, and only one is a beneficial species (predaceous *Phytoseiulus persimilis*).

A number of 21 alien pest species were unintentionally introduced, and only one, *Phytoseiulus persimilis*, was introduced intentionally.

The invasive species in Romania are considered *Heterodera schachtii*, *Globodera rostokiensis*, *Meloidogyne incognita*, *M. hapla*, *M. arenaria* (Nematoda Heteroderidae), *Ditylenchus destructor* (Nematoda Tylenchidae) and *Polyphagotarsonemus latus* (Acari Tarsonemidae) and *Bryobia rubrioculus* (Acari Bryobiidae). These invasive species are polyphagous and produce damage in field crops, greenhouses and orchards. The other 14 alien species are not yet, but in the future they could possibly become invasive species.

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AN ULTRASTRUCTURAL STUDY OF CARTILAGE ALTERATIONS IN HUMAN OSTEOARTHRITIS

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In this study, we describe the ultrastructure of chondrocyte subpopulations from hip cartilage of patients with clinical characteristics of osteoarthritis (OA), emphasizing the specific morphological features of cells which are localized in nonlesional and lesional OA areas. The articular cartilage is characterized by the presence of cells with a variable phenotype depending on the cartilage layer in which they are distributed. In the lesional cartilage areas were noticed cell alteration phenomena and modification of the extracellular matrix (ECM). Thus, a common characteristic of the OA lesional cartilage is the presence of chondrocyte clusters or "clones" that can represent the final attempt of chondrocytes to repair the ECM. Some chondrons are degenerated, their cells showing different features of the cell death. A hallmark of the lesional OA cartilage is represented by high frequency of the necrotic cells that could explain the reduction of the OA cartilage cellularity with the evolution of the degenerative process. The alteration of the ECM was more evident at the pericellular and territorial matrix levels where collagen network was disorganized or interrupted with the appearance of short fibrillar fragments. This finding suggested that chondrocytes represent the source of this alteration.

1. INTRODUCTION

Osteoarthritis is an articular degenerative chronic disease very common in the elderly. It is considered a primary disease of cartilage described as a loss of articular cartilage, with joint space narrowing accompanied by secondary bone changes [1]. Despite much research on this condition, the precise mechanisms involved in the disease are not known. The etiology of OA is unknown but it is generally agreed to be multifactorial including genetic factors, genetic malalignment, joint overload or trauma, obesity and aging. The primary cause of cartilage degradation involves the active proteinases degrading extracellular matrix (ECM) constituents (collagens and proteoglycans). The most prominent proteinases in OA are matrix metalloproteinases (MMPs). Of this family, the collagenases, the stromelysins and the gelatinases, have been identified as being elevated in OA [2, 3]. Their biologic activity is controlled physiologically by specific tissue inhibitors of metalloproteinases (TIMPs) or by their activation.

In the past years, important progresses concerning morphology of the human OA cartilage have been achieved [4-6]. However, the majority of the histologic studies

refer to the OA cartilage from animal sources (rats, rabbits, etc.) and less to the human cartilage. Moreover, the main approached problem regards the description of different cartilage zones [7] and secondarily the defining of the ultrastructural characteristics of different chondrocyte types.

2. MATERIALS AND METHODS

Patients. Human femoral cartilage specimens were obtained from twenty four patients with OA (11 men and 13 women between 52 and 68 years old) who underwent arthroplastic surgery. These patients had the typical clinical and radiologic signs of this disease, without signs of other inflammatory diseases.

Tissue selection. The study was performed on a series of 43 biopsy specimens. They were excised from cartilage areas around the OA lesions and from the more normal-appearing tissue distally located. Specimens isolated from the two different areas were considered to be lesional or nonlesional. Special care was taken not to include newly-formed cartilage at sites of osteophyte formation. Slices were diced into smaller pieces and processed for electron microscopic assay.

Transmission electron microscopic (TEM) study. For TEM studies, thin slices of cartilage from lesional and nonlesional areas were fixed immediately with 2.5% glutaraldehyde in 0.1 M cacodylate buffer (pH 7.4), cut into small pieces of about $1 \times 2 \mu\text{m}$, rinsed in buffer and postfixated with 1% osmium tetroxide in 0.1 M cacodylate buffer. They were dehydrated in a graded series of ethanol and embedded in Epon 812. The ultrathin sections were stained with uranyl acetate and lead citrate and were observed with a "Phillips" EM 208 S (1998) electron microscope.

3. RESULTS AND DISCUSSION

Articular cartilage is characterized by a small number of chondrocytes embedded in a specialized microenvironment which insulates them and physically separates the cell from direct interaction with extracellular matrix. The chondrocyte and its pericellular microenvironment together represent the chondron, that is considered the primary structural, functional and metabolic unit of hyaline cartilage [8].

Phenotype variability of human OA chondrocytes. A typical morphologic feature of the normal and OA cartilage is represented by the presence of cells with a variable phenotype depending on the cartilage zone in which they were distributed. Some aspects concerning the ultrastructure of human OA cartilage are still poorly understood, especially those related to the variability in its cellular phenotype. This constitutes an impediment in the study of the OA pathogeny.

Vertically sectioned articular cartilage is divided histologically into four regions: superficial, transitional, radial and calcified layers, with the latter abutting on subchondral bone [9]. The cells in the superficial zone are elongated and located in an ECM with a reduced amount of proteoglycans (PGs) and uniform layers of collagen fibers tangentially distributed with the articular surface. They are surrounded by a thin pericellular matrix, but not by a pericellular capsule. In addition to the cells with an appearance of normal chondrocytes (Fig. 1) characterized by multiple filopodias on their surface and a large euchromatic nucleus, cells with a slightly altered phenotype were noticed. They are characterized by a heterochromatic nucleus, an abundant endoplasmic reticulum, less filopodias and the absence of thin fibrillar pericellular network (Fig. 2). It is possible that these altered chondrocytes to display a high activity of degradation of ECM.

In the lesional cartilage area chondrocyte clusters or "clones" are observed. These cells display a phenotype characterized by a large size and a rounded shape, and by the presence of a large euchromatic nucleus (Fig. 3). In the cytoplasm, numerous mitochondria greatly degenerated, vacuoles, secretory vesicles and small particles are noticed. The phenotype alteration of the "clonal" chondrocytes suggests the alteration of their cytoskeleton. This alteration of cell shape could be associated with ongoing modification in the chondrocyte genotype, since in normal conditions this cell type maintains the regular turnover of the ECM of the cartilage. Pelletier and Martel-Pelletier [10] demonstrated that some cells in the OA cartilage increased the production of MMPs over their inhibitors, causing its degeneration. It is possible that these cells to be the "clonal" cells. Furthermore, in a previous study, Aggeler *et al.* [11] correlated the expression of the procollagenase gene with changes happening in the cytoskeleton and the corresponding cell shape.

The formation of these chondrocyte clones is not clear. However, taking into account that they appear in OA cartilage it can be considered that these structures are a pathologic marker of the cartilage degeneration. Poole *et al.* [12] suggested that these clones, common to the surface of fibrillated defects, can represent the final attempt of chondrocytes to repair the ECM before its disappearance, since fibrillation progressively erodes the cartilage toward its inferior limit.

In the transitional layer, the chondrocytes are distributed isolated or in pairs and display a round or oval shape. Each cell is surrounded by a pericellular matrix that in turn is separated from the adjacent territorial matrix by a pericellular capsule (Fig. 4). The plasma membrane presents numerous filopodias that extend into the pericellular matrix space but do not penetrate beyond the pericellular capsule.

The radial layer contains in its superior region individual chondrocytes with abundant cytoplasmic particles and thin perinuclear filaments, and a dilated endoplasmic reticulum (Fig. 5), while in its deep region chondrons exist containing 3-4 chondrocytes.

In the lesional OA cartilage, some chondrons are degenerated, their cells showing different features of the cell death (Fig. 6). In the pericellular matrix from their close vicinity, the structural network is destroyed suggesting the expression of an intense proteolytic activity by these chondrocytes. The pericellular capsule forms a densely compacted structure and continues with a "weave" of matrix vesicles of different sizes at the articular pole of the superior chondron and at the superior pole of the basal chondron. Matrix vesicles accumulate preferentially at the two chondron poles and are irregularly distributed in the territorial matrix. They are considered to be the result of the pull or physical displacement of cellular chondrocyte processes, of secretion of the synthesized products or of cell death by necrosis and/or apoptosis.

An important feature of the articular cartilage is the presence of the secretory chondrocytes, which vary phenotypically depending on the layer where they are located [12]. These cells could be responsible for regeneration of the injured cartilage, a process that fails during the OA pathogenesis having as a final result the tissue degeneration. They display heterochromatic nuclei, prominent endoplasmic reticulum with dilated cisternae, vesicles and vacuoles (Fig. 7).

Degenerating chondrocytes that are rare in the normal adult cartilage seem to be numerous in the OA cartilage and could represent the morphologic evidence of cartilage turnover which bends to the degenerating process. Although these cells were not identified in chondrocyte cultures, they represent a component of the histopathology of the degenerative process that occurs in the injured human tissue. They are present in different stages of degeneration at the level of all cartilage layers and display pycnotic nuclei, a greatly vacuolated cytoplasm and filiform cytoplasmic projections coming apart from the cell [Fig. 8A, 8B].

Reduction of the OA cartilage cellularity was considered the result of the chondrocyte loss by cell death [14, 15]. Our studies showed chondrocytes in different stages of necrosis such as vacuolated chondrocytes with a tendency of fragmentation, characterized by destruction of the structural network from ECM and the presence of numerous matrix vesicles in the territorial matrix (Fig. 9A), and cell debris that consist of a nuclear or cytoplasmic electro-dense material, delimited by plasma membrane (Fig. 9B). They accumulate inside the lacuna of the initial chondron near the matrix vesicles.

These cells are especially present in the superficial and transitional layers, cell death being a feature of hypertrophic chondrocytes that try to respond to the ECM degradation by the return to the fetal phenotype [16].

It is becoming increasingly apparent that chondrocyte necrosis plays an important role in the development of OA, but it is not known yet if this is the result or cause of cartilage degradation in OA. If chondrocyte death occurs, the synthesis of ECM components ceases and this results in the cartilage destruction. On the other hand, it is known that excepting sanguine cells, the majority of cells require

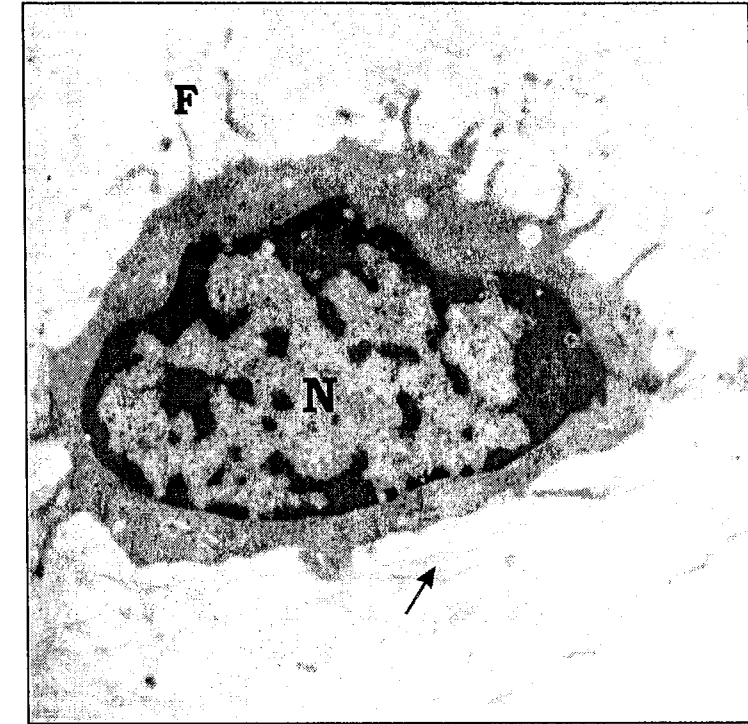


Fig. 1. – Normal chondrocyte of the superficial zone from the nonlesional OA cartilage area. Pericellular collagen fibers (arrow); nucleus (N); fillopodia (F). ($\times 12000$).

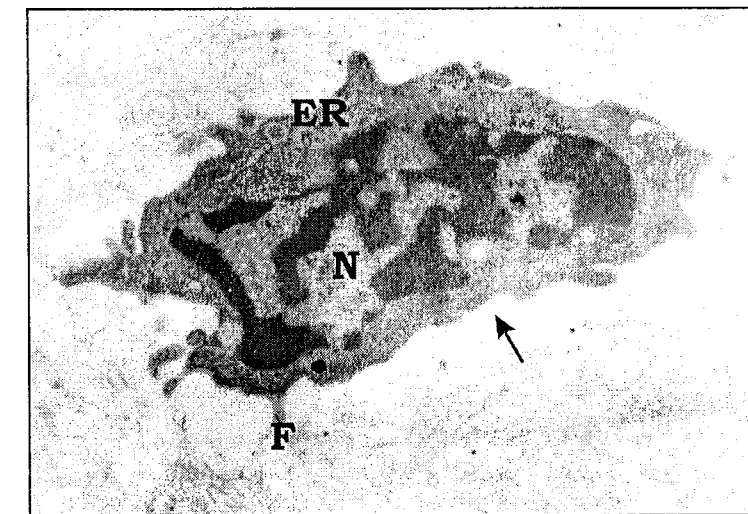


Fig. 2. – Chondrocyte with a slight modified phenotype from the OA cartilage. The absence of the fibrillar pericellular network (arrow); nucleus (N); fillopodia (F); endoplasmic reticulum (ER). ($\times 12000$).



Fig. 3. — Micrograph of a "clonal" chondrocyte. Mitochondria (Mt); vacuoles (v); nucleus (N). ($\times 10000$).

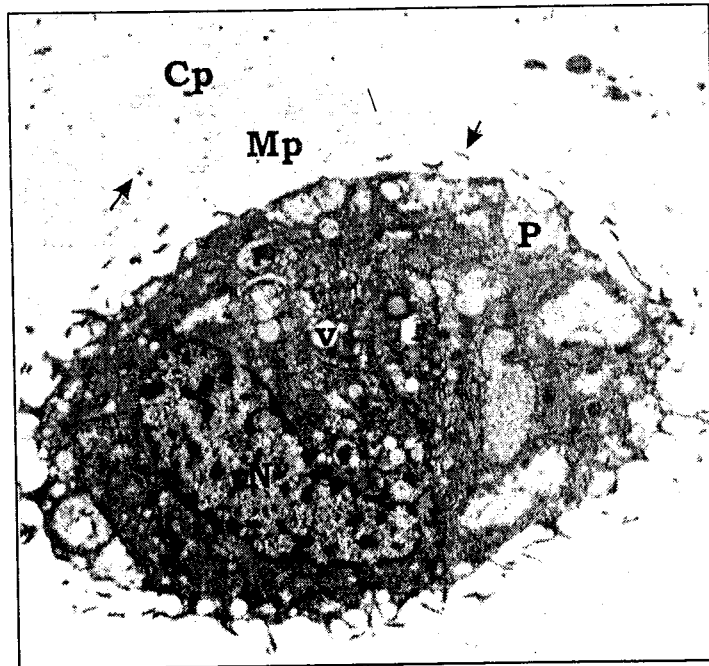


Fig. 4. — Chondrocyte of the transitional layer from the OA cartilage. Pericellular matrix (Mp); pericellular capsule (Cp); matrix vesicles (arrow); nucleus (N); vacuoles (v); small particles (P). ($\times 8000$).



Fig. 5. — Micrograph of a chondrocyte of the radial layer from the OA cartilage. Nucleus (N); perinuclear intermediate filaments (f); small intracytoplasmic particles (P). ($\times 10000$).

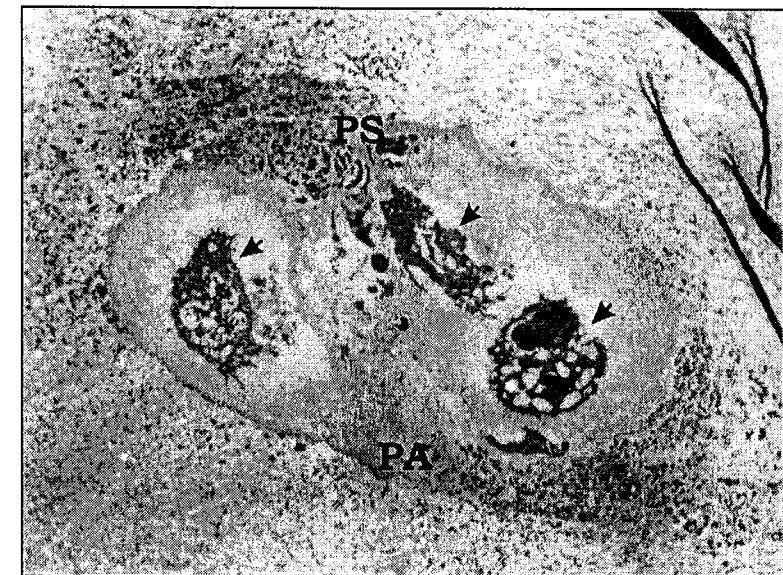


Fig. 6. — Organization of a degenerating chondron of the radial layer from the lesional OA cartilage. Superior pole of the basal chondron (PS); articular pole of the superior chondron (PA); necrotic cells (arrow). ($\times 5000$).

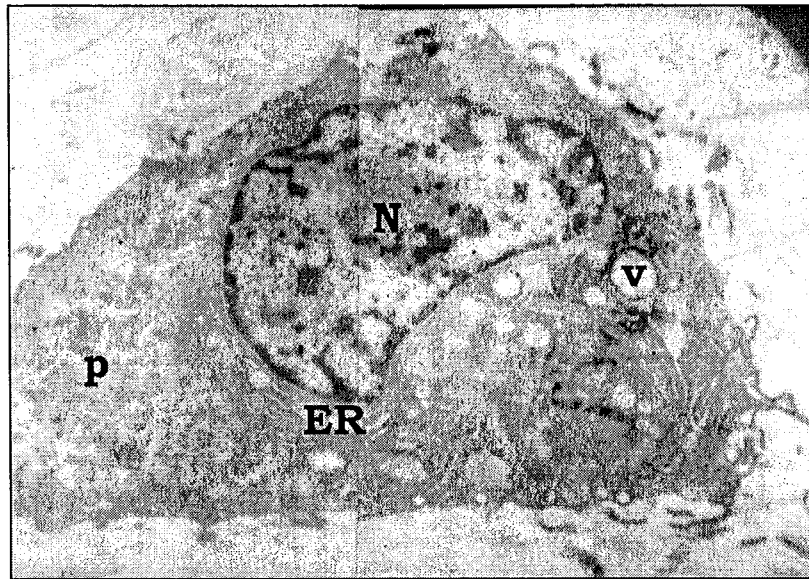


Fig. 7. – Secretory chondrocyte of the transitional layer from the OA cartilage. Nucleus (N); endoplasmic reticulum (ER); vacuoles (v); small intracytoplasmic particles (p); ($\times 16000$).

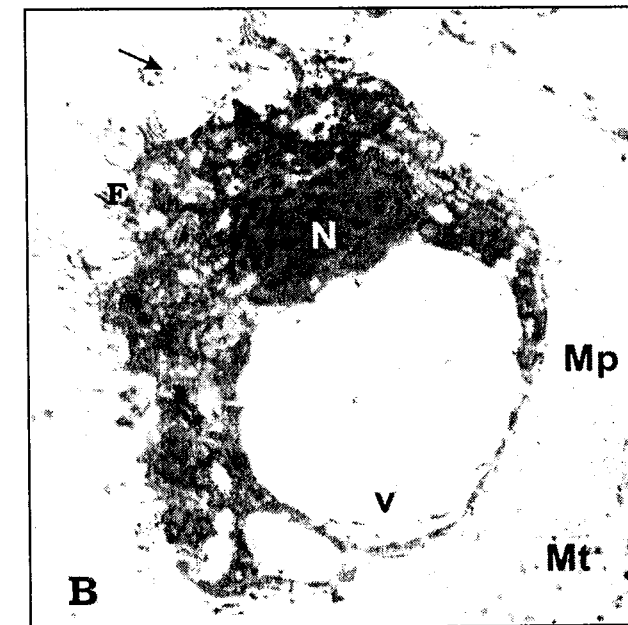
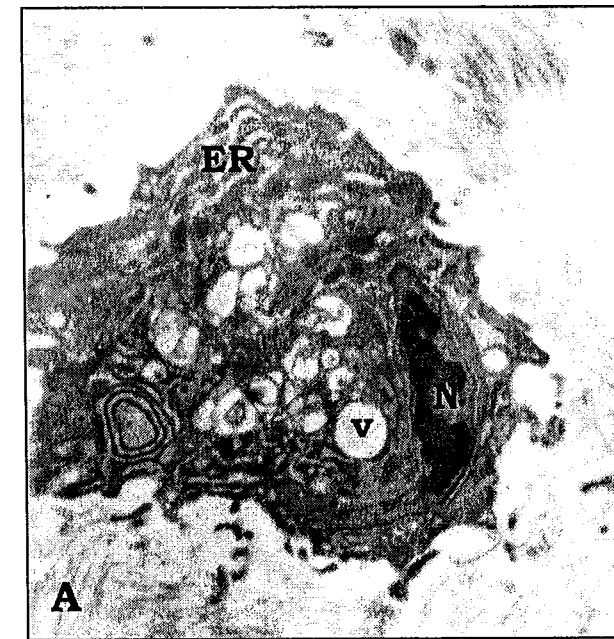


Fig. 8. – Micrograph of degenerating chondrocytes from the lesional OA cartilage. A) Degenerating chondrocyte of the superficial layer ($\times 10000$); B) Degenerating chondrocyte of the transitional layer ($\times 8000$). Nucleus (N); vacuoles (v); endoplasmic reticulum (ER); filopodia (F); pericellular matrix (Mp); territorial matrix (Mt). ($\times 8000$).

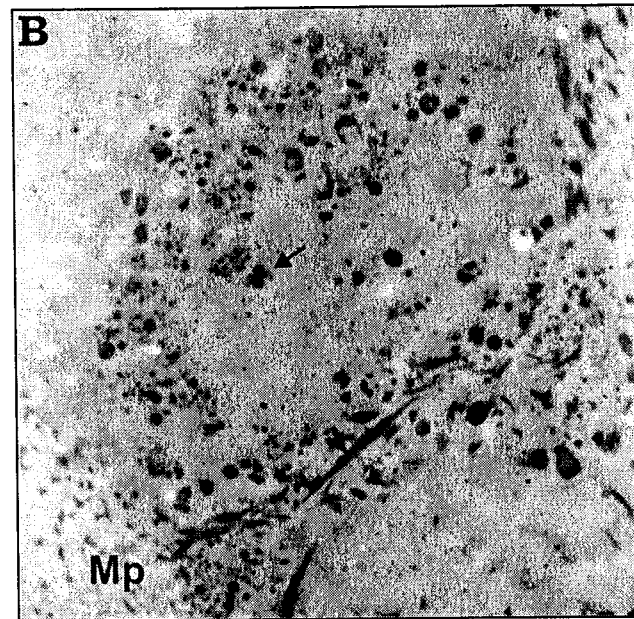
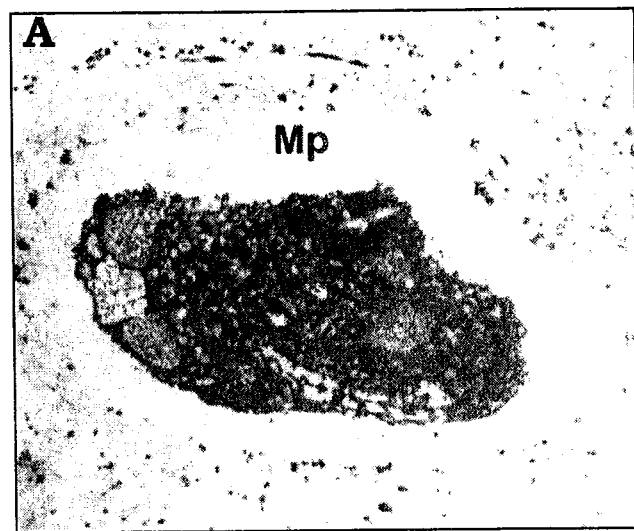


Fig. 9. - Micrograph of necrotic chondrocytes from the lesional OA cartilage. Pericellular matrix (Mp); cell debris (arrow). A) $\times 6500$; B) $\times 10000$.

Fig. 10. - The lacunae (arrow) in the fibrillar collagen network from the ECM of the lesional OA cartilage (superficial layer). ($\times 15000$).

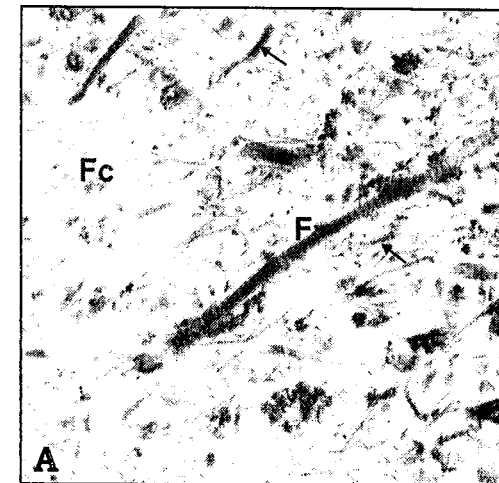
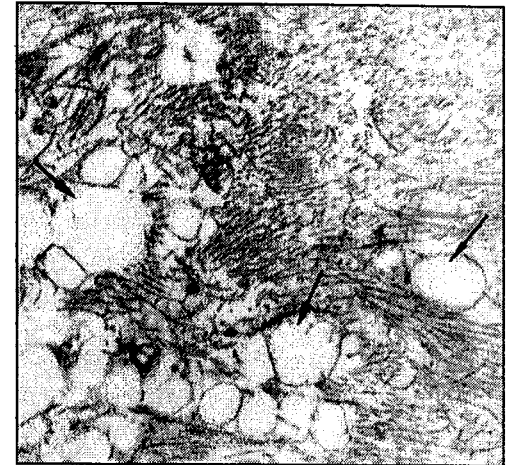


Fig. 11. - Electron micrograph illustrating the territorial matrix disorganization from the lesional OA cartilage. Collagen fibers tangentially distributed with the articular surface (Fc); Collagen fibers longitudinally distributed with the articular surface (F); atypical fibrillar network (Fa); collagen fragments (arrow). ($\times 25000$).

the attachment to other cells or to ECM for proper growth, function and survival [17]. As a main feature of pathologic degradation of the cartilaginous ECM, the proteoglycan loss and the alteration of the collagen network create a microenvironment for perturbation of the chondrocyte attachment to ECM.

The cartilage function is dependent on its ECM that is synthesized and maintained by chondrocytes. The functional properties of the ECM are mainly supplied by a fibrillar network of type II collagen and the major PG aggregate, aggrecan. In the normal adult cartilage, was found a high turnover of aggrecan while the collagen network is subject to a slight remodelling process [18].

Beside cell alterations, OA cartilage is subject to the modification of the ECM. Cartilage ECM consists of two basic components: a fibrillar and a nonfibrillar matrix. The fibrillar matrix is a network consisting mainly of type II collagen, together with other collagens, predominantly type IX and type XI [19]. The nonfibrillar component consists predominantly of highly sulfated aggrecan monomers, attached to hyaluronic acid and link protein that confer osmotic properties and elasticity of the ECM [20]. Cartilage matrix also contains a large number of other components that are important for matrix cohesion and for regulation of chondrocyte function. The small nonaggregating PGs decorin, biglycan and fibromodulin were all found in cartilage ECM [21]. Other proteins that were detected in cartilage matrix include fibronectin, cartilage matrix protein, and cartilage oligomeric protein that may be involved in the regulation of the cellular gene expression pattern and the chondrocyte phenotype [22].

Our ultrastructural studies showed the evidence of collagen alteration at the level of the chondrocyte pericellular microenvironment. Thus, the network of thin collagen fibrils displays interruptions with the appearance of small fibrillar fragments irregularly distributed (Fig. 10).

In the territorial matrix of the intact cartilage are present collagen fibrils which vary in diameter but are thicker than those from pericellular matrix. They exhibit an isotropic arrangement (indicated by the presence of both cross- and longitudinally sectioned elements). In the territorial matrix of the lesional OA cartilage, the collagen network is disorganized, with numerous free spaces and fragmented collagen fibers of different sizes (Fig. 11A) or it is interrupted by a thin fibrillar network, irregular and with evident lysis areas (Fig. 11B). This structure could result following the synthesis by the phenotypic altered OA chondrocytes of some cartilage atypical collagens, such as types I, III or/and V or as a result of the degradative process intensification.

The alteration of the pericellular collagen network could explain the loss of the matrix PGs. Aigner and Dudhia [23] showed by *in situ* hybridization experiments a suppression of the type II collagen and aggrecan expression that is important for the progression of the articular cartilage destruction. At this area level, hypoactive but not hyperactive chondrocytes were found. Phenotypic

characterization of these cells demonstrated that the reduction of their anabolic activity does not seem to be the result of their inactivation but could involve an OA chondrocyte specific differentiation pathway with the expression of atypical gene products such as type III collagen. These studies suggested a three-step evolution of cellular events during the OA cartilage degeneration: 1) an increase in collagen type II and aggrecan synthesis; 2) modulation of the chondrocyte phenotype with the expression of atypical gene products such as collagen type III; 3) suppression of aggrecan core protein and collagen type II (and III) mRNA expression with subsequent quantitative loss of aggrecan molecules from the ECM.

These observations do not preclude the importance of the proteolytic process and implicitly of MMPs as a pathogenic mechanism in the OA cartilage degeneration process, but rather serves to focus this on the damage to the collagen network. Mature collagen fibers are large, cross-linked rigid structures which are probably difficult to repair or replace. In contrast, PGs, with a faster turnover in normal cartilage, may more easily be replaced by newly synthesized molecules.

4. CONCLUSIONS

Our investigations demonstrated a phenotypic alteration of the OA cartilage cells correlated with the intensity of the degenerative process, suggesting thus their active participation to the OA pathogenesis. There were also demonstrated the interruption and disorganization of the collagen network within the ECM with the appearance of lacunae of different sizes and the loss of its compact fibrillar structure at the level and near the OA lesions.

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FLOW CYTOMETRIC ANALYSIS OF HUMAN CHONDROCYTE VIABILITY AND APOPTOSIS IN OSTEOARTHRITIS

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DANIELA BRATOSIN*

Osteoarthritis (OA) is an age-related disorder generally affecting articular cartilage. The ageing of whole organisms is linked to cellular senescence and parallels apoptosis as a cellular response to stress. To understand the mechanisms underlying the process of cell death in cartilage destruction, we investigated by flow cytometry cellular viability and apoptosis of human chondrocytes isolated from normal and osteoarthritic (OA) cartilage. Cell viability was evaluated by applying an original calcein-AM assay we recently proposed (Bratosin *et al.*, *Cytometry A*, 66A, 78–84, 2005) and apoptosis was quantified by changes in Light Scatter Properties, Annexin V-FITC/Propidium Iodide test and caspase-3 activity measurement using PhiPhiLux G₁D₂ as substrate. The results we obtained show that in this context apoptotic phenomena appear to be consistent and relevant and could contribute to cartilage degeneration.

Key words: Chondrocytes, apoptosis, flow-cytometry, viability, calcein-AM, phosphatidylserine exposure, caspase-3.

1. INTRODUCTION

Osteoarthritis (OA) is a chronic disease characterized by the progressive degeneration of cartilage and loss of articular function. In adult vertebrates articular cartilage is devoid of nerves, blood vessels or lymphatics and contains only one cell type, the chondrocyte. These resident cells are highly specialized and solely responsible for the maintenance and turnover of extracellular matrix macromolecules including type II collagen, aggregating proteoglycans and non-collagenous proteins (Muir, 1995). The biochemical performance of cartilage depends on the biochemical and biophysical properties of extracellular matrix macromolecules and thus on the normal metabolic activity and homeostatic status of chondrocytes (De Lise *et al.*, 2000). Consequently, the survival of chondrocytes is essential for normal cartilage function. A number of recently published findings have suggested a strong association between apoptosis, age-related diseases and aging (Warner, 1999; Zhang *et al.*, 2003).

From the phenotypic point of view, aging can be defined as the age-related progressive decline of physiological functions which may be caused by cell death.

There are two distinct patterns of cell death: necrosis and apoptosis. Massive cell injury, often accompanied by inflammation, can lead to necrosis. In contrast to the accidental death of cells, apoptosis, or programmed cell death (PCD), is an active process with specific morphological changes which are characterized by chromatin condensation, nuclear DNA fragmentation, cell shrinkage, plasma membrane blebbing and membrane-enclosed cell fragment (apoptosis bodies) formation. Apoptotic cells are phagocytosed by phagocytes thereby avoiding inflammation (Kerr *et al.*, 1972). Physiological apoptosis naturally occurs in all tissues and plays a vital role in many processes including tissue remodelling during development and cell transformation (Horton *et al.*, 1998). Apoptosis is responsible for balancing cell proliferation with cell death and for maintaining constant cell number in tissues (cellular homeostasis in adults). Furthermore, apoptosis involves specific initiating stimuli and intracellular signals and requires expression of a well-defined set of genes that accomplish the cellular suicide program. In general, apoptosis involves the sequential activation of a proteolytic cascade of enzymes called caspases which have been divided into initiators such as caspase - 8 and -9, and effectors such as caspases -1, -3, -6 and -7. Caspases cleave a number of target proteins as polyadenosine-diphosphate ribose polymerase, I-CAD (inhibitor of caspase-activated DNase) and cytoskeletal proteins.

Adult articular cartilage is thought to be a post-mitotic tissue and chondrocytes show a limited proliferative activity in response to injury, making chondrocyte survival critical for the preservation of cartilage structure and function (Aigner *et al.*, 2002). The decline in the number of articular chondrocytes with age was well documented by Mitrovic *et al.* (1983) and Stockwell (1991). Age-related decrease in chondrocyte number has been correlated with increased frequency of cartilage fibrillation in the human femoral head (Vignon *et al.*, 1976). With reference to the relation between apoptosis and cartilage degeneration in osteoarthritis, a number of recent studies have shown that apoptotic cell death takes place at an increased rate in osteoarthritic cartilage (Aigner *et al.*, 2004). Recent data have demonstrated a relationship between apoptosis and cartilage degeneration in human osteoarthritis (Blanco, 1998; Hashimoto *et al.*, 1998) as well as in experimentally induced animal osteoarthritis (Burton-Wurster *et al.*, 1982). However, in cartilage there are no macrophages and the fate of apoptotic bodies is uncertain. In those conditions, secondary necrosis would inevitably result with the disadvantage of uncontrolled release of lysosomal enzymes from apoptotic bodies causing serious damages to the extracellular matrix, including calcification, unless the cell had developed alternative mechanisms of destruction (Roach *et al.*, 2004). However, not all developmental cell deaths have the distinct morphological features of the "classical" apoptosis identified by Kerr *et al.* (1972). However, evidence has now accumulated from a variety of cell types, including neuronal cells (Fukuda *et al.*, 1999; Sperandio *et al.*, 2000), erythrocytes (Bratosin *et al.*,

2001) and chondrocytes (Roach *et al.*, 1999; 2000) that suggest the existence of more than one pathway for programmed cell death (Leist *et al.*, 2001; Bursch *et al.*, 2000). The aim of this study was to investigate by flow cytometry cellular viability and apoptosis of human chondrocytes isolated from normal and osteoarthritic cartilage.

2. MATERIALS AND METHODS

Materials

Dulbecco's modified Eagle's medium (DMEM) was obtained from Cambrex Bio Science (Verviers, Belgium), fetal calf serum (FCS), penicillin, streptomycin, amphotericin B and L-glutamine were from GIBCO BRL; hyaluronidase, trypsin and collagenase from *Clostridium histolyticum* were from Sigma (St. Louis, USA). Fluorescein-conjugated Annexin-V (Annexin V- FITC), Propidium Iodide (PI) and HEPES buffer were purchased from Pharmingen (San Diego, USA). PhiPhiLux G₁D₂ (OncoImmulin) was furnished by Calbiochem (La Jolla, USA) and calcein-AM by Sigma-Aldrich (St. Louis, USA). The flow cytometer was a Becton-Dickinson FACScan apparatus (San Jose, USA) with CellQuest Pro software for acquisition and analysis.

Isolation of Chondrocytes

Both normal and osteoarthritic articular cartilages, were obtained from patients with osteoarthritis under sterile techniques. Normal cartilage was obtained from the adjacent zone of normal area. Human articular chondrocytes were isolated as described elsewhere (Green WT Jr., 1971; Kuettner K.E., 1982) with a few modifications. All enzymatic solutions were prepared in Dulbecco's modified Eagle's medium (DMEM) supplemented with a mixture of antibiotics and antimycotics (penicillin 10 U/ml, streptomycin 10 mg/ml, amphotericin B 0.025 mg/ml), with L-glutamine 0.002M and with 10% of fetal calf serum (FCS). Articular cartilage (normal and osteoarthritic) was minced into small pieces and incubated with 1mg/ml of sheep testes hyaluronidase in DMEM medium for 20 min at 37° C. The pieces were washed with PBS (Phosphate Saline Buffer) pH 7.4 and maintained in a 0.25g% trypsin solution for 60 min at 37°C. The articular cartilage pieces were washed again with PSB buffer and solubilized overnight in 0.2g% collagenase from *Clostridium histolyticum* in DMEM medium with 10% FCS. Cells were then centrifuged for 15 min at 3000 g, washed with PBS buffer and then centrifuged for 15 min at 3000 g. The pellet was suspended in 2 ml PBS.

Flow cytometric analysis

Flow cytometric analyses were performed on a FACScan cytometer using Pro CellQuest software for acquisition and analysis. Cell in suspension in isotonic

PBS buffer pH 7.4, osmolality 320–330 mosmol kg⁻¹ were gated for the light scatter channels on linear gains, and the fluorescence channels were set on a logarithmic scale with a minimum of 10,000 cells analyzed in each condition.

Flow cytometric assay of cell viability using calcein-AM

Cell viability assessment was studied according to the procedure of Bratosin *et al.* (2005). The membrane-permeable dye calcein-AM was prepared as a stock solution of 10 mM in dimethylsulfoxide stored at -20°C and as a working solution of 100 μM in PBS buffer, pH 7.4. Chondrocytes, (4×10^5 in 200 μl PBS buffer), were incubated with 10 μl calcein-AM working solution (final concentration in calcein-AM: 5 μM) for 45 min at 37°C in the dark and then diluted in 0.5 ml of PBS buffer for immediate flow cytometric analysis of calcein fluorescence retention in cells. Experiments were performed at least three times with three replicates each time.

Cell death assays

Cell death was determined using an Annexin-V-FITC/PI apoptosis kit. Annexin-V is a Ca²⁺-dependent phospholipid-binding protein that has a high affinity for phosphatidylserine (PS) and is useful for identifying apoptotic cells with exposed PS. Propidium Iodide (PI) is a standard flow cytometric viability probe and is used to distinguish viable from nonviable cells. Viable cells with intact membranes exclude PI, whereas membranes of dead and damaged cells are permeable to PI. Cells that stain positive for Annexin V-FITC and negative for Propidium Iodide (PI) are undergoing apoptosis. Cells that stain positive for both Annexin V-FITC and PI are either in the end stage of apoptosis, undergoing necrosis, or are already dead. Cells that stain negative for both Annexin V-FITC and PI are alive and do not undergo measurable apoptosis.

Chondrocytes were washed with PBS buffer pH 7.4 and the cells (2×10^5) were resuspended in 100 μl of 1×binding buffer (10 mM HEPES/NaOH, 140 mM NaCl, 2.5 mM CaCl₂, pH 7.4). 10 μl Propidium Iodide and 5 μl Annexin V-FITC were added and incubated for 30 min at room temperature in the dark. After adding 400 μl of 1×binding buffer, the suspension was analysed in the flow cytometer and gated for biparametric histograms FL1 (FITC fluorescence) *versus* FL2 (PI fluorescence). The light scatter channels were set on linear gains and the fluorescence channels on a logarithmic scale. All studies were performed at least three times, with three replicates each time.

Caspase-3 activity determination

Caspase-3 activity was assessed using the fluorogenic peptide substrate PhiPhiLux G₁D₂ (green fluorescence). 10⁶ cells were labelled with 50 μl of

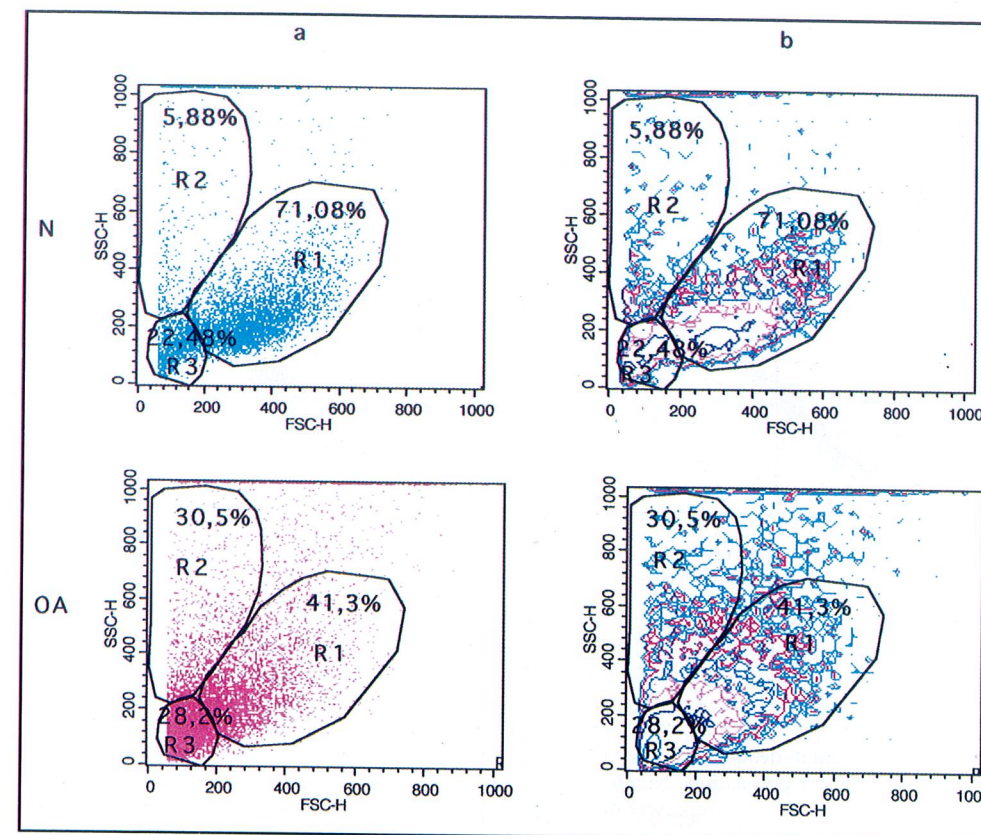


Fig. 1. – Comparative flow cytometric analysis of morphological changes of human normal (N) and osteoarthritic chondrocytes (OA). a: Dot-plot analysis FSC/SSC; b: Density plot analysis FSC/SSC of chondrocyte shape changes. Abscissae: forward scatter (cell size); ordinates: side scatter (cell density, granularity and refractiveness). Number of counted cells: 10,000. Results presented are from one representative experiment of three performed.

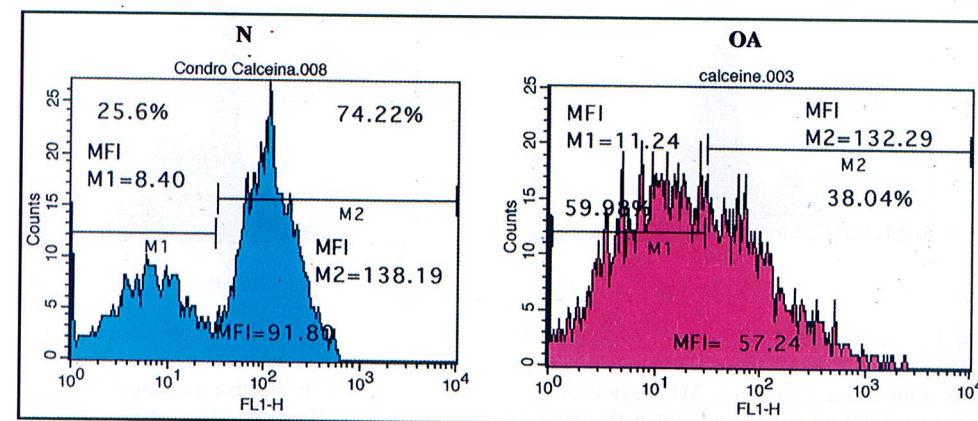


Fig. 2. – Comparative flow cytometric histogram analysis of calcein-AM cell viability of human normal (N) and osteoarthritic chondrocytes (OA). M2: region of fluorescent cells with intact membranes (living cells) and M1: region of nonfluorescent cells with damaged cell membranes (dead cells). Data are expressed as mean fluorescence intensity (MFI) and percentages of both regions M1 and M2. Abscissae: log scale green fluorescence intensity of calcein (FL-1). Ordinates: relative cell number. Number of counted cells: 10,000. Results presented are from one representative experiment of three performed.

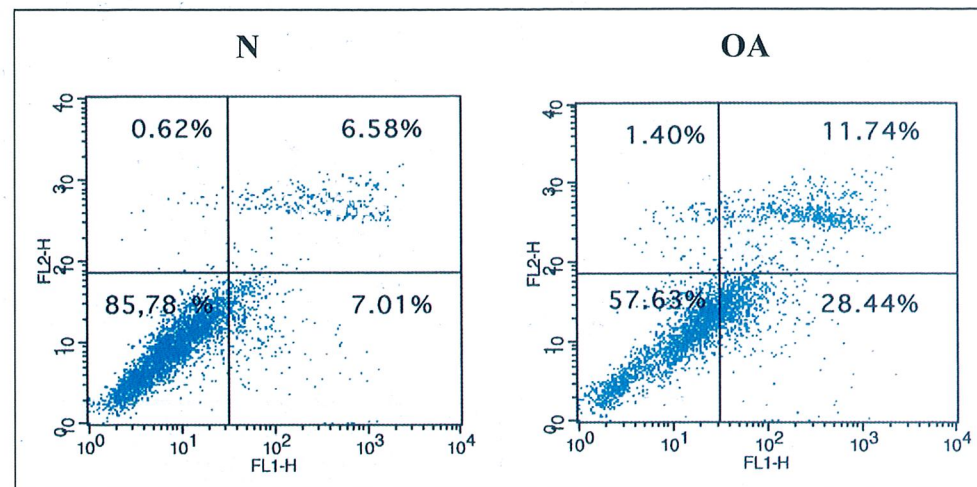


Fig. 3 – Comparative flow cytometric quadrant analysis of Annexin-V-FITC / Propidium Iodide double-stained of human normal (N) and osteoarthritic chondrocytes (OA). Abscissae: log scale green fluorescence intensity of Annexin-V-FITC (FL-1). Ordinates: log scale red fluorescence intensity of Propidium Iodide (FL-2). Low left quadrant: viable cells (Annexin-V and Propidium Iodide negative cells); low right quadrant: apoptotic cells (Annexin-V positive and Propidium Iodide negative cells); upper right quadrant: dead cells (Annexin-V and Propidium Iodide positive cells). % refers to the cell percentage of each population. Number of counted cells: 10,000. Results presented are from one representative experiment of three performed.

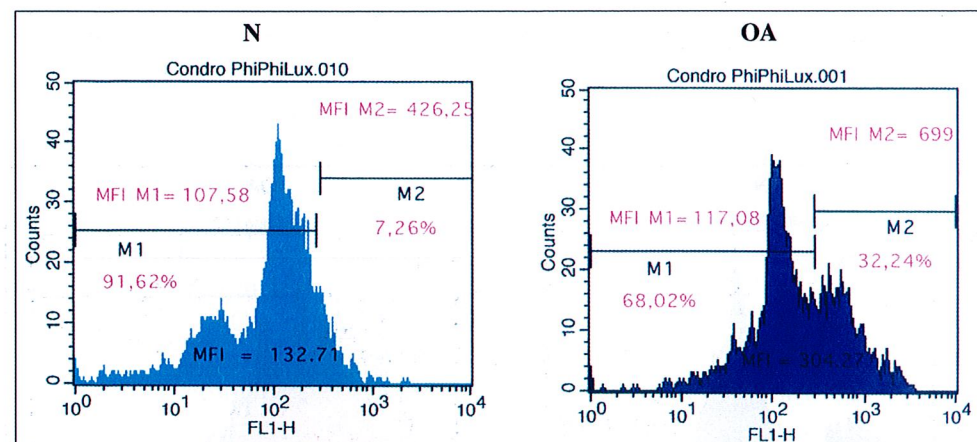


Fig. 4 – Comparative flow cytometric histogram analysis of caspase-3 activity in human normal (N) and osteoarthritic chondrocytes (OA) using PhiPhiLux G_1D_2 fluorescent substrate. M2: region of fluorescent cells with active caspase-3; M1: region of nonfluorescent cells with inactive caspase-3. Data are expressed as mean fluorescence intensity (MFI) and percentage of both regions M1 (caspase-3 negative cells) and M2 (caspase-3 positive cells). Abscissae: log scale green fluorescence intensity of PhiPhiLux G_1D_2 (FL-1). Ordinates: relative cell number. Number of counted cells: 10,000. Results presented are representative of three independent experiments giving similar results.

PhiPhiLux G_1D_2 (10 μ M) substrate solutions following the manufacturer's protocol Oncoimmunin. The suspensions were incubated in 5% CO_2 at 37°C for 1 h. Cleavage of the peptide linker of PhiPhiLux G_1D_2 (sequence DEVD) results in fluorescence detectable on the FL1 channel. After washings, they were analyzed by flow cytometry. Experiments were carried out in triplicate.

3. RESULTS AND DISCUSSIONS

Light scattering properties of chondrocytes in osteoarthritis

Analysis of the scattered light by flow cytometry provides information about cell size and structure. The intensity of light scattered in a forward direction correlates with cell size. The intensity of scattered light measured at a right angle to the laser beam (side scatter), on the other hand, correlates with granularity, refractiveness and the presence of intracellular structures that can reflect the light. The cell's ability to scatter light is expected to be altered during cell death, reflecting the morphological changes such as cell swelling or shrinkage, breakage of plasma membrane and, in the case of apoptosis, chromatin condensation, nuclear fragmentation and shedding of apoptotic bodies. During apoptosis, the decrease in forward light scatter (which is a result of cell shrinkage) is not initially paralleled by a decrease in side scatter. A transient increase in the right angle scatter can be seen during apoptosis in some cell systems. This may reflect an increased light reflectiveness by condensed chromatin and fragmented nuclei. However, in later stages of apoptosis, the intensity of light scattered at both forward and right angle directions decreases. Cell necrosis is associated with an initial increase and then a rapid decrease in the cell's ability to scatter light simultaneously in the forward and right angle direction. This is a reflection of an initial cell swelling followed by plasma membrane rupture and leakage of the cell's constituents (Darzynkiewicz *et al.*, 1997).

Figure 1 shows that morphological changes of human osteoarthritic chondrocytes were associated with cell shrinkage (decreased forward scatter and increased side scatter), one of characteristic features of apoptosis (Fig. 1, OAa and OAb). The percentage of cells in region R1 (increased FSC and decreased SSC) of osteoarthritic chondrocytes (Figs. 1 OAa and OAb) decreased drastically from 71% for normal chondrocytes (Figs. 1 Na and Nb) to 41%. Concomitantly, the proportion of osteoarthritic chondrocytes (region R2: decreased FSC and increased SSC) increased from 5.88% to 30.5% (Figs. 1 OAa and OAb). Region R3 is constituted of cell fragments which are present in equal proportions (22 to 28%) of total cell populations.

Cell viability calcein-AM assay of chondrocytes in osteoarthritis

We recently devised a new flow cytometric assay for the measurement of cells viability using calcein-AM (Bratosin *et al.*, 2005). The assay is based on the use of acetoxymethyl ester of calcein (calcein-AM), a fluorescein derivative and

nonfluorescent vital dye that passively crosses the cell membrane of viable cells and is converted by cytosolic esterases into green fluorescent calcein which is retained by cells with intact membranes (Bratosin *et al.*, 2005).

Application of this assay for analysing normal and osteoarthritic chondrocytes showed that two regions could be clearly and unambiguously defined: the region of fluorescent chondrocytes with intact membranes that is related to intracellular esterase activity and strongly correlated with the number of living cells (region M2) and the region of nonfluorescent dead cells with damaged cell membranes (region M1). As shown in Fig. 2, the number of viable cells (region M2) in osteoarthritic chondrocyte population decreased drastically (around 38%) as compared to normal chondrocyte population (around 74%). In this regard, it is important to mention that we have previously demonstrated that the loss of esterase activity was an early event that occurred before phosphatidylserine exposure (Bratosin *et al.*, 2005). A loss of chondrocyte viability may play a key role in the progression of the joint disease osteoarthritis in humans (Blanco *et al.*, 1998) and in a canine model (Burton-Wurster *et al.*, 1982).

Study of chondrocytes death by Annexin V-FITC and Propidium Iodide double-labelling

Normal and osteoarthritic chondrocytes were analyzed by flow cytometry for phosphatidylserine (PS) exposure (Annexin-V labelling) and membrane permeabilization (Propidium Iodide labelling). Phosphatidylserine residues are exposed in the external leaflet of cell membrane early during the process of apoptosis whereas the uptake of Propidium Iodide indicates a disrupted cellular membrane integrity generally observed during late apoptosis and cell necrosis.

Figure 3 shows comparative flow cytometric analyses of normal (N) and pathological chondrocytes (OA). The number of live cells (Annexin⁻/PI⁻) decreased drastically (86% and 57%, respectively). The proportions of Annexin-V positive and PI negative cells (Annexin⁺/PI⁻) were significantly increased (28.44%) for osteoarthritic chondrocytes contrary to 7% for normal chondrocytes. The proportions of Annexin-V positive and PI positive cells (Annexin⁺/PI⁺) indicating late apoptosis are almost similar for both cells: 6.58% for normal chondrocytes and 11.74% for osteoarthritic cells. In conclusion, approximately 30% of the chondrocytes were found to be apoptotic cells in human osteoarthritic cartilage, whilst very few of such cells were detected in normal cartilage, probably because of mechanical or enzymatic injury occurring during chondrocyte isolation.

Caspase-3 activity determination

Current knowledge of apoptosis has put caspase activation at the center of the apoptotic machinery. Multiple apoptosis-inducing factors directly or indirectly activate the cascade of caspases which are the executioners of death sentence by

catalyzing protein degradation so leading to cell death. In this viewpoint, we focused our experiment on the caspase-3, known as the executioner caspase of the so-called "nuclear apoptosis". Caspase-3 activation was assessed by flow cytometric analysis using the fluorogenic caspase-3 substrate PhiPhiLux G₁D₂. As shown in Figure 4, a significant cleavage of the fluorogenic substrate (M2 region) was detected in osteoarthritic chondrocytes. In fact, 32.24 % of these cells showed an MFI (mean fluorescence intensity) of 699 while only 7.26% of normal chondrocytes present a caspase-3 activity with an MFI of 426.25.

4. CONCLUSIONS

The results presented in this study demonstrate that chondrocytes constitutively express the death machinery capable of inducing apoptosis in osteoarthritis pathogenesis. In fact, we have shown by flow cytometric analyses that PCD of osteoarthritic chondrocytes display apoptotic features with morphological changes, loss of cell viability, phosphatidylserine exposure and significant caspase-3 activation. These results could represent in the future a potential therapeutic target in articular diseases to limit chondrocytic cell death.

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THE BIODIVERSITY OF THE SUBTERRANEAN ENVIRONMENTS OF ROMANIA

Introduction to the biology, ecology, conservation and protection of the terrestrial
and aquatic subterranean environments. Short dictionary of terms

ALEXANDRINA NEGREA, ȘT. NEGREA, A. ARDELEAN

The book represents the work of two of Romania's and the world's best known biospeleologists, Ștefan Negrea and Alexandrina Negrea, from the "Emil Racoviță" Speleological Institute in Bucharest and of Prof. Dr. Aurel Ardelean, a mecena of ecology and rector of the "Vasile Goldiș" University in Arad.

The 8 chapters, the selective references and the short original dictionary of biospeleological terms, form the first book of this kind in Romania, a world level synthesis of the faunistic, ecological and biogeographical researches on Romania's terrestrial and aquatic subterranean environment.

The first chapter presents a short history of the Romanian biospeleology, its development before and after Emil Racoviță and evokes the contributions of dedicated scientists.

This introductory chapter is followed by chapters on the subterranean flora and fauna of Romania and by very useful identification keys and pictures of the different terrestrial and aquatic fauna groups.

A third chapter is dedicated to the collection and conservation methods of the subterranean fauna for the scientific study of the characteristic biodiversity of the environment.

The authors have put together a very useful form that will help researches in getting a complete characterization of each cave and of its most important abiotical and biotical factors.

Chapter number four, the karstic and biospeleological regions of Romania, shows the evolutive stages in the karsts formation of the five biospeleological provinces of Romania, accompanied by maps illustrating the origin and evolution of the subterranean fauna. The five provinces have a very diversified subterranean fauna with many troglontic endemics belonging to the Insecta, Isopoda, Chilopoda, Aranea, Pseudoscorpionida, Collembola.

The fifth chapter, the Terrestrial and Aquatic Subterranean Ecosystems from Romania, contains the following sub-chapters: the classification of the subterranean environments, the characteristics of the subterranean environments, the ecologic categories and their adaptation to the subterranean life, the terrestrial and aquatic underground ecosystems and the ecology of the aquatic underground microorganisms.

An original schematic representation of the trophic cycles in the Romanian caves (alotrophic and chemotrophic) gives a full picture on the life in subterranean environment.

Chapter number six is dedicated to the conservation of the subterranean ecosystems from Romania and includes the following sub-chapters: the diversity of the subterranean environments and fauna; the causes for the deterioration of these environments, the causes for the deterioration of the subterranean animal populations, the subterranean environments and fauna and lastly methods for protection and conservation and legal aspects.

Romania has signed European and international conventions on the habitat and the subterranean species conservation. The authors have included a list of the applicable laws as well as legal recommendations for preserving the natural biospeleological patrimonies.

REV. ROUM. BIOL.-BIOL. ANIM., TOME 50, N°s 1-2, P. 115-116, BUCAREST, 2005

Chapter seven is a comprehensive study on the influence of the speleological environment on humans in general and in conditions of total isolation.

Although speotherapy is not one of the most popular therapy methods in Romania, such caves or salt mines like Peștera Urșilor, Ocna Dejului, Slănic Moldova, Târgu Ocna and Slănic Prahova have been used for a long time for the healing properties of their environment in respiratory diseases.

The last chapter, "Biodiversity of the Subterranean Environments According to the Modern Evolutionary Theories" includes comprehensive sub-chapters on the colonization, evolution and speciation in the speleic and interstitial environment. According to Șt. Negrea, the troglobitic and stygobitic species are actually the result of the adaptation and integration of some populations in the subterranean biocoenosis.

The references include the most important Romanian and international titles (210), out of which the authors synthesized a compendium of the latest biological news. The book contains both original illustrations and reproduced drawings from the bibliography.

The present study is an extremely useful learning instrument to a broad audience, from researchers and university students to ecologists and spelunkers. "The Biodiversity of the Subterranean Environments of Romania", with its broad perspective and scientific rigor at the same time, will remain for a long time the most important Speleology synthesis in Romania and one of the best works of this kind in the world.

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AVIS AUX COLLABORATEURS

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 plus récentes parutions dans la littérature.

Les auteurs sont priés de présenter leurs articles en double exemplaire, imprimés de préférence sur une imprimante laser et espacés à double interligne. Le contenu des articles sera introduit sur des disquettes dans un langage connu, préférablement Word 6.0. La composition et la mise en vedette seront faites selon l'usage de la revue: caractères de 11/13 points pour le texte, de 12/14 points pour le titre de l'article et de 9/11 pour les annexes (tableaux, bibliographie, explication des figures, notes, etc.) et le résumé en anglais de 10 lignes au maximum, qui sera placé au début de l'article. Il est obligatoire de spécifier sur les disquettes le nom des fichiers ainsi que le programme utilisé.

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Les textes ne doivent pas dépasser 10 pages (y compris les tableaux, la bibliographie et l'explication des figures).

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