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*Miloje ŠUNDIĆ and Igor PAJOVIĆ<sup>1</sup>***PHENETIC AFFINITIES AMONG THE *ERYTHRAEUS* AND  
*ABROLOPHUS* SPECIES FROM MONTENEGRO****SUMMARY**

Phenetic relationship among the *Erythraeus* and *Abrolophus* species from Montenegro have been made by cluster analysis using single linkage method and euclidian distance by applying computer programme Minitab 16 and depicted in the dendrograms. Comparison of characters and amalgamation steps are also given in tables. Already known 10 species of two different genera from Montenegro were included in these studied to derive a conclusion about the phylogenetic relationships. Based on the new species for science and new species for studied area conclusions and discussions regarding phylogenetic relationships and phenetic affinity between species of the same genus were made.

**Keywords:** Phenetic affinity, *Erythraeus*, *Abrolophus*, phylogenetic relationships.

**INTRODUCTION**

The Parasitengona (Acari: Prostigmata) constitute one of the most diverse taxa among the Acari with respect to species richness as well as with respect to inhabited biotopes and realised life styles. With the enclosure we wanted to represent the results of ecological and faunal researches of terrestrial Parasitengona in Montenegro, with a point to continue further researches that should clarify at least one part of variety of this extremely rich fauna. Terrestrial Parasitengona colonised a wide range of biotope types from xeric to hygic habitats and from lowland areas to the mountains. A high percentage of ubiquitous opportunists inhabiting particular alluvial floodplains and temporarily aquatic. Exceptional significance of terrestrial Parasitengona is in their regulation of number of other invertebrate groups which is expressed through obligatory parasitism of their larval stadiums. Their role in biological struggle is convincingly supported by a number of apomorphic characters, such as switch from predatory to parasitic life style of the larva, or the transformation of ancestrally active proto- and tritonymphs into calyptostatic instars. From the acarological point of view Montenegro is the most poorly-investigated territory of the Balkan Peninsula. Hitherto, on the area of Montenegro, fauna Parasitengona is a completely unexplored group of organisms. Although Parasitengona actively studied for two centuries, literary almost no data until publication *Erythraeus budapestensis* Fain & Ripka (1998), when for the first

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time described a species from this area. Until 2012. year there were only five types of family Erythraeidae, kohort Parasitengona: *Erythraeus budapestensis* Fain & Ripka, 1998; *Erythraeus (Zaracarus) ueckermanni* Saboori, Nowzari & Bagheri-Zenouz, 2004; *Erythraeus (Erythraeus) ankaraiicus* Saboori, Çobanoglu & Bayram, 2004; *Abrolophus pseudolongicollis kiejstuti* Haitlinger, 2006; *Hauptmannia kotorensis* Haitlinger, 2007; *Leptus (Leptus) josifovi*, Haitlinger, 2012.

## MATERIAL AND METHODS

We studied phenetic relationship among the *Erythraeus* and *Abrolophus* species from Montenegro. We used the computer programme Minitab 16 for cluster analysis and depiction of the dendrograms. The Parasitengona are sampled from May to August 2011 and stored in a small vials with 70% alcohol. In the laboratory, mite are cleared in Nesson's solution and mounted in Faure medium (Krantz and Walter, 2009). The terminology and abbreviations are adapted from Robaux (1974) and Southcott (1986).

## RESULTS AND DISCUSSION

Using a series of morpho-metric characters for genera, *Erythraeus* and *Abrolophus*, phenetic analysis is obtained, based on the general similarity of the two species of the genus. Units were grouped by cluster analysis into groups or classes with the aim of similar units (species) found in the same class (cluster). Clustering was performed based on the values calculated of the all variables for each unit (type) observed separately. As a final result, using this hierarchical method, a dendrogram was obtained. By the dendrogram, clusters (groups) are shown in the shape of plotted tree. First, calculate the distance of all the units (types) of each other, and then the groups are formed by joining techniques (agglomeration). Agglomeration method starts with a matrix of distances between the observation unit and assumes that each unit is itself a group of one. All units are in groups of size one, then the merging into larger groups that are close to each other. The close groups gradually merged until finally they find all the units (types) in each group. The two groups are combined into one if an observation unit of the group closest to a unit in the second group. If we have zero distance units are all individually and have as many groups as there are units of observation. The minimum distance between two units of the 2nd. For all groups (clusters) calculated the average distance (arithmetic center), or when the average distance between two groups of the lowest in comparison to other average distance between groups. Phenetic relationship of species have been made by cluster analysis using single linkage methods and, calculation of Euclidean distances and making dendrograms are displayed by the software program Minitab 16. Also presented and comparison of character and amalgamation steps. On the basis of new species of science and new species for the described area conclusions are drawn and given discussion about

phylogenetic relationships and phenetics affinity between species of the same genus.

### **Subfamilia Erythraeinae Southcott**

#### **Genus *Erythraeus* Latreille**

Erythraeinae are monophyletic groups within Erythraeoidea. Latreille described the genus *Erythraeus* in 1806. year and named *Acarus phalangoides* de Geer as typical species. Previously DeGeer (1778) collected a small red mites and marked them as' *Acarus phalangoides*. Name Erythraeidae family gave Robineau-Desvoidy (1828). Significant leap in the direction of research Erythraeoidea comes with papers A. Rent and A.C. Oudemans, which is not just describe the large number of species have already been created and modern systematics Acarina. The most important name of modern research Erythraeoidea certainly R. V. Southcott.

Already in his early works from the 1941. year he published several important revisions and gave the basis for modern systematics of this superfamily. Southcott (1995) divided the genus *Erythraeus* into two subgenus (*Erythraeus* and *Zaracarus*). This genus has a cosmopolitan distribution but the larvae are known mostly from Europe and Asia. From Montenegro is described 5 species of which 3 species belong to the subgenus *Erythraeus* (*Erythraeus*) and 2 species of subgenus *Erythraeus* (*Zaracarus*). Species were collected from different plants in different habitat types.

Dendrogram (fig. 1.) five species *Erythraeus* made on the basis of comparison of 38 characters (table 1.) shows that the *E. preciosus* and *E. Ueckermanni* have similarity level of 94.9% and constitute the first cluster A of the dendrogram. These two species belong to the subgenus *Zaracarus* in the group with a very short AM setae, and have been collected in habitats where there is similar environmental conditions. This shows that the similarity between these species could be caused by sharing of common genetic characters and similar ecological zones.

Species *E. ancaraikus* and *E. Southcott* make second cluster B at the level of 91.1% affinity. These species also share similar hosts and very close ecological habitats. A cluster is associated with cluster B and cluster C formed at the level of 84.4% affinity. Species *E. smolyanensis* joins the cluster C and D formed the last and largest cluster at the level of affinity 29.1% (table 2.). *E. smolyanensis* inhabit different ecological niches of other species and are thus distinctive phenetics relations of affinity (29.1%) compared to other species, probably supported by combined genetic characters.

Dendrogram shows different levels of affinities among species of the genus suggesting plesiomorphic properties that are separate from the ancestral line. Based on the data collected on the species becomes clear that the relationship between species are not necessarily expressed through ecological

affinity than mutual affinities can be supported by sharing of common genetic characters within the genus.

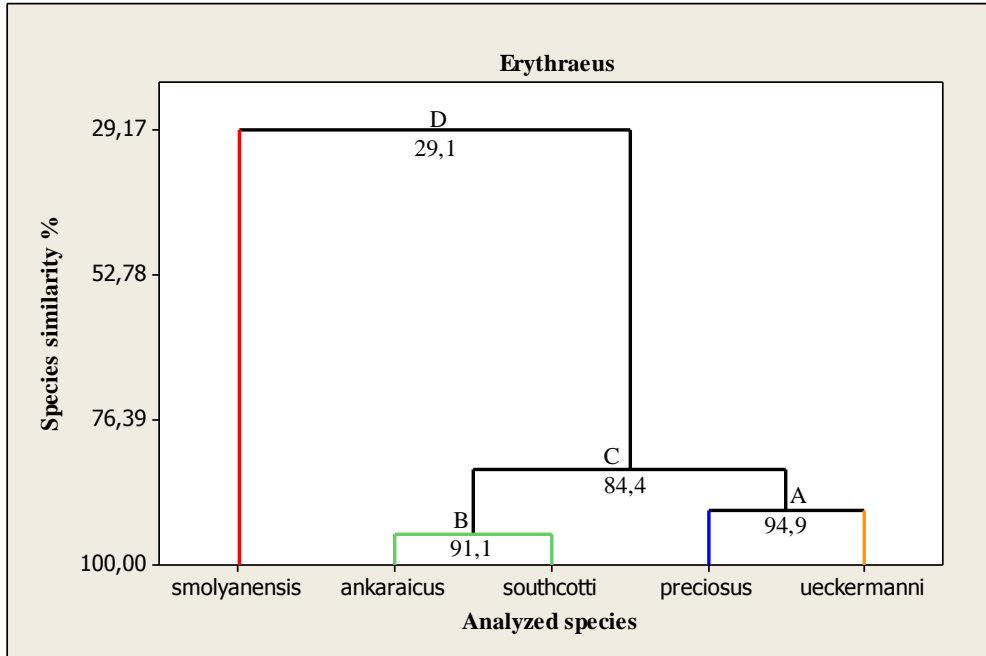


Figure1: Dendrogram of 5 *Erythraeus* Latreille species from Montenegro

Table 2: Amalgamation Steps of cluster in genus *Erythraeus* Latreille

Steps	Number of clusters	Similarity %	Distance Level	Clusters Jointed		New cluster	Number of Taxa in new Cluster
1	4	94,9251	201,62	2	3	A	2
2	3	91,0883	354,06	4	5	B	2
3	2	84,4577	617,48	2	4	C	4
4	1	29,1735	2813,88	1	2	D	5

### Subfamilia Balaustinae Southcott Genus *Abrolophus* Berlese

*Abrolophus* is a large genus within erythraeid mites but its taxonomy is still unstable and variable. The genus *Abrolophus*, *Hauptmannia*, *Marantelophus* and *Grandjeanella* is very similar and their unregulated taxonomy will become clear only through relationships postlarvae and larvae stage. Under such a

procedure involves collecting and rearing adult and their rearing in the laboratory in order to establish the basis of the larvae real correlation between the genus.

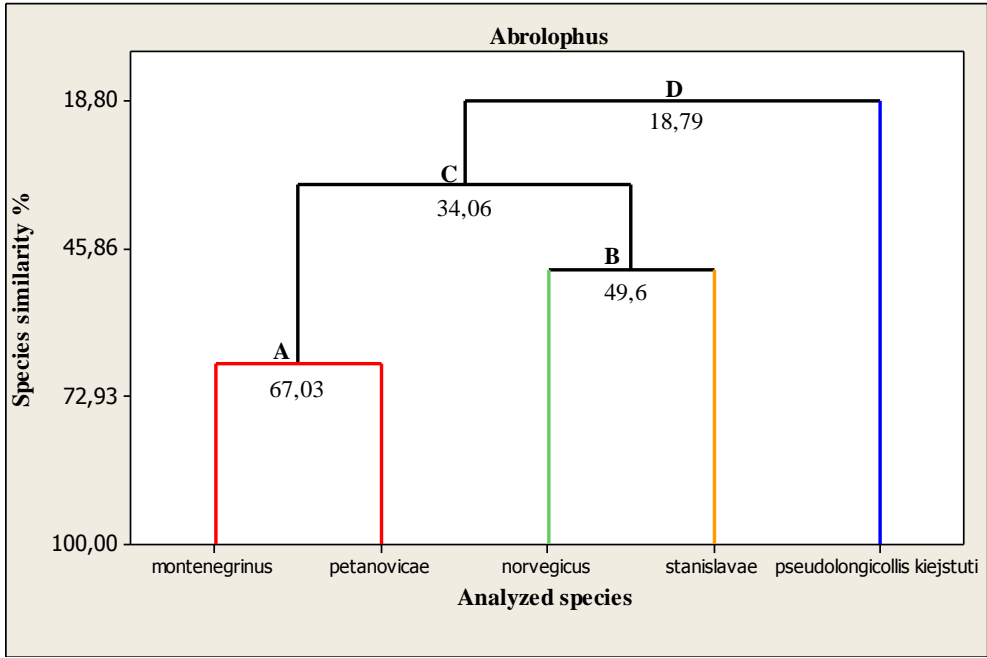


Figure 2: Dendrogram of 5 *Abrolophus* Berlese species from Montenegro

Tabele 4: Amalgamation Steps of cluster in genus *Abrolophus* Berlese

Steps	Number of clusters	Similarity %	Distance Level	Clusters Jointed		New cluster	Number of Taxa in new Cluster
1	4	67,0310	3,87298	1	2	A	2
2	3	49,6390	5,91608	3	5	B	2
3	2	34,0620	7,74597	1	3	C	4
4	1	18,7953	9,53939	1	4	D	5

Genus *Abrolophus* established Berlese 1891 and identified as *Abrolophus quisquiliarum* (Hermann) Berlese, 1891. This species was described on the level of adult and larvae. Until in 1996. year when Zhang and Goldarazena publicized *Abrolophus neobrevicollis* this genus has been described only at the adult level. Larvae *Abrolophus* species can be divided according to the presence or absence of comblike setae on the palptarsus. The following *Abrolophus* species have comblike setae on the palptarsus: A. *quisquiliarus* (Hermann, 1804); A.

*longicollis* Oudemans, 1910; *A. aitapensis* Southcott, 1948; *A. mortenseni* Southcott, 1994; *A. humberti* Haitlinger, 1996; *A. khanjanii* Haitlinger & Saboori, 1996; *A. tonsor* Southcott, 1996; *A. welbourni* Yao *et al.*, 2000; *A. benoni* Haitlinger, 2002; *A. bohmani* Haitlinger, 2003; *A. iraninejadi* Saboori & Hajiqanbar, 2005; *A. penelopae* Haitlinger, 2005; *A. basumtwiensis* Haitlinger, 2006; *A. unimiri* Haitlinger, 2006; *A. mirabelae* Haitlinger, 2007; *A. marinensis* Haitlinger, 2007; *A. crimensis* Haitlinger, 2008 and *A. nymindegabicus* Haitlinger, 2008.

Until we have described new species *Abrolophus montenegrinus* and *Abrolophus petanovicae* (Saboori, Šundić & Pešić sp. nov.), this genus, with comblike setae was not known in Montenegro. Until now, from Montenegro have been described 5 species of which 3 are new to science, and one of them is synonymized. Species were collected from different plants in different habitat types.

Dendrogram (fig. 2.) of five species *Abrolophus* made on the basis of comparison of 39 characters (table 3.) that were collected from ecologically quite similar habitats, with only deviates species *Abrolophus pseudolongicollis kiejstuti*, Haitlinger. Species *Abrolophus petanovicae* Saboori, Šundić & Pešić sp. nov., and *Abrolophus montenegrinus* Saboori, Šundić & Pešić sp. nov., form first cluster A at the level 67,03% similarity. These two species belong to the specified group of *Abrolophus* genus with comblike setae. If we look at all the other characters of the two species share, we can conclude that the similarity between these species to the greatest extent caused by common genetic characters, because environmental conditions in their natural habitats quite congruous. *Abrolophus norvegicus* (syn. *Hauptmannia striata*), Thor i *Abrolophus stanislavae*, Haitlinger were collected in the area of Lake Skadar, where there is a sub-Mediterranean climate and amphibian habitats where dominant character: flooded meadows and extremely humid belts ecosystems along the coast.

They are form cluster B with the degree of similarity of 46.9%. The four species form a common cluster C with the degree of similarity of 34.06%. *A. pseudolongicollis kiejstuti*, Haitlinger joins the cluster C with the degree of similarity of 18.79% and forms the last and largest cluster D (table 4.). The dominant morphological characters in species *A. stanislavae*, *A. pseudolongicollis kiejstuti* and *Abrolophus norvegicus* (syn. *Hauptmannia striata*) from the last three clusters is the absence of comblike setae on palptarsus what is on the dendrogram (fig. 2.) clearly defined. *A. stanislavae* and *A. norvegicus* form a medial cluster, what was expected because that cluster shared genetic and environmental factors with other species *A. pseudolongicollis kiejstuti* is from area where there is a somewhat different ecological conditions: moderate continental climate and less hygic habitats.

Considering the combination of genetic and environmental factors of this species we get a clear indication of this low degree of affinity with other species of this genus. Affinities of species in cluster A is mostly painted common genetic

traits and that's why we have a clear distinction from other clusters. Low levels of affinities among species of the genus *Abrolophus* indicates that plesiomorphic characters that are separated from ancestral lines.

Table 1: 38 characters in 5 species of the genus *Erythraeus* Latreille

I	(1, bases of ASE with strong cuticular structures; 2, ASE at its bases without strong cuticular structure;
II	(1, AL= DS ; 2, AL slightly longer than DS; 3, AL more the twice longer than DS; 4, AL more the three times longer then DS);
III	(1, Hy>S; 2, Hy<S);
IV	(1, coxala 1a less than twice the coxala 1b; 2, coxala 1a thrice than coxala 1b; 3, coxala 1a > 1b);
V	(1, AA>11; 2, AA< 11);
VI	comparison of lengths AM and S (1, S=AM; 2, S more than twice than AM; 3, S less than twice than AM; 4, S more than thrice coxala AM);
VII	(1, GL > 160); 2, GL = 120-160; 3, GL < 120).
VIII	dorsal body setae (fD) (1, 36-42; 2, 42- 64);
IX	ventral body setae (fV) (1, 20-39; 2, 12-19);
X	(1, ISD>50; 2, ISD< 50);
XI	setae on palpfemur;
XII	setae on palp genu;
XIII	setae on palptibia;
XIV	setae on palptarsus;
XV	comparison of lengths AL i PL (1, AL=PL; 2, AL slightly longer than PL; 3, AL 1.5 times longer than PL; 4, AL more than twice than PL);
XVI	setae behind coxa III on ventral body side (1, 14; 2, 12; 3, 10);
XVII	setae on basifemur I;
XXVIII	setae on telofemur I;
XIX	setae on genu I;
XX	setae on tibia I;
XXI	setae on tarsus I, (1,28; 2, 30 ; 3, 21 - 26);
XXII	setae on basifemur II;
XXIII	setae on telofemur II;
XXIV	setae on genu II;
XXV	setae on tibia II;
XXVI	setae on tarsus II, (1, 23-25; 2, 25-28, 3, 20-22);
XXVII	setae on basifemur III;
XXVIII	setae on telofemur III;

XIX	setae on genu III;
XXX	setae on tibia III;
XXXI	setae on tarsus III, (1, manje od 22; 2, više od 22);
XXXII	(1, AL enlarged near bases; 2, AL not enlarged near bases);
XXXIII	(1, W > 130; 2, W = 120-130; 3, W < 120);
XXXIV	(1, L > 80; 2, L = 50-80; 3, L < 50);
XXXV	length leg I in $\mu\text{m}$ ;
XXXVI	length leg II in $\mu\text{m}$ ;
XXXXVII	length leg III in $\mu\text{m}$ ;
XXXVIII	total length of the body in $\mu\text{m}$ .

Tabele 3: 39 characters in 5 species of the genus *Abrolophus* Berlese

I	(1, SD > 70; 2, SD < 60; 3 SD = 60-70);
II	palptarsus (1 with comb-like seta, 2 without comb-like seta);
III	gnathosoma ventral with (1, 4 pair of seta; 2, 3 pair of seta);
IV	(1, IP > 1150; 2, IP < 1150);
V	comparison of lengths of AL and PL (1, AL = PL; 2, AL < PL; 3, AL > PL);
VI	palptibia claw (1, with subterminal tooth; 2, without subterminal tooth);
VII	(1, AW = PW; 2, AW < PW; 3 AW > PW);
VIII	Palpal tibial claw (1, bifid; 2, entirely);
IX	(1, ISD = AL; 2, ISD < AL; 3, ISD > AL);
X	(1, AW twice than AP; 2, AW less than twice the AP; 3, AW = AP)
XI	setae on palpfemur;
XII	setae on palp genu;
XIII	setae on palptibia;
XIV	setae on palptarsus excluding solenidion and eupathidion;
XV	dorsal body setae (fD) (1, 42-46; 2, 46-64);
XVI	ventral body setae (fV) (1, 32-39; 2, 14-32);
XVII	setae on basifemur I;
XVIII	setae on telofemur I;
XIX	setae on genu I;
XX	setae on tibia I;
XXI	setae on tarsus I;
XXII	setae on basifemur II;
XXIII	setae on telofemur II;



XXIV	setae on genu II;
XXV	setae on tibia II;
XXVI	setae on tarsus II;
XXVII	setae on basifemur III;
XXVIII	setae on telofemur III;
XXIX	setae on genu III;
XXX	setae on tibia III;
XXXI	setae on tarsus III;
XXXII	(1, Supracoxal seta present on coxa I ; 2, Supracoxal seta absent on coxa I);
XXXIII	(1, eyes > 20; 2, eyes < 20);
XXXIV	(1, scutum as long as wide; 2, Scutum longer than wide);
XXXV	(1, Ti I > 70; 2, Ti I < 70);
XXXVI	(1, leg I longer than other legs; 2, leg I less than other legs);
XXXVII	(1, 1a > 40; 2, 1a =30-40; 3, 1a < 30);
XXXVIII	(1, 1b > 50; 2, 1b =40-50; 3, 1b < 40);
XXXIX	(1, GL > 120; 2, GL = 110-120; 3, GL < 110).

### CONCLUSIONS

Using a computer programme Minitab 16 we are make phenetic relationship among the *Erythraeus* and *Abrolophus* species from Montenegro throw comparison of characters and amalgamation steps which is depicted in the dendrograms. Similarity between species *E. preciosus* and *E. Ueckermanni* could be caused by sharing of common genetic characters and similar ecological zones. Phenetics relations of low level affinity for other species is probably supported by combined genetic characters. For the *Erythraeus* species dendrogram shows different levels of affinities among species of the genus suggesting plesiomorphic properties that are separate from the ancestral line. Species *Abrolophus petanovicæ* Saboori, Šundić & Pešić sp. nov., and *Abrolophus montenegrinus* Saboori, Šundić & Pešić sp. nov., have high level similarity, because this species belong to the specified group of *Abrolophus* genus with comblike setae and share common genetic characters. Others *Abrolophus* species shared poorly genetic and environmental factors and they have low degree of affinity among this genus. Species of genus *Erythraeus* and *Abrolophus* show quiet low level of affinity among them.

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**FENETIČKE SRODNOSTI IZMEĐU VRSTA *ERYTHRAEUS* I  
*ABROLOPHUS* IZ CRNE GORE**

**SAŽETAK**

Fenetički odnosi između *Erythraeus* i *Abrolophus* vrsta iz Crne Gore su urađeni klaster analizom koristeći single linkage metod i Euklidova rastojanja pomoću kompjuterskog programa Minitab 16 i prikazani su u dendogramima. Komparacija karaktera i aglomerativni koraci su dati u tabelama. Deset poznatih vrsta iz dva roda za područje Crne Gore su uključeni u ovu studiju da bi se izveli zaključci o filogenetskim odnosima. Na bazi novih vrsta za nauku i novih vrsta za opisivano područje izvedeni su i zaključci i data diskusija u vezi filogenetskih odnosa i fenetičkih afiniteta među vrstama istog roda.

**Ključne riječi:** Fenetičke srodnosti, *Erythraeus*, *Abrolophus*, filogenetske relacije.