Molecular Phylogenetics and Evolution 82 (2015) 324–329

Contents lists available at ScienceDirect

Molecular Phylogenetics and Evolution

journal homepage: www.elsevier.com/locate/ympev

Short Communication

Molecular phylogeny of the acorn barnacle family Tetraclitidae (Cirripedia: Balanomorpha: Tetraclitoidea): Validity of shell morphology and arthropodal characteristics in the systematics of Tetraclitid barnacles

Ling Ming Tsang [a], Ka Hou Chu [b], Yair Achituv [c], Benny Kwok Kan Chan [d,*]

[a] Institute of Marine Biology, National Taiwan Ocean University, Keelung, Taiwan
[b] Simon F. S. Li Marine Science Laboratory, School of Life Sciences, The Chinese University of Hong Kong, Shatin, Hong Kong
[c] The Mina and Everard Goodman Faculty of Life Sciences, Bar-Ilan University, Ramat-Gan 52900, Israel
[d] Biodiversity Research Center, Academia Sinica, Taipei 115, Taiwan

ARTICLE INFO

Article history:
Received 25 June 2014
Accepted 15 September 2014
Available online 27 September 2014

Keywords:
Molecular phylogeny
Shells
Barnacles
Molecular analysis

ABSTRACT

Shell structure is a crucial aspect of barnacle systematics. Within Tetraclitidae, the diametric and monometric growth patterns and number of rows of parietal tubes in the shells are key characteristics used to infer evolutionary trends. We used molecular analysis based on seven genes (mitochondrial COI, 16S and 12S rRNA, and nuclear EF1, RPII, H3, and 18S rRNA) to test two traditional phylogenetic hypothesis: (1) Tetraclitid barnacles are divided into two major lineages, which are distinguished according to monometric and diametric shell growth patterns, and (2) the evolutionary trend in shell parietal development began with a solid shell, which developed into a single tubiferous shell, which then developed into multitubiferous shells. The results indicated that Tetraclitinae and Newmanellinae are not monophyletic, but that Austrobalaninae and Tetraclitellinae are. The phylogram based on the genetic data suggested that Bathylasmatidae is nested within the Tetraclitidae, forming a sister relationship with the Austrobalaninae and Tetraclitinae/Newmanellinae clade. Within the Tetraclitinae/Newmanellinae clade, the genera *Tetraclita* (multitubiferous shell), *Tesseropora* (single tubiferous shell), and *Yamaguchiella* (multitubiferous shell) are polyphyletic. The results suggested that shell morphology and growth patterns do not reflect the evolutionary history of Tetraclitidae, whereas the arthropodal characteristics are informative.

1. Introduction

Acorn barnacles are one of the model organisms that were favored by Charles Darwin in his evolutionary studies (Darwin, 1854). They have a conical shell with an opercular opening at the summit (Fig. 1A). The number of shell plates varies among barnacles; hence, shell morphology plays a crucial role in barnacle identification and constitutes the basis of taxonomic and phylogenetic studies (e.g., Pilsbry, 1916; Newman and Ross, 1976; Newman, 1987). The Tetraclitidae family is characterized by a shell consisting of four tubiferous or solid plates (Fig. 1C, in *Epopella*), with a membranous or calcareous base that does not form a complex interdigitation with the wall (Ross, 1969; Newman and Ross, 1976; Ross and Perreault, 1999). Individuals in this family are common fauna in the rocky intertidal and shallow waters of tropical and subtropical regions (Newman and Ross, 1976).

Currently, Tetraclitidae comprises approximately 60 nominal species that are categorized into 10 genera and four subfamilies, namely Tetraclitinae, Tetraclitellinae, Austrobalaninae, and Newmanellinae, based on shell shape, shell and base structure, and rows of parietal tubes (Newman and Ross, 1976; Ross, 1999; Ross and Perreault, 1999). Traditionally, there are several controversial arguments regarding the evolutionary patterns of the Tetraclitidae. The family was divided into two main lineages (Ross, 1969) based primarily on the growth pattern of the parietes. Tetraclitinae exhibit dominantly monometric shell growth, whereas the shells of Tetraclitellinae grow dominantly diametrically (Ross, 1969; Fig. 1B). Monometric growth lineage is also known as the tesseroporan lineage (Ross, 1969), because it was hypothesized that the lineage began with a single tubiferous *Tesseropora* ancestor that evolved until it exhibited the multitubiferous state observed in *Tetraclita* (Fig. 1C). Ross (1970) suggested that the monometric growth lineage began with a solid-wall diametric-growth ancestor, which developed into a solid-wall monometric-growth *Epopella*-like ancestor, leading to *Tesseropora* with a single

* Corresponding author. Fax: +886 2 27872235.

http://dx.doi.org/10.1016/j.ympev.2014.09.015
1055-7903/

tubiferous shell, which evolved into *Tetraclita*. Anderson and Anderson (1985) corroborated this view and suggested that, because tetraclitellans exhibit the most plesiomorphic characteristics, they occupy a basal position in the *Epopella–Tesseropora–Tetraclita* line. By contrast, Foster (1978), Buckeridge (1983) and Foster and Buckeridge (1987) claimed that *Tetraclitella* is derived directly from *Tetraclita*. The validity of these taxonomic viewpoints has not been tested in a phylogenetic framework. A robust phylogeny of the Tetraclitidae family is crucial for testing these hypotheses.

Data provided by molecular phylogenetics can be used to refine the evolutionary patterns proposed based on comparative morphological analyses and provide new insights into the diversification history of acorn barnacle families (e.g., Pérez-Losada et al., 2008; Tsang et al., 2014). We used sequences of both mitochondrial and nuclear genes to construct a robust phylogeny for the Tetraclitidae family by including members from all four subfamilies. The inferred gene tree enables testing whether the morphological characteristics of shell structures and arthropodal appendages reflect the evolution of this family.

2. Materials and methods

2.1. Taxon sampling

We collected specimens from 6 of the 10 Tetraclitid genera, with representatives from all 4 subfamilies (Supplementary

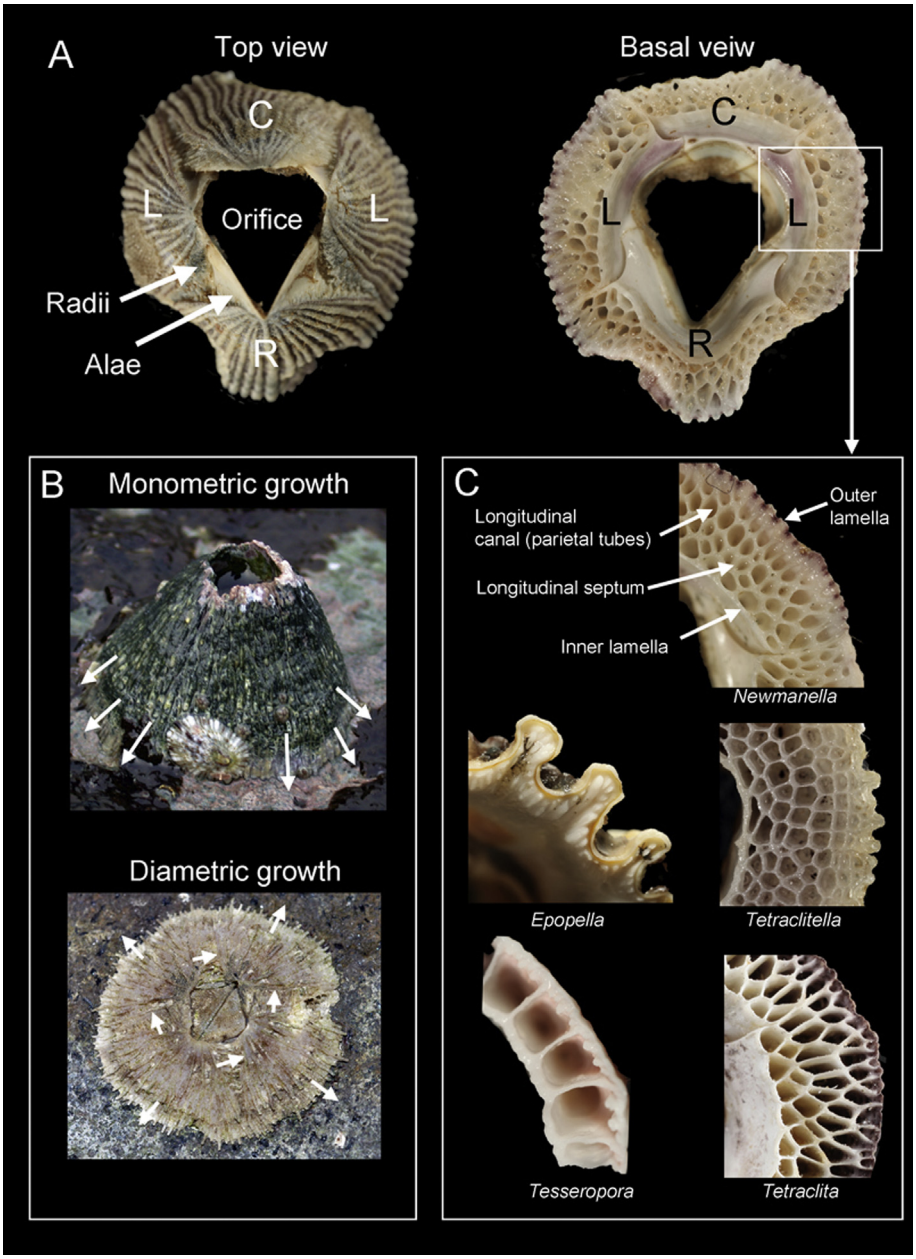

Fig. 1. Shell structure of Tetraclitid barnacles. (A) Top view and basal view of a *Newmanella* shell, showing that the shell is composed of 4 plates (Carina [C], Rostrum [R], and two Laterals [L]). (B) Two distinct growth patterns in Tetraclitid barnacles. Monometric growth involves accretion mainly at the basal areas of the shell and the orifice size increases by subsequent erosion in the natural environment (species shown, *Tetraclita squamosa*). Diametric growth involves growth occurring at the radii, alae (junctions of the shell plates) and basal region of the shell; such a growth pattern results in increase of the orifice size (species shown: *Tetraclitella karandae*). Arrows indicate the growth directions. (C) Variation in the shell structure amongst different genera in Tetraclitidae. *Newmanella*, *Tetraclitella*, and *Tetraclita* have multitubiferous shells. The shell of *Tesseropora* has a single row of tubes. The shell of *Epopella* lacks inner lamella; thus the shell is solid.