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Variation of Radula Characters of Thiaridae (*Molluscs: Gastropods*) in Various Types of Habitat in Papua

Suriani Br Surbakti¹, Yohanis Ngili²

¹Department of Biology, University of Cenderawasih, Jayapura, Indonesia ²Department of Chemistry, Biochemistry Research Group, University of Cenderawasih, Jayapura, Indonesia

ABSTRACT

Variation of 14 radula characters of 27 Thiaridae species from 5 habitat types in Papua has been investigated. The Thiaridae specimens were collected from Batanta Island, Sorong, Manokwari, Bintuni Gulf, Biak Island, Supiori Island and Jayapura. Scanning Electron Microscope (SEM) was used to observe microscopic Thiarids characters and cluster analysis was employed to group thiarids into 4 groups based on habitats and radula characters. Variations in habitat types (stony reef, sandy rubble, muddy sand, littered mud) had significantly affected the number of cusp (F=24.306, P=0.000), number of lateral tooth (F=9.598, P=0.000), number of tooth line (F=20.320, P=0.000) and length of tooth (F=67.038, P=0.000), and of the number of marginal tooth (F=23.612, P=0.264). Moreover, variations in habitats had also affected the types of radulae. For instance, stony reef habitats were dominated by Thiarids' with rectangular radula types, muddy sand by half-triangular types, sandy rubble by rounded types, and litter-mud by triangular types. Therefore, radula could not be used to identify Thiarids to species level due to its high character variation.

Keywords: Characters, Radula, Thiazidae, Variation, Papua-Indonesia

INTRODUCTION

Thiaridae have high diversity, reported there are four genus distributed there is Brotia, Sulcospira, Thiara, and Melanoides and several of them distributed throughout Papua. Thiaridae found in some types of waters, such as rivers, lakes, ponds, irrigation canals, rice paddies and stagnant water [1,2]. Thiaridae deployment in the region is associated with a high ability to adapt to the various conditions of abiotic and biotic. Conditions of abiotic consists of temperature, brightness, color, pH, dissolved oxygen, ammonia, nitrite, nitrate and phosphorus, depth, pattern of water flow, substrate type and season, as well as the presence of the organism is also influenced by biotic factors such as plants, detritus, algae and moss, as well as predators [3,4].

Radula characters could be used in the identification of the family. Saransin and Saransin and Bieler [5], Rintelen et al. [6] stated that the radula can be used to distinguish genus and species. Radula character is one character that has an important role in the identification [7,8]. Radula is a series of gears that are part of the digestive system and is present in the mouth are wrapped in the buccal mass. Chitramvong [9] explains that the radula consists of three parts: radula the front, middle and rear. Radula is used to scratch the substrate, cutting food and put food into osofagus.

Radula has 6 types, namely: the type rhipidoglossate, decoglossate, tanioglossate, rachiglossate, toxoglossate, stenoglossa and combined with toxoglossate rachiglossate type [10]. Genus Bellamyâ have taenioglossaate radula types. Likewise with Pachychilidae. Generally, gastropods and Family Thiaridae have radula type taenioglossaate. Thiaridae tooth structure rhipidoglossate type structure is marginal teeth, lateral teeth, median tooth can be described (1: 1: R: 1: 1). Generally distinguishing Family is the number of protrusions on lateral teeth, the number of rows of teeth, median tooth protrusion amount, the number of rows of teeth on the radula pocket, the number and shape of cusps and form ectones.

Some previous research reports, explaining the radula is a character that can be used to analyze the habitat types associated with the substrate. This is related to the type of food contained in the substrate. Different types of habitat can form different characters radula [8]. In this study will be explained about the biological processes and reveal the influence of habitat type on the Family Thiaridae radula character, then connected with different types of habitat in some areas of its distribution in Papua.

MATERIALS AND METHODS

Research sites

Research conducted in the most eastern part of the island of Indonesia. Thiaridae samples were collected from 10 study area in Papua are: Batanta, Sorong, Bintuni Bay, Manokwari, Biak, Supiori, Serui, Jayapura, Wamena and Merauke. Sampling is done on Lake Sentani and 29 rivers. Furthermore, to see the character based on habitat type of radula was observed in the laboratory.

The procedure of making preparations of radula

Making preparations radula begins with surgical specimens. Buccal mass located above the front foot and in between the tentacles removed and put into eppendorf (plastic tube) containing 70% alcohol. Observations carried out under the buccal mass brand stereo microscope Nikon SMZ-1B. Furthermore, prepared enzyme protease-K dissolved in Milli-Q as much as 10 mg and the protease-K in 1 ml of Milli-Q. The solution was stored in the refrigerator. Buccal mass removed from the alcohol 70% and soaked in an enzyme solution as much as 15-25 μ l and 300-500 μ l diggestive buffer microliter pipettes (pipetman) brand rainin with p20 (for enzyme). Further brands homogenized with a vortex mixer Crusher Thermolyne type 16700 and incubated for 5-6 h at a temperature of 50°C at AQUABATH TM brand Labline until the tissues and fat soluble so that what remains is a radula. Furthermore, the radula is cleaned in a concave glass dish containing distilled water and 70% alcohol by using a pin radula cleaned from the remnants of fat, the process is performed under a microscope. Furthermore, the radula taped stup who have been given a double adhesive, to the position facing the observer. Then, prepared with pure gold coating (ion coating) stubs plain 10 mm diameter ×10 mm with coater ion Sony Cyber-shot. Radula observed using SEM JEOL, JSM-5310LV models.

Matrix data of radula

Radula observed in the form of an image with a magnification of 150X, 200X, 250X, 350X, 500X, 750x, and 1000X. Characters are observed include: complete or not radula (teeth median, lateral and marginal), the shape of the teeth masocone median (triangular, rounded, rectangular, square). Comparison of size masocone, ectocones the teeth of the median, the number of cusps, outer marginal shape, number of teeth marginal in a row, the number of protrusions in the tooth lateral, number of rows of teeth and long radula. Other data covered is the type of substrate and the waters flow [5].

Qualitative and quantitative characters

Quantitative data includes: the amount of marginal teeth, median tooth, lateral teeth, marginal in a row, the number of protrusions in the lateral teeth, the number of rows of teeth on the radula pockets, and long radula. Radula qualitative data tabulated separately. Qualitative data includes: completeness radula, masocone shape and tooth ectocones median, lateral tooth shape, tooth shape to the median. Masocone size comparison and ectocones the median teeth. Another very important data are: type of water flow and type of substrate [5].

Analysis of the data by type of substrate

To determine the effect of substrate type of the character of the radula which covers the length of the radula of the substrate type, the number of rows of teeth and substrate type, and number of cusps with the type of substrate, shape mesocone, and the number of teeth median, lateral, and marginal to the type of substrate analyzed by ANOVA and to see the difference between the influence of substrate type done Test Analysis Significant difference. Data were analyzed by ANOVA performed with SPSS version 16.0.

Analysis of grouping characters\All data that have been identified radula character is converted into the data Nexus. Furthermore, the data were analyzed using SPSS 16.0. With Cluster Analysis done radula pengkalsifikasian and grouping characters into more homogeneous groups. The resemblance of character types is measured based on habitat type. To measure the similarity between types Thiaridae, used quadratic degree of similarity of characters. Squares level character similarity can be calculated by:

$$d(A,B) = \sum (A_n - B_n)^2 = (A_1 - B_1)^2 + (A_2 - B_2)^2 + \dots + (A_n - B_n)^2$$

Where, d(A,B)=Squared distance between the samples A and B, A_n =The character value (a) to-*n* on the type A, B_n = the character value (b) to-*n* on the type B

Values that have been obtained from the squared distance calculations will form a group. A small value will form a low value inequalities. This means that compared having a same type of great character so clumped. The next step is to calculate the distance of the first group with other jenisThiaridae (Purwanto et al., 2005). This grouping method is called Agglomerative (incorporation based on equal character). The results of the analysis in the show in the form of dendogram to look Thiaridae relationship types based on the type of substrate.

RESULTS AND DISCUSSION

Radula Analysis of of 27 types of Thiaridae derived from the results of 29 rivers and lakes exploration Sentani in Papua., Which has a different habitat types using a Scanning Electron Microscope. The results show that there are variations in the shape and size of the radula like shape ectocone and mesocones, long radula, the number of rows of teeth radula, the number of bumps on the teeth laterally, the number of bumps on the teeth of the median, the number of bumps on the teeth marginal and the number of cusps on each type.

The measurement results showed that *M. granifera* have the most long radula 2.18 mm, and the number of rows of teeth majority, namely 73 pieces. *S. flicaria* shortest 0.82 and the number of rows of teeth the least is *T. acantica* 33 in one pocket radula.

ANOVA Analysis of describes the difference in the number of cusps of the radula in any type of substrate, F=24,306, P=0.000. Tukey's Multiple Comparison test through Thiaridae known that live in sand substrate has the smallest number of cusps compared to rocks and coral substrate (P=0.000), gravel (P=0.011) and sludge (P=0.096). There are four groups of variations in the number of cusps on the type of substrate, the first group of rocks and coral substrate, the second group is a gravel substrate, the third group is mud, four groups are the type of substrate (Figure 1). It can be concluded that the type of substrate classifying the number of cusps with substrate type hard, medium and soft.



Figure 1: Diagram of variations in the number of cusps on the type of substrate

Results of analysis showed F=23,612, P=0.000. Through test of Multiple Comparison Tukey, known thiaridae that live in sand substrate having a number of teeth marginal least compared to the substrate of rocks and coral (P=0.000), gravel (P=0.001), and the sludge (P=0109) that the number of teeth marginal the radula cederung more diverse. There are four groups of varying the amount of marginal teeth on the type of substrate, the first group of rocks and coral substrate, the second group is a gravel substrate, the third group is mud, four groups are the type of substrate of sand there are differences in the number of marginal teeth on the type of substrate each habitat (Figure 2).



Figure 2: Diagram of variations in the number of teeth marginally against the type of substrate

The number of rows of teeth on the radula pocket shows the difference with F=20,320 test, P=0.000. Through Multiple Comparison Tukey test, known thiaridae who live in mud substrate has a number of rows of teeth at most compared to the substrate of rocks, gravel, rock (P=0.000) and sludge (P=0.001). There are three groups of varying the amount of the number of rows of teeth on the type of substrate. The first group of substrate rocks, gravel and sand. The second group is the type of substrate sludge, the third group is the reef substrate type. In other words, the number of tooth rows are grouped by the type of substrates that substrate type hard, medium and soft.

Results of the analysis showed radula tooth length F=67,038, P=0.000. Through Multiple Comparison Tukey test, known thiaridae that live in the mud has a radula longer than the substrate of rocks, gravel, sand, and rock (P=0.000). Radula long clumped in on the type of substrate, the substrate first group of rocks, gravel and sand, the group 2 is the substrate and the substrate of mud. Group 3 is a coral substrate (Figure 3).

Results of analysis of the lateral tooth difference on different substrates, F=37,278, P=0.000. Through test Multiple Comparison Tukey, known thiaridae who live in the substrate slurry has a number of teeth laterally at least in comparison with the substrate reefs (P=0.000), rock (P=0.001), gravel (P=0.003) and sludge (P=0177). Furthermore, substrate type classifying the number of teeth lateral to the habitat types into three groups, the first group of substrate rocks, gravel and rocks, the second group is a substrate of mud, the third group is the type of sand substrate (Figures 4 and 5).



Figure 3: Radula teeth variation in the length of the substrate type



Figure 4: Diagram lateral variations in the number of teeth on the type of substrate

Results grouping 27 types Thiaridae based character radula from 10 regions in Papua with spreading area Batanta, Sorong, Manokwari, Teluk Bintuni, Biak, Supiori, Serui, Jayapura, Wamena, Merauke, with the type of substrate varies as coral, rocks, gravel, sand, mud, litter and mix. Thiaridae character descriptions results were analyzed to form four large groups in general the same character. Based on the degree of similarity of characters seen that the group of the genus *Thiara (T. winteri, T. acanthica, T. mirifica, T. setosa), Terebia artecava, Stenomelania* sp. 2, *S. Flicaria* and *S. terebiformis* inhabit the same substrate that is a substrate of coral with flowing water. The groups showed similarities in mesocone rectangular (rectangular) and the number of teeth in a row. What is interesting is this group made up of three distinct genus but occupy the same substrate. Figure 6 shows the same radula characters on different genus.

Thiaridae groups that have similarities are: *T. mirifica, M. holandri, Stenomelania* sp. 10, *Stenomelania* sp. 4, *S. torulosa, S. funiculus, Stenomelania* sp. 7, *S. fulgurans* and *Stenomelania* sp. 6. The analysis showed similarities that show similarity in the number of teeth in a row, and form its mesocone. Substrate type partition form strong resemblance radula character mainly type substrates such as gravel, sand, gravel sand and calm waters that flow. This looks at the nine species that belong to the genus Thiaridae Melanoides and Stenomelania. Both of these genus live on the same substrate type and have a semblance of a radula. Rintelen [6] states that there is a tendency to cluster radula character based on the type of substrate (Figure 7).

Stenomelania sp. 3, Stenomelania sp. 5, S. rufesens shows the similarity in the shape its own mesocon rounded or half rounded shape and the number of teeth in a row. Substrate type where life is sandy gravel and water rushing (Figure 8).



Figure 5: Dendogram of Thiaridae radula character based on the type of substrate



Figure 6: The shape of the teeth median on rocky reef substrate (rectangular), scale=0.5 mm



T. plicaria (gravel substrate) M. Holandri (gravel substrate) Stenomelania sp.10



Figure 7: The shape of the teeth median on gravel substrate (rounded), scale=0.5 mm

Tiara scabra, Stenomelania sp. 8, *M. canalis, M. episcopalis, M. tuberculata, Stenomelania* sp. 9, *M. granifera, and M. holandri, spread in Papua mainland and island with mud berserasah substrate type, water type does not flow and there is stagnant. The genus Thiaridae third region occupies a different but have similar character mesocone mainly type triangular-shaped (Figure 9).*

The results of evaluation of the character types found thiaridae distribution which means, because of the analysis found in the same habitat types even though different jenisThiaride. If thiaridae occupy the same habitat and substrate type there is a tendency similarity of character. Conversely, if the same type occupy habitats with different substrate types the radula different characters. Under these conditions can be explained that the partition type of habitat will shape the character of different radula. Consistently found that the same type of habitat even though separated by distance it kind thiaridae distributed to different regions have a radula character types have in common. The type of environment influenced similarity [8].



Stenomelania sp 7 S. rufescens Stenomelania sp 5

Figure 8: The shape of the median teeth on a substrate of sand muddy (half rounded), scale=0.5 mm



T. scabra Stenomelania sp 8 M. tuberculata



Figure 9: Form median tooth on wet mud substrate (*trianguler*), scale=0.5 mm

Thiaridae that occupy the substrate reefs and rocks are similar in shape, mesocones and ectocone. Thiaridae types that occupy sandy gravel substrate has ectocone and mesocones rounded shape. While thiaridae who occupy the muddy sand substrate type and mesocones hemispheric ectocone (half raunded). Similarities ectocone and mesocones of radula is evidenced by the discovery of the type of thiaridae distributed in Supiori, Biak, Serui, Jayapura, Wamena and Merauke on the same substrate, namely mud berserasah have trianguler character.

Based on that discovery can be explained that the radula is plastic, resulting in ekofenotipe the radula. Ekofenotipe radula character is influenced by the type of substrate. Different response to each type of thiaridae in different habitats will come up with variations of the radula, which types are distributed in the same habitat will shape the character of the same. It is also argued by Bleakney that kind *Placidae dentritica* have variations radula characters in different habitats.

Thiaridae group which occupies the reef habitat show the character is almost similar to the substrate-shaped rock with a tendency mesocone rectanguler (rectangle), and a radula almost the same length. Andrade stated that the number of cusps of *Littoria falava* influenced by the type of substrate and water currents. In his study *L. falava*, whose initial habitat on the rocky coast moved to the muddy mangrove forests, within 40 days occurred cusp shape change, occurs ekofenotipe on the same type on different substrates.

Thiaridae groups that live in the river gravel, sand or a mixture of both and also bersubstrat river sand and mud, litter and mix shows, that turns thiaridae groups tend to have the same character. This is because of habitat bersubstrat disebapkan the same, have the same availability of feed. Michel explains the difference in the character of the radula on these kinds of *Lavigeria* in lake Tanganyika, Africa which consume different foods on different substrates.

Type *Tiara scabra*, *M. tuberculata*, *M. granifera*, *M. holandri*. *M. canalis*. *M. episcopalis*, has the same radula form that is trianguler ectocone. This group occupies the same substrate type that berserasah mud. This is supported by Rintelen et al. which states that, pachycilidae found in Sulawesi have identical radula. Padilla found variations in the group long radula teeth gastropod species Lakuna parva and *L. vincta* based on habitat type and the same food source.

Adaptation to the environment affect the type thiaridae radula character. This was seen in the taxon group of *M. tuberculata* and *T. scabra* living in the river bersubstrat stony coral, sand substrate and mud and living in the mixture tends to almost the same substrate. And vice versa, the taxon *T. scabra* found in the lake Sentani, *M. tuberculata* in Merauke, and *M. episcopalis* in the Buntuni Bay *Stenomelania* sp. 9 in Biak live and occupy the same substrate, namely wet mud. The group has a number cusp equation. Radula character distribution does not follow a fixed pattern. It is envisaged if the environment changes the type of radula will adjust. Radula volatile character at ambient pressure. This is evident in the genus *Tylomelania* have radula characters varied in different habitat types.

The diversity of radula morphology caused by different environmental conditions. Each environment will have different effects terhada thiaridae. Rivers that has a high substrate heterogeneity that is based on the cluster of coral, moss rock, berserasah waters and muddy, Thiaridae radula found a different character. The condition is supported by Sonia which states that the differences in conditions and environmental factors influence the type of radula types littoraria. In his research showed heterogeneity radula character is influenced by the diversity of algae.

In general, the structure of the radula evolved in different habitats or when consuming different foods. Thiaridae morphological variability in genetically influenced. The variability in the type caused by different genetic polymorphisms, phenotypic plasticity influenced by genotype to produce different morphologies. Radula the reproductive organs according to the function of these organs is useful as a tool and always berintraksi with the environment in utilizing the available food in the neighborhood, so change the character of the radula considered to be very close to a specific function as a tool unutuk take and cut and break and send food through osopagus to hull. Radula can not be used as a distinguishing species for special kinds of Thiaridae because of the high variation in the type, so that the character radula can only be used in the grouping until the family [11-30].

CONCLUSION

Substrate type determines the character variations on family thiaridae radula. Type the same substrate has a tendency similar characteristics as well. Although distributed at a great distance (speciation allopatrik). Thiaridae group have the same character on the condition of the river flowing, serene, and stagnate, especially on long character radula, the number of rows of teeth. The main characters are varied on the radula is the median tooth shape especially bulge its ectocone and mesocones. Effect of habitat types to the different variations of the radula real character in the analysis ANOVA and Tukey's Multiple Comparison. Radula can be used in the grouping at the family level, but radula can not be used as a distinguishing species because of the high variation in the level of species. There are 5 types of substrate (mud, gravel, rock, rocks, sand) that affects the character of the radula thiaridae, including the number of cusps, the number of teeth laterally, the number of rows of teeth, and long radula, and to influence the shape ectocone and mesocones but does not affect the number of teeth marginal. Flow continued research can be done on areas wider in order to give an idea more firmly, with a variety of substrate-specific habitats. Analysis of the stomach contents Thiaridae group is essential to connect the food type with the types of radula.

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