EcoTaxa UVP Guide

<u>Mesozooplankton</u>

Background:

The UVP5HD records ROIs larger than 600µm. However, it is very difficult to identify living particles which are smaller than 1mm. Read Lombard et al (2019) for details about imaging tools and their scope. Using UVP vignettes, it can be tempting to classify things which are not definite particles. The suggestive power of human imagination and AI-predications can be misleading. Thus, we take a conservative approach in identifying particles as living. While this may lead to underestimation, it can allow for more confidence in IDs. All categories have "inclusion criteria" which should be met to label a vignette.

Crustaceans:

The UVP reliable will image several types of pelagic crustaceans. Notably copepods, ostracods, euphausiids, decapods, and the occasional amphipod. Often it is fairly easy to identify taxa to something more specific than *Crustacea*. However, there is the rare vignette which can be difficult to distinguish into a specific group. In such a case, it should be labeled *Crustacea*.

1. Copepoda:

General Description

Copepods are the most abundant metazoan on the planet.

UVP Identification Techniques

Large copepods are fairly easy to recognize when in a favorable orientation. The easiest way to identify copepods is the presence of two antennae. Even on smaller copepods, large antennae are often visible. Alternatively, shape and grey patterns can help identify copepods. The typical copepod shape is ellipsoidal, which can be easily confused with many particles. However, grey color patterns can reveal gut structures of Copepods. This and partial appendage visibility can facilitate copepod identification.

The Best



Figure 1. Ideal images of *Copepoda*. These meet all inclusion criteria: antennae and additional appendages are clearly visible. Grey coloration shows copepod physical features inside their ellipsoidal shape.

The Average – antennae



Figure 2. Common instances of *Copepoda* identified by appendage visibility. Here, while the overall individual may be small, the presence of two large antennae clearly reveal these to be copepods.

The average – color & shape



Fig 3. Common *Copepoda* identified by body shape and coloration. No antennae are visible, yet these individuals have body shapes and colors consistent with what is expected of copepods. Partial appendages may be visible.

The Challenging



Fig 4. Challenges instances of *Copepoda*. In these, it may be difficult to ID the copepod. However, in all there are partial appendages visible or copepod-like coloration. Note that some of these may be too small to be included in analyses. However, it is still good to ID them when able. Viewing in a zoom can be helpful.

Inclusion Criteria:

All vignettes labeled as Copepoda should have either:

- Two clear antennae attached an ellipsoidal shape.
- An ellipsoidal shape with grey patterns consistent with copepod physiology.
- Partial antennae or tail with grey patterns suggestive of copepod physiology.

Often confused with:

Pteropoda, Acantharea, Ostracoda.

2. Eumalacostraca

Background:

With the UVP it can be challenging to distinguish between Euphausiids and Decapods.

UVP Identification Techniques:

Inclusion Criteria:

Often confused with:

5. Polychaeta

Background: UVP Identification Techniques: Inclusion Criteria: Often confused with:

Rhizaria

Background:

Rhizaria- Supergroup of a meboid protists who generally have mineral tests made of silica, $\mbox{SrSO}_4,$ or $\mbox{CaCO}_3.$

Rhizaria, as a taxonomic category, are a recently described clade whose members were previously thought to be spread across protozoan phylogenies. Refer to Burki & Keeling (2014)^[1] for a description of the recent description of Rhizaria. Rhizaria are extremely diverse, in both terms of evolutionary lineages and ecological roles. Within Rhizaria, the exact taxonomy is still under investigation. Currently, there are two key groups within the Rhizaria; Cercozoa and Retaria.

Cercozoa are an interesting phylum of protists who are extremely diverse. They include one of the two known photosynthetic Rhizaria lineages, the chlorarachniophytes. Additionally, the smallest known heterotrophic grazer, *Minorisa minuta*, belongs to the Cercozoa. However, most of these taxa are extraordinarily small and lack firm structures. The Phaeodarea are the only subclass of Cercozoa which we are able to study using the UVP. These ameboid protists are exclusively heterotrophic and thought to lack any symbiotic microalgae^[2].

Retaria includes the phyla Foraminifera and Radiolaria. The Foraminifera are very well studied due to their persistence in sediment record. Additionally, planktic foraminifera have successfully been cultured, allowing for a more detailed description of their life cycle. Their tests are made of Calcium Carbonate (similar to Coccolithophores) and form in a somewhat spiral shape. Some foraminifera contain photosynthetic microalgae in their pseudopodia. These algae both can provide a food source (typically consumed before reproduction) and locally raise pH, facilitating calcification of the foram's tests^[3].

The radiolarians are placed into two groups based on their mineral composition. Polycystinea are radiolarians whose tests are made of silica (glass-like). Primarily, we observe Collodaria, a very small yet colonial member of the polycystinea whose colonies can reach large sizes. Acantharea are the other group of Polycystine radiolarians, whose tests are made of strontium sulfate. Many of these taxa host photosymbionts, however they have recently been document to have larger rates of predation than previously thought

Broadly, Rhizaria are identified by their central tests and pseudopodia. With UVP sampling, we are capable of imaging those rhizaria whose total ESD are >500 μ m. However, phylum-specific identification is best with vignettes of Rhizaria >1000 μ m. While many protists can be difficult to identify with morphology alone, there are some key characteristics we can use to distinguish between Rhizaria groups.

Identifications Using the UVP

Rhizaria:

If you cannot identify rhizaria to a lower taxon, just label it as "Rhizaria X" and we can look into it later. Many rhizaria cannot be distinguished (particularly smaller cells) and thus we will eventually just classify them as Rhizaria.

1. Foraminifera:

General Description

As stated, above foraminifera are well identified by their somewhat spiral tests (F1a). Their pseudopodia are flexible and can be retracted when not feeding. These pseudopodia can also host symbionts which display a halo-like feature.



Figure 1. Left: Scanning electron microscopy of a fossilized planktic foraminifera (<u>https://www.nhm.ac.uk/our-science/our-work/biodiversity/planktonic-forminera.html</u>) Right: *Orbulina universa,* imaged under light microscopy with golden symbionts (https://www.sciencedirect.com/topics/earth-and-planetary-sciences/foraminifera).

UVP Identification Techniques

In UVP vignettes, I have only seen foraminifera with their pseudopodia extended. I believe those with retracted pseudopodia are likely indistinguishable from debris. Generally, they can be distinguished from other rhizaria by their amorphous-dark centers coupled with a loose, fuzzy array of pseudopodia.

The Best: Will be extremely large, with dark tests and long pseudopodia



Fig 2. Good images of foraminifera. Left to right: BATS 2019, CCELTER 2019, Tara oceans.

The Average: Often times, their pseudopodia can be bent or swirly. Additionally, frequently their entire pseudopodal networks is either not imaged or removed by segmentation.



The Challenging: When very small they can be tricky to distinguish from Acantharea. Additionally, the segmentation can often remove parts of their pseudopodia as their own vignettes. This would be a segmentation ("true") duplicate.



Fig 4. Small formanifera. These were identified by their dark centers and non-symmetric appendages. BATS2019(3),CCELTER2019.

المعتقدة مطلح בדרפ לפישה Fig 5. A poor segmentation of foraminifera pseudopods.

2. Radiolarians:

This group contains both the Collodaria and the Acantharea. These orders however are likely to not get confused with each other so we will not categorize anything at the level of radiolarian (if unsure it should be categorized as Rhizaria. *Note: I am somewhat concerned about the group Sprumellaria. These are polycystinea who appear to be morphologically similar to Acantharea. At this point, no UVP users identify Sprumellaria so it may be too small or something. However, if we are imaging Sprumellaria and calling it Acantharea, that provides an issue for estimating silica vs strontium.*

2.1 Acantharea:

General Description

Acantharea are radiolarians made of strontium sulfate ranging from tens to hundreds of microns^[4](F6). Their central capsule is surrounded by several, symmetric spines which intersect at the center. Additionally, the ectoplasm can extend beyond the central capsule and connect the spicules. This can be filled with algae and food and vary in color and transparency ^[4](F7). Fun fact, Acantharea host dozens of nuclei in a single cell.



Fig 6. Range of sizes of Acantharea imaged with an ISIIS near Japan. Figure 2 in Brisbin et al 2020.



Fig 7. Images of Acantharea with extended cytoplasm (periplasm). Left, light microscopy (4). Right, Video Plankton Recorder, scale bar 1000µm (Nakamura 2017).

UVP Identification Techniques

Acantharea are abundant in our UVP casts although they can be tricky to distinguish from other Rhizaria groups. The key attribute to look for is the presence of symmetric spines. The visibility of a central sphere is variable based on the extent of the ectoplasm.

The Best: Will have many, clearly symmetric spicules. I tend to think of these as "Sharp" in appearance.



The Average: Often vignettes will have only a few spicules and varying amounts of material on the center of the cell. These often push the lower limit of the UVP as well. Remember to look for straight & symmetry.



Figure 9. Common examples of Acantharea images. BATS(3), Exports, CCELTER(2).

The Challenging: Acantharea are challenging to identify at very small sizes or when there are larger centers which can be confused with a small foram. Additionally, cells with large amounts of ectoplasm and attached particles seem to have relatively smaller spines.



2.2 Collodaria:

General Description

These are the only colonial radiolarian, whose individual cells are small yet form large colonies, often several millimeters in length. The largest recorded colony was 3m in length. Some collodarian taxa are solitary however, forming large globule-shapes. These organisms host algal symbionts shared between cells and thrive in surface waters.



Figure 13. Light microscopy images of collodrians. Left two are solitary while rightmost is a colony. Images from Suzuki & Not (2015).



Figure 12. Collodaria recorded with video plankton recorder. Leftmost is a solitary organism while the other three are colonies. Scale bar 1000µm (Nakamura 2017).

UVP Identification Techniques

Collodaria will generally be fairly light and possibly confused with badfocus images. Some although will have dark centers if they are solitary. Colonial organisms are easy to identify. Here I'll show each type of collodaria. Solitary are described as either globule or dark based on their appearance.

 Collonial<Collodaria: These generally look like long, clusters of little specks in long or circular aggregations. Often these will be separated in segmentation or not full imaged.



 SolitaryGlobule: These are greyish in the center with large globish shapes surrounded by a more transparent halo



Figure 14. Solitary forms of collodaria displaying a globish appearance. BATS(2), GEOMAR 2015.

 Solitaryblack: These are best described from other rhizaria by their soft-looking halo around the dark center. These are possibly confused with castanellidae, the key distinguishing for black collodaria are their consistently dark centers and their circular halos.



3. Phaeodaria

Phaeodaria are an group of cercozoans who generally contain silicia skeletons. These were previousy considered a group within radiolarians but were moved to the cercozoans following 18s RNA sequencing. While they do have silica skeletons (a <u>scleracoma</u>), they are much more porous and fragile than the silicous radiolarians. The main features of all Phaeodaria include; a central capsule which is often too small to distinguish in our vignettes, a phaeodium which is a dark mass of particles, and the

silica lattice-like skeleton: the scleracoma. If a vignette cannot be identified to a lower taxonomic level, but is clearly some type of phaeodarian, we can ID it as unknown phaeodarian (F16)



Figure 16. Unidentified phaeodarian. It clearly has some phaeodium and a sclerocoma, however it does not match other vignettes. It looks possibly to be a polyhedral *Tuscaroridae* but that is not a confident identification.

3.1 Aulacanthidae

General Description

This family of phaeodarians have a spherical scleracoma with a mesh-like "spherical veil" and spines which exendt from the (typically) centrally located phaeodium to beyond the spherical veil (F17). These are easily confused with Aulosphaeridae and possibly formainifera, but the straight extension of the spines and central phaeodium are key distinguishing features.



Figure 17. Schematic illustration of a typical aulacanthidae. From Nakamura & Suzuki 2017.



Figure 18. Living images of aulicanthidae. Left, light microscopy images from Nakamura & Suzuki 2017. Right, Video plankton recorder, scale bar 1000µm from Nakamura et al. 2017.

UVP Identification Techniques

Aulacanthiae can have a wide range of sizes and some additional variation in morphology. There are some groups which we use to futher classify aulacanthidae. All groups are characterized by straight

spines with a dark, amorphous center, which is the phaeodium. The sphereical veil should be present but is variable in its visibility and opaqueness. The three labels to use are: the higher taxanomic category aulacanthidae, the genus Aulacantha, and the genus Aulographis.

Aulacanthidae, the good: The spherical veil is present, but often is fairly transparent with only edges visible. The spines are straight, extending well beyond the spherical veil with the ocasional dark tip.



Aulacanthidae, the challenging: Often the veil is less visible and spinre are not fully captured. These can easily be confused with foraminifera. A key difference for aulacanthadiae is the straight-ness of the spines while forams have more "loose" pseudopodia.



- Figure 20. Challenging images of Aulacanthidae. BATS, CCELTER2019(2)
- Aulacantha, the good: These are easy to distinguish by their more opaque spherical veil, and many straight spines which just barely extend beyond the spherical veil.



Figure 21. Characteristic vignettes of Aulacantha. BATS(2), GEOMAR2018, CCELTER 2019

 Aulacantha, the challenging: When very small or opaque, the spines are often indistinguishable. In these cases, key features are the dark, central phaeodium and spherical veil.



Figure 22. Dark or small Aulacantha with indistinguishable spines. BATS(2), CCELTER2019(2).

 Aulographis, the good: Aulographis are extremely similar to aulacantha. The key feature for aulographis are the oblong shape and often two phaeodium. Spines are often less visible than aulacatha



Figure 23. Characteristic images of Aulographis. CCELTER(2), EXPORTS

 Aulographis, the challenging: When vignettes are particularly light, aulographis may appear similar to Aulosphaeridae, or even a solitary globule. Key features to remember are the central location of aulographis' phaeodium.



3.2 Aulosphaeridae

General Description

A primarily spherical phaeodaria, the scleracoma in this taxa is typically lattice-like structure. Spine-like tubes extend from the edge of the scleracoma. Phaeodium is off-center, at times just slightly (F25) and others it can be near the edge of the scleracoma (F26a). Central capsule is often near the size of the phaeodium and can be visible in UVP photos. At times, they can form colonial arrangements.



Figure 25. Illustration of general aulosphaeridae strucutre. From Nakamura & Suzuki 2015.



Figure 26. Images of live Aulosphaeridae. Top, light microscopy from Nakamura & Suzuki 2015, Stukel et al. 2018. Bottom, video plankton recorder from Nakamura et al. 2017.

UVP Identification Techniques

UVP vignettes of Aulosphaeridae rarely show visible tubes. The circular scleracoma is often visible yet transparent and tubes can give it a fuzzy appearance. Phaeodium is typically smaller than aulacanthidae and is often slightly off-centered to all the way near edge of interior scleracoma. Capsule is visible in larger photos. Special labels include colonial forms and the genus *Aulotractus*, which takes a more torpedo shape.

The Good: vignettes show a light, yet visible scleracoma. Phaeodium and capsule are visible at times. Tubes give a slightly fuzzy appearance.



Figure 27. Characteristic images of aulosphaeridae. BATS(2),CCELTER2019(2).

The Average: Typical vignettes do not show tubes, particularly for smaller cells. Scleracoma is visible but not be visibly defined.



The challenging: At very small sizes, there are tough to distinguish from other phaeodarians, particularly small aulacantha. Key features to look for are smaller phaeodium (relative to aulacantha) and lack of spines.



Figure 29. Challenging vignettes of aulosphaeridae. BATS, EXPORTS(2), CCELTER2019.

Colonial: Easily distinguishable, large aulosphaeridae joined at the edge of their scleracomas.
Often more oblong in their shape and can include empty shells.



 Aulotractus: Singular aulosphaeridae with a more topedo shape. Can often include a little taillike pinch in the scleracoma.



3.3 Cannosphaeridae

General Description

Central phaeodium with a fragile scleracoma. Spine-like structures connect to center and form very porous scleracoma. Often very geometric in appearance.



Figure 32. Light microscopy of a cannosphaeridae. From Nakamura & Suzuki 2015.

UVP Identification Techniques

Typically, decently large (>2mm). Centrally located capsule and phaeodium and visible, although very faint, scleracoma. Geometric in appearance with polygon-looking surface. Can be colonial. (So far) observed in smaller numbers than other phaeodarian, as a result, no average-looking vignettes.

The good: Dark centers with hexagon-like lattice surrounding it. Often with debris or other particles on the surface.



Figure 33. Characteristic images of Cannosphaeridae. CCELTER2019, EXPORTS

The challenging: Often these are not fully imaged or cut in the segmentation process. Key features will still display a geometric pattern round the phaeodium. Additionally, scleracoma may be less visible.



Colonial: In colonial forms, often large and visible. Joined along some outer edge of the scleracoma.



3.4 Castanellidae

General Description

Though less described in other literature, this species can be in large numbers in some UVP projects. This taxon has an extremely opaque scleracoma. The phaeodium can be large relative to the size of the scleracoma and take up the majority of the extracapsular zone. While some have been imaged with spines, these are often absent or at least not visible. Generally observed between .2-1mm



Figure 35. Living images of Castanellidae via light microscopy. Left two, Canada institute of fisheries. Right Biard et al 2018 (scale = 500µm).

UVP Identification Techniques.

Due to their smaller size, these cells often appear simple dark spheres in UVP photos. However, the contrast in the central capsule/phaeodium region can give an identifiable shape – a greyish sphere with a dark, often dual-sided middle region. Often larger vignettes will have an amorphous surrounding grey area behind the central region.



The Average: Smaller individuals can appear very dark. Key identifying features here are a slightly light region in the sphere and at times an amorphous grey region

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Figure 37. Typical images of Castanellidae. BATS(3), CCELTER2019(2)				

The Challenging: Due to their small size alone, the average vignette may be difficult to distinguish from other rhizaria alone. If you are not confident in your ID, these can be labeled as Rhizaria or even phaeodarian unknown.

3.5 Coelodendridae

General Description

These large phaeodarian are easily distinguished from others. Their name comes from the branching-like structure of their scleracoma, although this feature is typically not visible in UVP vignettes (F36). The central capsule and phaeodium are dark and centered. The scleracoma is often not spherical but rather oblong or triangular. The core of the cell often spans a few hundred to a thousand microns. Coelodendridae also have styles, large appendages which can extend up to a few mm (F37). These appendages often are thought to be used for food capture, with recorded appearances of metazoans attached to the styles. As a result, they often can be filled with dark particles. Some colonies do form through multiple individuals attached via styles.



Figure 36. Illustration of Coelodendridae (Phaeodendrida). From Nakamura & Suzuki 2015.





UVP Identification Techniques

Alone, Coelodendridae can be difficult to distinguish from other phaeodarians. However, the presence of the long style makes them easy to identify. Can be identified to the genus level in the case of *Coelographis*. When styles are not visible (and is not coelographis), categorize as coelodendrum

 Coelodendridae, the good: Vignettes will be large due to the presence of the styles, dark centers and scleracoma easily visible. Typically, 2-5 styles although more at times.



Figure 38. Characteristic images of coelodendriae. BATS(2), Exports

 Coelodendridae, the average: Often the full extend of legs will not be imaged and/or segmented poorly.



 Coelodendriade, the challenging: When styles more frequent and straight, these can be confused with aulacanthidae. A key feature to look for is the length of the appendage, Coelodendridae will have their styles extend much further beyond their scleracoma than aulacanthidae spines. Additionally, Coelodendridae's scleracoma will be less spherical



Colonial: Multiple Coelodendridae attaching through their styles. Appear entangled often.



- Figure 41. Coelodendridae in colonial formation. CCELTER2019, BATS
- Coelodendrum: When styles are not present, but still clearly Coelodendridae. Branch-like skeleton may be visible



Figure 42. Characteristic images of Coelodendrum spp. TARA,CCELTER2019,BATS

 Coelographis, the good: Styles may or may not be present, although much shorter when present. Cell in a triangular/arrowhead shape.



 Coelographis, the average/challenging: When smaller, structures will be less visible and borders may not be well imaged. Key feature to distinguish from other phaeodarians are the triangular shape.



3.6 Medusettidae

General Description

Individuals are often smaller 100-500µm than other phaeodarians but can reach a few mm in size. Individuals have scleracoma with appendages extending from one side of the cell. Appearance is like a little head with swimmy feet. Phaeodium typically located towards the appendage base. While I haven't found a description of them as colonial, imaging suggests they may form aggregations. These clusters can be a few millimeters in size.



Figure 45. Medusettidae images. Left and middle, light microscopy from Nakamura & Suzuki 2015 and Biard & Ohman 2020. Right zoocam from Ohman et al 2018.

UVP Identification Techniques

Vignettes will often have a core center with appendages extending beyond on one side. In clusters they can appear as little clumps with legs extending around.

Solitary: Appendages small and extending from one side. Maybe fairly visible.



Colonial: Clustered at their sclerocomas, legs often extending in the same direction



3.7 Tuscaroridae

General Description

There individual cells have a morphology more similar Medusettidae than other phaeodarian. Individual cells have a globe-like scleracoma, large capsule, a disperse phaeodium, and bend appendages extending from one side. Individuals can reach several millimeters in size. These form colonies, attaching around a central lattice sphere made of silica.



Figure 48. Illustrations of Tuscaroridae. Left, individual cell; Right, colonial formation around a silica lattice sphere. From Nakamura & Suzuki 2015.



Figure 49. Individual light microscopy images of Tuscaroridae. Scale bar 1000µm. Ikenoue et al. 2019.

UVP Identification Techniques

These are primarily imaged in a colonial form. Colonies reach up to a few centimeters in size. To date, I have not recorded their presence at BATS. Central lattice can be spherical or polyhedral in shape.

Spherical Colony:



<u>ອີດທາຫຼັງ ອີດທີ່ສີດສັກກ</u> Figure 50. Spherical colonies of Tuscaroridae. Dark points are individual cells. GEOMAR2018, EXPORTS(2).







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