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# Characterisic soil mite's communities (Acari: Gamasina) for some natural forests from Bucegi Natural Park – Romania

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#### List of nonstandard abbreviations:

- OLF = litter-fermentation soil layer
- OH = humus soil layer
- = significance level D
- = rank correlation r

Key words: correlation, cluster, forest, gamasid, humus, litter.

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## ABSTRACT

Background and Purpose: One of the most important characteristics of a natural ecosystem is its stability, due to the species' and communities' diversity. Natural forest composition model constitute the main task of the present-day management plans. Soil mites are one of the most abundant edaphic communities, with an important direct and indirect role in decomposing, being considered bioindicators for terrestrial ecosystems. The aim of the paper was to identify characteristics of soil mites community's structure.

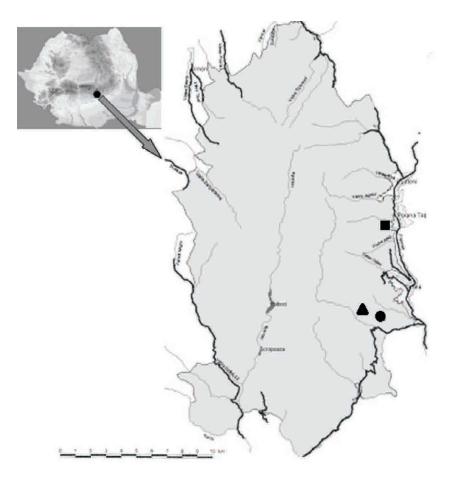
Materials and Methods: Soil mites community's structure (composition of the species assemblage, abundance of the species, species associations and interdependence between species) from three mature natural forest ecosystems, from Bucegi Natural Park -Romania, were analyzed using statistical analyses, which combine two different methods: cluster analysis and correlations.

Results: Two different species associations were described. One of them was identified as stable association in a hierarchical cluster and another, subset of the first, was composed by species pair wise positive correlate (monotonic associations). The number of species grouped in stable associations was similar in both soil layers (OLF and OH), but different in every type of ecosystems: lowest in fir forest, medium in spruce area and the highest in beech forest. 66.6% of species from stable associations were included in monotonic association. Veigaia nemorensis and Neopodocinum mrciaki were common species for all investigated ecosystems, in both soil layers, as well as for stable and monotonic associations.

**Conclusions:** The present study revealed that each type of ecosystem was characterised by a stable and monotonic associations. 24.74% from all identified species were grouped in these associations.

# INTRODUCTION

ATURAL forests are complex and stable ecosystems. Over time, the structure and function of a natural ecosystem should remain relatively stable, even in the face of disturbance. These characteristics are due to factors that provide ecosystem's stability, as: species diversity (interactions, life strategies), trophic complexity (food web structure) and nutrient or energy flux. On soil level, interrelations between soil microarthropods, influence physical, chemical and biological processes. These microarthropds are characterized by a great diversity, one of the most abundant groups being predatory soil mites (20, 21, 23, 39, 45).



**Figure 1**. Geographical position of Bucegi National Park, Romania ( $\blacksquare$  = ecosystem with Abies alba;  $\blacktriangle$  = ecosystem with Picea abies;  $\bullet$  = ecosystem with Fagus sylvatica).

Additional informations concerning one of the soils biological components of the terrestrial ecosystem constitute an important database for the forest conservation, especially in natural protected areas. Predatory soil mites (Mesostigmata: Gamasina) are the main regulatory mechanism for other soil invertebrates as i.e. springtails, nematodes, enchytreides, oribatids. In the soil trophic web, mites are considered consumers from the third level (18, 19, 20). They are very mobile arthropods, capable to migrate in different habitats, favorable according to their ecological requirements. Peculiar environmental conditions determine specific population structure. Any disturbance can induce quantitative (abundance) and qualitative (species composition) modifications on soil mite communities. These modifications are specific "signals" of the soil ecosystem changing, and gamasid mites could be consider as one of the bioindicator group, with potential to use in forestry practices. This is the reason that the gamasid could be considering one of bioindicators group for terrestrial ecosystems (7, 11, 26, 34, 38, 44). In Europe, as well as in Romania, studies of soil predatory mites from protected areas using population parameters (abundance, numerical density, constancy, dominance, species diversity) demonstrated that the environment conditions (abiotic factors: soil temperature, humidity, pH, organic matter; biotic factors as vegetal association) and human impacts, influenced the structure and dynamics of these invertebrates' communities (8, 9, 10, 13, 14, 27, 28, 39, 41).

The aim of the paper was to identify the structure of studied mite communities (composition of the species assemblage, abundance of the species, species associations and interdependence between species) from three mature natural forest ecosystems, from Romania, by combining two different methods: cluster analysis and Spearman correlations. This study provides valuable information concerning one of the most important soil invertebrate groups from terrestrial ecosystems, taking into account that a "natural forest" composition model is the main goal of present-day management plans.

## MATERIAL AND METHODS Investigated area

One of the most important protected areas from Romania, having a rich diversity of flora and fauna is Bucegi Natural Park (BNP). It is situated on the east side of

#### TABLE 1

Description of the investigated forest ecosystems from BNP.

Characteristics	Picea abies	Abies alba	Fagus sylvatica
Geographical	N = 45°21'09.15";	N = 45°23'50.20''	N = 45°20'56.45";
Coordinates	E = 25°31'10.59''	E = 25°32'00.07''	E = 25°31'24.9"
Altitude	1350 m	950–1000 m	1200 m
Exposure	SE	NV	S
Slope	45°	10°–15°	10°
Area (ha)	12.9	16	3
Litter	Continuous-very thin layer	Continuous, 3–4 cm	Continuous, 4–5 cm
Soil	Mull-moder humus	Rich in humus and sandy	Brown eumesobasic, with clayey-sandy fine texture
Vegetal association	Leucanthemum waldsteinii	Oxalis-Pleurozium	Oxalis-Dentaria-Asperula
Actual composition	9 spruce: 1 beech	10 fir	10 beech
Age (years)	110	150	110
Production class	4	2	3
Volume (mc/ha)	405	658	399
Growth (mc/ha)	3.5	5.9	4.6

Meridional Carpathians and includes whole Bucegi Massif. This is developed in an amphitheater shape with a wide south opening and limited by the peaks, which often reach to 1000 m altitude, in comparison with adjacent areas. The BNP has a total area by 32.663 ha (Figure 1).

The ecological investigation was made in 2001-2003, in three forest ecosystems from BNP. The investigated forest ecosystems are described below (Table 1) *(6, 27, 28)*.

#### **Mite samples**

The size of one sampling plot was  $10000 \text{ m}^2$  (1 ha). The soil samples were collected monthly, with a random stratification method, using a square metal core (1000 cm<sup>3</sup>). Stratified sampling entails partitioning the soil sample into two subsamples, taking account of the soil layers: litter - fermentation layer (OLF) and humus layer (OH). Each soil sample was divided into 500 cm3 subsamples. The surface of one soil sample was by 10\*10 cm. The depth of the sample was by 10 cm. In both years, 1008 soil samples were analyzed, divided in 2016 subsamples (OLF and OH). An equal number of soil samples were collected from all investigated ecosystems. The extraction of the mites was made in 10-14 days, by Berlese – Tullgren method, modified by Balogh (1972). The samples have been kept in refrigerator, till next extraction. In all ecosystems 97 species were identified, with 23441 individuals. The mite's numbering and identification were made by a Zeiss stereomicroscope and by Axioscope A1 Zeiss microscope, provided by the Carl Zeiss Instruments S.R.L. Some of the mites were mounted whole on glass slides in Hoyer's medium *(21)*. Several mite specimens were dissected under a stereoscopic microscope after clearing in lactic acid. Each body part was mounted in Hoyer's medium or polyvinyl alcohol–lactic acid mixture (PVA) medium. The conservation of the gamasids fauna was made in an ethyl alcohol (70%). All identified species are in mites' collection of the Institute of Biology – Ecological Stationary from Posada.

#### Data processing

To perform the cluster analysis we build up a data matrix on the base of site observations. At each moment of time there was 14 soil samples collected in each forest. One denotes by  $t_i$  i=1.24 the moment of time observations, by  $z_a$  a=1.14 the site of observation and by  $y^{\alpha}_{ai}$  the number of individuals of the  $\alpha$ -species recorded at the time  $t_i$  and at the site  $z_a$  then the entries of the data matrix  $y^i_{\alpha}$  are given by.

$$y_i^{\alpha} = \sum_a y_{ia}^{\alpha}$$

To investigate the species association we use two different method, cluster analyses and Spearman rank correlation. The cluster analysis is first performed to identify one of more homogeneous groups of species and then we study the correlations within each group. The second analysis is performed in order to augment the meaning of the clusters.

In performing cluster analysis one need a dissimilarity function and a method to make clusters. In our simulation the best results have been obtained by using the Hellinger dissimilarity function, in connexion with hierarchically clustering method and single-link method. The quality of the structure is quantified by a cluster validity index and there exist a lot of internal validity indexes. One of them is cophenetic correlation coefficient that is the Mantel normalized statistics of the cophenetic matrix associate to the dendrogram and the distance matrix. On the base of the cophenetic correlation coefficient one accept a dendrogram as valid if the index is closer to one. In our analyze we use the cophenetic correlation coefficient to test the validity of dendrogram obtained by using different distance function. By using different data matrixes we find that, on average, the cophenetic correlation coefficient is higher when one uses the Hellinger dissimilarity function. The Hellinger dissimilarity function perform itself a data transformation and if one use another data transformation there exists a danger to falsify the initial data structure (24).

$$d_{\alpha,\beta} = \sqrt{\sum_{i} \left( \sqrt{\frac{y_i^{\alpha}}{y^{\alpha}}} - \sqrt{\frac{y_i^{\beta}}{y^{\beta}}} \right)^2}$$

For more information about the applications of the cluster analysis in the ecological data the see e.g. Legendre and Legendre (25).

We inquire the similarities of the time series of abundance of different species. There is a basic asumption that motivate our demers, namely, if two time series are similar then the response of the two species to the external factors is similar. On the ground that this asumption is true we tried to put in evidence one or more groups whose members behave in common manner. The cluster analysis is an widely used method to partioned a set into homogeneous subset and we apply it. Related to the cluster analysis there are two major problems: the number of clusters from a given set and the evaluation of the clustering results (12). To solve the problems we investigate the stability of the clusters and then we analyse the pairwise rank correlation within stable associations (36)

Let O be the set of all species. A subset M of O is named stable association if it is a cluster of the hierarchical structure corresponding to a given data matrix and remain a cluster if one adds new entries to the data. Taking into account that the few species are all time present, we investigate the stable associations by considering variable number of species as they are recorded through sample period. We consider the cases of at least 6, 12 and 18 present records in the time series. For all habitats there was only one stable association per habitat (Figure 3).

Next step in validation was to analyse the rank correlation of the species in the stable association. If there exist a subset such that the species in each paire are positive Sperman rank correlate we call that subset monotonic association. All calculations were made by using a C home made soft, and the graphics was drawing by using Xfig and gnuplot programs from GNU. The algorithms in the soft are most similar with some of well known statistical package R and can be obtained by request from one of the authors (35).

# **RESULTS AND DISCUSSION**

#### **Community composition**

A total of 23.441 gamasid mites were counted, belonging to 97 species. The maximum number of species was recorded in fir ecosystems (80 species), followed by the beech forest (73 species) and the minimum value was obtained in spruce ecosystem (68 species) (Table 2).

Analysing on soil layers, the number of species ranges from 64 species to 73 species, in OLF, and from 52 species to 60 species in OH. In *Picea abies* forest was recorded the highest number of individuals, contrary to *Abies alba* ecosystem with lowest number if individuals.

On OLF layer, on *Abies alba* forest the highest values of abundances rang from 7 % to 23% and on frequences from 22% to 24% (Figure 2 A1). In *Picea abies* ecosystem, some species recorded the highest values of both populational parameters, as *Neopodocinum mrciaki, Pergamasus athiasae* and *Eviphis ostrinus* (Figure 2 B1).

In *Fagus sylvatica* area, *Neopodocinum mrciaki* obtained the most increased values of abundance (54%) and frequency (23%) (Figure 2 C1).

In OH, in all investigated forestry ecosystems, species abundance and frequency recorded lower values, in comparison with those obtained in OLF layer (Figure 2 A2, B2, C2).

#### **Species Associations**

Analysing the mite populations from OLF, the percents of species grouped in stable associations were different: from 8.21% in fir forest to 14.06% in spruce forest and to 17.39% in beech forest. In OH, these values were closed to those obtained by species identified in OLF: 9.67% in fir forest; 15% in beech forest and 15.38% in spruce forest (Table 2; Figure 2, 3).

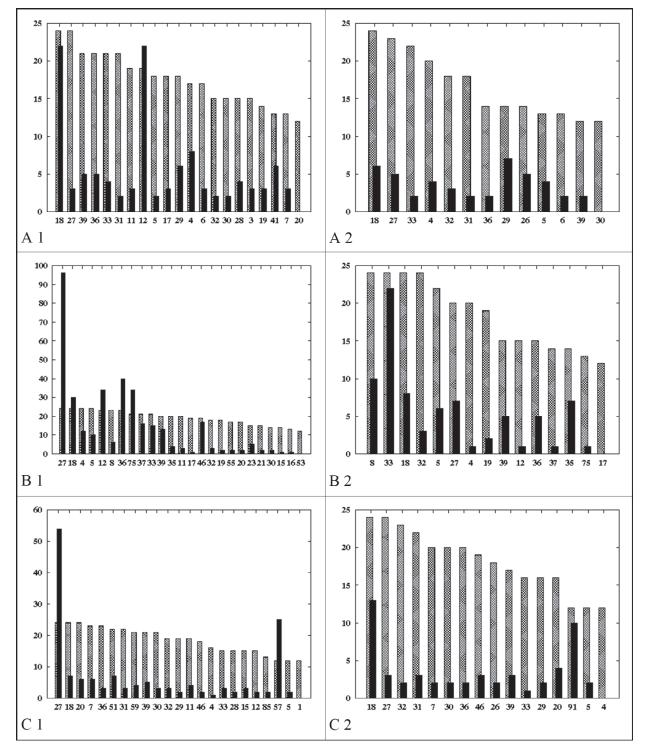
On the one hand, these associations were made by one common species for all ecosystems and soil layers as *Veigaia nemorensis* (ubiquitous species, predatory species) and only for OLF layer, as *Neopodocinum mrciaki* (edaphic detriticole). *Veigaia nemorensis* have a wide ecological plasticity, being capable to adapt on any type of habitats, while *Neopodocinum mrciaki* prefers coniferous forests, what is atypical for edaphically macrochelids (29, 30, 31, 40). On the other hand, in these stable associations were identified species characteristically for each type of ecosystems, as: *Paragamasus similis, Leptogamasus tectegynellus, Pergamasus athiasae, Pachyseius humeralis, Holoparasitus rotulifer* 

## TABLE 2

Mites species identified in soil of investigated forest ecosystem.

No	Species	Forests	No.	Species	Forest
1	Epicrius bureschi		50	Gamasodes spiniger	
2	Epicrius resinae		51	Veigaia transisalae	
3	Holoparasitus excipuliger		52	Zerconopsis remiger	
4	Leptogamasus parvulus		53	Arctoseius cetratus	
5	Paragamasus similis		54	Zercon peltadoides	
6	Paragamasus motasi		55	Pergamasus barbarus	
7	Lysigamasus neoruncatellus		56	Epicriopsis rivus	<b>.</b> •
8	Leptogamasus tectegynellus		57	Leitneria granulata	
9	Lysigamasus truncus		58	Vulgarogamasus remberti	
10	Pergamasus quisquiliarum		59	Veigaia paradoxa	
11	Pergamasus laetus		60	Lasioseius lawrencei	
12	Pergamasus athiasae		61	Prozercon sellnicki	
13	Paragamasus sp.		62	Olopachys vysotskajae	
14	Parasitus furcatus		63	Asca bicornis	
15	Vulgarogamasus kraepelini		64	Pachylaelaps imitans	
16	Vulgarogamasus oudemansi		65	Olopachys scutatus	
17	Vulgarogamasus zschokkei		66	Zercon carpathicus	
18	Veigaia nemorensis		67	Zercon arcuatus	<b>A</b> •
19	Veigaia exigua		68	Leptogamasus variabilis	
20	Veigaia cervus		69	Holoparasitus minimus	
21	Veigaia propinqua		70	Arctoseius brevichelis	
22	Leioseius magnanalis		71	Pachylaelaps magnus	
23	Melichares juradeus		72	Epicrius mollis	
24	Dendrolaelaps rotundus		73	Leptogamasus obesus	
25	Dendrolaelaps foveolatus		74	Zercon triangularis	
26	Rhodacarellus kreuzi		75	Holoparasitus rotulifer	
27	Neopodocinum mrciaki		76	Dendrolaelaps willmanni	
28	Geholaspis longispinosus		77	Proctolaelaps pomorum	
29	Macrocheles decoloratus		78	Gamasolaelaps excisus	•
30	Macrocheles montanus		79	Gamasolaelaps multidentatus	•
31	Pachylaelaps furcifer		80	Protogamasellus sp.	•
32	Pachyseius humeralis		81	Cheroseius sp.	•
33	Hypoaspis aculeifer		82	Eugamasus monticolus	<b>A</b> •
34	Hypospis nolli		83	Veigaia kochi	•
35	Hypoaspis oblonga		84	Macrocheles insignitus	•
36	Eviphis ostrinus		85	Iphidozercon venustulus	<b>A</b> •
37	Zercon fageticola		86	Dendrolaelaps samsinaki	•
38	Zercon romagniolus		87	Porrhostaspis lunulata	
39	Prozercon kochi		88	Rhodacarellus silesiacus	•
40	Arctoseius semiscissus		89	Pachylaelaps latior	<b>A</b> •
41	Arctoseius eremitus		90	Paragamasus vagabundus	•
42	Amblyseius sp.		91	Holoparasitus excisus	
43	Geholaspis mandibularis		92	Pergamasus alpinus	
44	Zercon peltatus		93	Pachylaelaps pectinifer	
45	Zercon pinicola		94	Zercon tatrensis	
46	Prozercon traegardhi		95	Zercon athiasi	
47	Prozercon fimbriatus		96	Hypospis montana	
48	Amblygamasus mirabilis		97	Leptogamasus doinae	
49	Eugamasus magnus				

■ = ecosystem with *Abies alba*;  $\blacktriangle$  = ecosystem with *Picea abies*;  $\bullet$  = ecosystem with *Fagus sylvatica*.

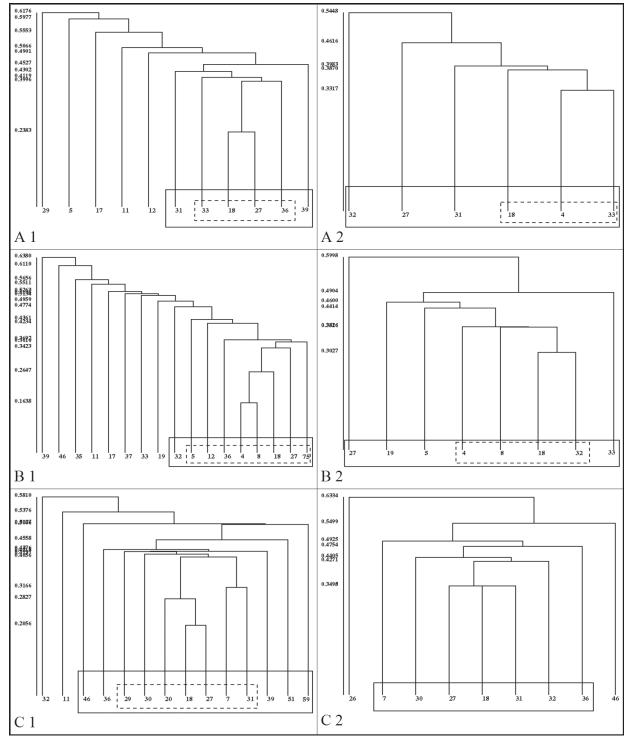


**Figure 2.** Histograms representing the frequencies - dashed bars and abundance - solid bars of the identified species, in soil layers of surveyed forests (A= Abies alba; B= Picea abies; C= Fagus sylvatica; 1=OLF, 2= OH).

for spruce forest and *Lysigamasus neoruncatellus, Veigaia* cervus, Macrocheles decoloratus, Macrocheles montanus, Pachylaelaps furcifer for beech forest.

Analyzing the frequency and abundance of soil mites, we can specify that in the OLF layer, species from stable

associations were grouped in two classes. First group was composed by species which recorded an increased frequency had a decreased abundance. This phenomen could be explained through migration (or spatial dynamics) of these invertebrates. Being very mobile and predator gamasids search all the area for food source.



**Figure 3.** Dendrograms presenting the results of cluster analyses (write used distance and method) of identified species, in soil layers (1=OLF, 2=OH) of surveyed forests (A= Abies alba; B= Picea abies; C= Fagus sylvatica). The solid box marks the stable association and dashed box marks the subset of species pair-wise monotonic associate.

The second group were species that had a decreased frequency and an increased abundance, this mite population possible to be in an equilibrium stage. In this case, favourable environmental conditions could determine stable gamasid populations, represented by a decreased number of species but with an increased number of individuals – abundance, characteristically phenomenon for a mature ecosystem (4, 27). In *Picea abies* forest, due to the high slope ( $45^{0}$ ), which determined existence of a very thin OLF layer, the majority of the gamasid species were

characterized by a lower number of individuals, but with high frequency. This species inhabits mainly soil substratum (needle litter, soil detritus, humus). The steep slope from this forest determined a substratum washing, destroying its favorable habitat, this species possible to migrate from adjacent areas (which explains the high frequencies).

In OH layer, the situation is similar. All species recorded high frequency, but decreased abundance. It is known that the OH layer consists of partially decomposed organic matter, which represents the main trophic reservoir for gamasid and represents a "refuge" for species of small dimension on unfavourable environmental conditions.

The mineralization processes that convert organic matter to the relatively stable substance that is humus, feeds the soil population, thus maintaining high and healthy levels of soil biodiversity *(19, 20, 32)*.

Once that were identified the stable associations, we observed (with exception of OH layer from beech forest) that species grouped in monotonic associations. 66.6% of species from stable associations were included in monotonic association. These species had the same increasing tendency of the two investigated parameters. This tendency could be explained by their aggregation, all these species being predators, very mobile, permanently looking for the food (having a binomial negative distribution) (42, 43).

In OLF layer, 5.47% from identified species in fir forest had the same increasing tendency of abundace and frequence, 10.14% from beech area and 12.5% from

OLF

spruce forest. Species, from OH layer, grouped in monotonic associations are less then in the OLF layer: 4.83% in fir forest, to 7.69% in beech area to 11.66% in spruce forest.

The percent of species which formed these association varied from one ecosystem to other, due to specifically environmental condition (abiotic factors: soil humidity, temperature, pH, organic matter or biotic factors: vegetation, other invertebrates groups as food source) for each area (3, 27, 28).

In OH layer the species which formed the monotonic associations was lower in comparison with OLF layer. We could consider that the OLF layer is a proper habitat for gamasid populations developing. It is known that the vertical distribution of microarthropods varies with changes in soil temperature and moisture content, and it is possible that the identified species from OH layer to migrate from litter. On the other hand, the structure of OLF layer is more porous and aerate in comparison with OH layer, permitting to the predatory mites to "hunt". Mite abundance is influenced by soil pore volume and we stress that soil structure should be considered as an explanatory variable when studying microarthropod communities (1, 33).

In beech forest, on OH layer, there were not identified any monotonic association. The presence of a brown eumesobasic soil, with clayey-sandy fine texture, which is harder to penetrate by the predator gamasids, could be a possible explanation.

OH

. 7		18	27	33	36									0.0	-
	18		.0020	.0007							-	4	18	33	-
	27				0.0008	3					4	0.05	.0005		1
	33	0.64	*		-						18	0.65	0.0	0.0007	
	36		0.64	*							33		0,6		
. 1	1	4	5	8	12	18	27	32	36	75					
	4		.0020	,0007	0.0120	,0000	) -		,0004	,0001					
	5	0.57		.001	-	.0000				-			0	10	00
	8	0.92	0.60			.0000	.0004	- 1	<b>A</b> 1	100		4	8	18	32
	12	0.50					-	-	-	-	4		-		.0000
	18	0.88	0.64	0.87	-		.0026	.0082	.0003	,0000	8	0.00	0.04	.0055	
	27			0.56	-	0.58			.0026	.0009	32	0.58	0.54	2	
	32	-			-	0.52					04	0,11	•		
	36	0.56	-		-	0.66				.0001					
	75	0.61				0.80	0.63	0.69	-						
c		7	18	20	27	29	30	31	51						
	7		-	•				.0003	-						
	18			-	.0001	,0000,	.0004								
	20	-	-		-	-	.0093	-	.0044						
	27	-	0.61	-		-	.0030		-						
	29	-	0.79				-	-	-						
	30		0.65	0.51	0.57				-						
	31	0.66	-		-	-									
	51		-	0.55	-		-	-							

**Figure 4**. Spearman rank correlation test for species belonging to monotonic association. (the values of the rank correlation-r are below the main diagonal and the p value above the main diagonal) (A= Abies alba; B= Picea abies; C= Fagus sylvatica).

The dependence between these groups of species, which formed monotonic associations, was tested using Spearman test. The values of "r" smaller that 0.5 were not be represented, meaning that species recorded a weak interdependence or they were independences.

Analyzing the rank correlation values, these varied from 0.5 to 0.92 (p-value = 0.0001- 0.0120) on species from OLF layer and from 0.54 to 0.77 (p-value = 0.0005- 0.0055), in OH (Figure 4 A, B, C).

The interdependences between species from monotonic associations could be explained taking account of their ecological preferences for the same type of habitat (being edaphic detriticole) and for the same type of food. Species from these families: Parasitidae (Leptogamasus parvulus, Leptogamasus tectegynellus, Vulgarogamasus zschokkei), Veigaidae (Veigaia nemorensis), Macrochelidae (Macrocheles decoloratus, Neopodocinum mrciaki) and Eviphididae (Eviphis ostrinus) are predators, having as trophically preferences some other soil invertebrates as: nematodes, springtails and dipterans larva. Only Hypoaspis aculeifer (Laelapidae family) is polyphagous (5, 16, 22, 45). These trophic similarities could be the main reason of their interdependence. A rich substrate in organic matter determined the presence of abundant soil fauna invertebrates, which represent the food source for predator mites.

## CONCLUSIONS

Using a hierarchical cluster algorithm (based on abundance and frequency), the gamasid mites were classified in stable associations. These stable associations were formed by common species as well as characteristically ones for each type of forest ecosystems. The percents of species grouped in stable associations were closed in both soil layers, but different in every type of ecosystems: lowest in fir forest, medium in spruce area and the highest in beech forest.

Species which had the same increasing tendency of abundance and frequency were constituted in monotonic associations. The interdependence between them had been analysed using Spearman correlation. This tendency of aggregation (frequent species with high number of individuals) could emphasize the gamasid's ecological and trophical preferences similarities. *Veigaia nemorensis* and *Neopodocinum mrciaki* were common species for all investigated ecosystems, in both soil layers, as well as for stable and monotonic associations. This fact demonstrates their affinity for mountainous ecosystems (especially for coniferous forests).

The present study wants to emphasize the each type of ecosystem was characterised by a stable and monotonic associations. 24.74% from all identified species were grouped in these associations. They could represent boindicators for studied ecosystems. **Acknowledgments:** This study was funded by project no. RO1567-IBB01/2014, from the Institute of Biology-Bucharest, of the Romanian Academy. We also thank to anonymous reviewer for their useful suggestions, constructive comments and publishing advices.

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