"Determination of Some Pacific Muricea species Using Morphology and Spicule Analysis" Michelle Hardee University Undergraduate Research Fellow, 1994-95 Texas A&M University Department of Science

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DETERMINATION OF SOME PACIFIC MURICEA SPECIES USING MORPHOLOGY AND SPICULE ANALYSIS

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Abstract.--Colony morphology and spicule analysis was used to further clarify the characteristics of Muricea fruticosa (Verrill, 1864) and refute the suggested synonymy of M. appressa (Verrill, 1869) and M. californica (Aurivillius, 1931).

Gorgonians, the horny corals, are familiar members of the class Anthozoa, subclass Alcyonaria, the octocorals, in the phylum Cnidaria. These animals are colonial invertebrates, the body being comprised of many eight-tentacled polyps surrounded by an outer rind, the coenenchyme, which covers a central collagenous axial rod. The coenenchyme is composed of calcareous spicules embedded in a mesogleal gel. The shape of these spicules, along with general colony shape and branching, is one of the most important characteristics used for identification to the specific level. Spicules function to limit compressibility of the outer coenenchyme and provide a protective cover for the polyps (Lewis and von Wallis, 1991). Muricea, the genus studied, is characterized by heavily branching colonies, mostly treeshaped, with the calyx (the lid covering the retracted polyp) formed by spicules and projecting away from the coenenchyme, producing a distinct lower lip. The polyps are scattered randomly across the branches, and do not form distinct rows. Muricea has been identified in both the Atlantic and Pacific oceans, and is widely distributed in tropical seas. As with all octocorals, the habitat of Muricea includes most latitudes and

depths; the polyps feed on plankton and detritus, allowing the organism to adapt well to a variety of environments.

There is a shortage of published literature on the Pacific Muricea, the descriptions of over 18 species arising from only a few researchers. Most of the Pacific species were described in 1869 by A. E. Verrill in his "Review of the corals and polyps of the West coast of America." His descriptions of Muricea were extremely detailed, tediously describing every spicule found in a colony, yet often not defining from which part of the coenenchyme the spicule originated. In 1924, W. Kukenthal briefly reviewed many of the Pacific species, providing more detail of spicule shape and origin, though without drawings or figures to assist the investigator in visual identification. Magnus Aurivillius, in 1931, described several new species of Muricea, stating that although they

"correspond in several characteristics with species already known, (they) have... nevertheless been provisionally determined as new ones by reason of my not having at hand any material of the species previously established, and since I do not find myself able to identify them only from existing descriptions" (Aurivillius 1931, p. 104).

A few investigators have attempted to clear up the confusion surrounding many *Muricea* species, yet the confusion continues to be compounded by improper identification in many books, which have based identification on polyp color, chemical properties,

and morphological variations. Both Aurivillius (1931) and Haderlie, Hand, and Gladfelter (1980) mention that M. californica may be synonymous with M. appressa, yet Haderlie, Hand, and Gladfelter only give references based on chemical properties, which is not sufficient evidence to establish synonymy, and Aurivillius presents no evidence at all for the comparison. Some descriptions of *M. californica* are based on yellow polyp color; this has proven to be a poor identification characteristic from pictures of M. californica showing a colony with both white and yellow polyps (Mary K. Wicksten, Texas A&M University). Other problems found in the published literature and identification keys of Muricea include striking differences in the description of basic colony shape, two identification keys, Cairns' Guide to the Commoner Shallow-water Gorgonians of Florida, the Gulf of Mexico, and the Caribbean Regions (1976), and Humann's Reef Coral Identification (1993), even differing in which branching pattern is designated lateral or dichotomous. These examples typify the poor repeatability in the work done on Muricea.

The purpose of this research was to explore the question of species synonymy between *M. californica* and *M. appressa*, and to enhance the identification of *M. fruticosa*, a species with some very distinctive characteristics. This goal was accomplished by carefully analyzing spicules and colony morphology.

All descriptions and observations were made from original type specimens, except for M. californica, of which a type specimen was not found. Type specimens of M. fruticosa were obtained on loan from the Peabody Museum of Natural History, Yale University, YPM No. 1574b and YPM No. 1574d. These dried specimens were fragments of a larger colony found in Pearl Islands, Panama, YPM No. 1574a, of which only a slide photograph was available for study. Type specimens of M. appressa and specimens identified as *M. californica* (but not type material) were obtained on loan from the Museum of Comparative Zoology, Harvard University. The two M. appressa specimens studied, MCZ No. 383 and MCZ No. 380, were dried, both originating from Panama. Both specimens were later renumbered and included in MCZ No. 3950. The M. californica specimen studied was found off Corona del Mar and preserved in ethanol. It is thought that these specimens may have been identified by E. Deichmann, a wellknown investigator of Muricea and other gorgonians.

Because some of the specimens were dried, making it difficult to remove spicules without destroying the surrounding branch, small pieces were removed from the specimens and placed in a vial of formalin, softening the mesoglea and allowing easier removal of spicules. This technique was first practiced on many *Muricea* specimens provided by Dr. Mary K. Wicksten, obtained during various dives in California. Spicules were carefully removed from the soaked pieces using an Olympus SZ-30 dissecting scope with a magnitude 10X. These spicules were then viewed using an Olympus CH-2 light microscope, using either 40X or 100X magnification depending on spicule size. Spicule sizes were measured to the nearest 0.1 mm. Pencil sketches, later retraced in ink, were made of both spicules and calyx shape in a sketchbook, along with full descriptions of spicule and colony morphology.

Results

Muricea fruticosa Verrill, 1869 Figs. 1-4

Type.--Peabody Museum of Natural History, Yale University (YPM), Nos. 1574b and 1574d, fragments of whole specimen No. 1574a, collected by pearl divers in Pearl Islands, Panama, between 11-14 meters, 1866.

Description of type.--Fragment branches average 5 mm width, colony dimensions: Height 28 cm; maximum width 38 cm. Colony is dense, bush-shaped, with a deep reddish-orange color, characteristically fading from reddish-orange in the tips and branches to a light yellow color in the main trunks and base. Axis is clear yellow at the tips, dark brown at the base. Branching is irregularly dichotomous, distinctly curving, especially the smaller branches near the ends. Terminal branches do not taper but either end evenly or are clavate. The calyx projects out 45 degrees from the branch when closed, near 90 degrees with the polyp extended. Lower lip is sharp and long with very large, distinct spicules, lying parallel to each other and sticking past the upper margin of the lower lip, giving the colony a prickly feel. The upper lip is small or barely noticeable. The calyces are close together but do not overlap, allowing the outer coenenchyme to be easily seen between the calyces. Calyx size and spacing varies with position on the colony--larger and closer towards the tips, more flat and spread apart near the base. Outer coenenchyme is characterized by extremely large spicules which can be seen by the naked eye, lying between the calyces and curving around them (Fig. 1B). These large spicules may not be seen nearer the base of the colony or in poorly preserved specimens. The outer coenenchyme spicules, reddish-orange colored and measuring up to 3 mm, can be stout, blunt and truncate at the ends, giving an almost rectangular shape, evenly covered by small tubercles; irregularly fusiform and covered with tubercles; hook-shaped on one end, both ends curving inward, or one end divided into a fork, the other end tapering to a point, these often covered on the ends with small, sparse, occasionally spiny tubercles, becoming more densely covered with tubercles toward the center (Fig. 2). The hook-shaped spicules are usually found around the base of the calyces. Inner coenenchyme spicules are distinctly smaller, their color ranging from yellow to white, and fusiform, often tapering to an a sharp point and covered with distinctly raised

tubercles (Fig. 3). Calyx spicules are long, up to 1.6 mm, very irregular and oddly shaped, most fusiform, with one end often forking slightly or very distinctly, evenly covered with distinct tubercles, occasionally with sharp spines projecting away from one side. The smaller calyx spicules are thin, fusiform, with ends either rounded or tapering to a point (Fig. 1).

Muricea californica Aurivillius, 1931 Fig. 5

Material examined.--Museum of Comparative Zoology, Harvard University (MCZ), large colony found dry, placed in ethanol, with fragments, collected by G. E. MacGinitie off Corona del Mar, at 18 m, July 25, 1935.

Description.--Dark reddish-orange colony with whitish-yellow polyps. Colony dimensions: width 10 cm; height 25 cm. Branches average width 5 mm. Colony primarily in one plane, some primary and secondary branches extend out of plane, branches curving to lie parallel to main branch. Branching lateral, terminals of even thickness or taper slightly. Axis reddish-brown at base, becomes light yellow-brown at tips. Calyx protrudes from 45 degrees to near 90 degrees away from branch when polyp is extended, very elongated and conical in shape. When polyp is not extended the calyx lies close and curves into the stem. Calyces are set close together but are not imbricated. Upper lip varies from no spicules noted to a definite lip present. Calyx spicules are small, up to 1.0 mm, closely set together, and do not project distally but are smoothly placed on the calyx. Spicules are club-shaped with large rounded spines or pointed tubercles projecting from the clavate end, the other end tapering, and covered with tubercles (Fig. 5A). The outer coenenchyme is mostly occluded by the calyces. It consists of small spicules, 0.5 mm the largest noted, with large spinulose processes on one end, often continuing along the side, the other end tapering and covered with tubercles (Fig 5B). Some spicules are fusiform, bent slightly in the middle, or have large processes in the middle projecting outward. Inner coenenchyme spicules are small, fusiform, and set with distinct tubercles.

Muricea appressa Verrill, 1864 Figs. 6-7

Types.--Museum of Comparative Zoology, Harvard University (MCZ), MCZ No. 380 (New No. 3950), dried, and MCZ No. 383 (New No. 3950), 3 specimens in ethanol, both collected in "shallow water" by P. H. Sternberg in Panama, July 1863. Specimens recently relabelled under one catalogue number.

Description of type.--Colony dimensions: height 9.2 cm; width 9.4 cm, terminal stem width 2.5 mm. Colony is a light brick red color. Growth is strictly in one plane, distinctly fan-shaped, branching pattern is mostly dichotomous with terminals often clavate and curved inward. Some terminal

branches are of even thickness with the rest of the branch. Several main trunks branch from the base, branches growing outward and eventually curving to lay parallel to the branch from which it originated. Calyces are small, crowded and overlapping, with very little outer coenenchyme showing through. Calyx tip is curved back into the stem, and the surface is covered with rough spicules, which do not project outward. Calyx spicules are small, up to 0.6 mm, the larger ones a reddish-orange and the smaller more yellow, and are rather orderly arranged on the calyx, becoming distinctly smaller as they near the calyx summit. Larger spicules are club-shaped, the swollen end projecting large sharp or blunted spinules, the other end acute and covered with tubercles (Figs. 6A, 7A). The small calyx spicules are clubshaped with enlarged spinules; fusiform and curved with spines projecting from the outer side; or are irregular with many spinulose processes (Fig. 6B). The outer coenenchyme, consisting mostly of the small amount seen through the calyces and the larger spicules ringing the calyx base, can be seen more easily by dissecting away the calyces. The spicules are small, the largest one 1.0 mm, and are irregular, curved, occasionally bluntly forked, or are fusiform with small distinct tubercles crowding the ends and larger, more spread out tubercles toward the middle (Figs. 6C, 7B).

Discussion

Muricea fruticosa

Both A. E. Verrill and W. Kukenthal have published descriptions of *M. fruticosa* along with many other Pacific *Muricea* species. Several problems which hamper clear and easy identification of *M. fruticosa* are present within both works, and are addressed here.

Verrill's description of *M. fruticosa* is extremely detailed and particular. He describes every possible variation of spicule shape, but never draws any general conclusions as to which spicules are distinctly characteristic for the species. He also fails to mention where certain shapes originated, whether they occur on the calyx, inner coenenchyme, or outer coenenchyme; instead he defines them as "larger", "longer", "stouter", "medium sized", or "smaller". Verrill also details every measurement of spicule size, yet he does not mention what spicule shape corresponds with the sizes, only giving the basic size--large, stout, etc. To improve upon Verrill's descriptions and adequately describe the species, the spicule size, shape, and origin must be noted, for the spicules are extremely important in determining Muricea to species. General sizes and ranges of possible sizes should be provided, but a detailed list of all spicule measurements is unnecessary. Verrill also goes into unnecessary specifics in describing the branching patterns of the colony, detailing branch point distances and whether they extend

some distance before dividing or if they simply divide regularly. Branching is determined by several factors, such as water current, and a colony's branching distances and regularity are too variable to designate as characteristic of a species. Designating branching as lateral or dichotomous is important, as is general formation (such as bush-shaped, fan-shaped, or distinct scarcity of branches) and branch curvature.

Highly characteristic of *M. fruticosa* is its color pattern. The colony fades from a reddish-orange in the distal branches and terminals to a light yellow, almost white color on the proximal main branches and inner trunk. Also distinctive of *M. fruticosa* is the arrangement of its outer coenenchyme spicules. Extremely large spicules, visible to the naked eye, lie prominently on the coenenchyme, often curving around the calyces. The calyx spicules are also very distinctive, projecting up away from the margin of the lower lip, arranged side-by-side, giving the calyx a ribbed look (Fig 1B). These characteristics should be given prominence in the species description; Verrill mentions the colony color but says nothing about the spicule pattern of the coenenchyme.

Kukenthal provides a key to the identification of many Pacific species (original manuscript in German, translated by Mary K. Wicksten) in his paper; although it is simple, it is difficult to follow and confidently key out a species. He determines the ultimate identification of *M. fruticosa* to be the "lower lip of the calyx short." Kukenthal does not provide a

basis for which to determine the length of this feature, nor are drawings provided to assist the identification process and support his key. The description mentions the color fading of the colony and the extremely large spicules, stating that they are "often curved and unusually formed." No more detail of spicule shape is provided, a key characteristic for this species identification. Completely opposite of Verrill, Kukenthal provides very little detail of colony branching, only mentioning "the cushion-shaped, richly branched colony has on all sides numerous short branches, which anastomose with each other".

To identify a species with any accuracy, descriptions and identification keys must determine what qualities of the species should be considered characteristic. This can be satisfactorily accomplished only by providing clear, distinct descriptions, consistently covering all characteristics, and accounting for the range of variation envisioned in a single word by many different investigators. By comparing the descriptions against other species, keeping the compared characteristics consistent, and setting standards of correlation for future investigators, it is possible to determine the range of morphological variation and provide a reliable basis of comparison. Because of the tremendous amount of work needed to be done, there is an absence of both manpower and interest in providing a reliable key for all the Pacific Muricea species, and few investigators are willing to tackle this problem. In an unpublished abstract, Homna and Patton (1994) have shown that Muricea may be ecologically

dominant, thus making it an important organism in an ecosystem and necessitating further study.

Muricea appressa and M. californica

The belief by two different investigators that *M. appressa* and *M. californica* might be synonymous with each other prompted the study of the morphology and spicules of these two species. Magnus Aurivillius, in "The gorgonians from Dr. Sixten Bock's expedition to Japan and the Bonin Islands, 1914," gives a possible synonymy with *M. appressa* Verrill, 1864 (p. 111); unfortunately, he does not provide any further detail. Haderlie, Hand, and Gladfelter in Morris, Abbott, and Haderlie's <u>Intertidal Invertebrates of California</u> (1980) mention that *M. californica* (Aurivillius 1931) may prove to be synonymous with *M. appressa* (Verrill 1864), yet only give references based on chemical properties (Grigg, 1970, 1972, 1974, 1975, 1976, 1977, cited in Haderlie, Hand, and Gladfelter, 1980).

Only one published description of *M. californica* was found, authored by Aurivillius (1931); both Verrill (1864) and Kukenthal (1924) provide descriptions of *M. appressa*. It is the opinion of this author that the two species are not synonymous but are distinctly different in many characters. Unfortunately, type specimens of *M. californica* could not be found. The following statements are based on descriptions and specimens that are not type material.

The distinction between the two species is determined more from colony morphology than spicule differences, although there are distinct differences between the spicule forms of these species (Table 1). M. appressa is typified by colony growth strictly in a single plane, and distinctly fan-shaped, with terminal branches often clavate and curved inward. M. californica, on the other hand, is not strictly planar; it has several branches which grow out of the plane of the colony. The terminal branches are of even thickness or taper slightly, not clavate, and branches curve away and eventually turn to run parallel to their branch of origin. In both species, the calyx curves inward and lies close to the stem when the polyp is retracted. However, in M. appressa the calyces are small, crowded, and distinctly overlapping, whereas in M. californica the calyces are not imbricated. In both species, the calyces cover the branch surface and allow little of the outer coenenchyme to show through. It is noted in M. appressa that the spicules of the calyx distinctly grow smaller as they near the distal edge of the lower lip.

Spicule analysis of the two species provides more differences. The calyx spicules of both species are usually club-shaped, the broader end often ending in spines or blunt projections. The dissimilarity, however, lies in the outer coenenchyme spicules. In *M. californica*, the spicules are all relatively small, their morphology much the same as the calyx spicules, showing the club shape with projections on one end (Fig. 5B). The outer coenenchyme spicules of *M. appressa* are more unusual. They are larger, up to 1.0 mm, and are irregular, bent, occasionally bluntly forked, or fusiform with small, distinct tubercles on the ends, blending to larger tubercles toward the middle (Fig. 6C). These differences, along with the morphological characters, appear to define *M. appressa* and *M. californica* as two distinct species, not synonymous. By using consistent comparisons and clarifying many of the descriptions, no doubt many other questions of species synonymy can be resolved.

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	H. appressa	M. californica	M. fruticosa
Colony color	light brick red.	dark reddish-orange.	deep reddish-orange in branches and tips, fades to a light yellow in main trunks and base.
Branching	strictly planar, distinctly fan-shaped, branching mostly dichotomous, terminals often clavate. Branches grow out, then curve back to parallel main branch.	primarily in one plane, some branches exted out of plane. Lateral branching, terminals even or tapering.	irregularly dichotomous, smaller branches and terminals distinctly curving. Terminals clavate or end evenly.
Calyx	small, crowded, overlapping. Calyx tip curves back to stem, and covered with rough spicules that do not project outward. Spicules distinctly smaller as near calyx summit.	projects 45 to 90 degrees when polyp extended. When closed, lies close and curves into stem. Calyces are not overlapping. Spicules do not project outward.	projects at least 45 degrees when closed, up to 90 degrees when polyp extended; they do not overlap. Lower lip sharp, long, with large spicules lying parallel and projecting past lip margin. Colony has a prickly feel.
Outer coenenchyme	very little showing through calyces.	mostly occluded by calyces.	very noticeable, large spicules, many lying between and curving around calyces.
Outer coenenchyme spicules	<pre>small; irregular, curved, occasionally bluntly forked; fusiform with small tubercles at ends, larger and more spread out in middle. Largest noted: 1.0 mm.</pre>	<pre>small; clavate with large spinulose processes on one end often continuing around one side, the other end tapering, with tubercles; some fusiform, bent in the middle, or with large processes in the middle projecting outward. Largest noted: 0.5 mm.</pre>	stout, blunt, truncate, covered with tubercles; irregularly fusiform, covered with tubercles; one end hook-shaped, both ends curved inward, or one end forked with the other end tapering, covered with small, sparse, and spiny or warty tubercles, becoming denser toward the middle. Largest noted: 3 mm.
Inner coenenchyme spicules	small, fusiform.	<pre>small, fusiform, distinct tubercles.</pre>	fusiform, many taper to sharp points, covered with distinctly raised tubercles.
Calyx spicules	larger ones are club-shaped, swollen end has large sharp or blunt spines, other end acute with tubercles. Small spicules are club-shaped with enlarged spines; fusi- form, curved, with spines on outer side; irregular with many spiny processes. Largest noted: 0.6 mm.	club-shaped with large rounded spines or pointed tubercles projecting from clavate end, other end tapering, covered with tubercles. Largest noted: 0.9 mm.	irregular, oddly-shaped, mostly fusiform, with one end forked. Covered evenly with tubercles or occasionally with spines on one side. Largest noted: 1.5 mm.

Table 1. Comparison of M. appressa, M. californica, and M. fruticosa identifying characteristics



Fig. 1. <u>Muricea</u> <u>fruticosa</u> Verrill, 1869. Type, YPM 1574b. 1866, 11-14 meters, Pearl Islands, Panama. A, calyx spicules; B, representation of spicule arrangement on a branch.



Fig. 2. <u>Muricea fruticosa</u> Verrill, 1869. Type, YPM 1574b. 1866, 11-14 meters, Pearl Islands, Panama. Outer coenenchyme spicules.















Fig. 3. <u>Muricea fruticosa</u> Verrill, 1869. Type, YPM 1574b. 1866, 11-14 meters, Pearl Islands, Panama. Inner coenenchyme spicules.



Fig. 4. <u>Muricea fruticosa</u> Verrill, 1869. Type, YPM 1574d. 1866, 11-14 meters, Pearl Islands, Panama. A, calyx spicules, viewed at 40X; B, smaller yellow calyx spicules, viewed at 100X; C, coenenchyme spicules and spicules surrounding calyx base, viewed at 40X and 10X, respectively.





0.2 mm

10.1 min

Fig. 5. <u>Muricea californica</u> Aurivillius, 1931. MCZ specimen. July 25, 1935, 18 meters, off Corona del Mar. A, calyx spicules, viewed at 100X; B, coenenchyme spicules, viewed at 100X.

0.2mm







Fig. 6. <u>Muricea</u> <u>appressa</u> Verrill, 1864. Type, MCZ No. 380 (New No. 3950). July 1863, shallow water, Panama. A, larger calyx spicules, viewed at 100X; B, small spicules at calyx summit, viewed at 400X; C, coenenchyme spicules, viewed at 100X.

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A



0.7 mm

Fig. 7. <u>Muricea appressa</u> Verrill, 1864. Type, MCZ No. 383 (New No. 3950). July 1863, shallow water, Panama. A, calyx spicules, viewed at 100X; B, coenenchyme spicules, viewed at 100X.

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