The Shell Characteristics of Land Snail *Eobania* Vermiculata (Müller, 1774) From Croatia

Biljana Rađa[1], Tonći Rađa[2], Mate Šantić[1]

[1] Faculty of Science, University of Split, Teslina 12/III, 21000 Split
phone: ++38521385133; fax. ++38521384086
radja@pmfst.hr, msantic@pmfst.hr

[2] Speleological Society „Spiljar“, Varaždinska 54, 21000 Split
tonci.radja@gmail.com

Abstract: The land snail genus *Eobania* P. Hesse, 1913 in Croatia consists of four taxons. The mean shell largest diameters (D) of the 13 samples ranged from 19,99 mm to 31,78 mm and the mean shell height (H) from 17,31 mm to 25,66 mm. The shell morphology confirms that all islands populations are smaller with examples of nanism on outside islands Palagruža and Sušac. Molecular analysis (16S rDNA and COI sequence data) showed no significant differences between the South – Adriatic population from the mainland and islands, thus providing no molecular–genetic evidences for classification of a single *E. vermiculata* species into separate subspecies. The observed differences could, probably, be attributed to ecological influences.The mean size of the largest population was approximately 1,5x larger than the mean size of the smallest population. The low variance between the size variation within populations and variation in shell shape were not expected.

Key words: Shell of land snaill, Eobania Vermiculata, Croatian Study

Introduction

Eobania vermiculata (Müller, 1774) or chocolate banded snail is a wide distributed land snail. The nearest countries to Croatia in which *Eobania vermiculata* is registered are Italy, Bulgaria (Dedov, 1998), Greece (Welter-Schultes & Willians, 1999) and Turkey (Örstan et al., 2005). By a review of the available literature we determined that *E. vermiculata* has been recorded in Croatia only in general terms in Dalmatia (Jaeckel et al., 1957), on The Island Dugi otok (Štamol 2004; Štamol & Kletečki, 2005) and as subspecies *Eobania vermiculata pelagosana* (Stossich, 1877) from the Islands of Palagruža and Sušac (Berberović 1963, 1967). Molecular phylogeny and biogeography for the species *E. vermiculata* from Croatia are unknown.
The previous studies on a terrestrial gastropods *Helix aspersa* and *Eobania vermiculata* (Sacchi, 1957; Berberović 1963, 1964, 1967) in Croatia were based only on morphometric characteristics of the shell. The results showed that all shells from the adriatic islands are smaller than the average values for land specimens. Those „nanism“ is especially expressed in populations of the Island of Palagruža and Sušac (open sea islands close to the italian seaside) so Berberović (1964) described new subspecies *Helix aspersa pelagosana* and Stossich (1877) *Eobania vermiculata pelagosana*.The basic survey on *Eobania vermiculata* shell morphometry made Berberović (1963) on land and island populations from the Middle Dalmatia (Croatia). He distinguished three separated groups: 1. group Palagruža and Sušac; 2. group „Biševo“ and 3. group „Lastovo“. The shell characteristics of group Palagruža and Sušac showed the nanism caused by geological origin of those islands, vegetation and abundance. The group Biševo is more similar to those from the Island of Gali (Gulf of Salerno). He also divided all examined populations in morphological groups in relation to populations from Palagruža and Sušac to „continental type“ (H/D ratio lower than Palagruža and Sušac; middle adriatic islands except island of Vis and lastovo archipelag). Lastovo archipelag and the Island of Vis are „lastovo type“ (H/D ratio same or bigger than Palagruža and Sušac) and „biševo type“ consists of populations from Island of Biševo, Korčula and Hvar (H/D ratio much lower than Palagruža and Sušac). The contact zone between types are Island of Vis with neighbouring islands. The land populations divided on populations north from the river Cetina, between the river Cetina and the river Neretva and south from the river Neretva. In this paper we examined the same shell parameters as Berberović (1964) and represent the morphometric characteristics of 15 different populations of *E. vermiculata*. The aim scope was to determine if any changes in shell size took place in a period from Berberović (1964) till today. Molecular analysis (16S rDNA and COI (Puizina et al., unpublished sequences GenBank Accession Numbers JF277380– JF277396 and JF802030 - JF802030) showed that there is no molecular–genetic evidences for classification of a single *E. vermiculata* species into separate subspecies.

Material and Methods

The sampling design we used for morphological and molecular analysis followed the pattern of Berberović (1963) (samples for Middle Adriatic) as well as from North and South Adriatic, thirteen sites in total (Figure 1).

Figure 1. Sampling sites along The Adriatic coast.

For shell measurement we collected 20 specimens per site and a total of 300 specimens with a reflected lip because this indicates cessation of growth and maturity of the snail. The measurements were made on the field.
The shell features we measured were shell high (H), shell width (D) and aperture high (h). After that we compared the results as relative shell high (H/D) (ratio between high and width in percents) and ratio between aperture high (h) and shell high (H) in percents. The intrapopulation variation was calculated with the coefficient of variation (CV) of all the morphometric characters of the examined populations. Geographical variation and qualitative data of the shell were subjected separately to principal component analysis (PCA). The UPGMA analysis was used to calculate similarities between the 13 populations based on morphometric data. All statistic calculations were made by Statistica 8.0 and XlstatPro softwares.

Results

The mean shell largest diameters (D) of the 13 samples ranged from 19,99 mm (sample from island of Sušac) to 31,78 mm (Šibenik, seaside) and the mean shell height (H) from 17,31 mm (Sušac) to 25,66 mm (Island of Šolta). The mean size of the largest population was approximately 1,5 x larger than the mean size of the smallest population. Coefficient of variation (CV%) values of *H*, *D* and *h* ranged from 11,02% to 11,72% and shows the low variance for the size variation within populations. The H/D ratio ranged from 0,78 (sample from Island of Palagruža) to 0,86 (sample from Island of Sušac). The low variance between the size variation within populations and variation in shell shape were not expected. The PCA analysis based on morphometric data of the shell characteristics of *Eobania vermiculata* confirmed that the populations from Islands of Palagruža and Sušac are clearly separated from the other populations (Figure 2).

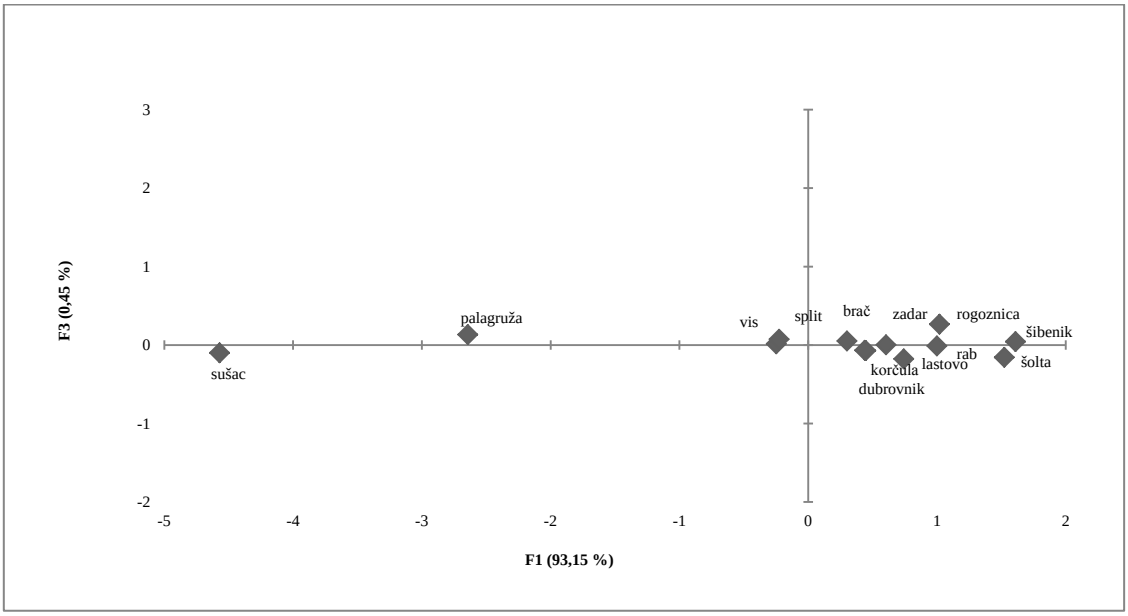

Figure 2. The diagram of PCA analysis.

The variables which mainly contributed to the first discriminant function (squared canonical correlation 0,98%) were shell hight (H) and for the second discriminant function (squared canonical correlation 0,94%) shell width (D). The populations from the island of Palagruža and Sušac constituted homogenous group (76,71%), populations from the island of Šolta and Šibenik (coast) constituted more or less homogenous group at the level of 13,57% and those from other sites were heterogenous (0-3%).The first factor (H, 93,15) and the second one (h, 0,45%) separated populations in seven groups according to their geographic proximity. The first group is characterized by two populations: Palagruža i Sušac; second group by population from the Island of Vis; third by Split (seaside); forth by the Island of Šolta and Šibenik (seaside); fifth by the Island of Rab and Rogoznica (seaside); sixth by the Island of Korčula; seventh is divided in three subgroups: Zadar (seaside), Island of Lastovo and Island of Brač and Dubrovnik (seaside) (Figure 2). UPGMA cluster analysis based on Euclidian distance (Figure 3) was consistent with PCA results.

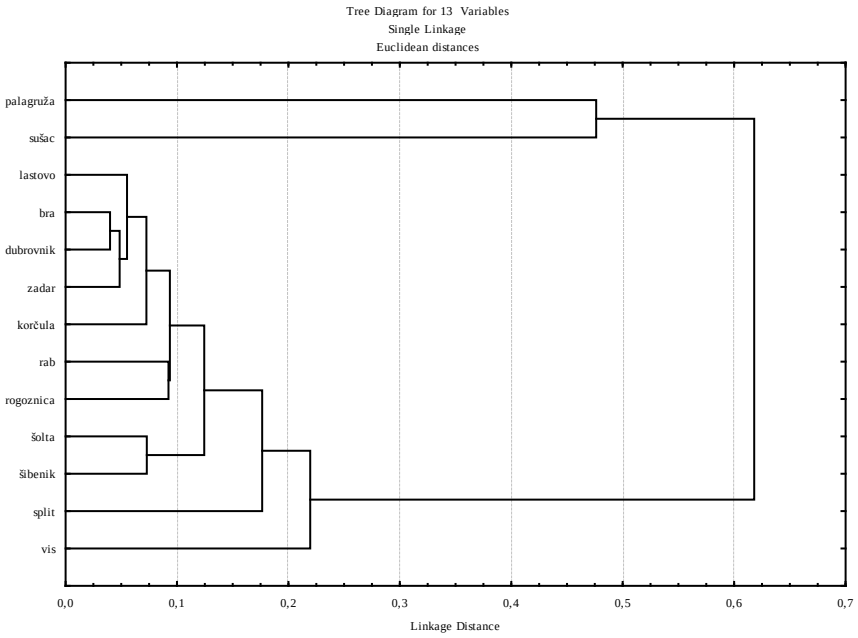

Figure 3. UPGMA cluster analysis.

Discussion

This study of morphometric characteristics of the shells revealed that populations exposed to antropogenic activity showed multivariate intrapopulation variation. The populations were grouped according to their geographical origin and cluster analysis support that thesis. Populations on open sea islands Palagruža (68 miles S from Split) and Sušac (25 km W from the Island of Lastovo) keep their „nanism“ as Berberović (1964) stated in his research. The main reason for “nanism” on those islands is geographic isolation (took place approximately 10 000 years ago) and extreme ecological conditions. Namely, according to Köppen the Island Palagruža is included in the Csa type of climate - Mediterranean climate with hot summers (only 309,7 mm of rain; high relative humidity of air of 74%-77% with low precipitation of 304 mm) (Trošić et al., 2003). Such climate characteristics are directly reflected on plant communities in which xerophyts are dominant floral component (Pavletić, 1978) and they are not suitable for snail ingestion because of essential oils and silicates in plant tissue. Compared to the starting morphological variation, our shells showed the greater similarity to the sizes of the islands Tremiti (Sacchi, 1957). Lazaridou et al. (1994) stated that the largest shell diameter and the aperture area are negatively related to the mean minimum annual monthly temperatures and Cook and O'Donald (1971) confirms that smaller snails survived better in unshaded and warmer conditions. Ecological conditions in the outer islands (limited amount of food, temperature and humidity) could be the reason of increased body size because those islands are known for extremely unfavourable environmental conditions. The mean annual air temperature is 16,7˚C. Total of 85,3 days per year minimum air temperature is 20˚C or higher. Annual precipitation is 289,5 mm (Trošić et al., 2003). So, this climate type known as mediterranean arid clymate type with long dry period (more than three months per year) with extremely high temperature cause longer hybernation in poikilotherms such as terrestrial snails than in mainland. Also, the resource availability is reduced. The *Eobania* snails are polyfagous herbivores and the amount of suitable plants (herbs and grasses) is limited (Trinajstić, 1973). The Island of Palagruža and Sušac are uninhabited offshore islands app. 68 miles away from the mainland and continuity of such contributions appears perfectly stable nanism. Taking into consideration the formation of these islands it is quite certain that the species *E. vermiculata* is native species here. The populations of inner islands showed grater morphological polymorphism in a way that some individuals reach the standard shell size, almost the same as continental populations. The geographic isolation of those islands is not so long as in outer ones, and the anthropogenic influence is obvious. The environmental characteristics are quite different from those on islands Palagruža i Sušac because the most of the land is agricultural; more water and higher humidity as well as unlimited quantity of food are the main reason why those individuals are bigger than on the islands of Palagruža and Sušac. The populations from the islands Lastovo and Korčula according Berberović (1963) were grouped together with continental populations south to the river Neretva. Our survey confirms such shell sizes as Berberović (1963) but in that group are also the populations from the island of Brač. The continental populations from that group are populations from Zadar and Dubrovnik. The continental populations north from the river Cetina are similar with populations from the island of Rab and Šolta which is in accordance with Bereberović (1963). The continental populations from the area between the rivers Cetina and Neretva are in the same group with populations from Split and island of Vis. The populations from the Island of Vis is according Berberović (1963) defined as the mixed zone of different types of shell sizes. Our survey confirm this statepoint. The reduction of shell size towards continent-inner islands-outer islands our survey did not confirmed. We found enlargment of shell sizes except on outer islands probably caused by antropogenic influence and changes in environmental conditions. Molecular analysis of two mitochondrial genes (Puizina et al. GenBank unpublished sequences) didn't support subspecies status of *E. vermiculata pelagosana* on molecular level so those samples could be define as ecotypes or forms.

References

Berberović, LJ. (1963). Mikroevolucija vrste *Eobania vermiculata* (Müll.) na srednjejadranskom primorju i ostrvima. *Godišnjak Biol. Instituta u Sarajevu*, 16 (1/2): 3- 76.

Berberović, LJ. (1964). Nova podvrsta kopnenih puţeva sa ostrva Palagruža u Jadranskom moru. *Godišnjak Biol. Instituta u Sarajevu*, 17: 3-12.

Bole, J. (1969). Subspecifična diferenciacija polžev na otokih in čereh ob zahodni obali Istre. Dissertationes classis IV. *Academia scientiarum et artium Slovenica*, 12 (3): 139-151.

Cook, L., & O'Donald, P. (1971). Shell size and natural selection in *Cepea nemoralis*. In: E.R. Creed, eds., *Ecological Genetics and Evolution*. Blackwell Scientific Publications, Oxford and Edinburgh. Pp. 93-108.

Cowie, R. H., Dillon, R. T., Robinson, D. G. & Smith, J. W.(2009). "Alien non-marine snails and slugs of priority quarantine importance in the United States: A preliminary risk assessment". *American Malacological Bulletin*, 27: 113-132.

Dedov, I.K. (1998). Annotated check-list of the Bulgarian terrestrial snails (Mollusca, Gastropoda). *Linzer Biol. Beitr*. 30/2: 745-765.

Filin, Z., & Ziv, Y. (2004). New theory of insular evolution: unifying the loss of dispersability and body-mass change. *Evolutionary Ecology Recearch*, 6: 115-124.

Jaeckel, S.G., Klemm,W., & Meise, W. (1957). Die land und Süsswasser mollusken der nördlichen Balkanhalbinsel. *Abh. Ber. Mus. Tierk.* Dresden., 23 (2): 141–205.

La Graeca, M., & Sacchi, C.F. (1957). Problemi del popolamento animale nelle piccole isole mediterranee. *Annuario dell' Ist. e Museo di zool. dell' Università di Napoli*, 9,3.

Lazaridou-Dimitriadou, M., Karakousis, Y., & Staikou, A. (1994). Geographical variation in shell morphology and isoenzymes of *Helix aspersa* Müller, 1774 (Gastropoda, Pulmonata), the edible land snail, from Greece and Cyprus. *Heredity*, 72: 23-35.

Örstan, A., Pearce, T.A., & Welter-Schultes, F. (2005). Land snail diversity in a threatened limestone district near Istanbul, Turkey. *Animal Biodiversity and Conservation*, 28(2): 181-188.

Palkovacs, E.P. (2003). Explaining adaptive shifts in body size on islands: a life history approach. *Oikos* **103**: 37-44.

Pavletić, Z. 1978. Pregled i analiza flore Palagruških otoka. *Biosistematika*, 4(1): 39-47.

Sacchi, C.F. (1957). Relazioni tra superficie insulare e mole corporea in Eobania vermiculata (Müll.) dell' Arcipelago toscano. *Bollettino di zoologia*, 24.

Stossich, M. (1877). Sulla geologia e zoologia dell'isola di Pelagosa. *Boll. Soc. Adriat. Sci.Nat.Trieste*, 3(2): 184–192.

Štamol, V. (2004). Terrestrial snails (Mollusca: Gastropoda terrestria) of the Telaščica Nature Park (Dugi otok, Croatia). *Natura Croatica*, 13 (2): 95–113.

Štamol, V., & Kletečki, E. (2005). Terrestrial snails (Mollusca: Gastropoda terrestria) of the Telaščica Nature Park (Dugi otok, Croatia). *Natura Croatica*, 14 (1): 1–28.

Štamol, V. (2010). A list of land snails (Mollusca: Gastropoda) of Croatia, with recommendations for their croatian names. *Natura Croaica*, 19 (1): 1–76.

Šegota, T. (1968). Morska razina u holocenu i mlađem Würmu. *Geografski glasnik*, 30: 15-39.

Šegota, T., & Filipčić. A. (1991). Arheološki i geološki pokazatelji holocenskog položaja razine mora na istočnoj obali Jadranskog mora. Rad HAZU, 458 (25): 149-172.

Trinajstić, I. (1973). O zoni sveze *Oleo-Ceratonion* u istočnojadranskom dijelu Balkanskog poluotoka. *Ekologija*, 8(2): 283-294.

Trošić, Ž., Jašić, D., & Marinković, V. (2003). Climatic features of Palagruţa island, Croatia. *Geoadria*, 1: 39-46.

Welter-Schultes, F.W., & Williams, M.R. (1999). History, island area and habitat availability determine land snail species richness of Aegean islands. *Journal of Biogeography*, 26: 239-249.