

Coralline algae of central New Zealand

An identification guide to common 'crustose' species

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Published by NIWA Wellington 2005

Edited and produced by Science Communication, NIWA, Private Bag 14901, Wellington, New Zealand

ISSN 1174-264X

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Citation:

Harvey, A.; Woelkerling, W.; Farr, T.; Neill, K.; Nelson, W. (2005). Coralline algae of central New Zealand: an identification guide to common 'crustose' species.

**NIWA Information Series No. 57. 145 p.

Cover: Non-geniculate coralline algae on subtidal rock wall. Photo by Sean Cooper, Department of Conservation.

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Preface

his project on non-geniculate coralline algae had its genesis more than a decade ago. In the early 1990s Bill Woelkerling and I discussed how we could begin to document New Zealand's non-geniculate flora and increase recognition of the importance of these species in coastal ecosystems. The gaps in our knowledge about the coralline algae of New Zealand had been obvious for many years but the technical difficulties involved in working with these calcified red algae, as well as the history of confused taxonomy and nomenclature for New Zealand species, meant that there were significant hurdles to be overcome.

The priority of this research work, which has been funded by the Ministry of Fisheries Biodiversity Programme (ZBD2001/05), has been to develop a reliable and durable reference, which can be used in fisheries and coastal management, as well as in marine resource protection and conservation. We wanted to

make information accessible to marine scientists and resource managers, and to improve understanding of these algae, both through the production of this guide and through improved reference collections lodged in the herbaria at Te Papa (WELT), Auckland Museum (AK), and Landcare Research (CHR).

The taxonomic research using morphological and anatomical characters which forms the basis of this guide has been complemented with data from molecular sequencing, carried out at the University of Otago by Dr Judy Broom and Darren Hart. While this work is not discussed in this guide, it forms a very important and exciting area of research making use of new tools and approaches to better understand species relationships and identity.

In treatments of coastal communities, even within recent publications, non-geniculate coralline algae

have been referred to as "pink paint", completely overlooking the diversity that may be present and the complex ecological roles particular species may play. We hope that the production of this guide and the underlying research that has been supported by the Ministry of Fisheries will cause people to look more closely, and to recognise and document coastal diversity including these key organisms, and that this will lead to a better understanding of the dynamic relationships between these algae and the coastal flora and fauna.

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Chapter 1. How to use this guide, and its limitations

dentifying non-geniculate corallines poses certain challenges, particularly for the non-specialist. Many species can vary considerably in growth form and external appearance, thereby making reliable sightrecognition of most species difficult or impossible. Two individuals belonging to the same species can look quite different depending on age, habitat, and other factors. Likewise, specimens of two species that are quite different taxonomically often cannot be told apart solely on the basis of growth form and external appearance. Thus, without further information, it generally is not possible to identify 17 of the 20 species included in this guide through immediate use of the keys (Chapters 8 & 10) or a quick scan of the species profiles (Chapter 12). Rather, coralline identification is based on vegetative and reproductive characters (detailed in Chapter 7), and use of the keys relies on observation of these important anatomical and morphological features.

ORGANISATION

The guide is divided into five major parts (A to E). These parts separate the various aspects of the guide and are intended to make it easier to use.

Part A – Using the guide

Part A (Chapter 1) outlines the organisation of the guide and the different ways of using it.

Part B - General information

Part B (Chapters 2 to 6) provides general information about the New Zealand coralline project (Chapter 2) and the nature and significance of non-geniculate

corallines in the marine environment (Chapter 3). Chapter 4 presents a brief summary of past work in New Zealand, and our current understanding of coralline classification is explained in Chapter 5. The collection and storage of specimens is dealt with in Chapter 6.

Part C - Coralline essentials

Part C (Chapter 7) covers basic coralline structure and reproduction.

Part D – Specimen identification

Part D (Chapters 8 to 11) deals with specimen identification, and details various laboratory procedures used to view important vegetative and reproductive characters. The guide allows for specimen identification to be achieved in two ways (using either 'simple' or 'more involved' methods), and is structured this way to permit people with a range of backgrounds and algal knowledge to use it.

Note that in some chapters – e.g., Chapters 9 and 11 – information from an earlier chapter is repeated, with the intention that each chapter serves as a complete, stand-alone reference for use in the lab or field.

Part E – Species profiles

Part E (Chapter 12) includes 'profiles' of the 20 species covered in this guide. These profiles include both field data (substrates, depth range, etc.) and detailed anatomical and taxonomic information for species found in central New Zealand during the present study.

HOW TO USE THIS GUIDE

1. Become familiar with coralline structure and reproduction

Coralline identification is based on vegetative and reproductive characters, and it is essential that users of the guide become familiar with coralline structure and reproduction (Chapter 7) before attempting identifications.

2. Identify specimens

Once the user is familiar with those vegetative and reproductive features, specimen identification can be achieved in two ways: 'With direct observation and/or simple lab procedures' (Chapters 8 and 9), or 'Using more involved lab procedures' (Chapters 10 and 11). The different chapters are intended for use by those with various degrees of taxonomic knowledge and access to specialised equipment.

The primary objective of this guide is to make coralline algal identification possible for a range of users, from professional phycologists to ecologists and non-professional naturalists interested in marine plants.

Chapters 8 and 9 are targeted at those with little algal knowledge and with access to basic lab facilities (i.e., slides, readily available chemicals, dissecting and compound microscopes). They contain a key for preliminary specimen identification (Chapter 8) and describe some simple lab procedures (e.g., the preparation of temporary slides containing whole mounts or squashes) that can be used to observe basic

features necessary for preliminary identification (Chapter 9).

In contrast, Chapters 10 and 11 are targeted at those with a more comprehensive knowledge of algae and with access to the necessary chemicals and equipment required for thin sectioning and more complete identification (e.g., L.R. White embedding resin, microtome for making micrometre-thick sections). Keys for definitive specimen identification are in Chapter 10, and the associated detailed lab procedures on how to embed and section material and make permanent slides are outlined in Chapter 11.

3. Go to species profiles

The species profiles (Chapter 12) provide detailed information on the 20 species found in central New Zealand during the present study. Identifications made using Chapters 8 or 10 can be supported (or rejected) by examining these profile figures.

ICONS ON PLATES

Three icons follow magnification data on the plates:

- depicts plants, etc., as seen with the naked eye
- depicts conceptacles, etc., as seen with a dissecting microscope or good hand lens
- depicts sections, whole mounts, etc., as seen with a compound microscope

When structures on photos have not been measured accurately, a magnification range is given, along with the appropriate icon.

GENERAL LIMITATIONS OF THE GUIDE

In using this guide, it is very important to remember its limitations.

- This guide is designed to help users identify specimens of non-geniculate coralline algae collected in central New Zealand. It is a prelude to monographic studies, but should not be considered a replacement or substitute.
- Most tetrasporangial (and bisporangial) specimens of the 20 species of non-geniculate corallines covered in this guide should be identifiable using the techniques and keys provided. Not all specimens, however, will be identifiable. Most sterile specimens (i.e., without reproductive structures) cannot be identified, and most male or female specimens cannot be fully identified in the absence of tetrasporangial (or bisporangial) individuals (see Figure 7.5). This limitation also applies to monographic studies.
- The sampling programme upon which this guide is based was extensive, but only a small fraction of the total central New Zealand coast was covered, and most sampling sites were visited only once. This means that other species occurring in the region may have escaped detection.
- This guide is designed and based on collections from central New Zealand. It has not been tested on specimens collected outside this region. If using the guide for specimens from other parts of New Zealand, keep in mind that such specimens

may not be identifiable because they represent species not included in the guide or not present in central New Zealand. Such specimens should **not** be discarded but rather should be deposited in a registered New Zealand herbarium with suitable notes (listed in item 14 on p. 28) so that they can be further assessed by specialists, particularly in the context of monographic studies.

- The characteristics of species given in the species profiles (Chapter 12) are based on specimens examined during the present study. New specimens may show greater variation in some characters (particularly measured characters such as conceptacle size) than has been recorded here.
- Published records for the entire New Zealand region for both geniculate and non-geniculate corallines have been summarised by Woelkerling & Nelson (2004), and are not repeated here. The accuracy of previously published records requires assessment in the context of monographic work, and experience in southern Australia (Woelkerling 1997) suggests that many of these previously published records are based on misidentifications or are otherwise untrustworthy.
- The names used for species in this guide have been based (with one exception) on the study of type material. Nevertheless, it is possible that older names for some of the species included here will be discovered during monographic studies, thus necessitating adoption of a different name.

Chapter 2. Introduction

ew Zealand shores are rich in coralline red algae. Most plants are easily recognised as corallines because they form calcified pinkish crusts or stony growths on a variety of substrates, including rocks, other algae, seagrasses, molluscs, and other animals. Throughout New Zealand, corallines are common and often conspicuous both in intertidal and subtidal habitats wherever suitable substrates occur. Some species can also grow unattached and can form extensive localised beds, made up of thousands of individuals, on the seafloor. Unattached plants are often called rhodoliths.

Some corallines have upright branches that consist of alternating calcified and uncalcified segments (Figure 7.2, A & B). Such corallines are said to be **geniculate**. Others lack branches or have branches that are entirely calcified and thus lack uncalcified joints or nodes (Figure 7.2, C–H). Such corallines are said to be **non-geniculate** and are commonly known as 'crustose' coralline algae. In this book, only non-geniculate (crustose) corallines are considered in detail.

There have been no monographic accounts of New Zealand coralline red algae. Eleven regional New

Zealand species lists have been produced since 1972 (see Woelkerling & Nelson 2004) but these lack keys. The floristic account of Chapman & Parkinson (1974), unfortunately, contained little original research (Parsons 1985), and the included keys are unreliable and essentially useless (Woelkerling & Nelson 2004). Thus the identification of New Zealand specimens has proven difficult.

The present guide was developed to provide a means of identifying non-geniculate corallines growing in the central New Zealand region (Figure 2.1) in a more modern context. It is not a replacement for a much

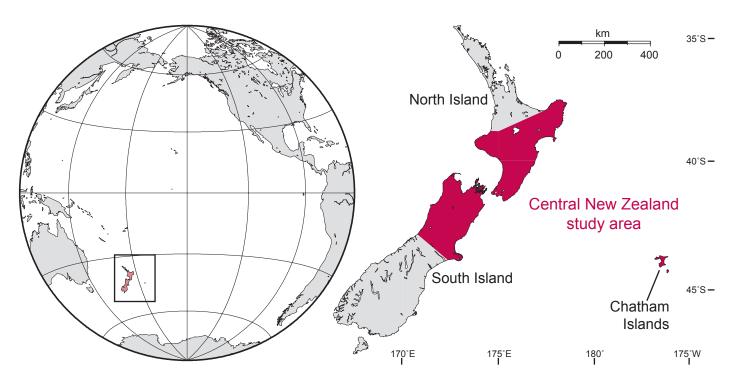


Figure 2.1: Map of the Pacific region, and the central New Zealand study area (shaded in red).

needed monographic account, but rather an interim mechanism for identifying species of coralline red algae that were encountered in the region over a 30-month period during 2002–04.

STUDY AREA AND SOURCES

In order to achieve progress as efficiently and as effectively as possible, the project focused on the central New Zealand region where about 40% of the total macroalgal flora is represented.

The guide is based almost entirely on new collections obtained from 87 'localities' (Figure 2.2), 73 of which lie between North Taranaki and Hokitika on the west coast of the North and South Islands respectively, and between East Cape and Banks Peninsula on the east coast. The biogeographically related Chatham Islands have been included in this 'central New Zealand' region. A few collections from an additional 14 localities outside the central New Zealand study area have also been included (see Appendix 1).

Twenty known species were found in the collections and are included in this guide. All species and the collection localities they were recorded from appear in Table A1 (Appendix 1).

While a number of collections examined during the present study are the subject of further taxonomic investigation, over 92% of useable collections fall within the 20 species included in this guide.

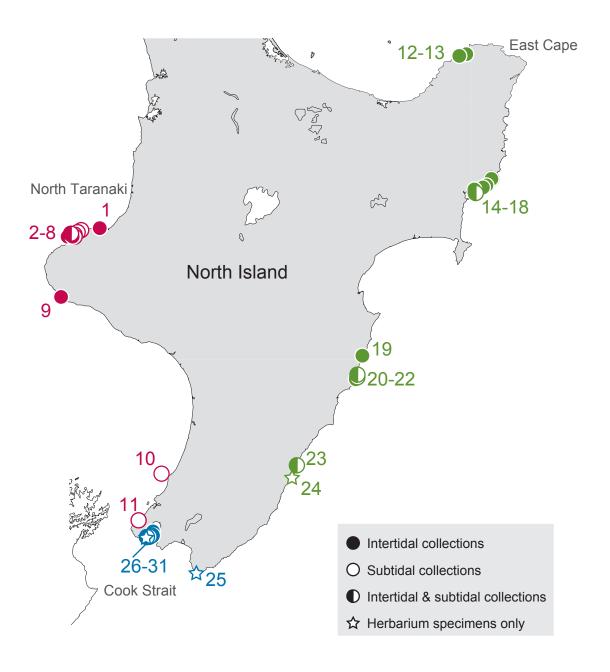
Names applied to the collections were determined by microscopic examination of vegetative and reproductive features using permanent slides of sectioned material.

Most of the identified, newly collected material (including permanent slides) has been deposited in the herbarium of the Museum of New Zealand Te Papa Tongarewa (WELT), with some specimens donated to the herbaria at Auckland Museum (AK) and Landcare Research (CHR). Identified

material loaned to WELT from other New Zealand herbaria has been returned (with permanent slides) to the herbarium of origin. Herbarium numbers of specimens used in the photos (except unidentified or non-New Zealand material) are listed in Table A2 (Appendix 2).

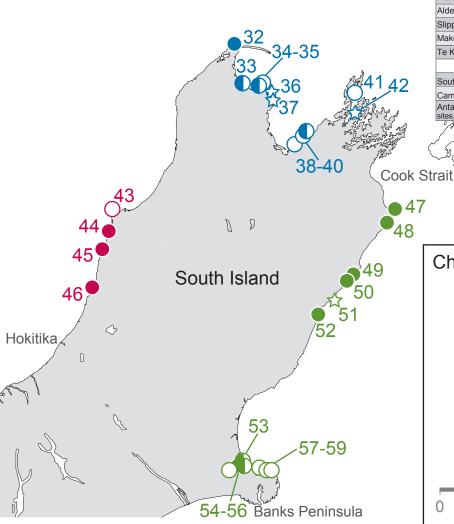
With the exception of *Lithophyllum carpophylli*, the type collections of all species included in this guide have been examined by the authors or by other investigators during previous studies, and recent papers containing accounts of the relevant species are listed in the profiles in Chapter 12. Examination of and comparison with type material provides assurance that the names are correctly applied. A lectotype has yet to be designated for *L. carpophylli*, a species with a distinctive appearance not reported outside the New Zealand region.

orth Islan	d, west co	past		
1		Parahaki Stream Reef	38°59'S	174°18'E
2		Radio Mast Boulders	39°00'S	174°07'E
3		Waiwhakaiho	39°01'S	174°05'E
4		Sugar Loaf Islands (2 collection sites)	39°02'S	174°00'E
5		Kawaroa Reef	39°02'S	174°03'E
6		Hapuka Rock	39°03'S	173°59'E
7		Belt Road Reef	39°03'S	174°03'E
8		Port Taranaki	39°03'S	174°02'E
9		Puketapu Road	39°31'S	173°55'E
10		Kapiti Island (3 collection sites)	40°52'S	174°55'E
11		Makara (4 collection sites)	41°13'S	174°42'E
orth Islan	d, east co	ast		
12		Te Rangiharu Bay	37°37'S	177°57'E
13		Raukokore	37°38'S	177°53'E
14		Te Tapuwae O Rongokako	38°36'S	178°12'E
15		Tatapouri	38°39'S	178°09'E
16		Makorori (2 collection sites)	38°40'S	178°07'E
17		Kaiti Beach	38°41'S	178°02'E
18		Tuamotu Island (2 collection sites)	38°42'S	178°03'E
19		Mangakuri Beach	39°58'S	176°55'E
20		Tuingara Point (3 collection sites)	40°07'S	176°52'E
21		Pourerere	40°08'S	176°52'E
22		Aramoana (2 collection sites)	40°09'S	176°51'E
23		Mataikona (4 collection sites)	40°48'S	176°16'E
24	☆	Castlepoint	40°54'S	176°13'E
orth Islan	d, Cook S	strait		
25	☆	Cape Palliser	41°37'S	175°16'E
26		Scorching Bay	41°18'S	174°50'E
27		Breaker Bay	41°20'S	174°50'E
28		Palmer Head (6 collection sites)	41°20'S	174°49'E
29		Island Bay (12 collection sites)	41°21'S	174°46'E
30		Princess Bay	41°21'S	174°47'E
31	₹	Owhiro Bay	41°21'S	174°46'E

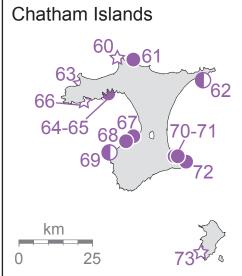


12

outh Islan	nd, Cook	Strait				
32		Wharariki Beach (2 collection sites)	40°30'S	172°41'E		
33		Patons Rock (2 collection sites)	40°47'S	172°46'E		
34		Wainui (2 collection sites)	40°48'S	172°56'E		
35		Golden Bay (4 collection sites)	40°47'S	172°58'E		
36	₩	Abel Head	40°51'S	173°03'E		
37	₩	Pinnacle I	40°55'S	173°04'E		
38	Ô	Nelson Haven	41°15'S	173°17'E		
39		Boulder Bank	41°11'S	173°22'E		
40		Cable Bay (4 collection sites)	41°09'S	3 173°24'E		
41	O	D'Urville Island (4 collection sites)	40°51'S	173°53'E		
42	₩	Maud Island	41°01'S	173°53'E		
outh Islan	nd, west c	coast				
43		Steeples (4 collection sites)	41°44'S	171°28'E		
44		Charleston	41°54'S	171°26'E		
45		Seal Rock Island	42°02'S	171°22'E		
46		Greigs	42°19'S	171°16'E		
outh Islar	nd, east c	oast				
47		Cape Campbell	41°44'S	174°17'E		
48		Chancet Rocks	41°50'S	174°12'E		
49		Maungamanu	42°13'S	173°52'E		
50		Rakautara (4 collection sites)	42°16'S	173°48'E		
51	☆	Kaikoura (3 collection sites)	42°25'S	173°41'E		
52		Oaro Reef	42°31'S	173°31'E		
53		Sumner (4 collection sites)	43°35'S	172°46'E		
54		Macintosh Bay (2 collection sites)	43°39'S	172°38'E		
55	0	Diamond Harbour	43°37'S	172°43'E		
56		Camp Bay	43°37'S	172°47'E		
57		Pigeon Bay	43°38'S	172°56'E		
58		Decanter Bay	43°39'S	173°00'E		
59		Raupo Bay	43°39'S	173°03'E		
hatham Is	slands					
60	₩	Cape Young	43°42'S	176°38'W		
61		Wharekauri	43°42'S	176°34'W		
62		Okawa Point (2 collection sites)	43°46'S	176°15'W		
63	☆	Waitangi West	43°47'S	176°49'W		
64 Whangatete Inlet 43°48'S						
65		43°49'S	176°42'W			
66 Cocean Beach 43°50'S 176						
67 Waitangi 43°57'S 176°34						
Heaphy Shoal (2 collection sites) 43°58'S 176°36"						
Point Durham (3 collection sites) 44°00'S 176°41'W						
70		Te One Creek	44°01'S	176°23'W		
71		Owenga Wharf	44°01'S	176°18'W		
72		Tommy Solomon (2 collection sites)	44°02'S	176°20'W		
73	57	Pitt Island (2 collection sites)	44°20'S	176°16'W		



North Island, north of study area							
Russell (2 collection sites)	35°16'S	174°07'E					
Poor Knights Is.	35°29'S	174°44'E					
Little Barrier I.	36°12'S	175°05'E					
Great Barrier I.	36°08'S	175°25'E					
Mangere I.	n/a	n/a					
Waiheke Channel	36°50'S	175°10'E					
Army Bay	36°36'S	174°49'E					
Little Manly	36°38'S	174°46'E					
Aldermen Is.	36°58'S	176°05'E					
Slipper Island	37°03'S	175°54'E					
Maketu	37°45'S	176°28'E					
Te Kaha	37°44'S	177°41'E					
South of study area							
Campbell Island	53°S	169°E					
Antarctica (5 collection sites, Ross Sea)	_	-					
°/20							



Chapter 3. Introducing coralline red algae

oralline red algae, or 'corallines', are members of the Rhodophyta (red algae) and are distinguished from other members of the phylum by the presence of cell walls impregnated with calcium carbonate (as calcite).

WHERE CORALLINES ARE FOUND

Corallines are distributed worldwide and are abundant in a number of diverse marine habitats, including rocky shores, seagrass meadows, tropical reefs, and rhodolith beds (or maerl).

They can be found growing in a range of temperatures, from the cold waters of the Arctic and Antarctic, through to the tropics. They also have a considerable depth range, occurring from the intertidal zone down to 270 m (Littler et al. 1985), and are the deepest known macroscopic plant life.

SIGNIFICANCE OF CORALLINES IN THE MARINE ENVIRONMENT

Corallines are important components of marine environments. The following are examples of just some of the ways in which non-geniculate coralline algae interact with, support, and provide habitat for animals and other algae in the marine environment.

The ecological role of corallines is both varied and significant, and detailed research into their ecology, physiology, biogeography, and conservation is dependent on the ability to identify specimens to species.

Corallines provide habitat, refuge, and grazing areas for numerous fish and invertebrates

Despite its apparent lack of appeal as a food source, a study in Western Australia (Jernakoff et al. 1993) found that the two major components in the diet of very young Western rock lobsters (juveniles within their first year after settlement) were coralline algae and molluscs. While the proportions of coralline algae and molluscs differed depending on the moult stage (premoult, intermoult, or postmoult), as much as 80% of the food in the foregut of postmoult lobsters was coralline algae.

A mutually beneficial relationship between a branching coralline and a crab occurs in seagrass beds off the coast of Florida (Stachowicz & Hay 1996). In this relationship the coralline alga *Neogoniolithon strictum* provides refuge to an herbivorous crab. Tethering experiments showed that fish rapidly ate any crabs left in the open, but crabs tethered close to an algal host were likely to survive. In turn, crabs are beneficial to the corallines as they eat epiphytic/fouling algae and apparently keep the surface of the coralline clean. After removal of the crabs the corallines were quickly overgrown by epiphytic seaweeds, and appeared pale in colour.

Corallines are important components of tropical reefs

Coralline algae are important and common components of tropical coral reefs worldwide. At Waikiki reef in Hawaii, non-geniculate coralline algae were found to cover 39% of the fringing reef (Littler 1973), and in southwest Japan, mean total cover was 33% (Iryu & Matsuda 1988). Overall cover on the reefs can be affected by sedimentation, with mean cover on the Great Barrier Reef highest (over 20%) on the clearer outer reefs, reducing to less than 1% in more sediment-affected environments (Fabricius & De'ath 2001).

Coral reefs are structured by wave action (Adey 1998) and depend on coralline algae to reinforce and maintain wave-resistant fronts. The calcium-hardened corallines fortify the reef by growing over the coral framework, creating a durable substrate that absorbs wave energy and helps reduce reef erosion (Björk et al. 1995, Littler & Littler 1995). Moreover, corallines are often the dominant components in areas of high wave action, such as the reef crest, where the more fragile corals get damaged (Adey 1998).

Corallines act as settlement inducers for marine invertebrates (including paua/ abalone, corals, and kina/sea urchins)

Paua (abalone, *Haliotis* spp.) larvae have cilia that enable them to swim through the water column. Seven days after fertilisation, however, the development of these motile planktonic larvae is arrested and they must find a suitable substrate to settle on before they can metamorphose into the sessile (attached) form (see Figures 3.1 and 3.2). This metamorphosis is controlled by a chemical associated with the surface of non-geniculate coralline red algae. The chemical is a small peptide that activates the larval nervous system, triggering the arrested larval stage to develop into the adult (Morse 1991). The 'morphogenic inducer' recognised by red paua (*Haliotis rufescens*) larvae

is a GABA-mimetic oligopeptide (Morse & Morse 1996). Twelve other species of abalone also show this dependence on coralline algae and a GABA-like inducer (Morse 1991).

Larvae of the Caribbean stony/hard coral *Agaricia humilis* are also motile and, like paua larvae, settle onto coralline algae before metamorphosis. Studies of this specific inducer have shown that it is part of the calcified cell wall of the corallines, but is a complex sulphated polysaccharide, and thus different from the inducer recognised by paua larvae (Morse & Morse 1991, 1996). A soft coral from Guam has also been shown to preferentially settle and produce polyps on coralline algae (Slattery et al. 1999).

These settlement responses can be specific, influencing the dispersion and recruitment of coral species and consequently the ecology and structure of coral reefs. Harrington et al. (2004) recently examined the substrate preferences of two Great Barrier Reef corals (Acropora tenuis and A. millepora). Given a choice of five coralline algae, coral larvae showed a preference for Lithophyllum prototypum (as Titanoderma) and were 15 times more likely to settle on this species than on the least favoured species, Neogoniolithon fosliei. The abundance and location of particular coralline species on tropical reefs, combined with this ranking process, will influence where some coral larvae settle and ultimately survive, and thus may be partly responsible for the fine-scale structure of coral reefs (see also Raimondi & Morse 2000).



Figure 3.1: Paua larva (arrow) just prior to settlement.

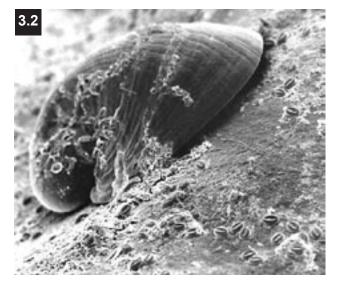


Figure 3.2: Paua larva days after settlement on the coralline alga *Sporolithon durum* (photo courtesy of Dr Sabine Daume, Department of Fisheries, Western Australia).

Corallines are the dominant component of rhodolith beds

Rhodolith beds (or maerl) are made up of numerous free-living (unattached) non-geniculate coralline algae (see Figure 3.3). These beds are common in the world's oceans, occurring in tropical waters through to polar regions, at a variety of depths (down to 130 m), and on a variety of substrates (fine mud to coarse pebbles) (Birkett et al. 1998) (see also Foster 2001).

Typically, rhodolith beds are found in protected habitats with strong currents, such as bays, inlets and sounds, where they are sheltered from the destructive forces of waves and where currents prevent them from being buried by sediment. The depth to which rhodoliths grow is largely related to water quality, with deeper beds occurring in clearer waters.

The rhodoliths are three-dimensional structures that provide an often intricate and stable habitat for other algae and invertebrates. As a result, a single bed can harbour an array of associated macroalgal and faunal species that includes scallops, brittle stars, sea anemones, and sea urchins (kina). Although most of these associated species also occur in other habitats, some are restricted to, or rarely found outside, rhodolith beds (Birkett et al. 1998)



Figure 3.3: Rhodoliths collected in the Marlborough Sounds (South I., NZ) (Mag. 0.6x).

Potential medical applications of coralline algae

Research into induction and settlement of paua/ abalone and coral larvae has revealed two potential medical applications of coralline algae.

The GABA inducer of abalone metamorphosis may prove useful for investigating human brain disorders. Preliminary research has found that this GABA oligopeptide is structurally similar to the neurotransmitter (gamma-aminobutyric acid) in the human brain and binds to GABA receptors in mammalian brains. This strong binding effect may ultimately lead to the development of new therapies for the treatment of GABA receptor disorders, such as epilepsy and depression (Morse 1991, Morse & Morse 1996).

The inducer recognised by *Agaricia* coral larvae may also ultimately help people battle immunodeficiency diseases or resist viral infections. Tissue culture experiments have shown that this polysaccharide inducer stimulates mammalian lymphocytes to divide and multiply, and this may prove useful in the development of new drugs that help stimulate the human immune system (Morse & Morse 1996).

Chapter 4. Coralline biodiversity in the greater New Zealand region

ost users of this guide will be concerned primarily with identifying specimens of nongeniculate species collected mainly in central New Zealand. The documented occurrence in central New Zealand of the 20 species covered here also represents a step towards elucidating the taxonomic biodiversity of coralline red algae for the greater New Zealand region.

The greater New Zealand region, as explained by Woelkerling & Nelson (2004), comprises 'mainland' New Zealand (including North Island, South Island, and Stewart Island), and eight offshore groups of islands in New Zealand territorial waters. These groups are the temperate or subtropical Chatham Islands, Kermadec Islands, and Three Kings Islands; and the subantarctic Antipodes Islands, Auckland Islands, Bounty Islands, Campbell Island, and Snares Islands.

The true taxonomic biodiversity of coralline red algae in the greater New Zealand region has yet to be rigorously determined. This determination is dependent on data obtained from new, intensive field work, a detailed assessment of past published records and of the herbarium specimens upon which these are based, the study of relevant type collections, and an understanding of species limits from both a morphological and a molecular perspective. As a consequence, fully determining the taxonomic biodiversity of coralline red algae for the greater New Zealand region represents a longer-term challenge. The question of where matters currently stand in

relation to this challenge is briefly considered in this chapter.

PAST RECORDS

Between 1821 and 2003, records of coralline red algae for the greater New Zealand region have appeared in over 300 scientific publications. A baseline summary and analysis of these records was made by Woelkerling & Nelson (2004), and the comments to follow are based on or derived from data in that publication.

Woelkerling & Nelson (2004) documented published records for 80 species and infraspecific taxa (formally described varieties and forms of species) for the greater New Zealand region: of the 80 species and infraspecific taxa, 34 are geniculate corallines and 46 are non-geniculate corallines. In addition, they provided information for each coralline on its basionym (i.e., the original name under which the coralline was described), the type locality, the type specimen and its location, published illustrations of type material, and comments, including nomenclatural notes.

Woelkerling & Nelson (2004) found that the records contained many uncertainties, and warned that it would be highly misleading to assume that they gave an accurate picture of true species biodiversity of coralline red algae in the greater New Zealand region. This warning stemmed in part from results of an analysis of coralline records in the southern Australian region (Woelkerling 1997).

Woelkerling (1997) compared southern Australian species databases generated before and after monographic studies, and found that 70% of the species records in the pre-monographic database were spurious, or involved taxa of uncertain status, or involved taxa that were synonyms of other taxa. He also found that half of the species included in the post-monographic database were not represented in the pre-monographic database. There have been no monographic studies of New Zealand region corallines, and thus any impression of species biodiversity suggested in the greater New Zealand database generated by Woelkerling & Nelson (2004) is highly likely to undergo substantial change as more modern and meaningful information becomes available.

TYPE SPECIMEN STUDIES

The scientific naming of species is governed by a set of rules known as the International Code of Botanical Nomenclature. The rules are revised at Nomenclature Section meetings at successive International Botanical Congresses. The revision edited by Greuter et al. (2000) was current in February 2005.

Whenever a new species or infraspecific taxon is described, the International Code of Botanical Nomenclature requires that a type specimen be designated so that there is a permanent reference point upon which the future application of the scientific name can be based. Each species or infraspecific taxon can have only one correct scientific name, and

the correct one is the oldest name that is in accord with the International Code of Botanical Nomenclature.

Because the type specimen is the reference point for the application of a scientific name, it is important to have modern knowledge of the morphology, anatomy, and other aspects of each type specimen so that the correct application of the scientific name occurs when accounts or descriptions of species are published and when keys are produced for specimen identification.

Accounts of 20 species of non-geniculate corallines found in central New Zealand are provided in this guide. Both the morphology and anatomy of the types of all but one (*Lithophyllum carpophylli*) of these have been studied in detail in a modern context, and thus the names for these 19 taxa can be applied with confidence. Central New Zealand specimens referred to *Lithophyllum carpophylli* agree morphologically with the only known type material, but the internal anatomy of that type material has not been studied in a modern context.

In the context of the greater New Zealand region, however, the situation is somewhat different. Of the 80 species and infraspecific taxa recorded, 29 are based on type specimens collected in the greater New Zealand region. Woelkerling & Nelson (2004) found, however, that the status of 20 of these taxa is uncertain because the types have not been reexamined in a modern context. This means that subsequent published records for these names also are uncertain and will require verification once the types have been more fully studied.

The type specimens of the remaining 51 species come from outside the New Zealand region, and 16 of these types also have yet to be studied in a modern context. This means that New Zealand records for these species also are surrounded by uncertainty.

To summarise, modern morphological and anatomical knowledge of type specimens is important to the correct and consistent application of scientific names to specimens and in publication. Such knowledge is lacking for a number of species and infraspecific taxa recorded from the greater New Zealand region, so many published records require verification once detailed studies of relevant types become available.

SPECIES LIMITS

In coralline red algae, as in other organisms, all species in a given genus have, theoretically, certain morphological and anatomical features in common. These common features form the basis for placement of these species in one genus. Within a genus, however, each species also has a unique set of morphological/anatomical features that allows it to be consistently differentiated from other species belonging to the same genus. Morphological/anatomical features that group species into genera, or that differentiate species from one another within a genus, are controlled by differences in gene sequences.

Our present understanding of species limits in coralline red algae is very uneven and falls far short of the theoretical ideal. We have no idea which genes control those differences in morphological and anatomical characters thought to separate species. Work is ongoing to establish the utility of molecular analyses in coralline algal taxonomy. In the meantime, our perception of species limits within genera of corallines is based entirely on morphological and anatomical information.

Many described species of coralline red algae are poorly understood. Their types have not been studied in a modern context, and often such species are known from only one or a very few collections. It is not possible in such cases to assess how much variability might occur in morphological and anatomical features that potentially could be used to separate poorly known species from one another or from better known species. A number of species reported to occur in the greater New Zealand region (see Woelkerling & Nelson 2004) fall into this category.

This situation for the central New Zealand species included in this guide is somewhat better but by no means ideal. Species such as *Choreonema thuretii*, *Pneophyllum coronatum*, and *Sporolithon durum* appear to be readily recognisable on morphological-anatomical grounds and provide no problems in terms of specimen identification.

On the other hand, the presumed morphologicalanatomical boundaries between *Lithophyllum corallinae* and *L. stictaeforme* are not always clearcut, and apparent intermediates occur in both central New Zealand and southern Australian collections. Further morphological-anatomical and molecular studies are required to determine whether one species or two are involved, and whether there are any other morphological-anatomical characters that might be diagnostic at species level.

Problems are sometimes encountered in specimen identification of *Mesophyllum engelhartii* and *Synarthrophyton patena*. The two genera appear distinct both on morphological/anatomical grounds and on molecular grounds, but to confidently differentiate between the two species, both tetrasporangial and male plants are required. Tetrasporangial plants of the two species (which are far more common than male plants) overlap in growth form and have tetrasporangial conceptacles with very similar anatomy. In the absence of male plants or

molecular data, it is usually not possible to tell which species tetrasporangial specimens belong to.

THE CHALLENGE AHEAD

To gain a better understanding of coralline red algal biodiversity in the greater New Zealand region requires considerable research.

The present study has focused on the analysis of over 1000 collections, mostly newly obtained, from central New Zealand. This approach needs to be extended to remaining parts of mainland New Zealand and to the offshore island groups.

In addition, the types of many taxa reported from the region need to be studied in a modern context, not only to determine how scientific names should apply but also to determine whether these names should be applied to New Zealand material and whether past use of names for New Zealand species is correct or not.

Once species occurrence throughout the New Zealand region is better understood, monographic studies of genera and families involving both morphological-anatomical data and molecular data can be completed to provide a more definitive and reliable resource for specimen identification than is possible with this guide.

Chapter 5. Coming to grips with coralline taxonomy

The taxonomy and classification of coralline red algae has had a long, interesting history. Before the time of Linnaeus, coralline red algae (and coral animals) were generally considered to be plants. Linnaeus, however, became convinced that coralline red algae were animals, and between the mid 1700s and the mid 1800s an ongoing and at times bitter controversy raged over the plant vs. animal nature of corallines. The issue was finally settled when the distinctly plant-like reproductive structures of coralline red algae were first studied in detail about 1840.

Between the 1840s and 1980s the coralline red algae were almost always treated as a distinct family of red algae, the Corallinaceae. As knowledge of corallines and other red algae increased, however, it became apparent that the corallines were better treated as a distinct order, the Corallinales. This treatment has subsequently been consistently supported by molecular studies.

At first, the Corallinales included only one family, the Corallinaceae, but in 1993 a second family, the

Sporolithaceae, was recognised and in 2003, a third family, the Hapalidiaceae, was recognised. This expansion from one family to three is supported by molecular studies and morphological data (see Harvey et al. 2003a).

At the present time (see Harvey et al. 2003a), the Corallinaceae is divided into four subfamilies (Corallinoideae, Lithophylloideae, Metagoniolithoideae, and Mastophoroideae) (Figure 5.1), two of which (the Lithophylloideae and Mastophoroideae) include non-geniculate genera. The Hapalidiaceae is divided into three subfamilies (the Austrolithoideae, Choreonematoideae, and Melobesioideae), all of which are non-geniculate. The Sporolithaceae is not divided into subfamilies and contains only non-geniculate corallines.

Harvey et al. (2003a) summarised the diagnostic characters of the families and subfamilies and list currently recognised genera. Taxonomy is a dynamic science, and changes in classification will continue to emerge as new data (both morphological and molecular) become available. Indeed, six months

after publication of the summary by Harvey et al. (2003a), a new genus of Austrolithoideae, *Epulo*, was discovered in eastern Australia (Townsend & Huisman 2004).

During the present study, non-geniculate corallines belonging to all three families of Corallinales were found in central New Zealand. These include members of the Lithophylloideae, Mastophoroideae, Choreonematoideae, Melobesioideae, and both known living (non-fossil) genera of Sporolithaceae.

The characters used to separate the families and subfamilies and genera of central New Zealand nongeniculate Corallinales are summarised in Tables 5.1 and 5.2. Some of these characters are observable using the simple lab procedures outlined in Chapter 9, but others can only be seen after material is embedded and sectioned and examined with a compound microscope (see Chapter 11). Tables 5.1 and 5.2 are intended as a guide to the classification of central New Zealand corallines and should not be used for specimen identification.

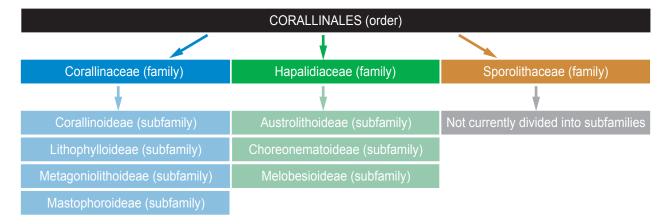


Figure 5.1: The order Corallinales and its families and subfamilies.

Table 5.1: Diagnostic characters of the families and subfamilies of Corallinales in this guide.

Corallinaceae		Hapalidiaceae		Sporolithaceae		
Tetrasporangia borne in co are uniporate.	onceptacles that	Tetrasporangia borne in c with a multiporate plate or	conceptacles roof.	Tetrasporangia borne in calcified compartments.		
are uniporate. Tetrasporangia with zon tetraspores.	ately arranged	Tetrasporangia with zonate tetraspores.	ely arranged	Tetrasporangia with cruciately arranged tetraspores.		
Apical plugs absent.		Apical plugs present.		Apical plugs present.		
Lithophylloideae		Choreonematoideae		Not currently divided into subfamilies		
Cells of adjacent filament by secondary pit connect	ions.	Conceptacles with an acel plate or roof ¹ . Cells of adjacent filaments	not linked by cell	Cells of adjacent filaments linked by either cell fusions or secondary pit connections or both.		
Lithophyllum carpophylli	Figure 12.1	fusions or secondary pit co		Heydrichia homalopasta Figure 12.19		
Lithophyllum corallinae	Figure 12.2	Choreonema thuretii	Figure 12.10	Sporolithon durum Figure 12.20		
Lithophyllum johansenii	Figure 12.3					
Lithophyllum pustulatum	Figure 12.4	Melobesioideae				
Lithophyllum stictaeforme Air Barbara Mastophoroideae Mastophoroideae	Figure 12.5	Conceptacles with a mul plate or roof. Cells of adjacent filaments cell fusions.	·			
Mastophoroideae		Melobesia membranacea	Figure 12.11			
Cells of adjacent filaments by cell fusions.	s linked principally	Mesophyllum engelhartii	Figure 12.12			
Hydrolithon improcerum	Figure 12.6	Mesophyllum erubescens	Figure 12.13			
Pneophyllum coronatum	Figure 12.7	Mesophyllum macroblastum	Figure 12.14			
Pneophyllum fragile	Figure 12.8	Mesophyllum printzianum	Figure 12.15			
Spongites yendoi	Figure 12.9	Phymatolithon repandum	Figure 12.16			
		Synarthrophyton patena	Figure 12.17	¹ Conceptacles look uniporate but have a hidden		
		Synarthrophyton schielianum	Figure 12.18	acellular multiporate plate beneath a single outer opening.		

Family and subfamily classification follows Harvey et al. (2003a) (see also Harvey et al. 2003b).

This table includes only those families and subfamilies of living Corallinales containing non-geniculate species and confirmed to occur in central New Zealand during the present study.

Table 5.2: Diagnostic characters separating genera of Corallinales in this guide.

	Vegetative thallus entirely endophytic	Connections between cells of adjacent filaments	Subepithallial initials: length relative to subtending cells	Thallus construction	Orientation of cells bordering tetrasporangial conceptacle pore canals	Tetrasporangial conceptacle roof formed by filaments	Spermatangial filaments	Involucre surrounding calcified compartments	
Corallinaceae (tetr	asporangia in u	niporate conceptad	cles)						
Lithophylloideae Lithophyllum		2º pits			varies	uncertain			
Mastophoroideae Hydrolithon		cell fusions			more or less perpendicu- lar to roof surface (vertical orientation)	peripheral & interspersed amongst sporangial initials			
Mastophoroideae Pneophyllum		cell fusions			more or less parallel to roof surface (horizontal orientation)	peripheral & interspersed amongst sporangial initials			
Mastophoroideae Spongites		cell fusions			more or less parallel to roof surface (horizontal orientation)	peripheral to the sporangial initials			
Hapalidiaceae (tet	rasporangia in r	nultiporate concept	tacles)						
Choreonematoideae Choreonema	yes	no connections		unconsolidated filaments			unbranched		
Melobesioideae Melobesia	no	cell fusions	uncertain (Harvey et al. 2003b, p.659)	dimerous			unbranched		
Melobesioideae Mesophyllum	no	cell fusions	as long or longer	monomerous			unbranched		
Melobesioideae Phymatolithon	no	cell fusions	as short or shorter	monomerous			both branched & unbranched		
Melobesioideae Synarthrophyton	no	cell fusions	as long or longer	monomerous			both branched & unbranched		
Sporolithaceae (te	Sporolithaceae (tetrasporangia in calcified compartments)								
Heydrichia		2º pits/cell fusions					branched or unbranched	present	
Sporolithon		2º pits/cell fusions					branched	absent	

Classification at family and subfamily level follows Harvey et al. (2003a). Generic classification follows Woelkerling (1996d, 1996e) for the Corallinaceae; Harvey et al. (2003b) for the Hapalidiaceae and Harvey et al. (2002) for the Sporolithaceae.

This table includes only those non-geniculate genera of the Corallinales confirmed to occur in central New Zealand during the present study.

Chapter 6. How to collect and store corallines

on-geniculate coralline algae can grow firmly attached to rocks, algae, or animals. As a result their collection and storage can be more difficult and somewhat different from that of other macroscopic algae.

This chapter outlines how to collect and store non-geniculate ('crustose') coralline red algae as permanent voucher material to ensure the ongoing value of specimens for scientific study.

CORALLINES IN SITU

Non-geniculate corallines occur in nearly all marine habitats where a solid substrate is present (e.g., rock, animals, seagrasses, or other macroscopic algae) (Table 6.1). At any given locality, they can be the dominant or sole macroscopic plant life present (covering up to 100% of the available substrate), or they can occur intermixed with other algae. Attached plants may be:

epilithic: growing on rocky reefs, boulders, or smaller stones (Figure 7.2, D).

epiphytic: growing on marine algae, including other corallines or on seagrasses (Figure 7.2, E).

epizoic: growing on molluscs, sponges, sea squirts, kina (sea urchins), corals, and some other animals (Figure 7.2, F).

epigenous: growing on glass, plastic, metal, and other refuse (Figure 7.2, G)

In both rocky and non-rocky habitats, **rhodoliths** can also be found forming populations and communities of free-living individuals (Figure 7.2, H).

A few species also occur as **parasites** or **semi-endophytes** (Figure 7.2, C) on other geniculate (Figure 7.2, A–C) or non-geniculate coralline algae. Most are very small and largely devoid of **pigment**.

Regardless of habitat or substrate, species can occur in **monospecific stands** or in **mixtures** of two or more species. Mixtures are most common amongst smaller species growing on algae, seagrasses, or mollusc shells.

Fully mature individuals of some species may be measured in millimetres: in other species individuals may be over 1 metre in size. As a result, (for coralline algae) a single 'collection' of a small species may contain thousands of individuals growing on a host alga or stone, while for some larger species, only part of a single individual may make up a collection. (This is an important difference from other algae, where a single plant often becomes a single 'sample'.)

Table 6.1: Habitats and common substrates.

Habitats	Common substrates
Intertidal reef tops and rocky surfaces Subtidal reefs and rocky surfaces Edges of reef platforms Boulder surfaces in cobble habitats Reef caves Reef pools Beneath rocky overhangs Sand or soft bottom areas may also harbour rhodoliths	Rock (including loose-lying pebbles and stones) Shells (live and dead molluscs) Other animals (e.g., kina/sea urchins, sponges) Geniculate corallines (may harbour epiphytic or parasitic species) Other non-geniculate coralline algae (may also harbour epiphytic or parasitic species) Holdfasts of larger algae (particularly brown algae) Other green, brown, or red algae (most commonly, the coralline epiphytes are smaller encrusting forms) Seagrasses Seabed refuse (bottles, plastic, etc.)

CORALLINE COLLECTING

General guidelines

- Try to obtain samples from all habitats (Table 6.1) at a sampling site. Habitat information should be recorded as soon as possible at or after collection.
- Major habitats often contain microhabitats or niches (e.g., higher light and lower light niches or different wave actions). All niches should be sampled within a habitat.
- Within each macro- and micro-habitat, try to obtain samples from all available substrates harbouring corallines (e.g., each species of host alga or mollusc shell represents a different substrate).
- If the substrate is a stone, another plant or animal, or a small object, include part or all of the substrate as part of the collection. This helps to keep the coralline plants intact and makes substrate identification easier.
- Stones or shells containing one apparent growthform are preferable to stones containing obvious mixtures of corallines. Each substrate (rock or shell) should have at least 50% or more cover of a single dominant plant.
- Whenever possible, collect several individuals of one apparent growth form. This will maximise the chances of obtaining female, male, and sporangial individuals. Avoid, however, collecting large numbers of one growth form to the exclusion of others.
- On each different substrate, collect samples of each different or potentially different growth form that may be present.

- In general
 - All individuals that superficially look the same (i.e., same colour and growth form) and occur on the same substrate should be grouped together as a single 'collection'.
 - Individuals that look the same but occur on different substrates (e.g., two host species of algae; on rock vs on glass) should be treated as separate collections.
- Similarly, individuals that clearly differ in growth form should be treated as separate collections unless there is an obvious gradient from growth form to growth form amongst the individuals in the sample.
- It is best to concentrate on collecting those corallines growing on rock first, then those on algae, then those on shell, etc. (i.e., develop a methodical manner).

Collecting equipment (other than dive gear)

Catch bag

Plastic bags: 'zip-lock', or bags with built-in wire ties ('Whirl-Pak'), various sizes (one per collection)

Mason's chisel (brick chisel or bolster), with blade 2–6 cm wide (see Figure 6.1)

Club hammer (i.e., one with a large flattened metal head for use with chisel)

Storage containers (e.g., large plastic barrels, Figure 6.2)

Waterproof paper

Waterproof marking pen

2B pencils

Formalin for general preservation (100% formalin, i.e., 40% formaldehyde solution, see Warning, p. 27)

Appropriate gloves (see Warning, p. 27)

For samples for molecular analysis: Ethanol (96–100%) and leak-proof vials or jars or small 'zip-lock' plastic bags, nappy liners or Chux cloths and silica gel

Heavy-duty plastic bags to combine collections for transport

Rubber bands





Figure 6.2: Black plastic barrel with secure lid, suitable for storage and transport of formalin-preserved coralline specimens from the field.

- Hammers and wide chisels (Figure 6.1) are used to remove corallines from rocky reef substrates. Try to get as much of the plant off as possible, and with the least number of fragments. Smaller stones, shells, or other algae can usually be collected by hand.
- Plants that are brittle and can easily fragment (i.e., those loosely attached to a substrate, or that are foliose, layered, or discoid in growth form) should be placed in plastic bags as they are collected, rather than added to already full catch bags.
- Ideally, put each collection in a separate plastic bag, as it is collected. Although time consuming and not always feasible, this keeps all fragments together and greatly facilitates subsequent processing and preservation.
- Keep an eye out for rhodoliths, particularly in subtidal areas where sand, pebbles, or broken shells occur. Rhodoliths also can occur beneath canopyforming plants on rocky bottoms and in deeper intertidal rock pools.
- When collecting in the subtidal, try to collect from different depth ranges (e.g., 5–10 m vs 10–15 m). Some species of corallines may be far more abundant at greater depths while others may be far more abundant at shallow depths. The depth chosen for collection is arbitrary, but it is more informative to restrict the depth range to about a 5 m span (as opposed to a 10 m span).

Do not collect

- Bleached plants.
- Plants with only old conceptacles or holes/cavities where reproductive structures used to be (see Figures 9.1 and 9.2).
- Tiny samples (see 'How much to collect' below).
- Lots of the same thing (when you have a good collection of what superficially appears to be the same thing/species, look for and collect only those things that are new or different).

How much to collect

Keep in mind that how much of each collection you need will depend on what you intend to do with each collection.

Epiphytes

Enough to fill 2 x 250 ml jars when excess host is trimmed/discarded.

Small snails/limpets

Minimum of 5. Optimum of 10–15 shells or enough to fill 2 jars. If possible kill and remove animals from shells before preservation.

Paua

Minimum of 1 large shell or 2 small shells. Optimum of 2–3 large shells or 5–6 smaller shells. Shells should have at least 50% cover of a clear dominant single plant.

Material on large rocks

If the rock easily breaks, place the entire collection in a separate bag on collecting. Minimum of 1 fertile piece 70 mm across. Optimum of enough material to fill two 250 ml jars.

Material on small pebbles/stones

The pebble/stone should have 50% or more cover of a clear dominant thing at least 70 mm across. Collect a minimum of 2 stones, optimally 6.

Rhodolith

Minimum of 10 individuals of a single growth form. Optimum of 20 individuals of a single growth form.

SAMPLES FOR SHORT-TERM STORAGE

Specimens for short-term storage can be kept as wet specimens (in flow-through tanks if possible) or airdried.

SAMPLES FOR LONG-TERM STORAGE

Specimens for long-term storage (without molecular analyses) should first be preserved in formalin and then preferably stored in an glycerol-ethanol solution (or air-dried). Formalin preserved air-dried material is not as useful for subsequent detailed morphological/anatomical study as liquid-preserved specimens.

If specimens are for long-term storage and future molecular analyses, they should be air-dried without prior preservation in formalin. Alternatively, small samples for molecular analysis can be taken before preservation of the remainder of the collection in formalin – see **DNA subsampling** later in this chapter.

PRINCIPLES OF HANDLING SPECIMENS

Process as quickly as possible

It is important to preserve samples as soon as possible after removal from the sea to avoid damage to the DNA and deterioration of non-calcified structures that are important for identification.

Keep cool and dark; pack loosely

Onboard ship, samples can be kept, if necessary, for up to 6 hours (in cool weather) before preservation, **provided** they are kept cool to cold in low light or darkness and are only **loosely** packed together. Live molluscs, sponges, and other animals should be kept separately from other material. Samples are best placed loosely in a black barrel (no more than half full), and just covered with seawater before placing the lid on the barrel.

Pack specimens to avoid damage

Specimens that are brittle or may easily fragment should not be mixed with more solid specimens, and it is best to place these two types of specimens in different barrels for storage and shipment. However, brittle and easily fragmented specimens can be mixed with other fleshy algae harbouring epiphytes as the fleshy algae will help cushion them.

PROCESSING SPECIMENS

WARNING: Parts of the preservation procedures outlined in this Chapter involve using formalin. Formalin is hazardous and should be used ONLY in a well ventilated area and with appropriate protective gear (safety glasses, nitrile [not latex] gloves). Consult your lab manager or workplace health and safety officer to confirm safe practices for working with and disposing of formalin. Ensure you conform to all regulations when transporting specimens in formalin.

- 1. Whenever possible, sort the material into three groups before preservation and placement in barrels:
 - a. rocks
 - b. molluscs, sponges, other animals
 - c. epiphytic corallines, delicate or easily fragmented corallines, geniculate corallines.
- 2. Remove DNA samples (if required) at this stage (see below).
- 3. Double bag each collection with heavy-duty plastic bags (this helps to prevent formalin leakage). Add labels containing the collection number and field information (on permanent/waterproof paper in **pencil**).
- 4. When all bags are ready, add about 10–15 ml of full strength formalin to each bag and **immediately** seal with rubber bands. Do not compact the bag or remove excess air when sealing. Wear appropriate gloves (nitrile) while doing this.
- 5. Place bags in screw-top black plastic barrels (Figure 6.2) (or similar) for storage and shipment.

DNA SUBSAMPLING

- 1. Only small amounts of material are required for DNA analysis. Usually one or several pieces 2–3 cm in greatest dimension is adequate for analysis, although more may be preserved if desired.
- 2. Select only healthy looking specimens that are not contaminated with other organisms (epiphytes, bits of host substrate, etc.). Reproductive structures (conceptacles) should be evident. Specimens must be large enough to allow for DNA preservation of part of the specimen and preservation of the remaining part (as a voucher) for the herbarium.
- 3. Select the portion to be preserved for DNA analysis and place in a screw cap container filled with 95–100% ethanol. Alternatively, wrap the coralline pieces in a small square of nappy liner or Chux cloth (to keep material together) and place in a 'zip-lock' bag with silica gel. Label the vial/bag.
- 4. Place a sticker and a drop of waterproof glue (e.g. Selley's *Multi-Grip*) or some brightly coloured nail polish over the exact area the DNA sample was taken to avoid any later confusion.
- 5. Store the vials/bags in a separate container from the formalin-preserved material. Do not transport samples for DNA analyses in barrels with formalin-preserved specimens as formalin vapour adversely affects the DNA.

PROCESSING FOR LONG-TERM STORAGE

While it is often possible to obtain the information required for identification from dried individuals, ethanol-preserved material always yields superior preparations and more readily interpreted slides.

- 1. Formalin should **not** be used for long-term preservation, because it is hazardous and both corallines and host material eventually become soft and fall apart when stored in formalin for long periods. However, specimens can be stored for several months in formalin before processing as detailed below.
- 2. After initial preservation in formalin, all material must be washed thoroughly in a fume hood. This is done by rinsing the material in a continuous flow of fresh water until all formalin is gone (2–3 hours is sufficient, although 24 h may be required under local regulations).
- 3. Material should already be sorted into separate 'collections' at the collecting stage. If not, rough-sorted material should now be sorted as detailed previously in this chapter.
- 4. Once fine sorting is completed, each collection can be further processed for long-term preservation in glycerol-ethanol and/or air-dried.
- 5. Each collection should include some liquidpreserved reproductive material for embedding,

sectioning, and identification. A 1:7:2 glycerol: ethanol:water ('glycerol-ethanol') solution is excellent for long-term storage. This is easily prepared by adding 100 ml of glycerol to 900 ml of 80% ethanol.

- 6. As a general rule, keep 2 x 250 ml (or even 2 x 500 ml) screw-cap jars worth of material and air-dry any excess material.
- 7. Collections on stones too big for the jars may have to be broken up at this stage (using a hammer, chisel, and cutting board always wear safety glasses). It is also a good idea at this time to get rid of any unnecessary algal/animal host material before putting the corallines in jars. Keep enough material to be able to identify the host.
- 8. Do not overcrowd or compress material in the jar as this inhibits penetration of the glycerol-ethanol solution and damages the coralline material.
- 9. Jar lids should be lined with an insert (rough side facing the jar contents) that creates a seal when screwed onto the jar as this will prevent or minimise evaporation.
- 10. Each collection is given an individual 'collection number' written on adhesive labels in pencil (ink will dissolve/run if the jar leaks). One is placed on the jar lid and one on the side of the jar. This helps when

several jars are open. (Note: the labels **will not** stick onto a jar covered in glycerol). Cover the labels with sticky tape to be certain they do not fall off.

- 11. A piece of permanent/waterproof paper with the collection number written in pencil is also placed inside the jar. This helps if the other labels drop off over time.
- 12. After the coralline material and labels are in the jar, completely fill the jar with the glycerol-ethanol solution. Jar lids are then screwed down and checked for leakage by turning on their side (note: check with manufacturer on lid material as some lids e.g. bakelite should not be closed tightly or cracking may occur. These lids will seal effectively without being tightly screwed shut).
- 13. In addition, a labelled box is prepared for each collection. This is used to store any air-dried material, scanning electron microscope stubs, embedded material, glass slides, notes, etc.
- 14. Labels should include such information as the date, place, and depth of collection, names of collectors, details on any associated substrate (on rock, name of host organism, etc.) and the collection number or code. Collection details may be printed from computer records, or written by hand.

STORAGE AND MAINTENANCE

Liquid-preserved material

Whenever possible, collections should be stored in glycerol-ethanol (see p. 28). This prevents the non-calcified structures from drying out or becoming distorted and also ensures longer-term preservation. The glycerol impregnates the algae and affords some protection from desiccation, even if complete ethanol evaporation occurs because of a faulty seal in the jar lid or a faulty thread on the jar.

Potential evaporation can be minimised if jars are completely filled with liquid before initial herbarium storage. If evaporation is detected, the material should be placed in a new jar with a new lid and reimmersed in glycerol-ethanol. The old jar and lid should be discarded.

WARNING: ETHANOL IS HIGHLY FLAMMABLE. ETHANOL AND SPECIMENS IN ETHANOL MUST BE STORED SAFELY.

LAWWABLE LIQUE

Consult your lab manager or workplace health and safety officer to confirm safe practices for storing, working with, and disposing of ethanol.

Air-dried specimens

Air-dried specimens are not as useful for subsequent detailed morphological-anatomical study as liquid-preserved specimens, for reasons already mentioned. However, air-dried material that has not been preserved in formalin can be used for molecular analyses, and any such material should be clearly labelled to indicate that it has not been preserved in formalin.

Air-dried material, unlike liquid-preserved material, will maintain its colour for a period of time if stored in darkness. Over time, however, the colour of many specimens can change and ultimately can be lost due to gradual pigment deterioration.

Air-drying generally is used for any excess material not preserved in glycerol-ethanol and for particularly large specimens to show the habit of intact individuals.

There is no advantage in storing air-dried material in silica gel over a long term; eventually the silica becomes saturated with moisture and this can then adversely affect the specimens.

Store all air-dried material in boxes with appropriate labels. Storage in boxes minimises fragmentation, and retains any fragments with the collection from which they originated. Such fragments are useful for scanning electron microscopy and, in the absence of liquid-preserved material, are also useful for embedding and sectioning.

Pressed specimens on herbarium sheets

Coralline red algae should **never** be stored as pressed specimens on herbarium sheets. Storage on herbarium sheets promotes fragmentation and renders many specimens useless in the longer term. This is particularly true for smaller epiphytic species and geniculate corallines.

Storage in cartons

All parts of a collection should be clearly and appropriately labelled (jars, accompanying box, slide boxes, SEM stubs, etc.) and, where possible, stored together in a single box representing a single herbarium preparation.

Chapter 7. Coming to grips with coralline structure and reproduction

Oralline identification is based on the vegetative and reproductive features outlined and illustrated in this chapter.

It is essential that the user becomes familiar with these features before attempting identifications using either simple lab procedures or more involved lab procedures.

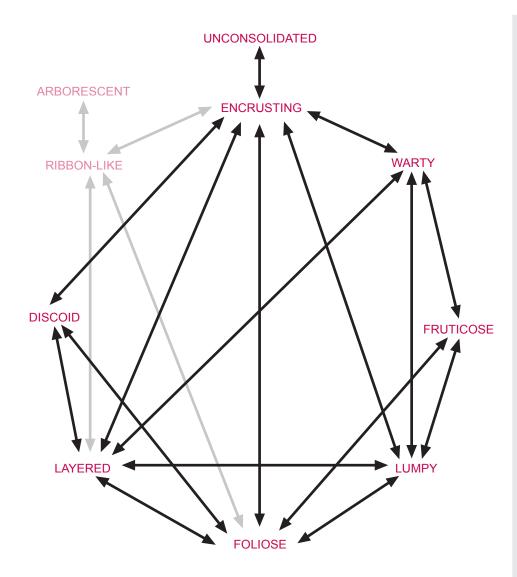
ORGANISATION

Coralline growth forms and substrates are defined and illustrated in Figures 7.1 and 7.2. Growth form terminology follows Woelkerling et al. (1993).

Figures 7.3 and 7.4 provide an introduction to the basic morphology and anatomy of the reproductive structures, which are described in more detail in Figures 7.6–7.9; important vegetative features

are illustrated in Figure 7.10. Morphological and anatomical terminology follows Woelkerling (1988).

A simplified sexual cycle of coralline algae is illustrated in Figure 7.5. The coralline algal life cycle involves three distinct phases. Usually, one important phase (the tetraspore-producing phase) is required for identification.



Arrows indicate intergrading growth forms.

Greyed out arrows and text indicate growth forms not found in central New Zealand to date.

Diagram and definitions adapted from Woelkerling, Irvine, & Harvey (1993).

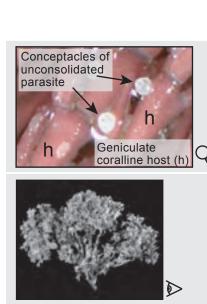
Non-geniculate coralline algae exhibit an intergrading network of growth forms with 10 focal points (see diagram at left). Plants with a growth form corresponding to a single focal point can be described with a single term (e.g., encrusting), and specimens intergrading between focal points can be described as lumpy to encrusting, encrusting to warty to fruticose, etc.

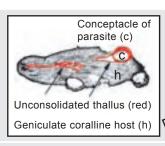
On a worldwide basis, a single species may show up to six different growth forms, any number of which may be exhibited by numerous other species. As a result, reliable sight-recognition of most species is difficult or impossible. Despite these limitations, growth forms are useful in a number of ways.

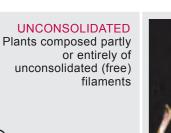
- 1. Growth forms are characteristic of certain genera. For example, *Mastophoropsis* and *Metamastophora* are arborescent. Neither occurs in central New Zealand. The vegetative thallus of *Choreonema*, which does occur in central New Zealand, is unconsolidated.
- Growth form is the most conspicuous feature of any coralline and can be used in conjunction with other characters to help in specimen identification. For example, *Melobesia membranacea* is the only very thin, encrusting, epiphytic, multiporate species known to occur in central New Zealand.

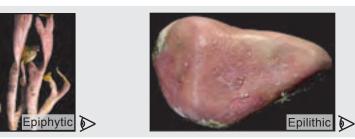
Definitions and images of the 10 growth form focal points are given below. The images are indicative only: considerable variation is encompassed within these terms. Wherever possible, growth forms have been illustrated using material collected from central New Zealand during the course of this study.

Note: this schema refers to non-geniculate coralline algae only, and is not applicable to geniculate coralline algae. Geniculate corallines are attached to rocks and other substrates by a crustose base/holdfast and often have a growth habit similar to the arborescent form described here. They can be distinguished from non-geniculate corallines by the presence of alternating calcified and non-calcified segments (see Figure 7.2, A & B).









ENCRUSTING Plants flat crusts or sleeve-like, without protuberances or branches



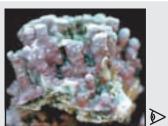
ARBORESCENT Plants tree-like, composed of a distinct holdfast and stipe bearing flattened, ribbon-like to fan shaped branches (not known in central NZ)



WARTY Plants with short (usually < 3 mm), unbranched protuberances



RIBBON-LIKE Plants composed of flat, ribbon-like branches and lacking a distinct holdfast (not known in central NZ)



FRUTICOSE Plants with cylindrical to compressed branches that are mostly > 3 mm long, do not look lumpy, and are free from one another or laterally



DISCOID Unbranched and largely unattached discs of various shapes, with or without struts



coherent **LUMPY**

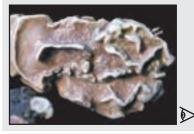


Plants with short, swollen protuberances that may vary in length, are often crowded and contiguous and rarely branched



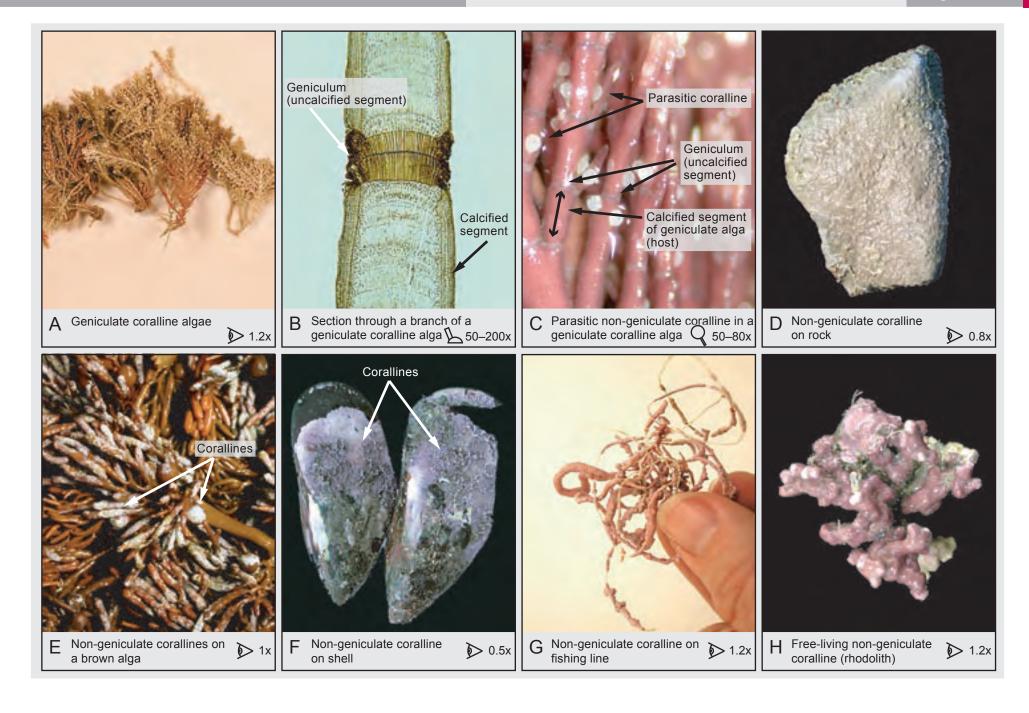


LAYERED Flattened branches forming horizontal layers overlapping one another, often giving the plant a terraced appearance in surface view





FOLIOSE Plants consisting of flattened plate-like branches arranged at various angles to one another, but not in horizontal layers



CORALLINE SUBSTRATES (Figure 7.2)

The order Corallinales includes both geniculate and non-geniculate taxa. Geniculate corallines are illustrated here for comparison, but only non-geniculate corallines are considered in detail in this guide.

Geniculate corallines have upright branches (A) consisting of alternating calcified and uncalcified segments (intergenicula and genicula respectively) (B). In contrast, non-geniculate corallines lack these uncalcified nodes and the thallus is entirely calcified (C–H).

Non-geniculate corallines

On a worldwide basis, a single non-geniculate species may occur on a number of substrates. Moreover, numerous other species may occur on the same substrates. As a result, **reliable** recognition of most species by substrate type alone is difficult or impossible.

Substrate type, however, is a conspicuous feature of any coralline and can be used in conjunction with other characters to help in specimen identification. For example:

- *Melobesia membranacea* is the only very thin encrusting, multiporate, epiphytic species known to occur in central New Zealand.
- Lithophyllum carpophylli is most commonly found growing on the brown alga Carpophyllum, and has a distinct foliose growth form. No other central New Zealand corallines show these features

Substrates

- Non-geniculates can be found as **attached** plants growing on a variety of substrates, including:
 - rocks (D) plants said to be **epilithic**.
 - brown, green, and red algae (E) including other coralline algae plants said to be **epiphytic**.
 - seagrasses plants said to be **epiphytic**.
 - animals such as sponges, kina, or molluscs (F)
 plants said to be epizoic.
 - glass, plastics, metal, and other refuse plants said to be **epigenous** (G).

Some species (e.g., *Choreonema*) may grow wholly or partly inside another alga – such plants are said to be **parasitic** or **semi-endophytic** (C).

• Some species can also grow as **unattached** (free-living) rhodoliths (H). Rhodoliths may be made up entirely of coralline plant material or may develop around a small stone or mollusc shell.

Figure 7.2

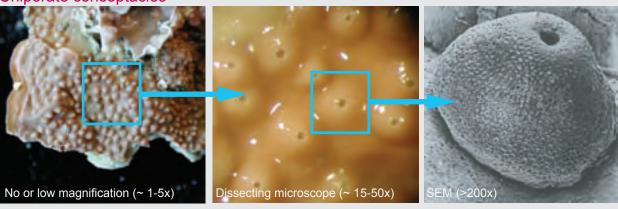
- A: Corallina sp.
- B: Unidentified geniculate coralline
- C: Choreonema thuretii
- D: Spongites yendoi
- E: Hydrolithon improcerum
- F: Spongites yendoi
- G: Unidentified non-geniculate coralline
- H: Sporolithon durum

Figure 7.3

Figures 7.3 & 7.4 introduce the reproductive structures present in non-geniculate coralline algae. It is important to become familiar with these features, as fertile specimens (those with reproductive structures) are usually required to identify non-geniculate coralline algae to species level.

There are three sorts of reproductive structures: uniporate conceptacles (Figure 7.3), multiporate conceptacles (Figure 7.4), and calcified compartments (Figure 7.4). The morphology and anatomy of these structures are introduced here (along with an indication of their size or scale), and described in more detail in the following pages. Definitions are given in the glossary (p. 131).

Uniporate conceptacles



Uniporate conceptacles are hollow chambers that house the reproductive bodies (spores or gametes). They have a single pore in their roof through which the spores or gametes are released. Uniporate conceptacles may contain either:

tetrasporangia (with zonately arranged tetraspores), male gametes (spermatia on spermatangial filaments), female gametes (carpogonia), or carposporophytes (with carposporangia).

Tetrasporangia

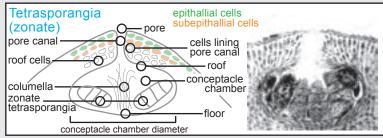
Tetrasporangia may occur in uniporate conceptacles, multiporate conceptacles, or calcified compartments. In uniporate conceptacles, the tetrasporangia usually contain four spores (the tetraspores) that are zonately arranged. Plants with tetrasporangial conceptacles are usually required for specimen identification.

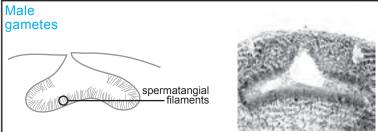
Male and female gametes

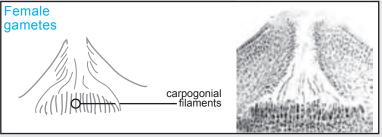
Male and female gametes occur in uniporate conceptacles only. Plants with male or female gametes only are usually unable to be identified to species level. Plants with male gametes, however, are used in conjunction with tetrasporangial plants to help identify certain species.

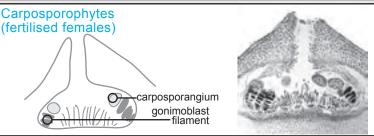
Carposporophytes bearing carposporangia

Carposporophytes arise from fertilised female gametes (carpogonia), and occur in uniporate conceptacles only. Plants with carposporophytes only are usually unable to be identified to species level.

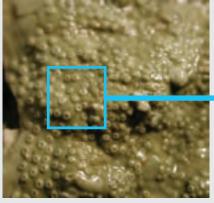


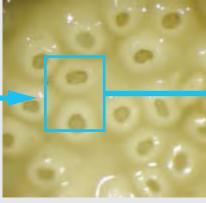


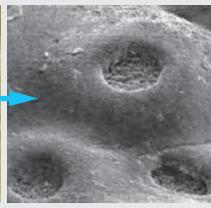


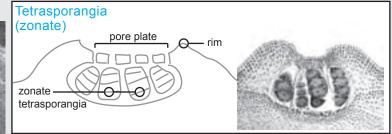


Multiporate conceptacles





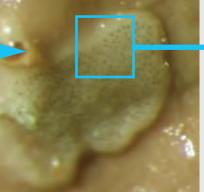


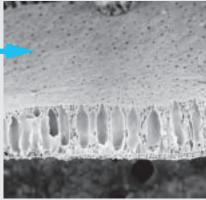


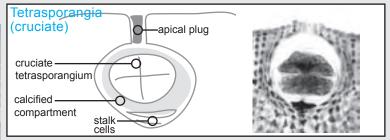
Multiporate conceptacles are hollow chambers that house tetrasporangia (never gametes). The tetrasporangia usually contain four spores that are zonately arranged, and the conceptacles have numerous pores in their roof through which these spores are released.

Calcified compartments (grouped into a sorus)









Calcified compartments are calcified, balloon-like structures that house tetrasporangia (never gametes). The tetrasporangia contain four spores that are cruciately arranged, and the compartments have a single pore in their roof through which these spores are released. Calcified compartments may be solitary and scattered across the thallus, or they may be grouped together in a cluster termed a sorus.

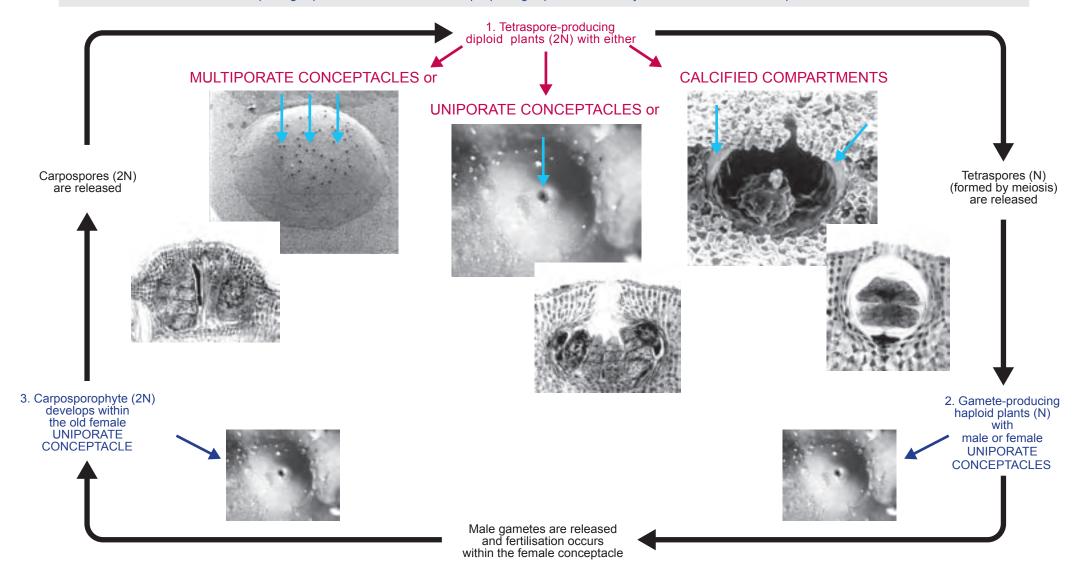
The type of reproductive structures in which tetrasporangia occur, and the arrangement of the tetraspores (zonate or cruciate), differ depending on the coralline family (see table at right). Male gametes, female gametes, and carposporophytes always occur in uniporate conceptacles in all coralline families.

Family	Tetrasporangia	Males, females, carposporophytes			
Corallinaceae	Uniporate conceptacle; zonate	Uniporate conceptacle			
Hapalidiaceae	Multiporate conceptacle; zonate	Uniporate conceptacle			
Sporolithaceae	Calcified compartment; cruciate	Uniporate conceptacle			

IMPORTANT NOTES

In most cases, tetrasporangial plants (plants producing tetraspores) are required for specimen identification.

- Tetraspores occur in multiporate conceptacles, uniporate conceptacles, or in calcified compartments.
- Multiporate conceptacles do not contain gametes or carposporophytes.
- Calcified compartments do not contain gametes or carposporophytes.
- Uniporate conceptacles can contain tetraspores, male gametes, female gametes, or carposporophytes.
- In the absence of tetrasporangial plants, male, female, and carposporangial plants are usually unable to be identified to species level.



SIMPLIFIED CORALLINE SEXUAL CYCLE (Figure 7.5)

The photos are representative only, and variations occur in other genera.

- The coralline algal sexual cycle involves three distinct phases:
 - the tetrasporangial (tetraspore-producing) phase
 - the male and female gamete-producing phase
 - the carposporangial (carpospore-producing) phase
- Usually, tetrasporangial plants are required for specimen identification (and males are also required in some cases).
- Tetraspores occur in:
 - uniporate conceptacles,
 - multiporate conceptacles, or
 - calcified compartments
- Bispores:
 - occur in the same kinds of conceptacles as tetraspores
 - may be part of the sexual cycle or, more usually, may be an asexual spore and not part of the sexual cycle
- Male gametes, female gametes, and carpospores occur only in uniporate conceptacles.

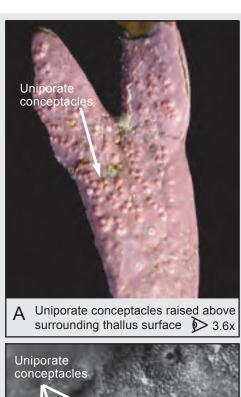
General notes

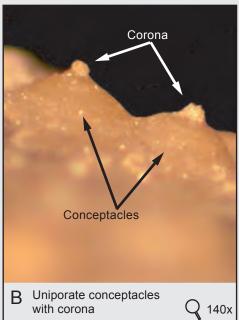
- 1. Tetraspore-producing phase (2N)
 - tetraspores are borne either in uniporate conceptacles (Figure 7.6), in multiporate conceptacles (Figure 7.7), or in calcified compartments (Figure 7.8)
 - at maturity, tetraspores are released and develop into haploid gamete-producing (male and female) plants
- 2. Male & female gamete-producing phase (N)
 - male gametes (spermatia, in structures called spermatangia) (Figure 7.9, B & D) and female gametes (carpogonia) (Figure 7.9, F) are borne in uniporate conceptacles (Figure 7.9, A, C & E)
 - at maturity, male gametes are released and passively carried to female conceptacles, where they attach to trichogynes (Figure 7.9, F) and fertilise carpogonia to produce a zygote
- 3. Carpospore-producing phase (2N)
 - after fertilisation, the zygote undergoes a series of developmental changes resulting in a much reduced, microscopic, diploid carposporophyte within the old female conceptacle (Figure 7.9, G)
 - the carposporophyte produces carposporangia, each of which contains a single carpospore (Figure 7.9, H)
 - at maturity, carpospores are released and develop into independent, diploid, tetraspore-producing plants

- Species that produce multiporate conceptacles will have uniporate conceptacle phases in their sexual cycle (i.e., containing males, females, or carposporophytes).
- Species that produce uniporate tetrasporangial conceptacles will have other uniporate conceptacle phases in their sexual cycle (i.e., containing males, females, or carposporophytes).
- Species that produce calcified compartments will also have uniporate conceptacle phases in their sexual cycle (i.e., containing males, females, or carposporophytes).

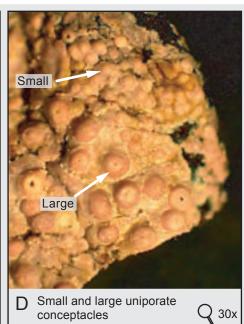
Field notes

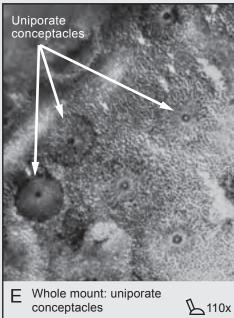
- Within a single species, diploid tetrasporeproducing plants and haploid gamete-producing plants are similar morphologically.
- Male and female conceptacles are most commonly borne on separate plants.



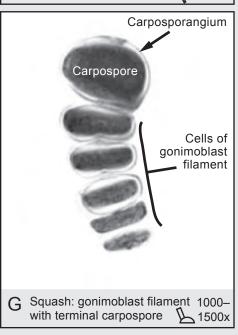


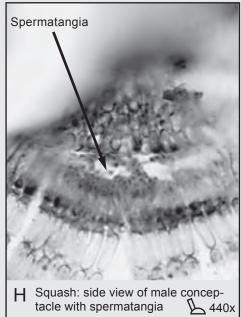












UNIPORATE CONCEPTACLES (Figure 7.6)

Features associated with uniporate tetrasporangial conceptacles are used to help identify coralline taxa.

General notes

- Uniporate conceptacles are enclosed chambers that may house/enclose:
 - tetrasporangia (with four **zonately** arranged tetraspores)
 - carposporangia (on a carposporophyte)
 - carpogonia (female gametes)
 - spermatia (male gametes in structures called spermatangia)
 - bisporangia (with two bispores)

(Compare with calcified compartments, which are chambers with a surrounding calcified structure.)

- For most identifications to species, specimens must be tetrasporangial (contain tetraspores).
- Uniporate conceptacles may be tetrasporangial, male, female, or carposporangial (see Figure 7.6).
- Male, female, and carposporangial plants are usually unable to be identified to species level in the absence of tetrasporangial plants.
- The roof of the conceptacle has a single pore through which the contents are released.
- All taxa in the family Corallinaceae possess uniporate tetrasporangial conceptacles.

Field notes

- Uniporate conceptacles may be raised above (A & B) or flush with (C) the surrounding thallus surface.
- Conceptacles can often be differentiated by their shape:
 - uniporates are often conical, and pores may be visible under a dissecting microscope (D)
 - multiporates are flat-topped, usually with no visible pores
- In epiphytic collections:
 - dozens of individuals commonly occur on a single host
 - multiporate and uniporate specimens may both be present
 - different sized uniporate conceptacles may be present
- Different sized uniporate conceptacles (D) may indicate:
 - different species
 - different stages in the sexual cycle of the same species (e.g., tetrasporic plants and male plants)
- The corona (B):
 - is a term for a group of filaments surrounding the pore and protruding above the thallus surface
 - can often be seen under a dissecting microscope (especially in side view)
 - is an important characteristic of *Pneophyllum* coronatum

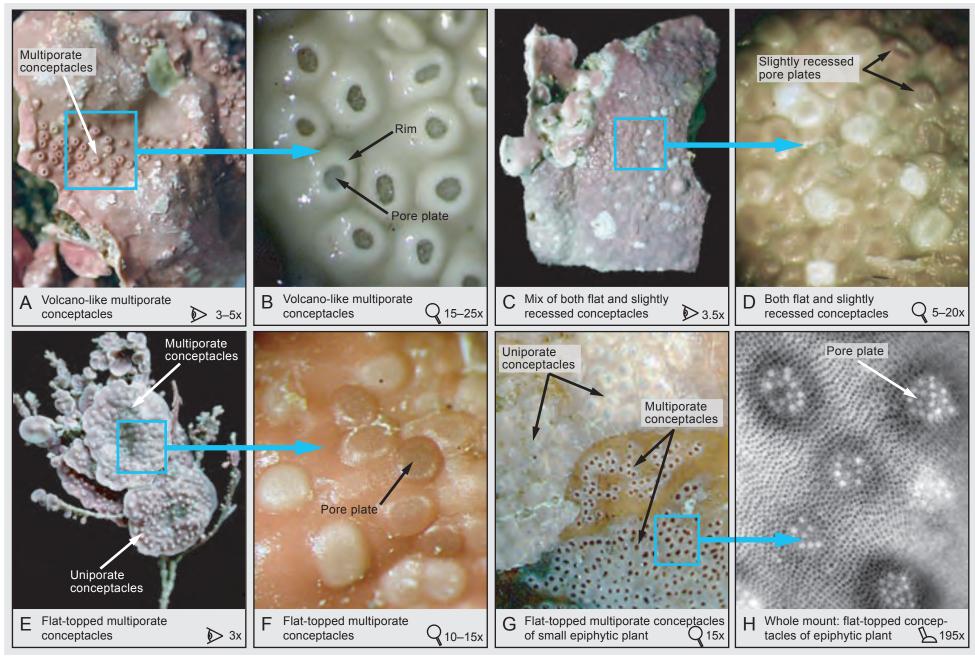
Helpful hints

- Simple lab procedures (Tables 9.1 & 9.2) can often confirm the presence of uniporate conceptacles on small epiphytic plants (E).
- Simple lab procedures (Tables 9.1 & 9.2) can also often confirm whether uniporate conceptacles house/enclose:
 - tetrasporangia (F) or bisporangia
 - carposporangia (G)
 - spermatangia (H) or
 - carpogonia (Figure 7.9, E & F)
- Volcano-like multiporate conceptacles (Figure 7.7, A) can be mistaken for uniporate conceptacles by the novice.

Figure 7.6

- A: Pneophyllum fragile
- B: Pneophyllum coronatum
- C: Spongites yendoi
- D: Lithophyllum sp.
- E: Lithophyllum carpophylli
- F: Unidentified coralline
- G: Unidentified coralline
- H: Lithophyllum carpophylli male conceptacle

All taxa in the family Hapalidiaceae possess multiporate tetrasporangial conceptacles



MULTIPORATE CONCEPTACLES (Figure 7.7)

Features associated with multiporate conceptacles are used to help identify coralline taxa.

General notes

- Multiporate conceptacles are enclosed chambers containing tetrasporangia. (Compare with calcified compartments, which are chambers with a surrounding calcified structure.)
- Each tetrasporangium contains four **zonately** arranged tetraspores (or sometimes two bispores).
- For most identifications to species, specimens must be tetrasporangial (contain tetraspores).
- Multiporate conceptacles have a pore plate with numerous pores/holes, through which the spores are released (H).
- The conceptacle roof may be:
 - volcano-like (A & B), or
 - flat-topped (E & F)
- Volcano-like conceptacles have a distinct rim and a central sunken pore plate (B).
- Flat-topped conceptacles may be mound-like (raised) or more or less level with the thallus surface, with a flat roof (i.e., lacking a distinct rim and central sunken pore plate) (F).
- Others may not easily fall within these two categories, i.e., some have only slightly sunken pore plates (D).

- Multiporate conceptacles commonly have a **multicellular** multiporate plate or roof (i.e., made up of cells with interspersed pores) (Figure 12.13, G & H).
- Multiporate conceptacles in *Choreonema* have an **acellular** multiporate plate (made up of a calcium carbonate matrix with interspersed pores) sunken beneath a single outer opening (Figure 12.10, F & G).
- All taxa in the family Hapalidiaceae possess multiporate tetrasporangial conceptacles (Figure 7.7).

Field notes

- In epiphytic (E & G) and epizoic collections:
 - dozens of individuals commonly occur on a single host, and
 - both multiporate and uniporate specimens are often present
- Pores of multiporate conceptacles are usually too small to be visible under a dissecting microscope.
- Conceptacles can often be differentiated by their shape:
 - multiporates are flat-topped or volcano-like
 - uniporates are often conical, and a single pore may be visible under a dissecting microscope (Figure 7.6, D)

- Volcano-like conceptacles (A) can be mistaken for uniporate conceptacles by the novice.
- In *Phymatolithon repandum*, multiporate conceptacles are commonly shed (Figure 12.16, C) leaving hollows (or cavities) that can easily be mistaken for intact conceptacles.

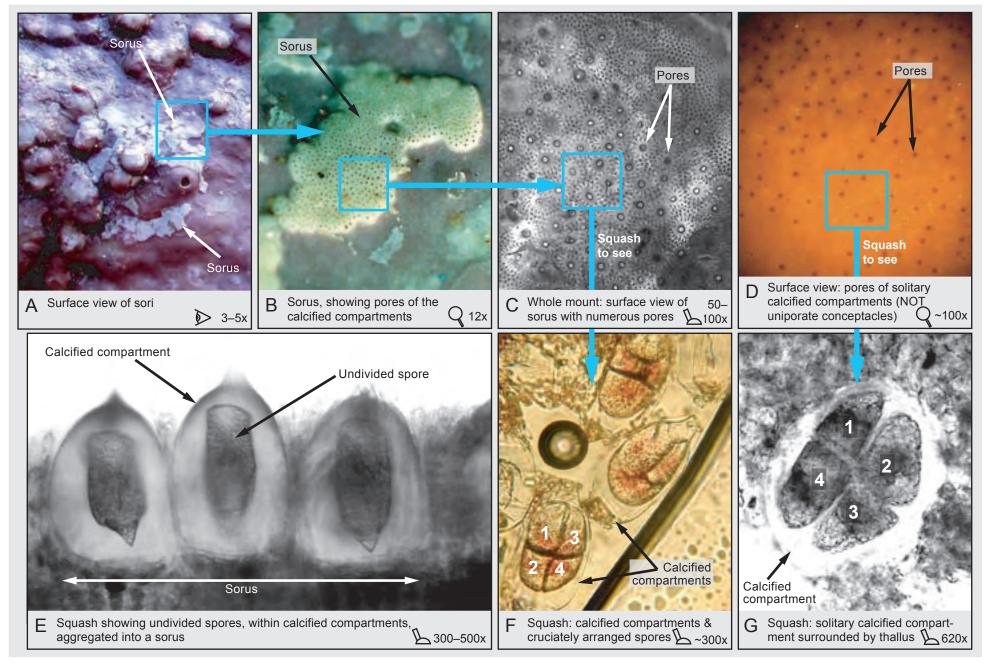
Helpful hints

- Pore plates of small epiphytic plants (e.g., *Melobesia*):
 - often appear as dark patches surrounded by a lighter thallus (G)
 - can often be confirmed by simple lab procedures (Tables 9.1 & 9.2) (H)

Figure 7.7

- A: Mesophyllum printzianum
- B: Synarthrophyton schielianum
- C: Mesophyllum erubescens
- D: Mesophyllum erubescens
- E: *Mesophyllum erubescens* epiphytic on a geniculate coralline
- F: Synarthrophyton patena
- G: *Melobesia membranacea* epiphytic on the red alga *Rhodymenia*
- H: Melobesia membranacea

All taxa in the family Sporolithaceae possess calcified compartments



CALCIFIED COMPARTMENTS (Figure 7.8)

Features associated with calcified compartments are used to help identify coralline taxa.

General notes

- Calcified compartments are calcified structures that contain/enclose a single tetrasporangium (E) (also Figure 7.5). (Compare with conceptacles, which are chambers without any surrounding calcified structure.)
- Each tetrasporangium contains four **cruciately** arranged tetraspores (F & G).
- For most identifications to species, specimens must be tetrasporangial (contain tetraspores).
- Compartments may be:
 - solitary (D), or
 - grouped into sori (A–C)
- Sori are irregular in shape and indefinite in size (A & B) (up to 25 mm across in collections from central New Zealand).
- All taxa in the family Sporolithaceae possess calcified tetrasporangial compartments (Figure 7.8).

Field notes

- Sori (A & B):
 - can often be seen by eye in the field or under a dissecting microscope
 - appear as irregularly shaped, slightly raised, pink or white patches on the plant surface
- Pores of sori (B) and solitary compartments (D) may be visible under a dissecting microscope.
- Numerous sori can be present, overall covering a large area of the plant.
- Size and reproductive maturity of individual sori on the same plant may vary greatly.

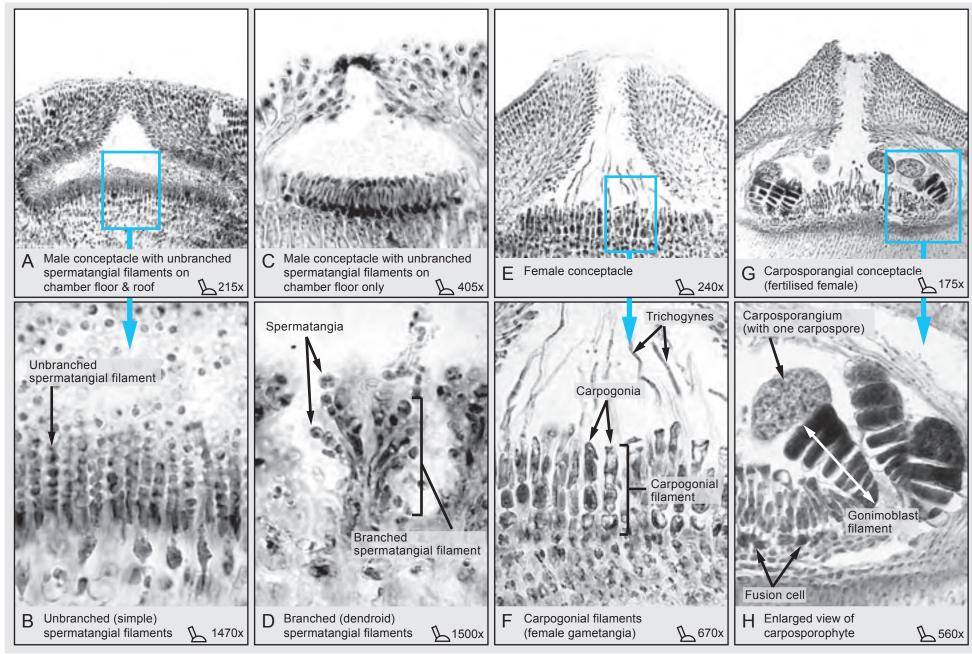
Helpful hints

- Epithallial cell sloughing, or other surface irregularities, may be mistaken for sori on initial inspection.
- Tetrasporangial plants of *Heydrichia homalopasta* have small and often inconspicuous pores at the surface (D) and may appear infertile or uniporate upon initial inspection.
- Simple lab procedures (Tables 9.1 & 9.2) can confirm the presence of :
 - sori (C)
 - calcified compartments (F)
 - cruciately arranged spores (G)
- Depending on the plane of view, four spores may not always be visible in every calcified compartment (compare the two compartments in F).

Figure 7.8

- A: Sporolithon durum
- B: Sporolithon durum
- C: Sporolithon durum
- D: Heydrichia homalopasta
- E: Sporolithon durum
- F: Sporolithon durum
- G: Heydrichia homalopasta

Photos are representative only and variations exist



MALE, FEMALE, AND CARPOSPORANGIAL CONCEPTACLES (Figure 7.9)

Tetrasporangial (tetraspore-producing) plants are usually required for specimen identification and males are also sometimes required. Thin sectioning, however, is required to see details in male conceptacles useful for identification.

Male, female, or carposporangial plants are usually unable to be identified to species level in the absence of tetrasporangial plants.

In general

- Male conceptacles are always uniporate and contain the male gametes (spermatia) in structures called spermatangia.
- Spermatangia:
 - are terminal on filaments that may be confined to the conceptacle floor (C) or may also occur on the conceptacle roof (A)
 - spermatangial filaments may be unbranched (simple) (B) or branched (dendroid) (D)
- Male gametes are usually very small and it is the spermatangia (housing the spermatia) that are usually evident in sections and squashes (D).
- Female conceptacles are always uniporate and contain the female gametes (or carpogonia).

- Carpogonia:
 - are terminal on filaments arising from the female conceptacle floor (E)
 - usually bear a trichogyne (F), to which the spermatia attach
- Carposporangial conceptacles are always uniporate and contain the carposporophyte.
- Carposporophytes develop within female conceptacles after fertilisation and when mature:
 - may include a large central fusion cell or a 'dissected/discontinuous' fusion cell; as well as gonimoblast filaments bearing terminal carposporangia (each containing one carpospore) (G & H), or
 - may apparently consist only of stalk cells bearing carposporangia (each containing one carpospore) (not shown)

Helpful hints

• When using simple lab procedures, use this figure in conjunction with Figure 9.11 (reproductive features seen with simple lab procedures) to determine if your plants are tetrasporangial, male, female, or carposporangial.

Further reading

When using more involved lab procedures, keep in mind photos are representative only and variation exists in genera other than those shown. For further reading and illustrations, see:

Family Corallinaceae
Subfamily Lithophylloideae
Woelkerling (1996d)
Subfamily Mastophoroideae
Woelkerling (1996e)
Penrose (1996a, 1996b, 1996c, 1996d)

Family Hapalidiaceae
Subfamily Austrolithoideae
Woelkerling & Harvey (1996)
Subfamily Choreonematoideae
Woelkerling (1996c)
Subfamily Melobesioideae
Woelkerling (1996b)

Family Sporolithaceae Woelkerling (1996a)

Figure 7.9

A: Mesophyllum engelhartii

B: Mesophyllum engelhartii

C: Lithophyllum carpophylli

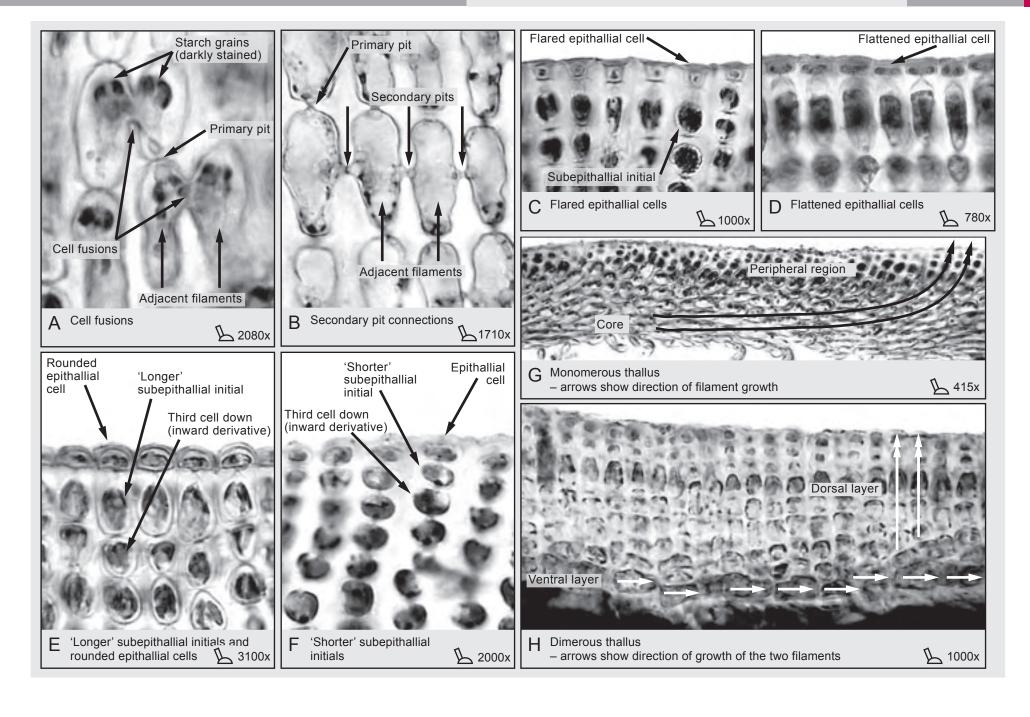
D: Synarthrophyton schielianum

E: M.engelhartii/S.patena

F: M.engelhartii/S.patena

G: M.engelhartii/S.patena

H: M.engelhartii/S.patena



VEGETATIVE CHARACTERS (Figure 7.10)

- Coralline identification is based on important features associated with the vegetative thallus and reproductive structures. Important vegetative features include the kinds of:
 - cell connections present
 - epithallial cells present
 - subepithallial initials present
 - thallus construction present
- Cell connections are the only vegetative features relatively easily observed with simple lab procedures (Figure 9.10).

General thallus anatomy

- Non-geniculate corallines are made up of many filaments that:
 - coalesce to form the thallus (G & H)
 - occasionally remain largely unconsolidated (free) (e.g., *Choreonema*) (Figure 7.1)
- Cells within a filament and cells between filaments are usually joined to one another by holes/ connections.
- Usually:
 - epithallial cells are on the ends of (terminate) most filaments (C–F)
 - subepithallial initials/cells occur just below the epithallial cells (E & F)

Cell connections (Figure 7.10, A & B)

Cell connections are important vegetative characters, and the kinds of cell connections present are used to help identify coralline taxa (see Table 5.2)

- Three main sorts of cell connections occur between
 Cell fusions usually are: filaments that make up the thallus:
 - primary pit connections
 - secondary pit connections (also termed secondary pits or 2° pits)
 - cell fusions
- These connections may occur between cells of the same filament or cells of adjacent filaments:
 - primary pit connections occur between cells of the same filament (A & B)
 - secondary pit connections occur between cells of adjacent filaments (B)
 - cell fusions occur between cells of adjacent filaments (A)
- Secondary pits and cell fusions are important vegetative characters and their presence/absence is used to help identify coralline taxa.
 - family Sporolithaceae have either cell fusions or secondary pit connections or both
 - subfamily Melobesioideae have cell fusions only
 - subfamily Choreonematoideae have neither cell fusion nor secondary pit connections
 - subfamily Lithophylloideae have secondary pit connections only
 - subfamily Mastophoroideae have cell fusions only
- A cell fusion is a linkage between two cells in which portions of the cells' walls break down and the protoplasts fuse.

- - larger then secondary pit connections
 - occur between some (not all) of the cells of adjacent filaments (A)
- A secondary pit is a linkage between two cells by means of a small opening in the cell wall (pit) filled by a plug (pit plug). The protoplasts do not fuse in secondary pit connections.
- Secondary pit connections usually are:
 - smaller then cell fusions
 - occur between most (often all) of the cells of adjacent filaments (B)
- Cells of non-geniculate corallines occasionally are not linked by either cell fusions or secondary pit connections (e.g., Choreonema).
- Cell connections can be observed with simple lab procedures (Tables 9.1 & 9.2) or thin sectioning (see Chapter 11).
- Primary pit connections occur in all non-geniculate coralline algae (A & B) and cannot be used to identify coralline taxa

Figure 7.10

- A: Mesophyllum printzianum
- B: Lithophyllum sp.
- C: Sporolithon durum
- D: Lithophyllum stictaeforme
- E: Mesophyllum macroblastum
- F: Phymatolithon repandum
- G: Synarthrophyton patena
- H: Melobesia membranacea

Epithallial cells (Figure 7.10, C-E)

- Epithallial cells are important vegetative characters, and the kind of epithallial cell present is used to help identify coralline taxa.
- Observation of epithallial cells requires thin sectioning (see Chapter 11).
- Filaments commonly are terminated at the thallus surface by epithallial cells.
- Two sorts of epithallial cells occur in non-geniculate coralline algae:
 - flared
 - rounded or flattened
- Flared epithallial cells have the flattened outermost wall extended laterally to form a distinct rim (C).
- Rounded or flattened epithallial cells lack a lateral extension and distinct rim (D & E).

Subepithallial initials (Figure 7.10, C, E & F)

- Subepithallial initials are important vegetative characters and the kind of initial present is used to help identify coralline taxa (see Table 5.2).
- Observation of subepithallial initials requires thin sectioning (see Chapter 11).
- Subepithallial initials:
 - occur directly below the epithallial cells, and
 - are meristematic actively dividing to give rise to new epithallial cells outwardly and new vegetative cells inwardly (the immediate inward derivative or third cell down)
- Three main sorts of subepithallial initials occur in non-geniculate coralline algae:
 - subepithallial initials that are 'as long or longer' than their immediate inward derivatives (or third cell down) (E)
 - subepithallial initials that are 'as short or shorter' than their immediate inward derivatives (or third cell down) (F)
 - subepithallial initials that are 'mostly the same size' as their immediate inward derivatives (or third cell down) (C)

Thallus construction (Figure 7.10, G & H)

- Thallus construction is an important vegetative character, and the kind of thallus construction present is used to help identify coralline taxa (see Table 5.2).
- Observation of thallus construction requires thin sectioning (see Chapter 11).
- Encrusting portions of plants/thallus may be:
 - monomerous, or
 - dimerous
- Monomerous thalli consist of a single system of filaments that contribute to:
 - a core, that runs more or less parallel to the thallus surface, and
 - a peripheral region, formed when portions of the core filaments curve outwards towards the thallus surface (G)
- Dimerous thalli consist of two groups of filaments that are oriented more or less at right angles to one another:
 - the first group of filaments forms a single ventral layer that contributes to thallus width
 - the second group of filaments arises at right angles from the ventral layer and contributes to thallus thickness (H)

Chapter 8. Specimen identification with direct observation and/or simple lab procedures

ome coralline specimens can be identified from LIMITATIONS OF SPECIMEN IDENTIFICATION direct observations with a dissecting microscope Definitive identification of coralline algae requires lab procedures. Other specimens, however, may also require some simple lab procedures (e.g., the mounts or squashes – see Chapter 9), and vet others can be fully identified only after more involved lab procedures such as examination of slides made from embedded and sectioned material (see Chapter 11 for detailed methods).

This chapter outlines the steps involved in the preliminary identification of specimens, and is intended for use by people with little or no algal taxonomy training and with access to limited laboratory equipment (dissecting and compound microscopes, slides and coverslips). The methods can be applied to either fresh or air-dried specimens or specimens preserved in glycerol-ethanol solution (after initial formalin preservation). It includes a Flow chart key (Figures 8.2A, 8.2B & 8.2C) illustrating important characters and indicating the methods (whole mounts or squashes) used to observe these features. If simple lab procedures are required to observe certain features. Chapter 9 details those methods.

or hand lens and without the need for any further embedding, sectioning, and microscopic examination of important vegetative and reproductive features. As a consequence, identifications made with simple preparation of temporary slides containing whole lab procedures should be regarded as less than 100% certain, or preliminary, and ultimately should be confirmed with more involved lab procedures, where possible.

> Although the sampling programme upon which this guide is based was extensive, other species may occur in the region which could key out (albeit incorrectly) in the flow chart key.

STEPS IN SPECIMEN IDENTIFICATION

The steps involved in the identification process are listed below. The best way to determine if simple lab procedures are required to identify your specimen is to start filling in a copy of Table 8.1. Keep in mind that not every specimen will be identifiable, and misidentifications are possible.

- 1 Become familiar with coralline structure and function (Chapter 7), including
 - growth forms and substrates
 - sexual cycle
 - reproductive structures
 - cell fusions and secondary pit connections

This is essential.

2. For most identifications to species, specimens must be fertile and the reproductive structures should be tetra/bisporangial (contain tetra/bispores) (see Coralline sexual cycle, Figure 7.5).

Remember:

- multiporate conceptacles are always tetrasporangial (see Figure 7.7) or bisporangial
- calcified compartments are always tetrasporangial (see Figure 7.8)
- uniporate conceptacles may be tetrasporangial, male, female, or carposporangial (see Figure 7.6)
- male, female, and carposporangial plants are usually unable to be identified to species level in the absence of tetrasporangial plants
- sterile plants are usually unable to be identified to species level
- misidentifications are also possible (see Limitations of the guide in Chapter 1)

- 3. Corallines often grow intermixed (different plants very close to one another on the same substrate). As a result, even though every effort is made to keep only one thing/species, there can often be a number of plants in a single collection.
 - The rule of thumb is to identify whatever appears to be the major or most common plant/species in a collection (see Figure 8.1), and/or denote the 'identified' plant using nail polish or similar.

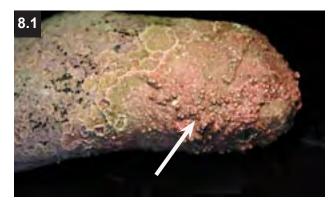


Figure 8.1: Rock with mixed coralline cover. Major plant in collection is arrowed.

- 4. Fill in a copy of Table 8.1 for each specimen. Circle or highlight only one answer as appropriate in parts A D. For easier use, the important characters in each alternative statement are in **bold**.
- 5. If simple lab procedures are required:
 - use the simple lab procedures detailed in Chapter 9 to observe the required features
 - see Tables 9.1 and 9.2 for the correct lab procedure to use for each feature
- 6. Once you have filled in Table 8.1, undertake a preliminary identification using the Flow chart key (Figures 8.2A, 8.2B & 8.2C). Start with Figure 8.2A.

- 7. Use the species profiles to support or reject your identification.
 - If the flow chart key leads to a single species, use the species profiles to support or reject your preliminary identification (Chapter 12)
 - If the flow chart key leads to a group of 2–4 species, full identification to species is not possible without slides of embedded and sectioned material (Chapter 11)

Note

It is important to follow through these steps, and the various keys, as directed. Jumping ahead and/or skipping steps to match a feature or features you can easily recognise may lead to misidentification.

Table 8.1: Specimen information that may be needed for identification using the FLOW CHART KEY (Figures 8.2A, 8.2B, & 8.2C)

- 1. Photocopy this table for each specimen.
- 2. Examine collection carefully is it mixed? Which plant are you identifying? You may need to mark this plant somehow.
- 3. Use dissecting microscope or simple lab procedures (outlined in Tables 9.1 & 9.2 and HELPFUL HINTS) to answer questions A to D below.
- 4. Circle or highlight only one answer in Parts A, B, C, and D, as directed.
- 5. Then use the Flow chart key (Figures 8.2A, 8.2B, 8.2C) to identify specimens.

A: WHICH GROWTH FORM OCCURS? (seen by eve or with a dissecting microscope)

- 1. **Encrusting** and forming a **thin** filmy coating/layer (usually less than 0.2 mm thick) commonly on other algae but also found on other substrates (Figure 12.11, A–D)
- 2. **Foliose** with upright ridge-like branches arising from an encrusting base growing on the brown alga Carpophyllum (Figure 12.1, A & B)
- 3. Encrusting or layered plant with strut-like branches arising from the bottom of each laver (Figure 12.18, B & C)
- 4. Plate-like or disc-like and loosely attached to geniculate corallines (Figure 12.13, C)
- 5. Growing IN geniculate corallines with only tiny whitish conceptacles protruding above calcified host surface (viewed with dissecting microscope) (Figure 12.10, B & C)
- 6. A form other than the above five. Such plants may be warty, lumpy, fruticose, foliose, or some combination of these and either:

6a - grow on various substrates.

6b - occur as rhodoliths

GO TO PART B

B: WHICH REPRODUCTIVE STRUCTURES OCCUR?

(use a dissecting microscope or simple lab procedures – see Chapter 9)

1. Apparent uniporate conceptacles (without a corona) (Figure 7.6, A & C) (Figure 12.19, C & D) GO TO PART C

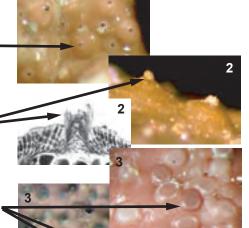
2. Apparent uniporate conceptacles with a corona (Figure 7.6, B) (Figure 12.7, D & E) GO TO FIGURE 8.2A

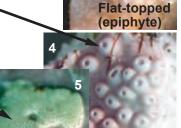
3. Flat-topped multiporate conceptacles (lacking a distinct rim and a central sunken pore plate) (Figure 7.7) (Figure 12.17, D & E) GO TO FIGURE 8.2A

4. Volcano-like multiporate conceptacles (each has a distinct rim and a central sunken pore plate) (Figure 7.7, A & B) (Figure 12.18, D & E) GO TO FIGURE 8.2A

5. Calcified compartments grouped into **sori** (Figure 7.8) (Figure 12.20, C & D) GO TO FIGURE 8.2A

6. Plants appear sterile: sori not evident (Figure 7.8) conceptacles not evident (Figures 7.6 & 7.7)GÓ TO FIGURE 8.2A



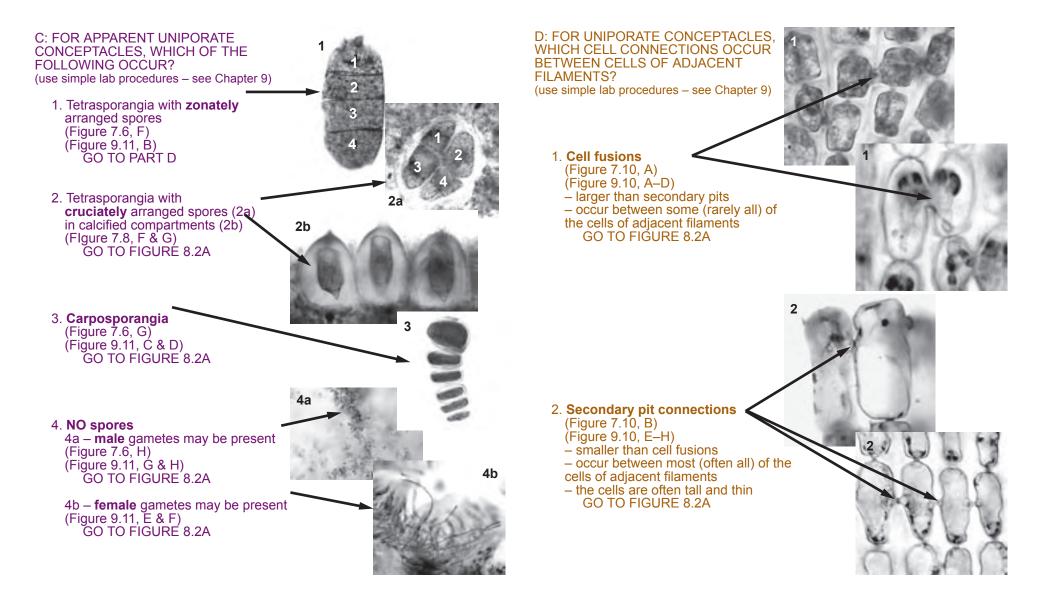


Flat-topped

Flat-topped



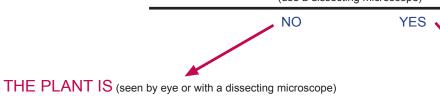
Table 8.1: continued



START HERE

ARE REPRODUCTIVE STRUCTURES PRESENT?

(use a dissecting microscope)



on various substrates

Further ID not

possible

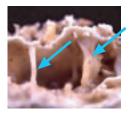
NOTE: Specimens not belonging to listed species may key out (albeit incorrectly) in this key.

NOTE: It is always recommended that identifications are ultimately confirmed with more involved lab procedures (see Chapters 10 and 11).

Encrusting or layered with

strut-like branches arising from the bottom of each layer

(Figure 12.18, B & C)



Synarthrophyton schielianum

(Go to Figure 12.18)

Foliose with upright ridge-like Of various growth forms branches arising from an encrusting base on the brown alga Carpophyllum

(Figure 12.1, A & B)



Lithophyllum carpophylli

(Go to Figure 12.1)

A: Apparent uniporate conceptacles (Figure 7.6)



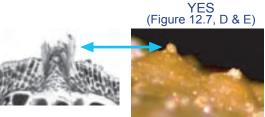
NO

GO TO FLOW CHART KEY

FIGURE 8.2B

IS A CORONA PRESENT?

(use a dissecting microscope or simple lab procedures)



Pneophyllum coronatum

(Go to Figure 12.7)

B: Flat-topped or volcano-like multiporate conceptacles (Figure 7.7)

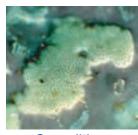
THE SPECIMEN HAS (use a dissecting microscope or simple lab procedures)



FIGURE 8.2C

Calcified compartments grouped into sori (Figure 7.8)

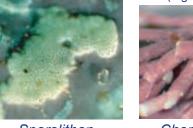
C:



Sporolithon durum

Tiny whitish conceptacles protruding from geniculate coralline algal host (Figure 12.10)

D:

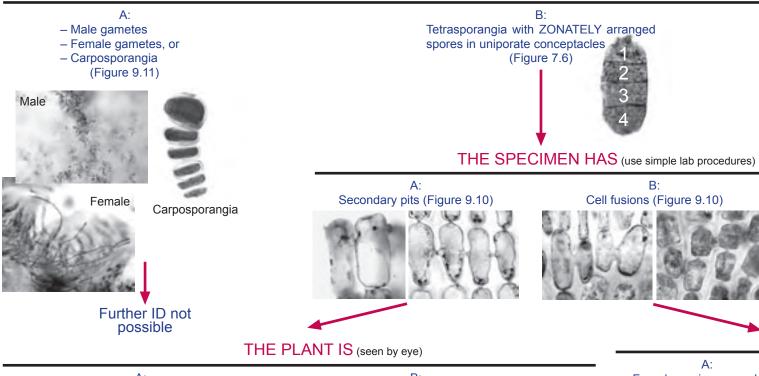


Choreonema thuretii

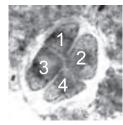
(Go to Figure 12.20) (Go to Figure 12.10)



APPARENT UNIPORATE CONCEPTACLES CONTAIN (use simple lab procedures)



C: Tetrasporangia with CRUCIATELY arranged spores in calcified compartments (Figure 7.8)

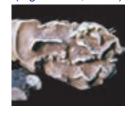


Heydrichia homalopasta (Go to Figure 12.19)

THE PLANTS ARE (seen by eye)

/ ose with unright ridge

Foliose with upright ridge-like branches arising from an encrusting base on the brown alga *Carpophyllum* (Figure 12.1, A & B)



Lithophyllum carpophylli
(Go to Figure 12.1)

B:
Of various sizes and growth forms,
not growing on *Carpophyllum*fronds

Lithophyllum corallinae (Figure 12.2)
Lithophyllum johansenii (Figure 12.3)
Lithophyllum pustulatum (Figure 12.4)
Lithophyllum stictaeforme (Figure 12.5)
(L. stictaeforme may occur on Carpophyllum holdfasts)

THIN SECTIONING REQUIRED FOR FURTHER ID (See Tabular key)

A:
Found growing on rocks or
shells (mussels, limpets, etc)
(Figure 12.9, A–C)



Spongites yendoi (Go to Figure 12.9)

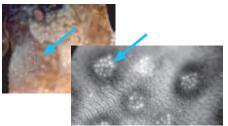
Found growing on other algae (Figure 12.8, A–C)



Pneophyllum fragile (Figure 12.8)
Hydrolithon improcerum (Figure 12.6)
THIN SECTIONING REQUIRED FOR
FURTHER ID (see Tabular key)

THE PLANTS ARE (use dissecting microscope or simple lab procedures)

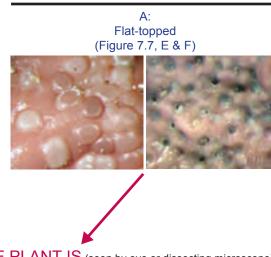
Encrusting, forming a thin layer on other algae and with tiny multiporate conceptacles (Figure 12.11, A-E)



Melobesia membranacea (Go to Figure 12.11)

Not as in A

THE MULTIPORATE CONCEPTACLES ARE (use simple lab procedures)



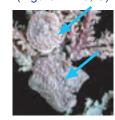
THE PLANT IS (seen by eye or dissecting microscope)

On rock/pebbles, with rounded crater-like depressions where conceptacles once occurred (Figure 12.16, A & C)



Phymatolithon repandum (Go to Figure 12.16)

B: Plate-like and loosely attached to geniculate coralline (Figure 12.13, C)

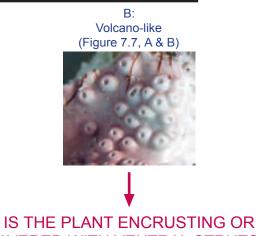


Mesophyllum erubescens (Go to Figure 12.13)

On various substrates, but without rounded crater-like depressions where conceptacles once occurred

Mesophyllum engelhartii (Figure 12.12) Mesophyllum erubescens (Figure 12.13) Synarthrophyton patena (Figure 12.17) Phymatolithon repandum (Figure 12.16)

> THIN SECTIONING REQUIRED FOR **FURTHER ID** (see Tabular key)



LAYERED WITH VENTRAL STRUTS? (seen by eye or dissecting microscope)

NO

Mesophyllum macroblastum (Go to Figure 12.14)

Mesophyllum printzianum (Go to Figure 12.15)

THIN SECTIONING REQUIRED FOR **FURTHER ID** (see Tabular key)

YES (Figure 12.18, A-C)



Synarthrophyton schielianum

(Go to Figure 12.18)

Chapter 9. Simple lab procedures

his chapter details the simple lab procedures that may be required to complete Table 8.1 and help identify specimens. Procedures generally involve the preparation of temporary slides of whole mounts or squashes of the thallus and reproductive structures.

CHEMICALS AND EQUIPMENT

General equipment for 'picking off'

dissecting microscope, adjustable light source, single edge razor blades (or craft knife or similar), small glass vials, fine forceps, drip tray, pencil, stickers, sellotape, nail polish or small brightly coloured stickers and glue (optional), disposable gloves (optional)

Extra equipment for preparing temporary slides slides, coverslips, pipettes and bulbs, tissues, compound microscope

General chemicals

Aniline blue, water soluble (also called acid blue 22) or methyl blue (also called acid blue 93) stain

Aniline blue, water soluble, Gurr Prod. 34003 Methyl blue, Sigma M6900

Working solution: 0.1 to 1% aqueous solution Note: either stain will do.

Nitric acid

Nitric acid 70%, Pronalys AR BSPNL733.2.5 Working solution: 0.6 M solution

SOLUTION PREPARATION

Before making whole mounts and squashes, prepare the two required solutions.

Aniline blue or methyl blue

A 1% aqueous solution is prepared by adding 1 g of stain powder to 100 ml distilled water. Slightly warm the water and stir to help the stain dissolve. Either stain will do.

(For those without access to the above stains, green or blue food colouring may be substituted, but staining times and effects may vary).

0.6 M nitric acid solution for decalcification

Slowly add 27 ml of 70% nitric acid to 1 l of water. Gently swirl to mix. (See Warning, below left, and always add acid to water when diluting from concentrated stock.)

(For those without access to nitric acid, household vinegar may be substituted for dilute nitric acid, but decalcification times may vary).

WARNING: CONCENTRATED (70%) **NITRIC** ACID IS CORROSIVE. IT MUST BE STORED AND HANDLED SAFELY. Store concentrated nitric acid flammable separately from goods. Consult your lab manager or workplace health and safety officer to confirm safe practices for storing and working with nitric acid.

PICKING OFF FERTILE FRAGMENTS

- 1. Familiarise yourself with coralline structure and reproduction. **This is essential.** The features you will be looking for are illustrated in Figures 7.6–7.10 and their appearance in whole mounts or squashes are also shown in Figures 9.10 & 9.11.
- 2. Examine the specimen with a dissecting microscope, and make appropriate notes.
 - Is there a mixture of plants in the collection? (see Figure 9.7)
 - Is there a mixture of uniporate conceptacles, multiporate conceptacles or sori in the collection?
 - Are there any features that can be **misinterpreted** as healthy/whole conceptacles or sori? e.g., old conceptacles with white roofs (see Figure 9.1), broken conceptacles (see Figure 9.2), worm/borer holes (see Figure 9.3), peeling/sloughing of the thallus surface (see Figure 9.4).
 - Does the material appear sterile?

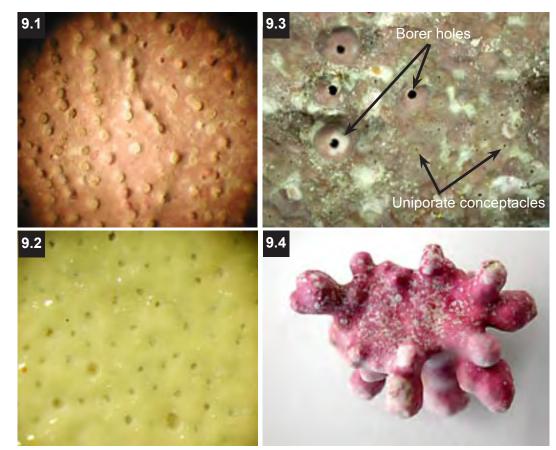


Figure 9.1: Old (white) conceptacles. Figure 9.2: Broken conceptacle roofs.

Figure 9.3: Borer/worm holes.

Figure 9.4: Thallus surface sloughing (white patches).

3. Decide what features you need to observe to complete Table 8.1 and the best method of observing them (see Tables 9.1 & 9.2). In general:

Whole mount

Make a whole mount to view the surface of conceptacles (are they uniporate or multiporate?).

Squash

Make a squash to see cell connections or the contents of conceptacles (are they male, female, or tetrasporangial?).

Flick tops off conceptacles

Flick tops off conceptacles, then whole mount to see the contents (are they male, female, or tetrasporangial?).

Scrape very thin corallines off algae/rock

Scrape very thin corallines off host algae or rock (after decalcifying) to see cell connections or the contents of conceptacles/compartments.

4. Remove pieces of coralline with whole/intact conceptacles/sori.

For very thin corallines growing on other algae

Cut the host (with coralline attached) into small squares about 4–10 mm long.

For very thin corallines growing on rock

Break off pieces of rock about 10–20 mm long (with coralline attached). Use razor blades, small chisel, or fingers, depending on the job.

For relatively thick corallines growing on rock or algae

Either:

A: **CAREFULLY** prise off pieces of coralline with whole/intact conceptacles. Use razor blades, small chisel, or fingers, depending on the job (see Figure 9.5). Remove a fertile piece of coralline about 4–10 mm long.

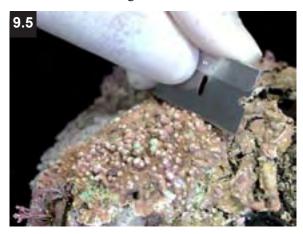


Figure 9.5: Using razor blade to prise coralline from substrate.

or

B: **CAREFULLY** prise off pieces of coralline with whole/intact conceptacles. Remove a fertile piece of coralline about 12–15 mm long. Flick the tops off (decapitate) conceptacles with the tip of a razor blade (see Figure 9.6).

Most of the contents should remain (for subsequent viewing) even after decalcification and staining.







Figure 9.6: Flicking the top off (decapitating) a conceptacle.

- 5. Note which sort of conceptacle you picked (uniporate, multiporate, sori) and identify the area you picked from by:
 - attaching a small brightly coloured sticker (using alcohol-resistant glue, e.g., Selley's Multi-Grip (NZ) or Tarzan's Grip (Australia)), or
- adding a spot of nail polish (see Figure 9.7) This can be invaluable if there is later confusion.

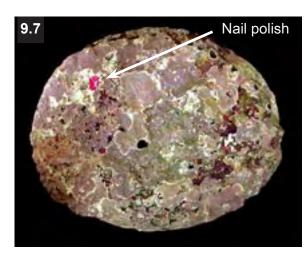


Figure 9.7: Mixed coralline cover, with 'picked' coralline marked with nail polish.

6. When picking:

- pick off at least 4 fertile fragments per collection (you may need to use a number of techniques)
- part of the thallus will also be needed for specimen identification this is usually picked off with the reproductive structures
- pieces should be small enough and thin enough to fit under a coverslip. (As a general rule about 4–10 mm long)
- pieces should also be free of sand, rock or unnecessary sterile thallus
- 7. Place fragments in small (about 22 mm diameter) glass vials.
- 8. Write the specimen number and other useful information IN PENCIL (pencil is alcohol resistant) on a sticker, and use clear tape to secure the label to the glass vial.

DECALCIFYING FRAGMENTS

- 1. Half fill glass vials containing specimen fragments with 0.6 M nitric acid.
- 2. Leave coralline in acid until gas bubbles cease forming. This may take 30–60 min for thin material and 2–3 h for somewhat thicker material.
- 3. Change acid every 30–60 min as needed. A lack of gas bubbles can be caused by an exhausted acid supply, so always replace with fresh acid for a few minutes before assuming the sample is decalcified.
- 4. Remove acid from vial with a pipette or suction device. Tilt the vial and remove acid from the upper edge of the bottom of the vial or small pieces may be sucked away.
- 5. Rinse **gently** with water.

SLIDE PREPARATION

Be patient

- You won't always see what you are looking for in the first slide.
- A number of different techniques may be needed.
- Allow for variation in how things appear compared to photos.
- Look carefully at the slides; some features can be quite small and difficult to see initially.
- Some features can be easily seen without staining, others may require staining.
- Before making slides, read the helpful hints (over page).

Whole mounts

- 1. It is often useful to look at the fertile fragments with a dissecting microscope first to see where and how many conceptacles there are.
- 2. Place the whole specimen on a slide (add water and a coverslip) i.e., whole mount specimen (see Figure 9.8).

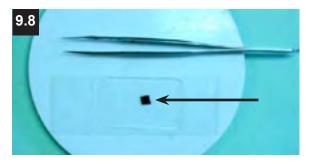


Figure 9.8: Stained specimen (arrow) on microscope slide under coverslip.

- 3. View with a microscope.
- 4. If staining required stain a fertile fragment for
- 5–20 seconds, **gently** rinse with water, and repeat the above process.

Squashing to see contents of conceptacles

- 1. It is often useful to look at the fertile fragments with a dissecting microscope first to see where and how many conceptacles there are.
- 2. Place specimen on a slide (add water and a coverslip).
- 3. View with a microscope.
- 4. Find conceptacles and squash by pressing on coverslip with forceps or pencil (see Figure 9.9).

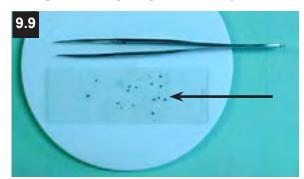


Figure 9.9: Squashed specimen fragments (arrow) on microscope slide under coverslip.

- 5. View with a microscope.
- 6. If staining required stain a new fertile fragment for 5–20 seconds, **gently** rinse with water, and repeat the above process.

Squashing to see cell connections

Cell connections are best viewed in **side view** (not surface view) and must be **one cell thick**.

- 1. Either:
 - place specimen on a slide (add water and a coverslip) and roll the thallus on its side as you squash it to one cell thick

OR

- make thin sections of the thallus first with a razor blade, then place these on their side on a slide and squash to one cell thick
- 2. View with a microscope.
- 3. If staining required stain a new fragment for 5–20 seconds, **gently** rinse with water, and repeat the above process.

Whole mounts of 'decapitated' conceptacles

- 1. It is often useful to look at the fertile fragments with a dissecting microscope first to see where and how many 'decapitated' conceptacles there are.
- 2. Place specimen on a slide (add water and a coverslip).
- 3. View with a microscope.
- 4. If staining required stain a new fertile fragment for 5–20 seconds, **gently** rinse with water, and repeat the above process.

Scraping thin corallines off algae or rock

- 1. Scrape thin coralline off the host alga or rock and place scrapings on a slide (add water and a coverslip).
- 2. View with a microscope.
- 3. If staining required stain a new fertile fragment for 5–20 seconds, **gently** rinse with water, and repeat the above process.

HELPFUL HINTS & CAUTIONS

Reproductive structures

- View in surface view to see if uniporate or multiporate.
- Squash or knock tops off to view contents of uniporate conceptacles (are they male, female, or tetrasporangial?).
- Try to have a number of reproductive structures on each fertile fragment.

Cell connections

- Best viewed in side view (not surface view).
- Best viewed under x100 lens using immersion oil.
- Squashes need to be flat.
- Squashes need to be **one cell thick**.

Cell fusions

- Larger than secondary pit connections.
- Occur between some (not all) of the cells of **adjacent** filaments (Figure 9.10, B).
- The cells are often short and squat (Figure 9.10, B).
- May give the cells a 'dumbbell' or 'H' appearance (Figure 7.10, A; Figure 9.10, B–D).

Secondary pit connections

- Smaller than cell fusions.
- Occur between most (often all) of the cells of **adjacent** filaments (Figure 9.10, E).
- The cells are often tall and thin (Figure 9.10, G & H).
- Look for tiny 'arms/outgrowths' (secondary pits) joining the cells (Figure 9.10, F).
- Finding and recognising secondary pit connections requires patience, and adjusting the focus.

Thin epiphytic corallines

Turn the light on the microscope up high, and open the iris diaphragm. This will allow light to shine through the host so you can see the epiphytic coralline.

Columella

The columella (central filaments) of uniporate tetrasporangial conceptacles can look similar to trichogynes and carpogonia of female conceptacles.

Starch grains

Starch grains (darkly stained bodies) may be observed in cells when looking for cell connections (Figure 7.10, A).

TABLES 9.1 & 9.2

Tables 9.1 and 9.2 (pp. 66–67) indicate the ways in which particular information can be obtained using the procedures in this chapter. The tables list examples of figures where each feature is illustrated (often Figures 9.10 and 9.11, but also other places throughout the guide).

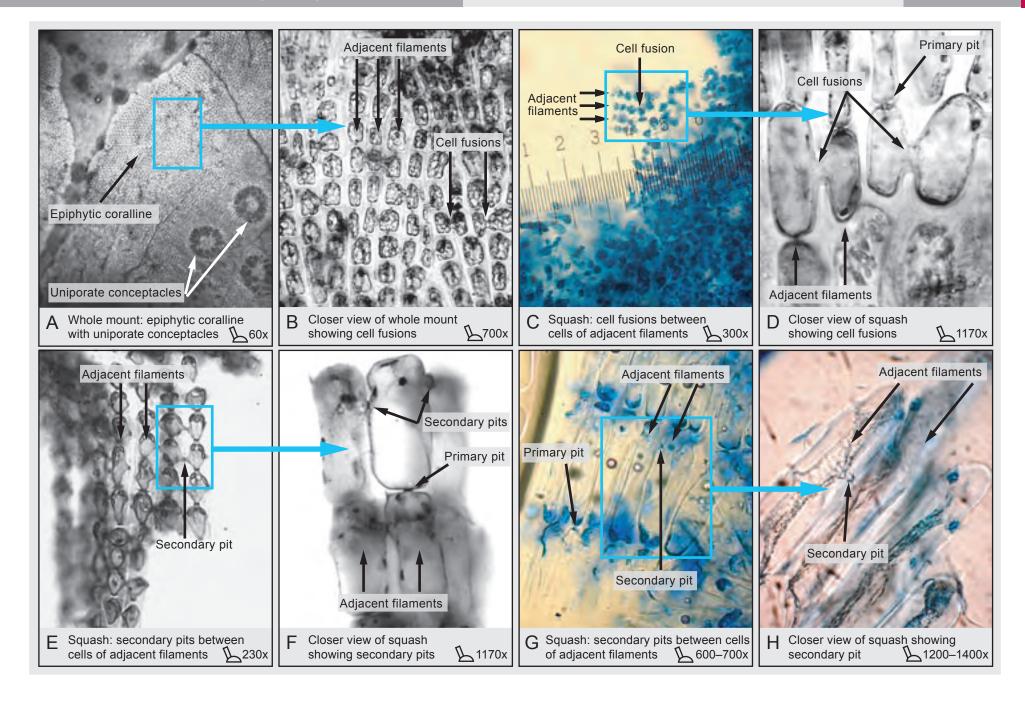
Table 9.1: Simple lab procedures used to observe features in THICKER plants with large conceptacles, with reference to relevant illustrations.

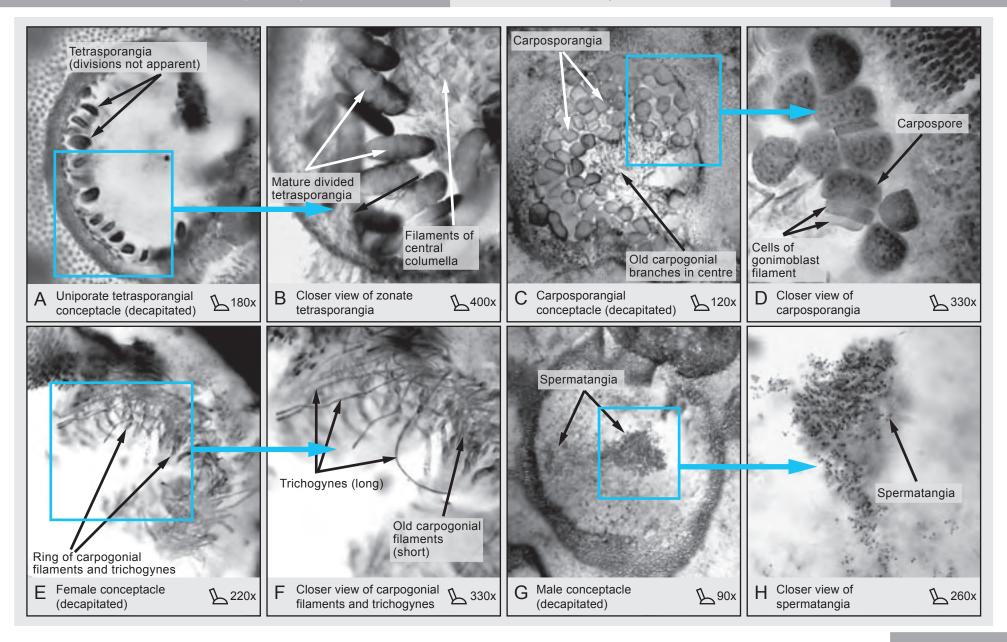
	Apparent uniporate conceptacles	Flat-topped multiporate conceptacles	Volcano-like multiporate conceptacles	Sori	Calcified compartments	Zonately (Z) or cruciately (C) arranged tetraspores	Carposporophyte (gonimoblast filaments and carpospores)	Carpogonia (females)	Spermatangia (males)	Cell fusions	2º pits
Dissecting microscope	Figure 7.6, A–D	Figure 7.7, E & F Figure 12.17, D & E	Figure 7.7, A & B Figure 12.18, D & E	Figure 7.8, A & B Figure 12.20, C & D	NO (only pores of possible compartments) Figure 7.8, D Figure 12.19, C & D						
Knock tops off conceptacles and whole mount – or whole mount sori				Figure 7.8, C		Figure 9.11, B (Z)	Figure 9.11, C & D	Figure 9.11, E & F	Figure 9.11, G & H		
Lightly squash sample				Figure 7.8, E & F	Figure 7.8, E–G	Figure 7.6, F (Z) Figure 7.8, F & G (C)	Figure 7.6, G		Figure 7.6, H	Figure 9.10, C & D	Figure 9.10, E–H

Table 9.2: Simple lab procedures used to observe features in VERY THIN plants with small conceptacles, with reference to relevant illustrations.

	Apparent uniporate conceptacles	Multiporate conceptacles	External conceptacles of Choreonema	Sori	Calcified compartments	Zonately (Z) or cruciately (C) arranged tetraspores	Carposporophyte and carpogonia	Corona	Spermatangia	Cell fusions	2º pits
Dissecting microscope	Figure 12.6, C & D Figure 7.6, D	Figure 7.7, G Figure 12.11, D & E	Figure 12.10, B & C	Figure 7.8, A & B	NO (only pores of possible compartments) Figure 7.8, D Figure 12.19, C & D		Difficult to see in very thin plants with small conceptacles	Figure 7.6, B	Difficult to see in very thin plants with small conceptacles		
Whole mount	Figure 7.6, E	Figure 7.7, H Figure 12.11,	Figure 12.10, D	Figure 7.8, C			Difficult to see in very thin plants with small conceptacles	Figure 12.7, E	Difficult to see in very thin plants with small conceptacles	Figure 9.10, A & B	Hard to see
Whole mount and lightly squash		Figure 12.16, E		Figure 7.8, E	Figure 7.8, E–G	Figure 7.6, F (Z) Figure 7.8, F & G (C)	Difficult to see in very thin plants with small conceptacles		Difficult to see in very thin plants with small conceptacles	Figure 9.10, C & D	Figure 9.10, E–H
Scrape off with razor after decalcifying					Figure 7.8, E–G	Figure 7.6, F (Z) Figure 7.8, F & G (C)	Difficult to see in very thin plants with small conceptacles		Difficult to see in very thin plants with small conceptacles	Figure 9.10, C & D	Figure 9.10, E-H

Columns in Table 9.2 with coloured backgrounds denote a column heading differing from that in Table 9.1. Note that there are 2 columns detailing multiporate conceptacles in Table 9.1, but only one in Table 9.2.





Chapter 10. Specimen identification – using more involved lab procedures

Slides prepared from embedded and sectioned material (see Chapter 11) allow complete and definite identification.

This chapter deals with the identification of specimens using the Dichotomous and Tabular keys set out below. Before using these keys it is first necessary to embed and thin section specimens with a microtome (see Chapter 11) to allow detailed examination of the vegetative and reproductive anatomy. These related chapters (Chapters 10 & 11) are intended for use by those with some algal taxonomy training and access to specialised chemicals and equipment (L.R. White embedding resin, microtome, etc.), although people using simple lab procedures can use the keys, with limitations.

Where text and images are pertinent to both simple and more involved lab procedures, they are repeated in both sections so that each chapter works as a standalone resource

LIMITATIONS OF SPECIMEN IDENTIFICATION

Although the sampling programme upon which this guide is based was extensive, only a small fraction of the **total** central New Zealand coast was covered, and most sampling sites were visited only once. This means that other species occurring in the region may have escaped detection, and any such species (even after embedding and sectioning with a microtome) will not be able to be identified using the keys below.

Good quality fertile specimens (especially tetrasporangial or bisporangial plants) that cannot be readily identified should **not** be discarded. Rather, they should be deposited in a registered New Zealand herbarium with suitable notes (see Chapter 6) so that they can be further assessed by specialists, particularly in the context of monographic studies.

STEPS IN SPECIMEN IDENTIFICATION

The steps involved in the identification process are listed below. Keep in mind that not every specimen will be identifiable, and misidentifications are possible.

- 1. Become familiar with coralline structure and function (Chapter 7), including
 - growth forms and substrates
 - sexual cycle
 - reproductive structures
 - cell fusions and secondary pit connections

This is essential.

2. For most identifications to species, specimens must be fertile and the reproductive structures should be tetra/bisporangial (contain tetra/bispores) (see Coralline sexual cycle, Figure 7.5).

Remember:

- multiporate conceptacles are always tetrasporangial (see Figure 7.7) or bisporangial
- calcified compartments are always tetrasporangial (see Figure 7.8)
- uniporate conceptacles may be tetrasporangial, male, female, or carposporangial (see Figure 7.6)
- male, female, and carposporangial plants are usually unable to be identified to species level in the absence of tetrasporangial plants
- sterile plants are usually unable to be identified to species level
- misidentifications are also possible (see Limitations of the guide in Chapter 1)

- 3. Corallines often grow intermixed (different plants very close to one another on the same substrate). As a result, even though every effort is made to keep only one thing/species, there can often be a number of plants in a single collection.
 - The rule of thumb is to identify whatever appears to be the major or most common plant/species in a collection (see Figure 10.1), and/or denote the 'identified' plant using nail polish or similar.



Figure 10.1: Rock with mixed coralline cover. Major plant in collection is arrowed.

- 4. Information for specimen identification will need to be obtained both from direct observation of plants using a dissecting microscope, and the examination of the prepared slides.
- 5. Study the dichotomous and tabular keys in this chapter to:
 - determine which characters can be assessed through direct examination of the specimen with the unaided eye or with a dissecting microscope (in **black** text in the keys)

- determine which characters can be assessed only from examination of the prepared slides with a compound microscope (in red text in the keys)
- 6. Examine, embed, and section material as outlined in Chapter 11. You will usually need to section:
 - conceptacles or calcified compartments for important reproductive features
 - the thallus for important vegetative features
- 7. Use either the Dichotomous key OR the Tabular key (see below) to identify the specimen.
- 8. Use the Species profiles to support or reject your identification (Chapter 12).

KEYS

The keys are based on the same information, but differ in how they are used. While many people will be familiar with dichotomous keys, a significant advantage of tabular keys is that the user is less likely to get stuck – if a particular character cannot be seen in a specimen you can easily move on to another character and eventually come to a final identification.

Although the dichotomous and tabular keys include many features observable only in sectioned material (red text), a number of characters can be observed using simple lab procedures (black text) and people without specialised equipment and prepared slides can use these keys, with limitations.

Using the Dichotomous key (10.1)

- 1. Start with the first decision pair.
- 2. The answer to this couplet will lead to a further decision pair in a set order until a final identification is reached.
- 3. The relevant Species profile in Chapter 12 should then be consulted to support or reject the identification.

Using the Tabular keys (10.2 & 10.3)

- 1. First ascertain which key you will need to use (Key 10.2 for specimens with uniporate tetrasporangial conceptacles and Key 10.3 for specimens with multiporate tetrasporangial conceptacles, calcified compartments, or which are endophytic).
- 2. Start with any feature listed in the appropriate key and determine which species possess that character state.
- 3. Then choose another character and repeat the process, using only those species flagged using the previous character(s).
- 4. Continue until all but one species is eliminated. The relevant Species profile in Chapter 12 should then be consulted to support or reject the identification.

Key 10.1: Dichotomous key.

Note: Plants with tetrasporangial conceptacles are essential for identification and plants with male conceptacles are also required in one case (11. *Synarthrophyton patena* cf *Mesophyllum engelhartii*). Characters evident using simple lab procedures are in black; characters evident only in thin sections are in red. Because emphasis is placed on characters observable with simple lab procedures, not all species in a given genus are necessarily grouped together. The characters employed are useful for identifying specimens, but are not necessarily diagnostic of the species or genus.

1. 1.	Vegetative thallus entirely endophytic in geniculate corallines, with external colourless conceptacles (Figure 12.10, B & C)
2. 2.	Tetrasporangia with cruciately arranged spores, borne in calcified compartments that may be grouped into sori or individually scattered (Figure 7.8)3 Tetrasporangia with zonately arranged spores, borne in conceptacles (Figures 7.6 & 7.7)
3.3.	Calcified tetrasporangial compartments grouped into sori (Figure 7.8, A–C & E); compartments not surrounded by an involucre , sporangia on a single-celled stalk (Figure 12.20, E)
4. 4.	Tetrasporangial conceptacles with multiporate roofs (Figure 7.7)
5. 5.	Conceptacles volcano-like (with a distinct rim surrounding a central sunken pore plate) (Figure 7.7, A & B)
6.6.	Thallus producing struts from the ventral surface (Figure 12.18, B & C); male conceptacles with both branched and unbranched spermatangial filaments in the same conceptacle (Figure 7.9, B & D). Synarthrophyton schielianum Thallus not producing struts from the ventral surface; male conceptacles with unbranched spermatangial filaments only (Figure 7.9, A & B)
7. 7.	Tetrasporangial conceptacle pore canals bordered by cells (especially near the base of the canal) that are more elongate than other cells in the conceptacle roof (Figure 12.15, G). Mesophyllum printzianum Tetrasporangial conceptacle pore canals bordered by cells that are similar in size and shape to other cells in the conceptacle roof (Figure 12.14, G). Mesophyllum macroblastum

Key 10.1: Dichotomous key (continued)

8.	Mature thallus 'small', 'thin' and encrusting, growing on other algae (epiphytic) (Figure 12.11, A–D); thallus construction dimerous (Figure 7.10, H)
8.	Mature thallus 'larger' and 'thicker', of various growth forms and various substrates; thallus construction monomerous (Figure 7.10, G)9
9.	Tetrasporangial conceptacle pore canals bordered by cells (especially near the base of the canal) that are more elongate than other cells in the conceptacle roof (Figure 12.13, H)
9.	Tetrasporangial conceptacle pore canals bordered by cells that are similar in size and shape to other cells in the conceptacle roof (Figure 12.12, E)10
10. 10.	Subepithallial initials as short as or shorter than the cells immediately subtending them (Figure 7.10, F). Phymatolithon repandum Subepithallial initials as long as or longer than the cells immediately subtending them (Figure 7.10, E). 11
11.	Male conceptacles with both branched and unbranched spermatangial filaments in the same conceptacle (Figure 7.9, B & D) Synarthrophyton patena
	Male conceptacles with unbranched spermatangial filaments only (Figure 7.9, A & B)
	Cells of adjacent vegetative filaments connected by secondary pits (Figure 7.10, B; Figure 9.10, E–H)
13.	Plants with distinct foliose growth form (upright branches arising from an encrusting base) (Figure 12.1, A & B); in central New Zealand found on fronds of <i>Carpophyllum</i> spp
13.	Plants varying in growth form and substrates, not foliose and not growing on <i>Carpophyllum</i> fronds
	Tetrasporangial conceptacle pore canals completely blocked by 2 or 4 enlarged cells (Figure 12.3, E & F); conceptacle chambers usually 95–145 μm in diameter. Lithophyllum johansenii
14.	diameter. Tetrasporangial conceptacle pore canals not completely blocked by enlarged cells (Figure 12.4, E & F); conceptacle chambers usually over 185 μm in diameter. 15

Key 10.1: Dichotomous key (continued)

	Floors of tetrasporangial conceptacle chambers usually situated 3–4 (6) cell layers below surrounding thallus surface; conceptacle roofs commonly 1–2 cells thick above chamber (Figure 12.4, E & F). Lithophyllum pustulatum Floors of tetrasporangial conceptacle chambers usually situated 6 or more layers below the surrounding thallus surface; conceptacle roofs commonly 3–6 cells thick above chamber (Figure 12.5, F & G). 16
	Tetrasporangial conceptacle chambers usually 200–240 μm in diameter; conceptacle roofs commonly 3–4 cells thick above chamber (Figure 12.2, E & F). Lithophyllum corallinae Tetrasporangial conceptacle chambers usually 290–420 (450) μm in diameter; conceptacle roofs commonly 3–6 cells thick above chamber (Figure 12.5, F & G). Lithophyllum stictaeforme
	Plants growing on rock or shells (Figure 12.9, A–C) (ancillary character); tetrasporangial conceptacle roofs formed by filaments peripheral to sporangial initials (developmental feature not often seen – see taxonomic notes, Figure 12.9). Spongites yendoi Plants growing on other algae (Figure 12.7, A–C) (ancillary character); tetrasporangial conceptacle roofs formed by filaments peripheral to and interspersed amongst sporangial initials (developmental feature not often seen – see taxonomic notes, Figures 12.7 & 12.8). 18
	Tetrasporangial conceptacle pores with a corona of filaments that protrude above the thallus surface (Figure 12.7, D–F)
19.	Tetrasporangial conceptacle pore canals lined by cells that are orientated more or less perpendicular to the thallus surface (vertical orientation) and do not protrude laterally into the pore canal (Figure 12.6, E & F); thallus composed of overlapping layers that are commonly two cells thick (Figure 12.6, G)
19.	Tetrasporangial conceptacle pore canals lined by cells that are orientated more or less parallel to the thallus surface (horizontal orientation) and protrude laterally into the pore canal (Figure 12.8, G); thallus lacking overlapping layers that are two cells thick

Key 10.2: Tabular key to species of Corallinaceae (subfamilies Lithophylloideae and Mastophoroideae) in this guide.

Note: this key is for plants for which BOTH of the following are true: A. NOT endophytic, and B. possess uniporate tetrasporangial conceptacles.

Characters evident using simple lab procedures are in black; those requiring thin sections are in red. Characters are useful for identification but are not necessarily diagnostic of the listed species. Specimens not belonging to listed species will not key out satisfactorily.

5. possess uniporate tetrasporarigiar conceptacies.												
	Connections between cells of adjacent filaments (Figure 7.10)	Plants with distinct foliose growth form on Carpophyllum fronds (Figure 12.1)	Plants usually thin, encrusting and epiphytic (see individual species profiles)	Tetrasp. conc. pore with a corona protruding above the thallus (Figure 12.7)	Tetrasp. conc. pore canals completely blocked by enlarged cells (Figure 12.3)	Orientation of cells bordering tetrasp. conc. pore canals (see individual species profiles)	Diameter of tetrasp. conc. chambers in μm	Tetrasp. conc. roof formed by filaments (developmental feature not often seen)	Thallus consists of overlapping layers that are two cells thick (Figure 12.6)	Floors of tetrasp. conc. x cell layers below surface	Roofs of tetrasp. conc. x cell layers thick (see individual species profiles)	
Corallinaceae												
Lithophylloideae												
Lithophyllum carpophylli	2º pits	yes	no	no	no	n/a	185–270	uncertain	no	4–6	1–2	
Lithophyllum corallinae	2º pits	no	no	no	no	n/a	200–240	uncertain	no	6 or more	3–4	
Lithophyllum johansenii	2º pits	no	no	no	yes	n/a	95–145	uncertain	no	6–8	1–2	
Lithophyllum pustulatum	2º pits	no	yes	no	no	n/a	185–300	uncertain	no	3–4 (6)	1–2	
Lithophyllum stictaeforme	2º pits	no	no	no	no	n/a	290–420 (450)	uncertain	no	6 or more	3–6	
Corallinaceae												
Mastophoroide	ae											
Hydrolithon improcerum	cell fusions	no	yes	no	no	more or less perpendicular to roof surface	135–180	peripheral & interspersed amongst sporangial initials	yes	n/a	n/a	
Pneophyllum coronatum	cell fusions	no	yes	yes	sometimes by the corona	more or less parallel to roof surface	120–250 (345)	peripheral & interspersed amongst spor- angial initials	no	n/a	n/a	
Pneophyllum fragile	cell fusions	no	yes	no	no	more or less parallel to roof surface	55–205	peripheral & interspersed amongst sporangial initials	no	n/a	n/a	
Spongites yendoi	cell fusions	no	no	no	no	more or less parallel to roof surface	120–255	peripheral to sporangial initials	no	n/a	n/a	

n/a = not applicable

Key 10.3: Tabular key to species of Hapalidiaceae and Sporolithaceae in this guide.

Note: this key is for plants that possess ONE of the following features: A. an endophytic thallus with external colourless conceptacles, or B. multiporate tetrasporangial conceptacles, or C. calcified compartments that are scattered or grouped into sori.

Characters evident using simple lab procedures are in black; those requiring thin sections are in red. In several cases, both tetrasporangial and male plants are needed for full identification. Characters are useful for identification but are not necessarily diagnostic of the listed species. Specimens not belonging to listed species will not key out satisfactorily.

	Vegetative thallus entirely endophytic (external conceptacles) (Figure 12.10)	Thallus with ventral struts (Figure 12.18)	Mature thallus small, thin and epiphytic (Figure 12.11)	Spores zonate or cruciate (Figures 7.6 & 7.8)	Tetrasp. housed in: (Figures 7.7 & 7.8)	Tetrasp. conc. roof/plate morphology (Figures 7.7 & 12.10)	Thallus construction (Figures 7.1 & 7.10)	Subepithallial initials compared to subtending cells (Figure 7.10)	Tetrasp. conc. pore canal cells compared to other roof cells (see species profiles)	Sperm. filaments (Figure 7.9)	Calcified comp. with: (Figures 12.19 & 12.20)
Hapalidiaceae											
Choreonema thureti	yes	no	no	zonate	conceptacles	multiporate plate hidden beneath single outer opening	unconsolidated filaments	n/a	n/a	unbranched	n/a
Melobesia membranacea	no	no	yes	zonate	conceptacles	flat-topped	dimerous	uncertain	all cells similar	unbranched	n/a
Mesophyllum engelhartii	no	no	no	zonate	conceptacles	flat-topped	monomerous	as long or longer	all cells similar	unbranched	n/a
Mesophyllum erubescens	no	no	no	zonate	conceptacles	flat-topped	monomerous	as long or longer	basal cells elongate	unbranched	n/a
Mesophyllum macroblastum	no	no	no	zonate	conceptacles	volcano-like	monomerous	as long or longer	all cells similar	unbranched	n/a
Mesophyllum printzianum	no	no	no	zonate	conceptacles	volcano-like	monomerous	as long or longer	basal cells elongate	unbranched	n/a
Phymatolithon repandum	no	no	no	zonate	conceptacles	flat-topped	monomerous	as short or shorter	all cells similar	branched & unbranched	n/a
Synarthrophyton patena	no	no	no	zonate	conceptacles	flat-topped	monomerous	as long or longer	all cells similar	branched & unbranched	n/a
Synarthrophyton schielianum	no	yes	no	zonate	conceptacles	volcano-like	monomerous	as long or longer	all cells similar	branched & unbranched	n/a
Sporolithaceae											
Heydrichia homalopasta	no	no	no	cruciate	calcified compartments scattered (not in sori	n/a	monomerous	mostly similar in size	n/a	unbranched	5-celled stalk and involucre
Sporolithon durum	no	no	no	cruciate	calcified compartments grouped into sori	n/a	monomerous	mostly similar in size	n/a	branched	single-celled stalk & lacking involucre

Chapter 11. More involved lab procedures - thin (microtome) sectioning and permanent slides

This chapter details more involved lab procedures used to thin (microtome) section material and prepare permanent slides to help identify specimens using the keys in Chapter 10. These methods assume you'll be working with specimens preserved in glycerol-ethanol solution after initial formalin preservation (detailed in Chapter 6), but can also be applied to fresh or air-dried specimens.

The procedures describing how to pick off and decalcify fertile fragments are similar to – but slightly different from – those in Chapter 9. They are repeated here for completeness and clarity.

CHEMICALS AND EQUIPMENT

General equipment for 'picking off'

dissecting microscope, adjustable light source, single edge razor blades (or craft knife or similar), glass vials (about 22 mm diameter at base), fine forceps, drip tray, pencil, stickers, sellotape, nail polish or small brightly coloured stickers and glue (optional), disposable gloves (optional)

Extra equipment for embedding and sectioning

fume hood, oven (set at 60–65 °C), wooden boards (to support blood trays in the oven), slides, coverslips, pipettes and bulbs, tissues, compound microsope, hotplate, brass weights, red heat lamp (250 W), small paint brush

Baskets with mesh inserts

plastic tubes (about 40 mm long, about 10 mm diameter) with mesh bases
Baskets have proved difficult to source commercially.

They are not essential, but help retain and isolate fragments of specimen within glass vials.

Blood trays

PVC, about 250 μm thick, with circular wells (depressions) about 20 mm wide and 7 mm deep Blood trays have proved difficult to source commercially, and we have had them purpose-made by plastics manufacturers, from bespoke moulds, in both Australia (c. 1995) and New Zealand (Award Plastics, Christchurch, 2005). Commercially available histological moulds approppriate for resin embedding and curing at 60 °C may be an alternative.

Melinex coverslips

Melinex 539, 100 μm thick Sourced from Award Plastics, Christchurch, NZ.

Microtome

While any microtome that allows $8-12~\mu m$ sections is suitable for involved lab procedures, the microtome used to section corallines in this study was an American Optical Corporation (Buffalo, New York, USA) Sledge microtome, Model 860.

General chemicals

Potassium permanganate (KMnO₄)

Working solution: 5% aqueous solution

Nitric acid

Nitric acid 70%, analytical reagent Working solution: 0.6 M solution

Ethanol

Working solutions: various – see text

L.R. White resin, medium grade

London Resin Co., Reading, Berkshire, England

Histo-Clear

National Diagnostics, Atlanta, Georgia

Eukitt mounting medium

O. Kindler, Freiburg, Germany

SOLUTION PREPARATION

Before starting, prepare all solutions.

0.6 M nitric acid solution for decalcification Slowly add 27 ml of 70% nitric acid to 1 l of water.

Slowly add 27 ml of 70% nitric acid to 1 l of water Gently swirl to mix.

5% aqueous potassium permanganate (KMnO₄)

Add 5 g of stain powder to 100 ml distilled water. Slightly warm the water and stir to help the stain dissolve

30%, 60%, 90%, and 100% ethanol solutions

All ethanol solutions are v/v in water.

WARNING: CHEMICALS AND EQUIPMENT USED IN THESE PROCEDURES MAY BE HAZARDOUS. CHEMICALS MUST BE STORED AND HANDLED SAFELY.

Consult your lab manager or workplace health and safety officer to confirm safe practices for storing and working with chemicals and equipment.

PICKING OFF FERTILE FRAGMENTS

- 1. Familiarise yourself with coralline structure and reproduction. (The reproductive features to look for are illustrated on Figures 7.6–7.8). **This is essential.**
- 2. Examine the specimen with a dissecting microscope, and make appropriate notes.
 - Is there a mixture of plants in the collection? (see Figure 11.6)
 - Is there a mixture of uniporate conceptacles, multiporate conceptacles or sori in the collection?
 - Are there any features that can be **misinterpreted** as healthy/whole conceptacles or sori? e.g., old conceptacles with white roofs (see Figure 11.1), broken conceptacles (see Figure 11.2), worm/borer holes (see Figure 11.3), peeling/sloughing of the thallus surface (see Figure 11.4).
 - Does the material appear sterile?

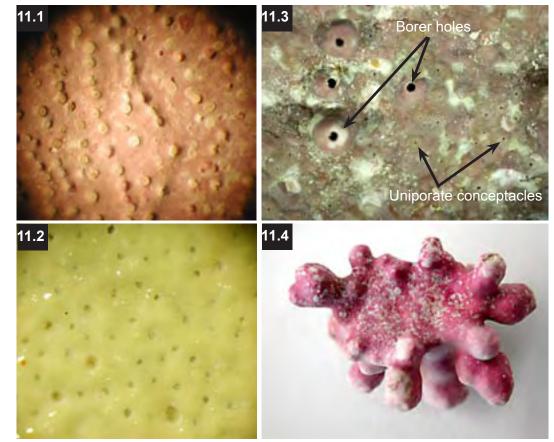


Figure 11.1: Old (white) conceptacles. Figure 11.2: Broken conceptacle roofs.

Figure 11.3: Borer/worm holes.

Figure 11.4: Thallus surface sloughing (white patches).

3. Remove pieces of coralline with whole/intact conceptacles/sori. The features you will need to observe relate to reproductive structures and the associated vegetative thallus.

For very thin corallines growing on other algae

Cut the host (with coralline attached) into small squares about 4–10 mm long.

For very thin corallines growing on rock

Scrape off the coralline (with whole/intact conceptacles) using a razor blade as best you can. Remove enough coralline to work with – you will lose much of it during processing.

For relatively thick corallines growing on rock or algae

CAREFULLY prise off pieces of coralline with whole/intact conceptacles or sori (see Figure 11.5). Use razor blades, small chisel, or fingers. Remove a fertile piece of coralline about 4–10 mm long.



Figure 11.5: Using razor blade to prise coralline from substrate.

- 4. Note which sort of conceptacle you picked (uniporate, multiporate, sori) and identify the area you picked from by:
 - attaching a small brightly coloured sticker (using alcohol-resistant glue, e.g., Selley's Multi-Grip (NZ) or Tarzan's Grip Super Glue (Australia)), or
- adding a spot of nail polish (see Figure 11.6) This can be invaluable if there is later confusion.



Figure 11.6: Mixed coralline cover, with 'picked' coralline marked with nail polish.

- 5. When picking:
 - pick off at least 4 fertile fragments per collection
 - part of the thallus will also be needed for specimen identification – this is usually picked off and sectioned next to/with the reproductive structures
 - pieces should be about 4–10 mm long
 - pieces should also be free of sand, rock, or unnecessary sterile thallus

6. Place fragments in small baskets with a mesh/strainer on one end (see Figure 11.7), and place the baskets inside small (about 22 mm diameter) glass vials. If baskets are unavailable, place specimens directly into glass vials. About half fill vials with 0.6 M nitric acid.



Figure 11.7: Baskets (plastic, ~40 mm long and ~10 mm diameter) with mesh/strainer inserts.

- 7. Write the specimen number and other useful information IN PENCIL (pencil is ethanol resistant) on a sticker and use clear tape to secure the label to the glass vial.
- 8. Refill the specimen jar with glycerol-ethanol if any has evaporated.

DECALCIFYING FRAGMENTS

- 1. Make sure vials are about half full of 0.6 M nitric acid.
- 2. Leave coralline in acid until gas bubbles cease forming. This may take 30–60 min for thin material and 2–3 h for thicker material
- 3. Change acid every 30–60 min as needed. A lack of gas bubbles can be caused by an exhausted acid supply, so always replace with fresh acid for a few minutes before assuming the sample is decalcified.
- 4. Remove acid from vial with a pipette or suction device. Tilt the vial and remove acid from the upper edge of the bottom of the vial; otherwise small pieces may be sucked away. If you are using baskets inside the vials, these will help retain the specimen.
- 5. Rinse **gently** with water.

STAINING

- 1. Remove water from vial.
- 2. Cover specimens with 5% potassium permanganate for 20 min.
- 3. Remove stain with a pipette or suction device. Rinse **gently** with water.

DEHYDRATION

- 1. Remove water from vial.
- 2. Add 30% ethanol solution. Remove after 30 min.
- 3. Add 60% ethanol solution. Remove after 30 min.

- 4. Add 90% ethanol solution. Remove after 30 min.
- 5. Add 100% ethanol solution.
- 6. The specimen is now ready for L.R. White embedding.

NOTE: Do not leave specimens in higher concentrations of ethanol in a vial without a stopper for long periods. Once dried, the material is unsuitable for microscopy. Do not leave in acid for excessive amounts of time, as this will also destroy the tissue.

EMBEDDING IN L.R. WHITE RESIN

- 1. Remove 100% ethanol solution.
- 2. Leaving specimens in place in vials (with baskets, if using), cover specimens with L.R White resin. Leave overnight at 4 °C. The resin will remain liquid.
- 3. Take vials to fume hood. Place blood trays (PVC sheets with small wells) on a wooden board. Fill wells to top with fresh L.R. White (fill alternate wells otherwise trays tend to warp) (see Figure 11.8).
- 4. Label wells (on the wooden support board see Figure 11.8) and, using plastic sticks with flattened ends to carefully extract the specimen from its basket/vial, place one specimen in each resin-filled well.
- 5. Slide small melinex squares/coverslips (cut to size from larger sheets) over each well, ensuring no air bubbles are present by simultaneously sliding the melinex squares across and using a pipette to fill wells to the rim (see Figure 11.8). Oxygen must be excluded for the resin to harden.

- 6. Place in a standard lab oven at 60–65 °C for 3–4 hours.
- 7. Remove specimens from oven, peel off Melinex squares, wash with water, and dry with paper towel. Press specimens out of plastic wells, taking care to match each embedded specimen with its label from the wooden board.
- 8. Specimens are now ready for sectioning. Embedded specimens can be stored at room temperature indefinitely in labelled paper envelopes or small ziplock plastic bags.

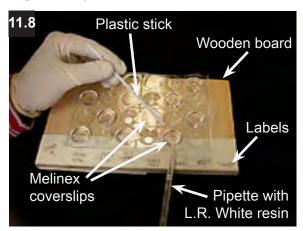


Figure 11.8: Blood tray (with L.R. White resin in alternate wells).

NOTE: If using baskets, after removing specimens soak resin-covered baskets in 100% acetone until clean. Dry and then re-use.

SECTIONING USING A MICROTOME

Use of microtomes will vary from machine to machine; however, there are a few important rules.

- **1. Angle is extremely important.** Sections must be cut at 90° to the thallus/conceptacle surface. Slight variations from this can cause the diagnostic features to be unclear.
- 2. Cut sections 8–12 µm thick.
- 3. Blocks of L.R. White-embedded specimens can be easily trimmed to fit into a chuck (see Figure 11.9). Place the resin block under a red heat lamp for about 1 min. to soften the resin, then trim with a razor blade.

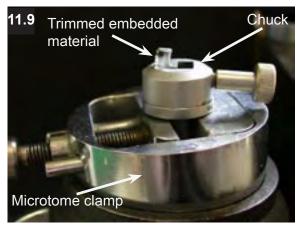


Figure 11.9: Microtome specimen holder, with trimmed embedded material in place.

MAKING PERMANENT SLIDES

- 1. Make sections of the reproductive structures and thallus. Try and keep sections in the same order they come off the block. This can help when analysing pore canals.
- 2. Place sections on a slide and add a drop of Histo-Clear (histological clearing agent), the less used the better. Tilt the slide to run the Histo-Clear over the line of sections on the slide.
- 3. Run a thin line of Eukitt mounting medium over sections, using a wooden stick. Place a coverslip on top and press down gently using blunt end of a small paintbrush. Check the slide under a compound microscope. It is important that the sections are completely flat.
- 4. Place slide on a hotplate set to warm (i.e., not too hot to touch with your palm for 5 s). Place weights on top to keep the sections flat (see Figure 11.10). Leave slides overnight to harden.
- 5. Slides are now ready to examine under a compound microscope.

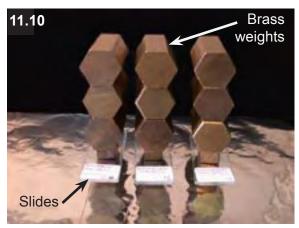


Figure 11.10: Permanent slides on foil-covered hotplate, with brass weights.

Chapter 12. Species profiles

The species profiles in this chapter provide information on the 20 species found in central New Zealand during the present study. The information for each species occurs on two facing pages, and is presented under simple headings in a consistent format. Species are grouped into families, subfamilies, and genera (see Table 12.1).

Field profile

Field profiles give field information and photographs showing entire specimens and external features for each species. Habit, growth forms, and observable reproductive features are illustrated. Information on occurrence in central New Zealand, important field characters, similar species found in central New Zealand, and relevant field notes are also included.

Anatomical and taxonomic profile

Anatomical and taxonomic profiles contain taxonomic information and photographs showing internal structures for each species. Important vegetative and reproductive characteristics, reference specimens held in New Zealand herbaria, and selected references for additional information are provided. Diagnostic features are illustrated and any taxonomic problems noted. Herbarium abbreviations follow Holmgren et al. (1990).

Habit and growth form images



Information on occurrence in central New Zealand, important field characters, similar species found in central New Zealand, and relevant field notes are included

Photographs show reproductive structures for the species

Taxonomic profile for the species

Important vegetative and reproductive characteristics for the species

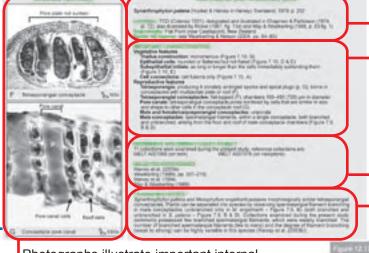
Reference specimens held in New Zealand herbaria, and selected references for additional information

Any taxonomic problems or issues for the species are noted

Anatomical and taxonomic profile

Field

profile

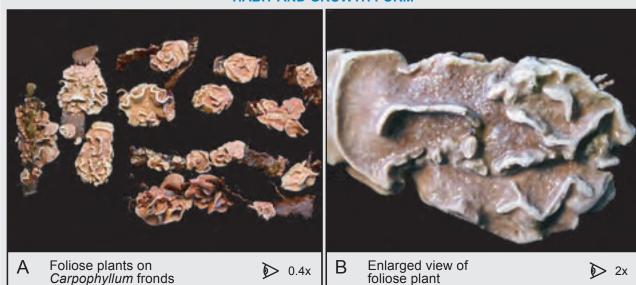


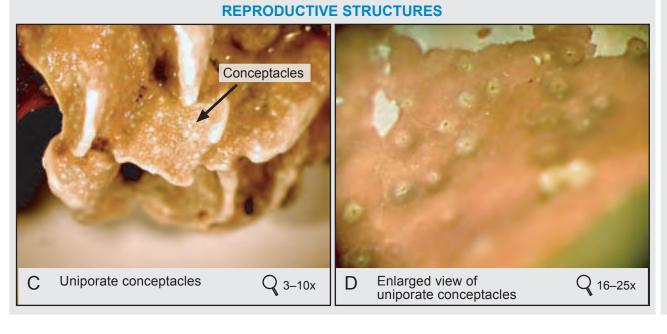
Photographs illustrate important internal structures for each species

Table 12.1: Species found in central New Zealand.

Species profiles are grouped into families, subfamilies, and genera, then alphabetically by species within each genus.

Family	Subfamily	Species name	Species complex note where applicable	Figure number	Pages
Corallinaceae	Lithophylloideae	Lithophyllum carpophylli		12.1	86 & 87
Corallinaceae	Lithophylloideae	Lithophyllum corallinae	forms a species complex with <i>L. stictaeforme</i>	12.2	88 & 89
Corallinaceae	Lithophylloideae	Lithophyllum johansenii		12.3	90 & 91
Corallinaceae	Lithophylloideae	Lithophyllum pustulatum		12.4	92 & 93
Corallinaceae	Lithophylloideae	Lithophyllum stictaeforme	forms a species complex with <i>L. corallinae</i>	12.5	94 & 95
Corallinaceae	Mastophoroideae	Hydrolithon improcerum		12.6	96 & 97
Corallinaceae	Mastophoroideae	Pneophyllum coronatum		12.7	98 & 99
Corallinaceae	Mastophoroideae	Pneophyllum fragile		12.8	100 & 101
Corallinaceae	Mastophoroideae	Spongites yendoi		12.9	102 & 103
Hapalidiaceae	Choreonematoideae	Choreonema thuretii		12.10	104 & 105
Hapalidiaceae	Melobesioideae	Melobesia membranacea		12.11	106 & 107
Hapalidiaceae	Melobesioideae	Mesophyllum engelhartii		12.12	108 & 109
Hapalidiaceae	Melobesioideae	Mesophyllum erubescens	forms a species complex with <i>M. printzianum</i>	12.13	110 & 111
Hapalidiaceae	Melobesioideae	Mesophyllum macroblastum		12.14	112 & 113
Hapalidiaceae	Melobesioideae	Mesophyllum printzianum	forms a species complex with <i>M. erubescens</i>	12.15	114 & 115
Hapalidiaceae	Melobesioideae	Phymatolithon repandum		12.16	116 & 117
Hapalidiaceae	Melobesioideae	Synarthrophyton patena		12.17	118 & 119
Hapalidiaceae	Melobesioideae	Synarthrophyton schielianum		12.18	120 & 121
Sporolithaceae		Heydrichia homalopasta		12.19	122 & 123
Sporolithaceae		Sporolithon durum		12.20	124 & 125





FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 9 of 87 collection localities (including 1 outside the central NZ study area) (Appendix 1)

Depth range: intertidal and subtidal to at least 6 m

FIELD CHARACTERS

Size: plants up to 45 mm across Substrates: Carpophyllum fronds Growth form: foliose (A & B)

Tetrasporangial conceptacles: uniporate (C & D)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles and can be tentatively identified to species level by the substrate and growth form.

COMPARISONS WITH SIMILAR SPECIES

Lithophyllum carpophylli has uniporate tetrasporangial conceptacles and secondary pits. Plants are relatively large and thick, are found growing on Carpophyllum fronds, and have a distinct foliose growth form (upright ridge-like branches arising from an encrusting base). No other known central NZ corallines show these features.

(See also Taxonomic Notes below)

FIELD NOTES

Plants with distinct foliose growth form (upright ridge-like branches arising from an encrusting base) growing on *Carpophyllum* fronds.

INTERNAL FEATURES 4-6 cell layers 1 220x Tetrasporangial conceptacle Mucilagenous material 1-2 cell layers, Cells projecting into pore canal Conceptacle pore **№** 40x Thallus

ANATOMICAL AND TAXONOMIC DATA

Lithophyllum carpophylli (Heydrich) Heydrich, 1897a, p. 52

Syntype: TRH (Foslie Herbarium, A19-1248); illustrated in Printz (1929, Pl. LXXII, fig. 15)

Type locality: Bay of Islands, New Zealand

Earlier NZ reports: see Woelkerling & Nelson (2004, pp. 69–70)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous or dimerous (Figure 7.10, G & H), lacking branches with two central rows of elongate cells situated at an acute angle to one another (G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: secondary pits only (Figure 7.10, B)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent); borne in conceptacles that are uniporate (E)

Tetrasporangial conceptacles: floors of conceptacles usually situated 4–6 cell layers below surrounding thallus surface (E), chambers 185–270 µm in diameter, roofs commonly 1–2 cells thick above the chamber (F)

Pore canals: lined with cells that may project somewhat into, but do not completely block, the canal (although the canal may appear blocked by mucilaginous material) (F)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

13 collections were examined during the present study; reference collections are: WELT A026947 (on *Carpophyllum*) WELT A026948 (on *Carpophyllum*)

SELECTED REFERENCES

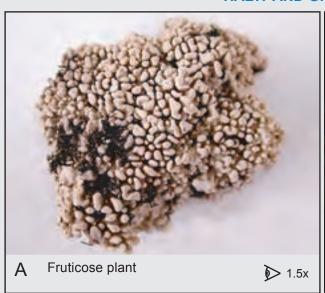
Adams (1994) (as *Tenarea carpophylli*) Nelson et al. (1992) (as *Tenarea carpophylli*) Nelson et al. (1991) (as *Tenarea carpophylli*)

TAXONOMIC NOTES

The type of *Lithophyllum carpophylli* has not been examined in a modern context. Its generic placement is uncertain and its status as a distinct species remains uncertain (Woelkerling & Nelson 2004). However, 'carpophylli' plants are very distinctive in the field (upright branches arising from an encrusting base on *Carpophyllum* fronds – see Field Data, above) and thus are illustrated here as a separate species. Although often identified as *Tenarea carpophylli*, the plants vegetatively agree with *Lithophyllum* as defined by Woelkerling, Sartoni & Boddi (2002) (lacking branches with two central rows of elongate cells situated at an acute angle to one another) (G) and thus are placed in this genus pending further taxonomic studies.

Lithophylloideae

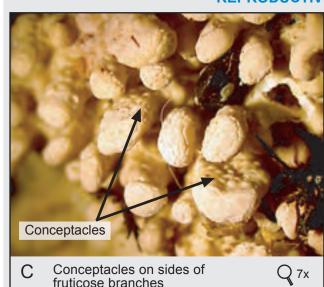
fruticose plant

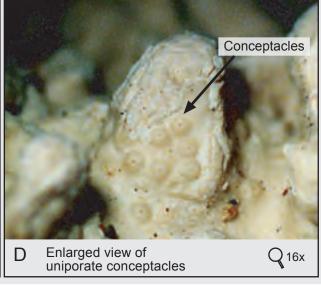


SPECIES PROFILE



REPRODUCTIVE STRUCTURES





FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 1 of 87 collection localities (this location outside the central NZ study area) (Appendix 1)

Depth range: intertidal (known to occur subtidally in southern Australia)

FIELD CHARACTERS

Size: plants up to 40 mm across

Substrates: rocks

Growth form: fruticose (A & B)

Tetrasporangial conceptacles: uniporate (D)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

COMPARISONS WITH SIMILAR SPECIES

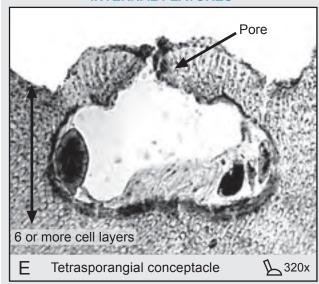
Lithophyllum corallinae has uniporate tetrasporangial conceptacles and secondary pits. Lithophyllum johansenii (Figure 12.3), Lithophyllum pustulatum (Figure 12.4) and Lithophyllum stictaeforme (Figure 12.5) also show these features, but differ in various reproductive characters (see Tabular key).

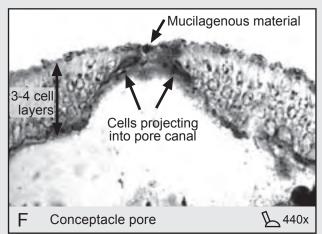
The presence of tetrasporangia and secondary pits can often be confirmed with simple lab procedures (see Tables 9.1 & 9.2).

FIELD NOTES

One collection of *L. corallinae* was confirmed to occur in NZ (this site was outside the central NZ study area - see Appendix 1). Seven other collections from within the study area, however, were intermediate between L. corallinae and L. stictaeforme (see Taxonomic Notes below). Lithophyllum corallinae, therefore, may be more common and more variable in NZ than recorded in this guide.

INTERNAL FEATURES





ANATOMICAL AND TAXONOMIC DATA

Lithophyllum corallinae (Crouan & Crouan) Heydrich, 1897b, p. 47

Lectotype: CO (unnumbered); designated and illustrated in Chamberlain (1991, p. 67, fig. 208 as *Titanoderma*); also illustrated in Woelkerling & Campbell (1992, fig. 22A as *Lithophyllum*)

Type locality: Brest, France

Earlier NZ reports: see Woelkerling & Nelson (2004, pp. 71–72)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous or dimerous (Figure 7.10, G & H) Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: secondary pits only (Figure 7.10, B)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent);

borne in conceptacles that are uniporate (E)

Tetrasporangial conceptacles: floors of conceptacles usually situated 6 or more cell layers below surrounding thallus surface (E), chambers 200–240 μm in diameter, roofs commonly 3–4 cells thick above the chamber (F)

Pore canals: lined with cells that may project somewhat into, but do not completely block, the canal (although the canal may appear blocked by mucilaginous material) (F)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

1 collection was examined during the present study; reference collection is: WELT A3210 (on rock)

SELECTED REFERENCES

Woelkerling (1996d, pp. 231–233)

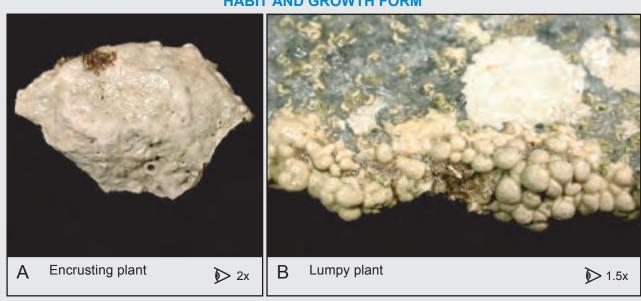
Chamberlain & Irvine (1994) (as *Titanoderma corallinae*)

Woelkerling & Campbell (1992) (as Lithophyllum corallinae)

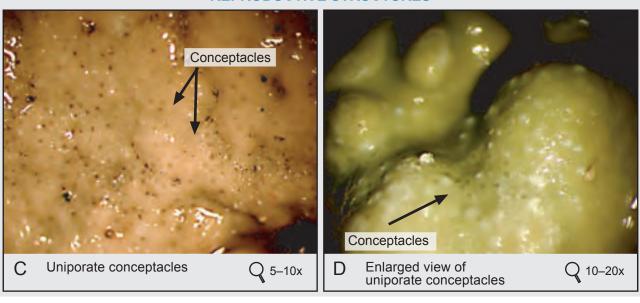
TAXONOMIC NOTES

Lithophyllum corallinae and L. stictaeforme are distinguished from one another on differences in tetrasporangial conceptacle chamber diameter and roof anatomy (see Tabular Key). Seven collections from central NZ, however, possessed conceptacles that were intermediate between the two. As a result, these two species have been placed together in a Lithophyllum stictaeforme – L. corallinae complex, and the two distinct forms are illustrated in this guide pending further work. Intermediate collections have been previously reported from southern Australia (Woelkerling & Campbell 1992, p. 97).

Lithophylloideae



REPRODUCTIVE STRUCTURES



FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 4 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 8 m

FIELD CHARACTERS

Size: individual plants up to 90 mm across

Substrates: rock

Growth form: encrusting to warty to lumpy (A & B) Bisporangial/tetrasporangial conceptacles: uniporate (C & D) (see Taxonomic Notes below)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate bisporangial/tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

COMPARISONS WITH SIMILAR SPECIES

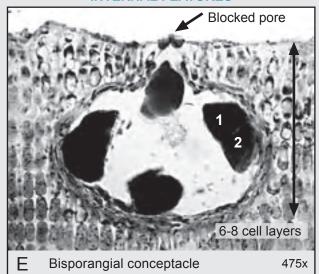
Lithophyllum johansenii has uniporate bisporangial/tetrasporangial conceptacles and secondary pits. Lithophyllum corallinae (Figure 12.2). *Lithophyllum pustulatum* (Figure 12.4), and **Lithophyllum stictaeforme** (Figure 12.5) also show these features, but differ in various reproductive characters (see Tabular key).

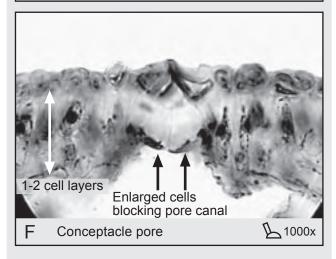
The presence of bisporangia/tetrasporangia and secondary pits can often be confirmed with simple lab procedures (see Tables 9.1 & 9.2).

FIELD NOTES

Found growing on rock in central NZ (known to occur on rock and brown algal holdfasts in southern Australia – see Woelkerling (1996d)).

INTERNAL FEATURES





ANATOMICAL AND TAXONOMIC DATA

Lithophyllum johansenii Woelkerling & Campbell, 1992, p. 61

Holotype: LTB (11724); illustrated in Woelkerling & Campbell (1992, fig. 36A)

Type locality: Port Fairy, Victoria, Australia Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: dimerous (Figure 7.10, H)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: secondary pits only (Figure 7.10, B)

Reproductive features

Bisporangia/tetrasporangia: producing 4 zonately arranged spores (bisporangia producing 2 spores) (apical plugs absent); borne in conceptacles that are uniporate (E)

Bisporangial/tetrasporangial conceptacles: floors of conceptacles usually situated 6–8 cell layers below surrounding thallus surface (E), chambers 95–145 µm in diameter, roofs commonly 1–2 cells thick above the chamber (F)

Pore canals: completely blocked by 2 or 4 enlarged cells (F) Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the

floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

5 collections were examined during the present study; reference collections are: WELT A026950 (on rock) WELT A026952 (on rock)

SELECTED REFERENCES

Woelkerling (1996d, pp. 217–219) Woelkerling & Campbell (1992)

TAXONOMIC NOTES

Collections of this species found in central NZ during the present study possessed conceptacles with bispores only (both tetraspores and bispores occur in southern Australian plants – see Woelkerling (1996d)). Bispores are presumably diploid spores sometimes formed instead of haploid tetraspores. Bisporangial and tetrasporangial conceptacles are morphologically similar and plants can be identified to species using the vegetative and reproductive characters detailed above.

Figure 12.4

HABIT AND GROWTH FORM

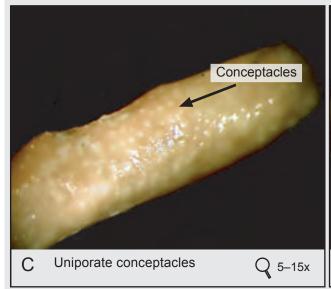


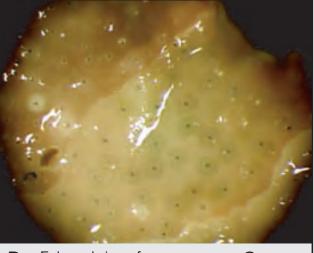


Encrusting plants on Pterocladia

2x

REPRODUCTIVE STRUCTURES





Enlarged view of uniporate conceptacles

Q 15–25x

FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 6 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 5 m

FIELD CHARACTERS

Size: individual plants up to 20 mm across Substrates: other algae (e.g., Gymnogongrus,

Pterocladia, Cladophora) Growth form: encrusting (A & B)

Tetrasporangial conceptacles: uniporate (C & D)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

COMPARISONS WITH SIMILAR SPECIES

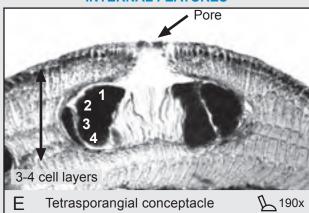
Lithophyllum pustulatum has uniporate tetrasporangial conceptacles and secondary pits. Lithophyllum corallinae (Figure 12.2), Lithophyllum johansenii (Figure 12.3), and Lithophyllum stictaeforme (Figure 12.5) also show these features, but differ in various other characters (see Tabular key).

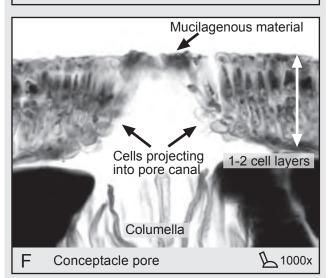
The presence of tetrasporangia and secondary pits can often be confirmed with simple lab procedures (see Tables 9.1 & 9.2).

FIELD NOTES

An encrusting species with uniporate conceptacles found growing on various algae in central NZ (known to also occur on rock, sponges and shells in southern Australia – see Woelkerling (1996d)).

INTERNAL FEATURES





ANATOMICAL AND TAXONOMIC DATA

Lithophyllum pustulatum (Lamouroux) Foslie, 1904b, p. 8

Lectotype: CN (Herb. Lamouroux) (unnumbered); designated and illustrated in Woelkerling, Chamberlain & Silva (1985, p. 325, fig. 29); also illustrated in

Woelkerling & Campbell (1992, fig. 50A)

Type locality: from an unknown locality in France

Earlier NZ reports: see Woelkerling & Nelson (2004, p. 85)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: dimerous (Figure 7.10, H)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: secondary pits only (Figure 7.10, B)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent); borne in conceptacles that are uniporate (E)

Tetrasporangial conceptacles: floors of conceptacles usually situated 3–4 (6) cell layers below surrounding thallus surface (E), chambers 185–300 μm in diameter. roofs commonly 1–2 cells thick above the chamber (F)

Pore canals: lined with cells that may project somewhat into, but do not completely block, the canal (although the canal may appear blocked by mucilaginous material) (F)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

7 collections were examined during the present study; reference collections are: WELT A027016 (on *Pterocladia*) WELT A027019 (on *Gymnogongrus*)

SELECTED REFERENCES

Ringeltaube & Harvey (2000)

Woelkerling (1996d, pp. 227-229)

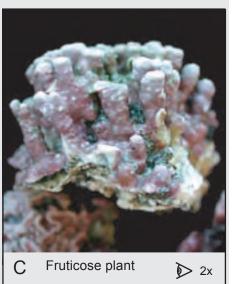
Chamberlain & Irvine (1994) (as *Titanoderma pustulatum*) Woelkerling & Campbell (1992) (as *Lithophyllum pustulatum*)

TAXONOMIC NOTES

Lithophyllum pustulatum is treated as a species of *Titanoderma* by some authors (e.g., Chamberlain & Irvine (1994), Littler & Littler (2003)). Although molecular data imply the two genera are phylogenetically distinct, they cannot be unequivocally separated on morphological or anatomical grounds (see Woelkerling et al. (2002) for more details). *Lithophyllum* and *Titanoderma* are not considered distinct genera for the purposes of this guide and this species has been placed in *Lithophyllum* pending further study.







REPRODUCTIVE STRUCTURES





FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 16 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 12 m

FIELD CHARACTERS

Size: plants up to 100 mm across

Substrates: rocks, shells (e.g., mussels, paua), sea tulips, other algae (e.g., Carpophyllum bases) Growth form: encrusting to warty to lumpy or

fruticose (A–C)

Tetrasporangial conceptacles: uniporate (D & E)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

COMPARISONS WITH SIMILAR SPECIES

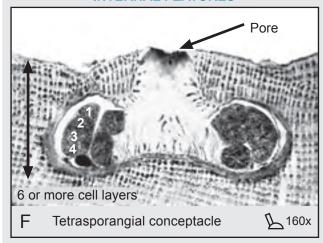
Lithophyllum stictaeforme has uniporate tetrasporangial conceptacles and secondary pits. Lithophyllum corallinae (Figure 12.2), Lithophyllum johansenii (Figure 12.3) and Lithophyllum pustulatum (Figure 12.4) also show these features, but differ in various reproductive characters (see Tabular key).

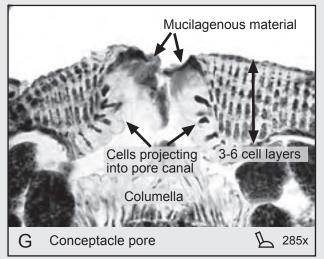
The presence of tetrasporangia and secondary pits can often be confirmed with simple lab procedures (see Table 9.1 & 9.2).

FIELD NOTES

A highly variable species in growth form with uniporate conceptacles.

INTERNAL FEATURES





ANATOMICAL AND TAXONOMIC DATA

Lithophyllum stictaeforme (Areschoug in J. Agardh) Hauck, 1877, p. 292

Lectotype: S (unnumbered); designated and illustrated in Athanasiadis (1999, p.

736, fig. 1)

Type locality: Mediterranean Sea Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous or dimerous (Figure 7.10, G & H) Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: secondary pits only (Figure 7.10, B)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent); borne in conceptacles that are uniporate (F)

Tetrasporangial conceptacles: floors of conceptacles usually situated 6 or more cell layers below surrounding thallus surface (F), chambers 290–420 (450) µm in diameter, roofs commonly 3–6 cells thick above the chamber (G)

Pore canals: lined with cells that may project somewhat into, but do not completely block, the canal (although the canal may appear blocked by mucilaginous material) (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

31 collections were examined during the present study; reference collections are:

WELT A027057 (on shell & rock) WELT A027022 (on rock) WELT A027020 (on rock)

WELT A027058 (on shell & rock) WELT A027056 (on Carpophyllum holdfasts)

SELECTED REFERENCES

Ringeltaube & Harvey (2000) (as Lithophyllum frondosum)

Athanasiadis (1999) (Lithophyllum frondosum is considered a heterotypic synonym of L. stictaeforme)

Woelkerling (1996d, pp. 233–237) (as Lithophyllum frondosum)

Furnari et al. (1996) (as Lithophyllum frondosum)

TAXONOMIC NOTES

Lithophyllum stictaeforme and L. corallinae are distinguished from one another on differences in tetrasporangial conceptacle chamber diameter and roof anatomy (see Tabular key). Seven collections from central NZ, however, possessed conceptacles that were intermediate between the two. As a result these two species have been placed together in a Lithophyllum stictaeforme - L. corallinae complex, and the two distinct forms are illustrated in this guide pending further study. Intermediate collections have been reported previously from southern Australia (Woelkerling & Campbell 1992, p. 97).

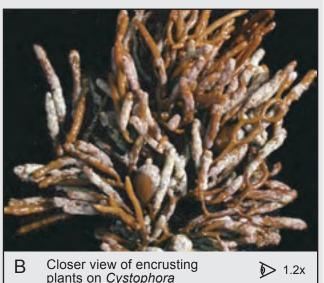
Figure 12.6

HABIT AND GROWTH FORM

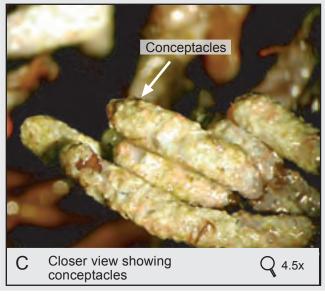
Corallinaceae

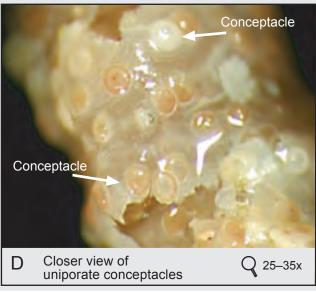


SPECIES PROFILE



REPRODUCTIVE STRUCTURES





FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 6 of 87 collection localities (Appendix

1)

Depth range: intertidal & subtidal to at least 1 m

FIELD CHARACTERS

Size: individual plants up to 15 mm across

Substrates: Cystophora

Growth form: encrusting (A–C)

Tetrasporangial conceptacles: uniporate (D)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate tetrasporangial conceptacle pore canals (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

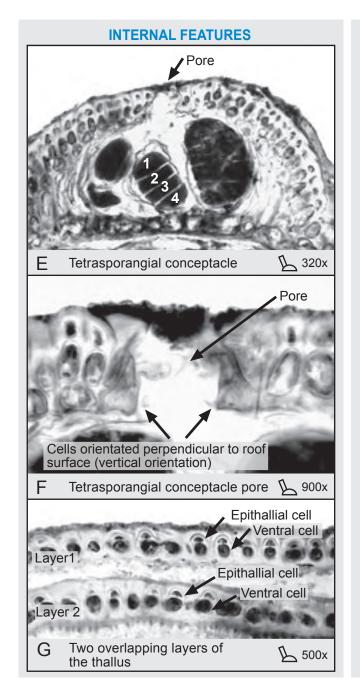
COMPARISONS WITH SIMILAR SPECIES

Hydrolithon improcerum is a small, thin, epiphytic plant with uniporate tetrasporangial conceptacles and cell fusions. Pneophyllum coronatum (Figure 12.7) and Pneophyllum fragile (Figure 12.8) also show these features, but differ in various vegetative and reproductive characters (see Tabular key).

The presence of tetrasporangia and cell fusions can often be confirmed with simple lab procedures (see Tables 9.1 & 9. 2).

FIELD NOTES

Hydrolithon improcerum was found only as small, thin encrusting plants on the brown algae Cystophora torulosa and C. retroflexa in central NZ (it is known to occur on various brown and red algae in southern Australia – see Penrose (1996a)). In the present study, H. improcerum was collected only from localities on the east coast of the North Island.



ANATOMICAL AND TAXONOMIC DATA

Hydrolithon improcerum (Foslie & Howe in Foslie) Foslie, 1909, p. 55

Holotype: TRH (Foslie Herbarium, A14–752); illustrated in Townsend & Adey

(1990)

Type locality: Montego Bay, Jamaica Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: dimerous (Figure 7.10, H), composed of a number of overlapping layers, each commonly 2 cells thick (consisting of ventral cells terminated by epithallial cells) (G)

Epithallial cells: rounded or flattened but not flared (C & Figure 7.10, D & E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent); borne in conceptacles that are uniporate (E)

Tetrasporangial conceptacles: roofs formed by filaments surrounding (peripheral to) and interspersed amongst sporangial initials, chambers 135–180 µm in diameter

Pore canals: lined with cells that are orientated more or less perpendicular to the roof surface (vertical orientation) and do not protrude laterally into the pore canal (F)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

8 collections were examined during the present study; reference collections are: WELT A027029 (on *Cystophora*) WELT A026945 (on *Cystophora*) WELT A027059 (on *Cystophora*)

SELECTED REFERENCES

Penrose (1996a, pp. 258-259)

Townsend & Adey (1990) (as Goniolithon improcerum)

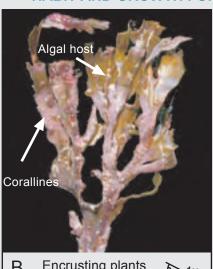
TAXONOMIC NOTES

None

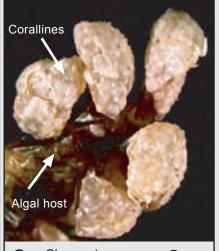
Mastophoroideae



on Caulerpa



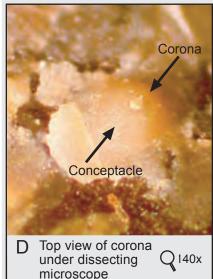


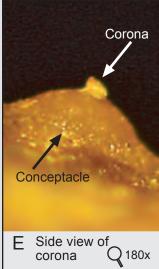


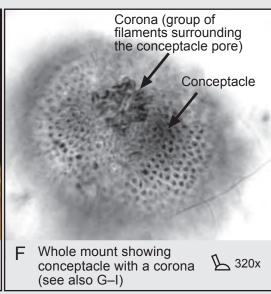
Closer view, on Caulerpa

Q_{4x}

REPRODUCTIVE STRUCTURES







FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 15 of 87 collection localities (including 1 outside the central NZ study area) (Appendix 1) Depth range: intertidal & subtidal to at least 8 m

FIELD CHARACTERS

Size: plants up to 20 mm across (plants often become confluent and form extensive coverings on the algal host)

Substrates: red, green, and brown algae (e.g., Caulerpa, Zonaria, Lessonia)

Growth form: encrusting (A-C)

Tetrasporangial conceptacles: uniporate (E)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate tetrasporangial (or carposporangial) conceptacles for a corona (group of filaments surrounding the pore and protruding above the thallus surface) (see Tabular key). Male plants have uniporate conceptacles and are difficult to identify to species level.

COMPARISONS WITH SIMILAR SPECIES

Pneophyllum coronatum is a small, thin, epiphytic plant with uniporate tetrasporangial conceptacles and cell fusions. Hydrolithon improcerum (Figure 12.6) and Pneophyllum fragile (Figure 12.8) also show these features, but differ in various reproductive characters, most notably the lack of a corona (see Tabular key).

Pneophyllum coronatum is the only epiphytic species found during this study with a corona. This can often be seen with simple lab procedures (D-F) (see Tables 9.1 & 9.2).

FIELD NOTES

Pneophyllum coronatum often occurs as small, dense, mixed populations on various algae (A-C).

INTERNAL FEATURES Corona Tetrasporangial conceptacle √ 310x Corona Cells orientated parallel to the roof surface (horizontal orientation) Tetrasporangial conceptacle pore \$\square\$ 365x Carposporangial conceptacle ∑ 350x

ANATOMICAL AND TAXONOMIC DATA

Pneophyllum coronatum (Rosanoff) Penrose in Chamberlain, 1994a, p. 141

Holotype: CN (unnumbered); illustrated in Rosanoff (1866, pl. 4, fig. 9)

Type locality: Port Phillip Bay, Victoria, Australia

Earlier NZ reports: see Woelkerling & Nelson (2004, p. 72)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: dimerous (Figure 7.10, H)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent); borne in conceptacles that are uniporate (G)

Tetrasporangial conceptacles: roofs formed by filaments surrounding (peripheral to) and interspersed amongst sporangial initials, chambers (90) 120–250 (345) μm in diameter

Pore canals: lined with cells orientated more or less parallel to the roof surface and protruding laterally into the pore canal (horizontal orientation) (H) (these horizontal filaments become more vertically oriented as they elongate and form the corona, H)

Female/carposporangial conceptacles: uniporate, mature conceptacle pores surrounded by a corona of filaments that protrude above the thallus surface (I)

Male conceptacles: uniporate, spermatangial filaments unbranched, arising from the

Male conceptacles: uniporate, spermatangial filaments unbranched, arising from the floor of male conceptacle chambers (Figure 7.9, C); pores lacking a corona of filaments

REFERENCE SPECIMENS LODGED AT WELT

34 collections were examined during the present study; reference collections are:

WELT A026977 (on Caulerpa) WELT A026978 (on Zonaria)

WELT A026982 (on Zonaria) WELT A027060 (on Carpophyllum)

SELECTED REFERENCES

Penrose (1996b, pp. 267-269)

Chamberlain (1994b) (Penrosé in Chamberlain (1994b) considered *Pneophyllum caulerpae* to be a heterotypic synonym of *P. coronatum*)

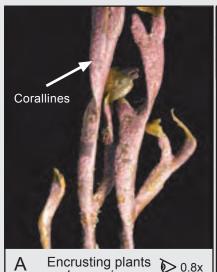
Chamberlain (1994a) (as *Pneophyllum caulerpae*)

TAXONOMIC NOTES

Woelkerling (1996e, p. 238) uses mode of conceptacle development to separate *Pneophyllum* from *Spongites* (see Tabular key). Because this requires developmental stages that are often missing in collections, an ancillary character has been used to help identify plants of these two genera in New Zealand (and southern Australia – see Woelkerling (1996e)) (epiphytic for *Pneophyllum* versus epilithic, epizoic, or unattached for *Spongites*). *Pneophyllum coronatum*, however, has been reported to occur on rock, glass, shells, and other algae in the British Isles (Chamberlain (1994a, p. 134) as *P. caulerpae*).

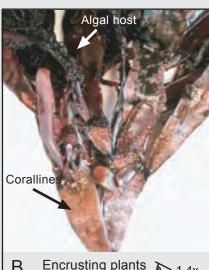
Figure 12.8

HABIT AND GROWTH FORM

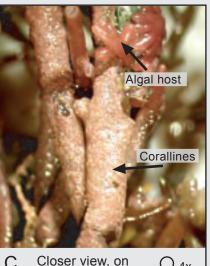


on Lessonia

SPECIES PROFILE



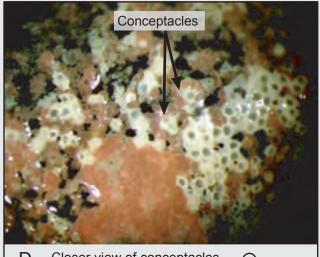


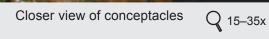


Champia

 Q_{4x}

REPRODUCTIVE STRUCTURES







FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 12 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 12 m

FIELD CHARACTERS

Size: plants up to 15 mm across (plants often become confluent and form extensive coverings on the algal host)

Substrates: various red and brown algae (e.g., Champia, Adamsiella, Lessonia)

Growth form: encrusting (A–C)

Tetrasporangial conceptacles: uniporate (E)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and uniporate tetrasporangial conceptacle pore canals (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

COMPARISONS WITH SIMILAR SPECIES

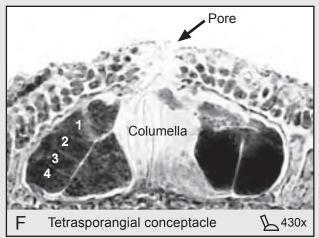
Pneophyllum fragile is a small, thin, epiphytic plant with uniporate tetrasporangial conceptacles and cell fusions. *Hydrolithon improcerum* (Figure 12.6) and **Pneophyllum coronatum** (Figure 12.7) also show these features, but differ in various reproductive characters (see Tabular key).

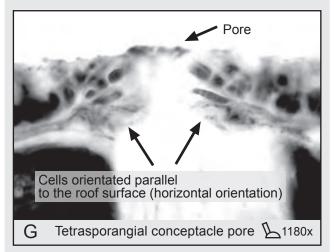
The presence of tetrasporangia and cell fusions can often be confirmed with simple lab procedures (see Tables 9.1 & 9.2).

FIELD NOTES

Pneophyllum fragile often occurs as small, dense, mixed populations epiphytic on various algae (A–D).

INTERNAL FEATURES





ANATOMICAL AND TAXONOMIC DATA

Pneophyllum fragile Kützing, 1843, p. 385

Holotype: L (941.241–152); illustrated in Chamberlain (1983, figs 24–27) and

Penrose & Woelkerling (1991, figs 1–8)

Type locality: unknown locality in the Mediterranean Sea

Earlier NZ reports: see Woelkerling & Nelson (2004, pp. 76–77)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: dimerous (Figure 7.10, H)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent);

borne in conceptacles that are uniporate (F)

Tetrasporangial conceptacles: roofs formed by filaments surrounding (peripheral to) and interspersed amongst sporangial initials, chambers

55–205 µm in diameter

Pore canals: lined with cells orientated more or less parallel to the roof surface and protruding laterally into the pore canal (horizontal orientation) (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the

floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

20 collections were examined during the present study; reference collections are:

WELT A026983 (on Pterocladiella)

WELT A027061 (on Champia)

WELT A026985 (on Adamsiella)

WELT A026986 (on Lessonia)

WELT A027062 (on Champia)

WELT A027063 (on Champia)

SELECTED REFERENCES

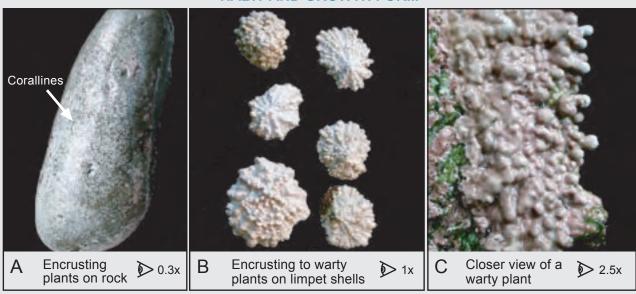
Penrose (1996b, pp. 269-271)

Chamberlain (1994b)

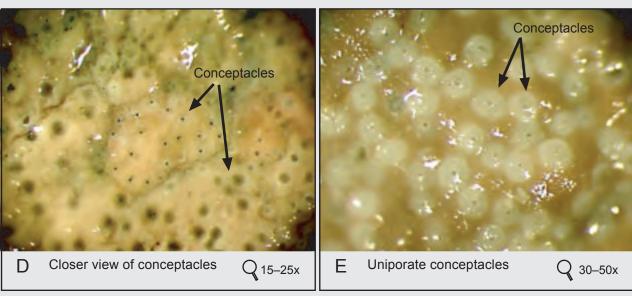
Penrose & Woelkerling (1991)

TAXONOMIC NOTES

Woelkerling (1996e, p. 238) uses mode of conceptacle development to separate *Pneophyllum* from *Spongites* (see Tabular key). Because this requires developmental stages that are often missing in collections, an ancillary character has been used to help identify plants of these two genera in New Zealand (and southern Australia – see Woelkerling (1996e)) (epiphytic for *Pneophyllum* versus epilithic, epizoic, or unattached for *Spongites*). *Pneophyllum fragile*, however, has been reported to occur "very occasionally" on rock in the British Isles (Chamberlain 1994b, p.143).



REPRODUCTIVE STRUCTURES



FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 32 of 87 collection localities (including 1 outside the central NZ study area) (Appendix 1) Depth range: intertidal & subtidal to at least 15 m

FIELD CHARACTERS

Size: plants up to 140 mm across (plants often become confluent and form extensive coverings on the substrate)

Substrates: rock and shells (e.g., limpets and mussels)

Growth form: encrusting to warty (A–C) Tetrasporangial conceptacles: uniporate (E)

IDENTIFICATION

Definitive identification (see Tabular key) requires sectioning and microscopic examination of the thallus and both mature and developmental tetrasporangial conceptacles (although the substrate is often used as an ancillary character instead of developmental conceptacles – see Taxonomic Notes below). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

COMPARISONS WITH SIMILAR SPECIES

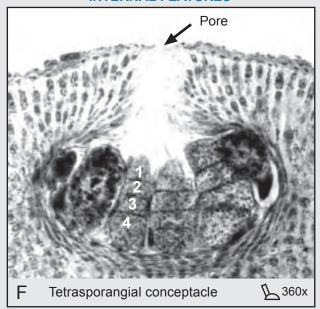
Spongites yendoi has cell fusions, uniporate tetrasporangial conceptacles, and is found growing on rocks and shells. No other known central NZ coralline shows these features.

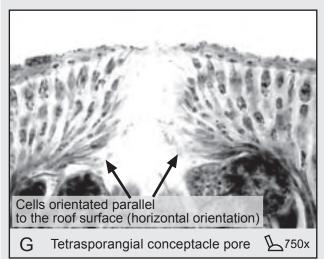
The presence of tetrasporangia and cell fusions can often be confirmed with simple lab procedures (Tables 9.1 & 9.2).

FIELD NOTES

Spongites yendoi appears to be a common intertidal and shallow subtidal (0-1 m) alga on rocks and shells in central NZ. It is not, however, restricted to these depths (collected down to 15 m depth during the present study).

INTERNAL FEATURES





ANATOMICAL AND TAXONOMIC DATA

Spongites yendoi (Foslie) Chamberlain, 1993, p. 100

Lectotype: TRH (Foslie Herbarium, A1-53); designated by Foslie (1904a); illustrated in

Chamberlain (1993, fig. 29)

Type locality: Shimoda Harbour, Izul, Japan Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous or dimerous (Figure 7.10, G & H) **Epithallial cells:** rounded or flattened but not flared (Figure 7.10, D & E)

Cell connections: cell fusions only (Figure 7.10. A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores (apical plugs absent);

borne in conceptacles that are uniporate (F)

Tetrasporangial conceptacles: roofs formed by filaments surrounding (peripheral to) the sporangial initials, chambers 120-255 µm in diameter

Pore canals: lined with cells orientated more or less parallel to the roof surface and protruding laterally into the pore canal (horizontal orientation) (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor of male conceptacle chambers (Figure 7.9, C)

REFERENCE SPECIMENS LODGED AT WELT

80 collections were examined during the present study; reference collections are:

WELT A027013 (on shell, with extra thallus at surface) WELT A027010 (on shell) WELT A027015 (on rock) WELT A027011 (on rock, with extra thallus at surface)

WELT A027064 (on shell) WELT A027065 (on rock)

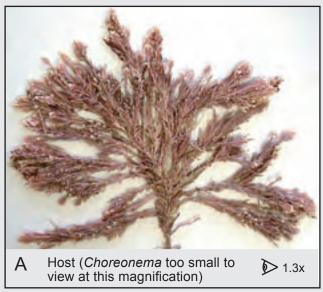
SELECTED REFERENCES

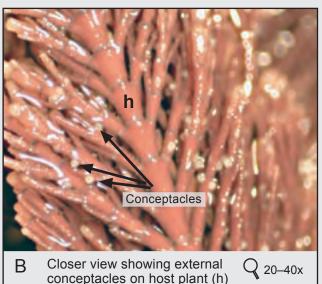
Penrose (1996c, pp. 279-280)

Chamberlain (1993) Keats et al. (1993)

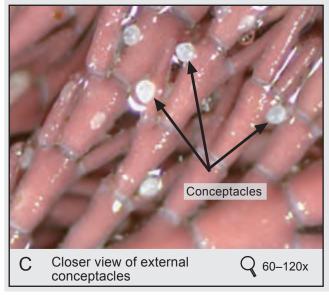
TAXONOMIC NOTES

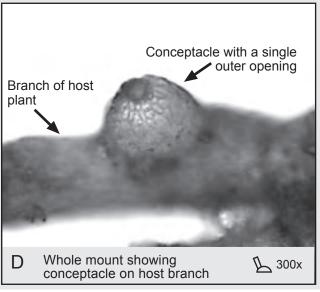
Woelklering (1996e, p. 238) uses mode of conceptacle development to separate Pneophyllum from Spongites (see Tabular Key). Because this requires developmental stages that are often missing, an ancillary character has been used to help identify plants of these two genera in New Zealand (and southern Australia) (epiphytic for Pneophyllum vs epilithic, epizoic, or unattached for Spongites). Central NZ collections of S. yendoi were monomerous or dimerous and no differences in the tetrasporangial conceptacles could be found between these collections. Spongites yendoi from southern Australia (Woelkerling 1996e) is known to be monomerous only. Twenty-two collections also possessed up to 5 layers of elongate to squat, lightly stained cells at the thallus surface. No differences in the tetrasporangial conceptacles could be found between these collections and further study is required to ascertain any taxonomic significance of this feature.





REPRODUCTIVE STRUCTURES





FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 4 of 87 collection localities (Appendix

Depth range: intertidal & subtidal

FIELD CHARACTERS

Size: conceptacles up to 95 μm across
Substrates: geniculate corallines (Haliptilon) (A)
Growth form: unconsolidated endophytic vegetative thallus with external conceptacles (whole plant is semi-endophytic) (B–D)

Tetrasporangial conceptacles: with an acellular multiporate plate sunken beneath a single outer opening (F & G)

IDENTIFICATION

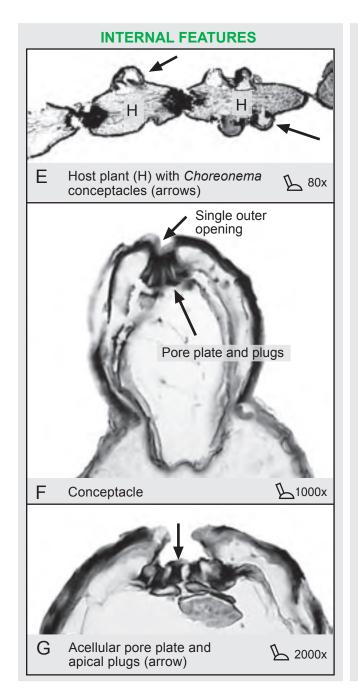
Definitive identification requires sectioning and microscopic examination of the thallus and tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles.

COMPARISONS WITH SIMILAR SPECIES

Choreonema thuretii is a tiny (µm across) parasitic alga found only in geniculate corallines. The vegetative thallus is unconsolidated and entirely endophytic (growing within the host), while conceptacles are external and colourless (due to an absence of chloroplasts and photosynthetic pigments). No other known central NZ corallines show these features.

FIELD NOTES

Choreonema may be found by collecting and examining host plants for conceptacles which can often be seen with a dissecting microscope (B & C) or with simple lab procedures (D). Choreonema was found in Haliptilon during the present study, but it has also been reported in Corallina (Adams 1972), Jania (Adams et al. 1974, Chapman & Parkinson 1974), 'Cornicularia' (South & Adams 1976) and Haliptilon (Nelson et al. 1991) in New Zealand.



ANATOMICAL AND TAXONOMIC DATA

Choreonema thuretii (Bornet in Thuret and Bornet) Schmitz, 1889, p. 455

Lectotype: PC (unnumbered); designated and illustrated in Woelkerling (1987, p.

113, fig. 1); also illustrated in Woelkerling (1998, fig. 341)

Type locality: Pointe de Querqueville, Francè

Earlier NZ reports: see Woelkerling & Nelson (2004, pp. 86–87)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: diffuse, consisting of largely unconsolidated filaments that may become partially consolidated in areas of conceptacle production **Epithallial cells:** absent on vegetative portions of plant, rounded or flattened but not flared on conceptacles (often difficult to see)

Cell connections: cell fusions and secondary pit connections absent

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores and apical plugs; borne in conceptacles with an acellular multiporate plate sunken beneath a single outer opening (F & G)

Tetrasporangial conceptacles: largely external to host (E); chambers 45–70 µm in diameter

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched (Figure 7.9, B), arising from the floor and roof of male conceptacle chambers

REFERENCE SPECIMENS LODGED AT WELT

4 collections were examined during the present study; reference collections are: WELT A027066 (in *Haliptilon* WELT A6543) WELT A027067 (in *Haliptilon* WELT A753)

SELECTED REFERENCES

Harvey et al. (2003b) Harvey et al. (2003a) Broadwater et al. (2002) Broadwater & LaPointe (1997) Woelkerling (1996c, pp. 212–214)

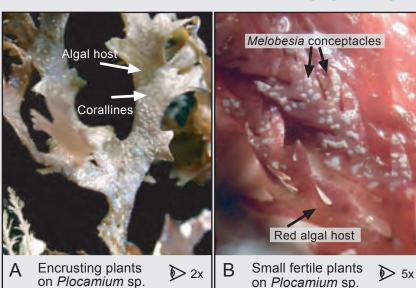
TAXONOMIC NOTES

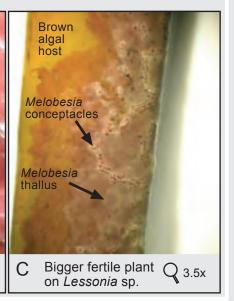
None

Figure 12.11

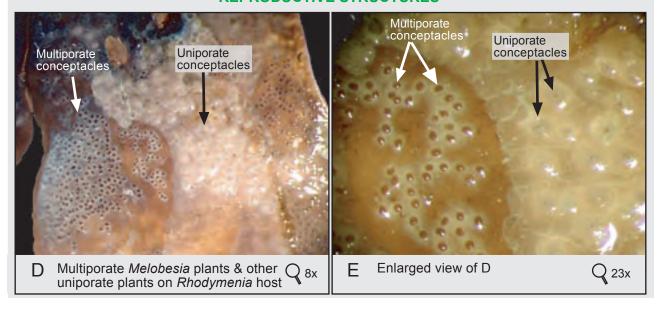
Hapalidiaceae

HABIT AND GROWTH FORM





REPRODUCTIVE STRUCTURES



FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 12 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 5 m

FIELD CHARACTERS

Size: plants up to 35 mm across

Substrates: various red and brown algae (e.g., Laurencia, Lessonia, Rhodymenia, Plocamium,

Pterocladia, Chondria) **Growth form:** encrusting (A–C)

Tetrasporangial conceptacles: multiporate,

flat-topped (D–F)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and multiporate tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles.

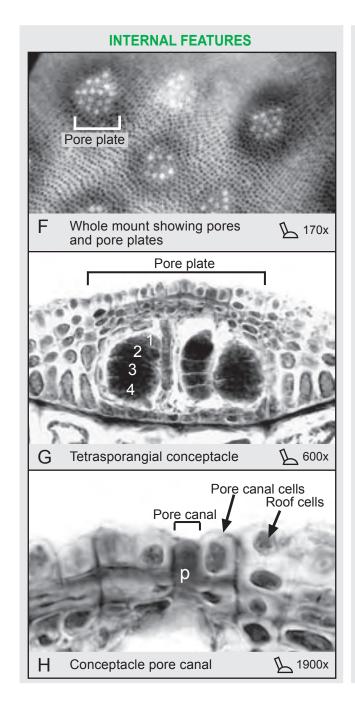
COMPARISONS WITH SIMILAR SPECIES

M. membranacea is a very small and thin epiphytic multiporate coralline found on various red and brown algae. No other known central NZ coralline shows these features. Other encrusting, epiphytic, multiporate species are generally much more robust (larger & thicker when mature) with bigger conceptacles (see Figures 12.13 & 12.17).

The presence of multiporate tetrasporangial conceptacles can often be confirmed with simple lab procedures (see Tables 9.1 & 9.2).

FIELD NOTES

Melobesia membranacea occurs commonly on a variety of red and brown algae, and is relatively small and thin when fertile (<35 mm across). Multiporate conceptacles often appear as dark patches surrounded by a lighter thallus (D & E).



ANATOMICAL AND TAXONOMIC DATA

Melobesia membranacea (Esper) Lamouroux, 1812, p. 186

Neotype: CN (unnumbered); designated and illustrated in Chamberlain (1985, p.

677, fig. 2); also illustrated in Wilks & Woelkerling (1991, fig. 1)

Type locality: an unspecified locality in France Earlier NZ reports: see Woelkerling & Nelson (2004, p. 81)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: dimerous (Figure 7.10, H)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Subepithallial initials: cell elongation characteristics uncertain

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores and apical plugs (p. H); borne in conceptacles with multiporate plate or roof (F & G)

Tetrasporangial conceptacles: flat-topped (G), chambers 40–165 µm in diameter

Pore canals: tetrasporangial conceptacle pores bordered by cells that are similar in size and shape to other cells in the conceptacle roof (H)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched (Figure 7.9, B),

arising from the floor and roof of male conceptacle chambers

REFERENCE SPECIMENS LODGED AT WELT

25 collections were examined during the present study; reference collections are:

WELT A026963 (on a red alga) WELT A026965 (on Laurencia)

WELT A027068 (on Lessonia) WELT A027069 (on Pterocladia)

SELECTED REFERENCES

Harvey et al. (2003b)

Woelkerling (1996b, pp. 168-171)

Wilks & Woelkerling (1991)

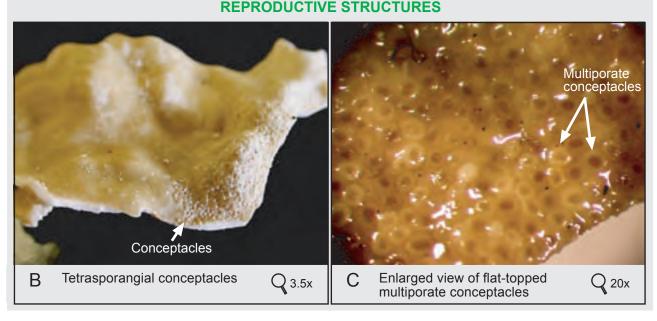
TAXONOMIC NOTES

None

HABIT AND GROWTH FORM



DEDDODUCTIVE OTDUCTUDEO



FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: see Field Notes below Depth range: subtidal to at least 10 m

FIELD CHARACTERS

Size: plants up to 45 mm across

Substrates: rock

Growth form: encrusting to warty (A)

Tetrasporangial conceptacles: multiporate,

flat-topped (C)

IDENTIFICATION

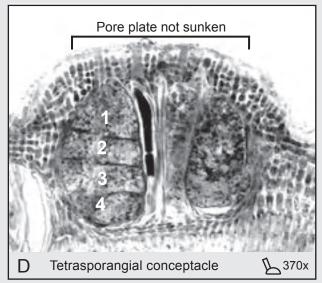
Definitive identification requires sectioning and microscopic examination of the thallus, tetrasporangial conceptacle pore canals, and male conceptacles (for filament branching) (see Tabular key). Male and female plants have uniporate conceptacles; male conceptacles are required for definitive identification (see Taxonomic Notes, below).

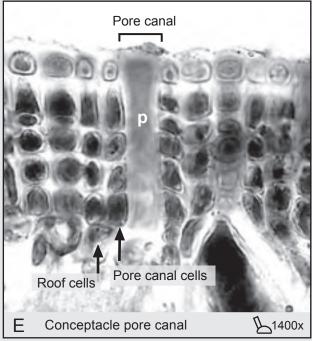
COMPARISONS WITH SIMILAR SPECIES

Mesophyllum engelhartii has flat-topped multiporate conceptacles. Mesophyllum erubescens (Figure 12.13), Phymatolithon repandum (Figure 12.16), and Synarthrophyton patena (Figure 12.17) also show this feature, but differ in various vegetative and reproductive characters (see Tabular key).

FIELD NOTES

Mesophyllum engelhartii & Synarthrophyton patena possess tetrasporangial conceptacles that are morphologically alike (Woelkerling 1996b) (see also Figure 12.17). Although only one collection of M. engelhartii was confirmed to occur in NZ (see Appendix 1), 67 collections possessed these sorts of tetrasporangial conceptacles but lacked males (used to separate the genera), and thus unequivocal identification to species was not possible. Mesophyllum engelhartii (and S. patena) therefore may be more common and more variable in NZ than recorded in this guide.





ANATOMICAL AND TAXONOMIC DATA

Mesophyllum engelhartii (Foslie) Adey, 1970, p. 23

Holotype: TRH (Foslie Herbarium, B18-2595); designated by Adey in Adey & Lebednik (1967, p. 69); illustrated in Printz (1929, fig. 14) and Woelkerling & Harvey (1993, figs 1 & 2)

Type locality: Cape Jaffa, South Australia, Australia

Earlier NZ reports: see Woelkerling & Nelson (2004, p. 75)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E) **Subepithallial initials:** as long or longer than the cells immediately subtending them (Figure 7.10, E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores and apical plugs (p, G); borne in conceptacles with multiporate plate or roof (D)

Tetrasporangial conceptacles: flat-topped (D), chambers 125–180 μm in diameter

Pore canals: tetrasporangial conceptacle pores bordered by cells that are similar in size and shape to other cells in the conceptacle roof (E)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the

floor and roof of male conceptacle chambers (Figure 7.9, A & B)

REFERENCE SPECIMENS LODGED AT WELT

1 collection was examined during the present study; reference collection is: WELT A026958 (on rock)

SELECTED REFERENCES

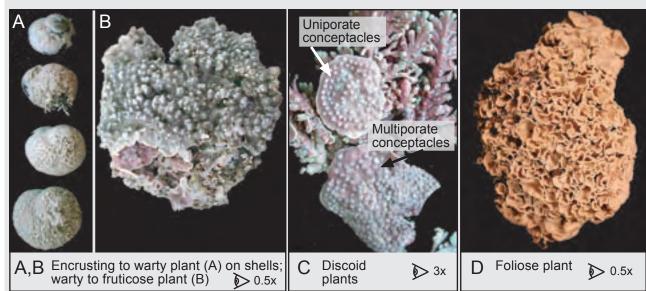
Woelkerling (1996b, pp. 193–197) Barry & Woelkerling (1995) Chamberlain & Keats (1995) Woelkerling & Harvey (1993)

TAXONOMIC NOTES

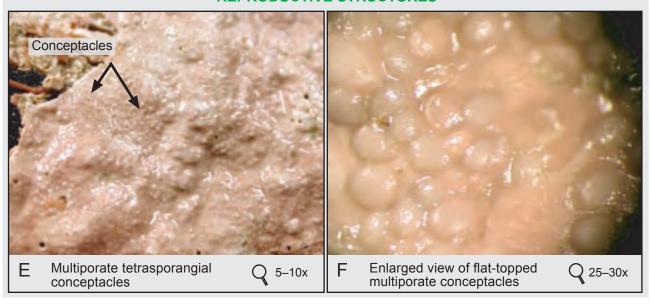
Mesophyllum engelhartii and Synarthrophyton patena possess morphologically similar tetrasporangial conceptacles (Woelkerling 1996b). Plants can be separated to species by observing spermatangial filament branching in male conceptacles (unbranched only in M. engelhartii – Figure 7.9, B) (both branched and unbranched in S. patena – Figure 7.9, B & D).

HABIT AND GROWTH FORM

SPECIES PROFILE



REPRODUCTIVE STRUCTURES



FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 50 of 87 collection localities (including 3 outside the central NZ study area) (Appendix 1)

Depth range: intertidal & subtidal to at least 25 m

FIELD CHARACTERS

Size: plants up to 115 mm across

Substrates: rocks, shells (e.g., paua, barnacles, oysters, Cookia) and other algae (e.g., geniculate corallines, Carpophyllum holdfasts)

Growth form: encrusting to warty to lumpy to fruticose or discoid to layered to foliose (A–D)
Tetrasporangial conceptacles: multiporate,

flat-topped (F)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and tetrasporangial conceptacle pore canals (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

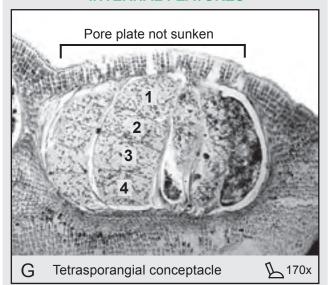
COMPARISONS WITH SIMILAR SPECIES

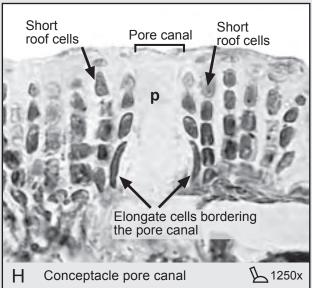
Mesophyllum erubescens has flat-topped multiporate conceptacles. Mesophyllum engelhartii (Flgure12.12), Phymatolithon repandum (Figure 12.16), and Synarthrophyton patena (Figure 12.17) also show this feature, but differ in various vegetative and reproductive characters (see Tabular key).

Mesophyllum erubescens also forms a species complex with **M. printzianum** (see Taxonomic Notes below).

FIELD NOTES

A highly variable species in growth form with flat-topped multiporate tetrasporangial conceptacles.





ANATOMICAL AND TAXONOMIC DATA

Mesophyllum erubescens (Foslie) Lemoine, 1928, p. 252

Holotype: TRH (Foslie Herbarium, C15-3112); illustrated in Foslie (1904a, pl. 3, fig. 20 – as

Lithothamnion); also illustrated in Keats & Chamberlain (1994, figs 26-34)

Type locality: Chaloup Bay, Fernando do Noronha Island, Brasil

Earlier NZ reports: see Woelkerling & Nelson (2004, p. 78) (as incisa and incisum)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Subepithallial initials: as long or longer than the cells immediately subtending them

(Figure 7.10, E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing four zonately arranged spores and apical plugs (p, H); borne

in conceptacles with multiporate plate or roof (G)

Tetrasporangial conceptacles: flat-topped (G), chambers 140–700 µm in diameter

Pore canals: tetrasporangial conceptacle pores bordered by cells that are more elongate,

especially near the base of the pore, than other cells in the conceptacle roof (H)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor and roof

of male conceptacle chambers (Figure 7.9, A & B)

REFERENCE SPECIMENS LODGED AT WELT

177 collections were examined during the present study; reference collections are:

WELT A027070 (on rock) WELT A026955 (on rock) WELT A026956 (on geniculate coralline)

WELT A027026 (on shells) WELT A027071 (on rock) WELT A027072 (on rock)

SELECTED REFERENCES

Harvey et al. (2003b) (Mesophyllum incisum is considered to be a heterotypic synonym of Mesophyllum erubescens)

Woelkerling (1996b, pp. 196–199) (as *Mesophyllum incisum*)

Keats & Chamberlain (1994) (as Mesophyllum erubescens)

Woelkerling & Harvey (1993) (as Mesophyllum incisum)

Woelkerling & Harvey (1992) (as *Mesophyllum incisum*)

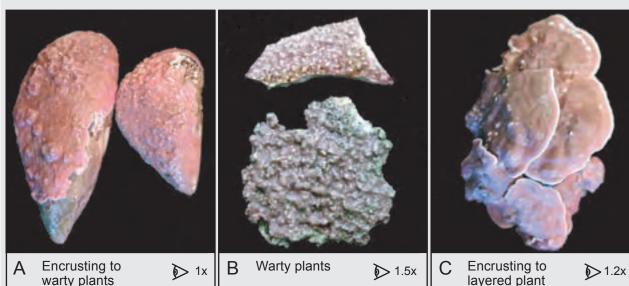
TAXONOMIC NOTES

This species shows similarities to *Mesophyllum printzianum* – both have pore canals bordered by cells that are more elongate, especially near the base of the pore, than other cells in the conceptacle roof (H). While two distinct forms of tetrasporangial conceptacles are often recognisable (flat-topped for *M. erubescens* vs volcano-like for *M. printzianum*), some specimens do not easily fall into one or other category – see Figure 7.7. As a result, these two species have been placed together in a *Mesophyllum erubescens–M. printzianum* complex and the two distinct forms are illustrated in this guide pending further taxonomic work.

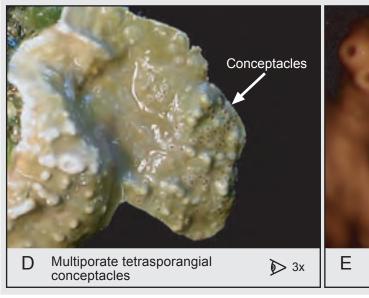
Figure 12.14

HABIT AND GROWTH FORM

Melobesioideae



REPRODUCTIVE STRUCTURES





FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 18 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 15 m

FIELD CHARACTERS

Size: plants up to 105 mm across

Substrates: rocks, shells (e.g., paua, mussels, *Turbo* sp.), ascidians, sponges and other algae (e.g., Ecklonia holdfasts)

Growth form: encrusting to warty or layered (A–C) Tetrasporangial conceptacles: multiporate. volcano-like (with a distinct rim surrounding a central

sunken pore plate) (E)

IDENTIFICATION

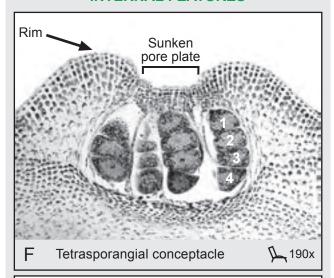
Definitive identification requires sectioning and microscopic examination of the thallus and multiporate tetrasporangial conceptacle pore canals (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

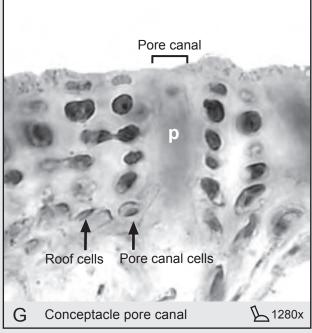
COMPARISONS WITH SIMILAR SPECIES

Mesophyllum macroblastum has volcano-like multiporate conceptacles. Mesophyllum printzianum (Figure 12.15) and Synathrophyton schielianum (Figure 12.18) also show this feature, but differ in various reproductive and vegetative characters (see Tabular key).

FIELD NOTES

A highly variable species in growth form with volcano-like multiporate tetrasporangial conceptacles.





ANATOMICAL AND TAXONOMIC DATA

Mesophyllum macroblastum (Foslie) Adey, 1970, p. 25

Holotype: TRH (Foslie Herbarium, B16-2435); illustrated in Woelkerling & Harvey (1993, figs

17-18); and Woelkerling (1996b, fig. 87A)

Type locality: Gulf of Naples, Italy Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Subepithallial initials: as long or longer than the cells immediately subtending them

(Figure 7.10, E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores and apical plugs (p, G); borne in conceptacles with multiporate plate or roof (F)

Tetrasporangial conceptacles: volcano-like (with a distinct rim surrounding a central sunken pore plate) (F), chambers 165–355 µm in diameter

Pore canals: tetrasporangial conceptacle pores bordered by cells that are similar in size and shape to other cells in the conceptacle roof (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the floor and roof of male conceptacle chambers (Figure 7.9, A & B)

REFERENCE SPECIMENS LODGED AT WELT

43 collections were examined during the present study; reference collections are: WELT A026959 (on mussel shells) WELT A026961 (on sponge) WELT A027046 (on rock) WELT A027073 (on rock))

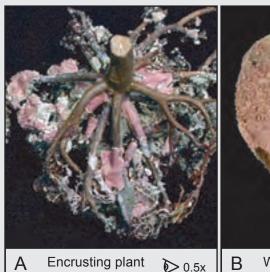
SELECTED REFERENCES

Cabioch & Mendoza (2003) Harvey et al. (2003b) Woelkerling (1996b, pp. 199–202) Woelkerling & Harvey (1993)

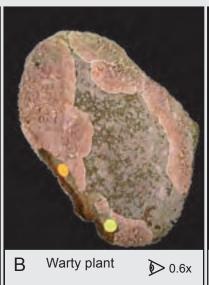
TAXONOMIC NOTES

None

HABIT AND GROWTH FORM

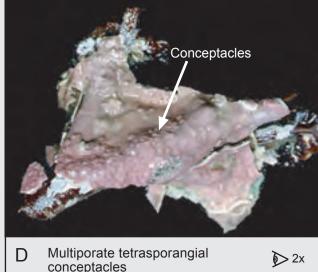


SPECIES PROFILE





REPRODUCTIVE STRUCTURES





FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 28 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 25 m

FIELD CHARACTERS

Size: plants up to 145 mm across

Substrates: rocks, shells (e.g., paua, mussels, Turbo sp.), sponges, other algae (e.g., *Ecklonia* & *Lessonia* holdfasts), plastic

Growth form: encrusting to warty to fruticose (A–C)

Tetrasporangial conceptacles: multiporate, volcano-like (with a distinct rim surrounding a central sunken pore plate) (D & E)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of the thallus and tetrasporangial conceptacle pore canals (see Tabular key). Male and female plants have uniporate conceptacles and are difficult to identify to species level.

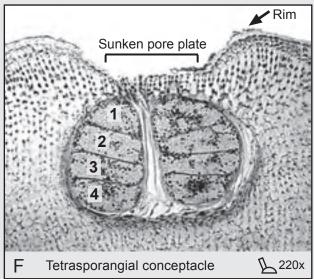
COMPARISONS WITH SIMILAR SPECIES

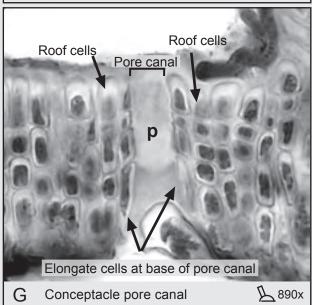
Mesophyllum printzianum has volcano-like multiporate conceptacles. Mesophyllum macroblastum (Figure 12.14) and Synarthrophyton schielianum (Figure 12.18) also show this feature but differ in various vegetative and reproductive characters (see Tabular key).

Mesophyllum printzianum also forms a species complex with M. erubescens (see Taxonomic Notes below).

FIELD NOTES

A highly variable species in growth form with volcano-like multiporate tetrasporangial conceptacles.





ANATOMICAL AND TAXONOMIC DATA

Mesophyllum printzianum Woelkerling & A. Harvey, 1993, p. 593

Holotype: LTB 15249; illustrated in Woelkerling & Harvey (1993, fig. 24A); also

illustrated in Woelkerling (1996b, fig. 89D)

Type locality: Blanket Bay, Otway National Park, Victoria, Australia

Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Subepithallial initials: as long or longer than the cells immediately subtending

them (Figure 7.10, E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores and apical plugs (p,

G); borne in conceptacles with multiporate plate or roof (F)

Tetrasporangial conceptacles: volcano-like (with a distinct rim surrounding a central sunken pore plate) (F), chambers 220–440 µm in diameter

Pore canals: tetrasporangial conceptacle pores bordered by cells that are more elongate, especially near the base of the pore, than other cells in the conceptacle roof (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched, arising from the

floor and roof of male conceptacle chambers (Figure 7.9, A & B)

REFERENCE SPECIMENS LODGED AT WELT

72 collections were examined during the present study; reference collections are:

WELT A027074 (on rock) WELT A027075 (on rock) WELT A026969 (on *Ecklonia* holdfasts)

WELT A026971 (on rock) WELT A027076 (on shells) WELT A026968 (on rock)

SELECTED REFERENCES

Harvey et al. (2003b)

Woelkerling (1996b, pp. 204-205)

Woelkerling & Harvey (1993)

TAXONOMIC NOTES

This species shows similarities to *Mesophyllum erubescens* – both have pore canals bordered by cells that are more elongate, especially near the base of the pore, than other cells in the conceptacle roof (G). While two distinct forms of tetrasporangial conceptacles are often recognisable (flat-topped for *M. erubescens* vs volcano-like for *M. printzianum*), some specimens do not easily fall into one or other category – see Figure 7.7. As a result, these two species have been placed together in a *Mesophyllum erubescens–M. printzianum* complex and the two distinct forms are illustrated in this guide pending further taxonomic work.



Encrusting

plants

1.4x

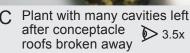
SPECIES PROFILE

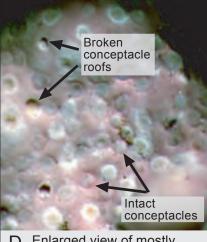


Melobesioideae

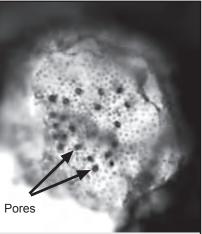
REPRODUCTIVE STRUCTURES







Enlarged view of mostly intact multiporate Q _{15-20x} conceptacles



Squash showing roof of a multiporate ∑290x conceptacle

FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 36 of 87 collection localities (including 1 outside the central NZ study area) (Appendix 1) Depth range: intertidal & subtidal to at least 18 m

FIELD CHARACTERS

Size: plants up to 120 mm across

Substrates: rocks/cobbles and a single epizoic collection (on old shells) (epiphytic thalli not found) Growth form: encrusting to warty (A & B)

Tetrasporangial conceptacles: multiporate, flat-topped, often leaving obvious cavities on the thallus surface as the conceptacle roofs break away (A, C & D)

IDENTIFICATION

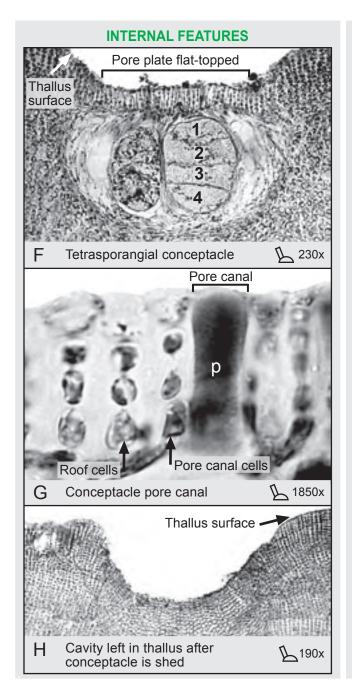
Definitive identification requires sectioning and microscopic examination of the thallus and multiporate tetrasporangial conceptacles (see Tabular key). Male and female plants have uniporate conceptacles and can often be identified to species level by using thallus characters (see Tabular key).

COMPARISONS WITH SIMILAR SPECIES

Phymatolithon repandum has flat-topped multiporate conceptacles. Mesophyllum engelhartii (Figure 12.12), Mesophyllum erubescens (Figure 12.13), and Synarthrophyton patena (Figure 12.17) also show this feature, but differ in various vegetative and reproductive characters (see Tabular key).

FIELD NOTES

Plants commonly found on rocks/cobbles. Plant surface often has cavities/holes where conceptacle roofs have broken away (C). These cavities or crater-like depressions can be mistaken for intact conceptacles. Tentative field identifications made on this basis need to be confirmed by sectioning. Simple lab procedures (Tables 9.1 & 9.2) can often confirm the presence of intact multiporaté conceptacles (E).



ANATOMICAL AND TAXONOMIC DATA

Phymatolithon repandum (Foslie) Wilks & Woelkerling, 1994, p. 190

Lectotype: TRH (Foslie Herbarium, C18-3354); designated by Adey *in* Adey & Lebednik (1967, p. 83); illustrated in Printz (1929, pl. 1, fig.10) and Wilks & Woelkerling (1994, fig.1)

Type locality: Halfmoon Bay, Port Phillip Bay, Victoria, Australia Earlier NZ reports: see Woelkerling & Nelson (2004, p. 85)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E) **Subepithallial initials:** as short or shorter than the cells immediately

subtending them (Figure 7.10, F)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing four zonately arranged spores and apical plugs (p,

G); borne in conceptacles with multiporate plate or roof (F)

Tetrasporangial conceptacles: flat-topped (F), chambers 120–280 μm in diameter

Pore canals: tetrasporangial conceptacle pores bordered by cells that are similar in size and shape to other cells in the conceptacle roof (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments, within a single conceptacle, both branched and unbranched, arising from the floor and roof of male conceptacle chambers (Figure 7.9, B & D)

REFERENCE SPECIMENS LODGED AT WELT

67 collections were examined during the present study; reference collections are:

SELECTED REFERENCES

Harvey et al. (2003b) Woelkerling (1996b, pp. 187–191) Wilks & Woelkerling (1994)

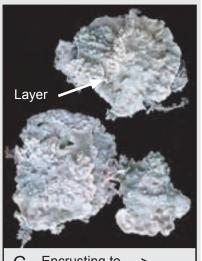
TAXONOMIC NOTES

Tetrasporangial conceptacles in this species are flat-topped (D). The roof of the conceptacle in F is slightly below the surrounding thallus surface (not volcano-like). Old tetrasporangial conceptacles do not become buried within the thallus. Instead, the roofs disintegrate and slough off, leaving obvious cavities or crater-like depressions in the thallus surface (H) that can often be observed in the field (A & C). Tentative field identifications made on this basis need to be confirmed by sectioning.





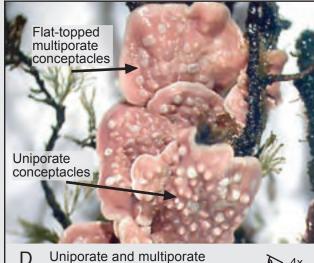
Encrusting to discoid **6**> 0.6x plants on sponge



Encrusting to lavered plants

⊘ 0.5x

REPRODUCTIVE STRUCTURES







multiporate conceptacles

FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 7 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 10 m

FIELD CHARACTERS

Size: plants up to 55 mm across

Substrates: rocks, sponges, and other algae (e.g., Halopteris, Landsbergia, Ballia & Gymnogongrus) **Growth form:** encrusting to discoid to layered (A–D) Tetrasporangial conceptacles: multiporate.

flat-topped (E)

IDENTIFICATION

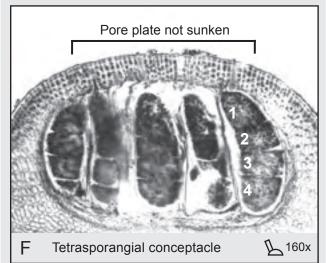
Definitive identification requires sectioning and microscopic examination of the thallus, tetrasporangial conceptacle pore canals, and male conceptacles (for filament branching) (see Tabular key). Male and female plants have uniporate conceptacles; males are required for definitive identification.

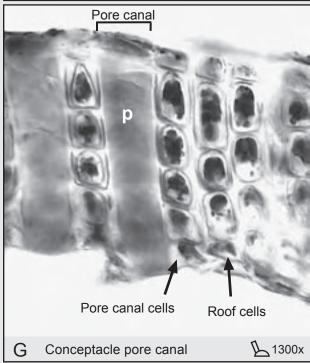
COMPARISONS WITH SIMILAR SPECIES

Synarthrophyton patena has flat-topped multiporate conceptacles. Mesophyllum engelhartii (Figure 12.12), Mesophyllum erubescens (Figure 12.13), and Phymatolithon repandum (Figure 12.16) also show this feature, but differ in various vegetative and reproductive characters (see Tabular key).

FIELD NOTES

Of the 11 collections examined during the present study, only one was found growing on rock; most collections were found growing on sponges and various algae. In these collections dozens of individuals commonly occur (A, B, & D) and both male and tetrasporangial conceptacles are often present, allowing definitive identification. Synarthrophyton patena may, however, be more common and more variable in central NZ than recorded in this guide (see Figure 12.12, Field Notes for Mesophyllum engelhartii).





ANATOMICAL AND TAXONOMIC DATA

Synarthrophyton patena (Hooker & Harvey in Harvey) Townsend, 1979, p. 252

Lectotype: TCD (Colenso 1331); designated and illustrated in Chapman & Parkinson (1974, pl. 72); also illustrated by Ricker (1987, fig. 73d) and May & Woelkerling (1988, p. 53 fig. 1)

Type locality: Flat Point (near Castlepoint), New Zealand

Earlier NZ reports: see Woelkerling & Nelson (2004, pp. 84–85)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Subepithallial initials: as long or longer than the cells immediately subtending them (Figure 7.10, E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing 4 zonately arranged spores and apical plugs (p, G); borne in conceptacles with multiporate plate or roof (F)

Tetrasporangial conceptacles: flat-topped (F), chambers 165–480 (700) µm in diameter **Pore canals:** tetrasporangial conceptacle pores bordered by cells that are similar in size and shape to other cells in the conceptacle roof (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments, within a single conceptacle, both branched and unbranched, arising from the floor and roof of male conceptacle chambers (Figure 7.9, B & D)

REFERENCE SPECIMENS LODGED AT WELT

11 collections were examined during the present study; reference collections are: WELT A027005 (on rock) WELT A027079 (on *Halopteris*)

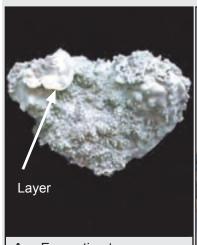
SELECTED REFERENCES

Harvey et al. (2003b) Woelkerling (1996b, pp. 207–210) Harvey et al. (1994) May & Woelkerling (1988)

TAXONOMIC NOTES

Synarthrophyton patena and Mesophyllum engelhartii possess morphologically similar tetrasporangial conceptacles. Plants can be separated into species by observing spermatangial filament branching in male conceptacles (unbranched only in *M. engelhartii* – Figure 7.9, B) (both branched and unbranched in *S. patena* – Figure 7.9, B & D). Collections examined during the present study commonly possessed few branched spermatangial filaments, which were weakly branched. The number of branched spermatangial filaments (few to many) and the degree of filament branching (weak to strong) can be highly variable in this species (Harvey et al. 2003b).

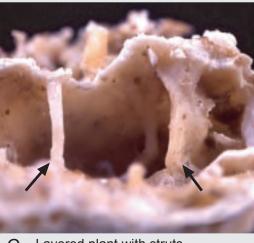
HABIT AND GROWTH FORM



Encrusting to layered plant



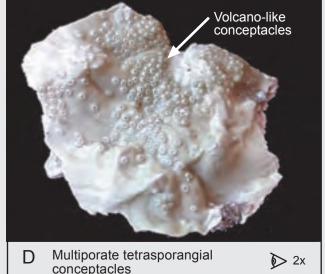
Encrusting plant with ventral struts > 7x abutting substrate



Layered plant with struts (arrows) abutting dorsal surface of the lower laver



REPRODUCTIVE STRUCTURES



♦ 1.7x



multiporate conceptacles

Q 5–10x

FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 4 of 87 collection localities (Appendix

Depth range: subtidal to at least 22 m

FIELD CHARACTERS

Size: plants up to 55 mm across

Substrates: rocks, sponges, and other algae (e.g.,

holdfasts of brown algae)

Growth form: encrusting to layered with ventral struts that either abut the substrate or dorsal surface of lower layers (A-C)

Tetrasporangial conceptacles: multiporate, volcano-like (with a distinct rim surrounding a central

sunken pore plate) (D & E)

IDENTIFICATION

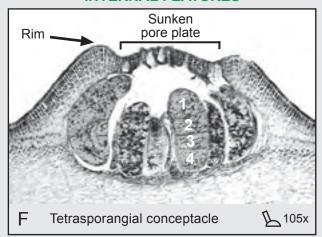
Definitive identification requires sectioning and microscopic examination of the thallus, tetrasporangial conceptacle pore canals, and male conceptacles (for filament branching) (see Tabular key). Male and female plants are uniporate with an encrusting to layered habit and ventral struts.

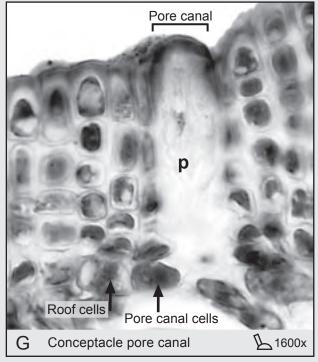
COMPARISONS WITH SIMILAR SPECIES

Synarthrophyton schielianum has volcano-like multiporate conceptacles and ventral struts. Mesophyllum macroblastum (Figure 12.14) and Mesophyllum printzianum (Figure 12.15) both have volcano-like multiporate conceptacles, but lack ventral struts. They also differ in various other vegetative and reproductive characters (see Tabular key).

FIELD NOTES

The presence of very large volcano-like tetrasporangial conceptacles and ventral struts are indicative of this species in the field.





ANATOMICAL AND TAXONOMIC DATA

Synarthrophyton schielianum Woelkerling & Foster, 1989, p. 40

Holotype: WELT A17854; illustrated in Woelkerling & Foster (1989, fig. 1B) Type locality: Waihere Bay, Pitt Island, Chatham Islands (New Zealand) Earlier NZ reports: see Woelkerling & Nelson (2004, p. 86)

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: rounded or flattened but not flared (Figure 7.10, D & E)

Subepithallial initials: as long or longer than the cells immediately subtending them (Figure 7.10. E)

Cell connections: cell fusions only (Figure 7.10, A)

Reproductive features

Tetrasporangia: producing four zonately arranged spores and apical plugs (p, G); borne in conceptacles with multiporate plate or roof (F)

Tetrasporangial conceptacles: volcano-like (with a distinct rim surrounding a central sunken pore plate) (F), chambers 420–815 µm in diameter

Pore canals: tetrasporangial conceptacle pores bordered by cells that are similar in size and shape to other cells in the conceptacle roof (G)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments, within a single conceptacle, both branched and unbranched, arising from the floor and roof of male conceptacle chambers (Figure 7.9, B & D)

REFERENCE SPECIMENS LODGED AT WELT

7 collections were examined during the present study; reference collection is: WELT A027004 (on sponge)

SELECTED REFERENCES

Woelkerling & Foster (1989)

TAXONOMIC NOTES

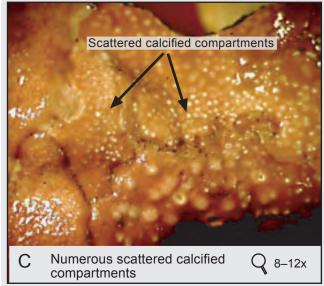
Ventral struts observed during the present study were simple or branched up to 7 mm long and 1.3 mm wide. These struts were generally smaller and narrower than struts seen in holotype material and previous collections (up to 10 mm long and 3 mm wide – see Woelkerling & Foster (1989)).

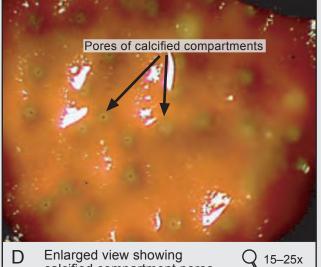
HABIT AND GROWTH FORM





REPRODUCTIVE STRUCTURES





calcified compartment pores

FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Collected at: 8 of 87 collection localities (Appendix

Depth range: intertidal & subtidal to at least 15 m

FIELD CHARACTERS

Size: plants up to 170 mm across

Substrates: rocks/cobbles (epiphytic and epizoic

plants not encountered)

Growth form: encrusting (A & B) Tetrasporangial compartments: solitary and

scattered throughout the thallus (never grouped

into sori) (C & D)

IDENTIFICATION

Definitive identification requires sectioning and microscopic examination of calcified compartments (see Tabular key). Male and female plants have uniporate conceptacles.

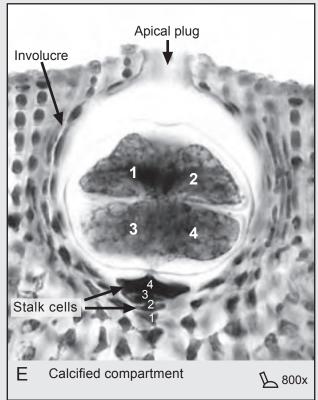
COMPARISONS WITH SIMILAR SPECIES

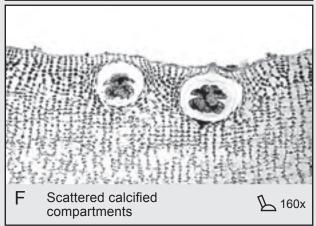
Heydrichia homalopasta has cruciately-arranged spores in calcified compartments scattered throughout the thallus. No other known central NZ corallines show these features.

The presence of cruciately arranged spores and calcified compartments (Figure 7.8, G) can often be confirmed with simple lab procedures (see Tables 9.1 & 9.2).

FIELD NOTES

Found only as encrusting plants on rocks/cobbles. Fertile tetrasporangial plants have inconspicuous holes scattered across the surface, and may appear infertile or uniporate (C & D) upon initial inspection.





ANATOMICAL AND TAXONOMIC DATA

Heydrichia homalopasta Townsend & Borowitzka, 2001, p. 237

Holotype: NSW 485140 (Townsend BB7902); illustrated in Townsend & Borowitzka (2001,

fig. 1)

Type locality: Bongin Bongin Bay, Mona Vale, New South Wales, Australia

Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: flared (Figure 7.10, C)

Subepithallial initials: mostly the same size as cells immediately subtending them

(Figure 7.10, C)

Cell connections: secondary pits only seen in NZ collections (Figure 7.10, B), although cell fusions previously reported in some Australian collections (Townsend & Borowitzka

(2001))

Reproductive features

Tetrasporangia: tetrasporangia terminal on a 5-celled stalk; producing 4 cruciately arranged spores and apical plugs; borne in calcified compartments (E & Figure 7.8, G) **Calcified tetrasporangial compartments:** surrounded by an involucre (E); solitary and scattered throughout the thallus (not grouped into sori) (F); compartments 70–130 µm in diameter. The term involucre applies to the vertically flattened sterile filaments surrounding the tetrasporangial compartments (E)

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments unbranched (Figure 7.9, B), arising from the floor and roof of male conceptacle chambers

REFERENCE SPECIMENS LODGED AT WELT

19 collections were examined during the present study; reference collections are: WELT A026941 (on rock) WELT A026942 (on rock) WELT A027080 (on rock)

SELECTED REFERENCES

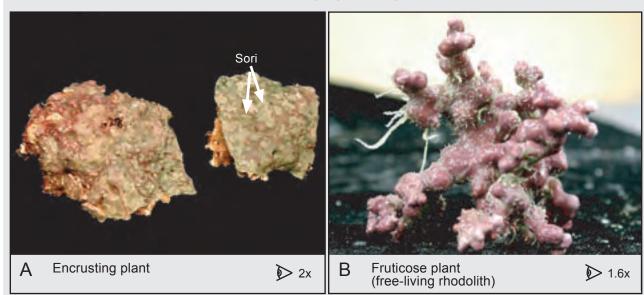
Harvey et al. (2002)

Townsend & Borowitzka (2001)

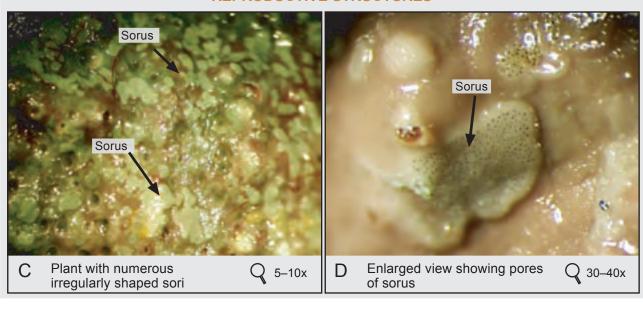
TAXONOMIC NOTES

A 5-celled stalk beneath the tetrasporangium can usually be seen in developing compartments. These stalk cells become extremely compressed as the sporangium matures and enlarges, and fewer than 5 stalk cells may be evident in the mature compartment (E). The cruciate divisions in the spore in E are not easy to see because of the way individual spores are oriented. The cruciate arrangement is clearer in Figure 7.8 (F & G).

HABIT AND GROWTH FORM



REPRODUCTIVE STRUCTURES



FIELD DATA FOR CENTRAL NZ

OCCURRENCE

Attached plants collected at: 7 of 87 collection localities (including 1 outside the central NZ study area) (Appendix 1)

Rhodoliths collected at: 2 of 87 collection localities (including 1 outside the central NZ study area)

Depth range: intertidal & subtidal to at least 19 m

FIELD CHARACTERS

Size: attached plants up to 120 mm across, rhodoliths up to 90 mm across

Substrates: rocks/cobbles and as free-living rhodoliths (A & B)

Growth form: encrusting to warty (attached plants) or lumpy to fruticose (rhodoliths) (A & B)

Tetrasporangial compartments: grouped into sori that are irregular in shape and indefinite in size (A, C, & D); sori occasional or common

IDENTIFICATION

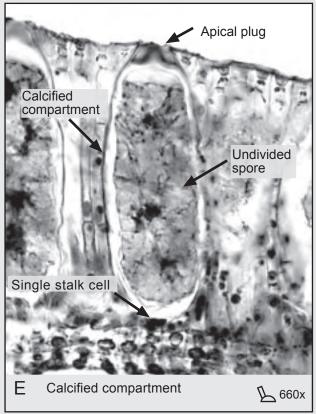
Definitive identification requires sectioning and microscopic examination of calcified compartments (see Tabular key). Male and female plants have uniporate conceptacles.

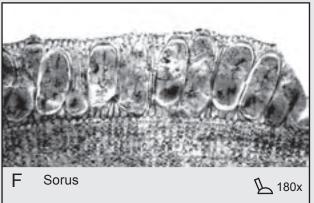
COMPARISONS WITH SIMILAR SPECIES

Sporolithon durum has cruciately arranged spores in calcified compartments grouped into sori. No other known central NZ corallines show these features, which can often be confirmed with simple lab procedures (see Tables 9.1 & 9.2).

FIELD NOTES

Sori appear as irregularly shaped, slightly raised, pink or white patches on the plant surface that can often be seen by eye or with a dissecting microscope (A & C). Ten (of 18) collections were found as attached plants; 8 were unattached rhodoliths. Twelve sterile rhodolith collections were also found. These could not be unequivocally identified, but vegetatively were either *Sporolithon* or *Lithothamnion* (see Appendix 1).





ANATOMICAL AND TAXONOMIC DATA

Sporolithon durum (Foslie) Townsend & Woelkerling in Townsend et al., 1995, p. 86

Lectotype: TRH (Foslie Herbarium, C19-3381); designated by Adey *in* Adey & Lebednik (1967, p.84); illustrated in Printz (1929, figs 1–3 as *Archaeolithothamnion*) and

Townsend et al. (1995, fig. 1A)

Type locality: Cape Jaffa, South Australia, Australia

Earlier NZ reports: no prior records

IMPORTANT CHARACTERISTICS

Vegetative features

Thallus construction: monomerous (Figure 7.10, G)

Epithallial cells: flared (Figure 7.10, C)

Subepithallial initials: mostly the same size as cells immediately subtending them

(Figure 7.10, C)

Cell connections: cell fusions only or both cell fusions and secondary pits (Figure

7.10, A & B)

Reproductive features

Tetrasporangia: terminal on a single-celled stalk; producing 4 cruciately arranged spores and apical plugs; borne in calcified compartments (E & Figure 7.8, E & F) **Calcified tetrasporangial compartments:** not surrounded by an involucre (E; see Figure 12.19 for involucre); usually grouped into sori (F) that are irregular in shape and indefinite in size (C & D); compartments 40–60 µm in diameter.

Male and female/carposporangial conceptacles: uniporate

Male conceptacles: spermatangial filaments branched (Figure 7.9, D), arising from the floor and roof of male conceptacle chambers

REFERENCE SPECIMENS LODGED AT WELT

18 collections were examined during the present study; reference collections are: WELT A027043 (on rock) WELT A026997 (on rock) WELT A026999 (rhodolith) WELT A027081 (rhodolith)

SELECTED REFERENCES

Harvey et al. (2002) Woelkerling (1996a, pp. 155–157) Townsend et al. (1995)

TAXONOMIC NOTES

None

Acknowledgments

We acknowledge the foresight and generosity of the New Zealand Ministry of Fisheries for funding this project (ZBD200105).

Numerous people played important and integral roles in the project and we gratefully acknowledge their enthusiastic and crucial participation. Sincere thanks to all of the following – this project wouldn't have happened without your help.

At La Trobe University, Natalie Short and Mia Miasari sectioned hundreds of coralline algae specimens for the project, providing invaluable assistance. Judy Broom and Darren Hart (University of Otago) have, in parallel with the taxonomic work collated in this guide, carried out ground-breaking work in obtaining molecular data for crustose coralline algae. Pat Brownsey and Jenn Dalen (Museum of New Zealand Te Papa Tongarewa) and staff in the Botany section provided invaluable support to the smooth running of this project, including laboratory and storage space.

Clinton Duffy (Department of Conservation) has supported the project from its inception.

Many thanks to Jean Woelkerling and Rob Harvey, who read the manuscript during preparation.

COLLECTIONS AND WORKSHOPS

The following people have all played a vital role, collecting (or facilitating the collection of) specimens, and/or attending collecting and identification training workshops. The comments and feedback many of them provided on preliminary drafts of this guide contributed enormously to the final product.

Many NIWA staff contributed their help, advice and expertise. They include: Owen Anderson, Anna Bradley, Stephen Brown, Russell Cole, Niki Davey, Ralph Dickson, Marty Flanagan, Jeff Forman, Nick Gust, Ian Hawes, Nicole Hancock, Andy Hill, Dave Kelly, Kerstin Kroeger, Anne-Nina Loerz, Chazz Marriot, Steve Mercer, Sheryl Miller, Mark Morrison, Reyn Naylor, Alf Norkko, Pete Notman, Mike Page, Rob Stewart, and Dean Stotter.

Staff from the Department of Conservation have also contributed their time, equipment and expertise: Sean Cooper, Clinton Duffy, Duncan Ferguson, Debbie Freeman, Nicky Gibbs, Lou Hunt, Al Hutt, Mark Martini, Don Neale, Heath Priest, Jamie Quirk, Kala Sivaguru, and Bryan Williams.

Thanks are due to staff and students of Canterbury University (Robyn Dunmore, Katie Lotterhos, Michelle Mei, Roly Russell, Dave Schiel, David Taylor and Spencer Wood), University of Otago (Judy Broom, Darren Hart and Wyn Jones), Victoria University of Wellington (Joe Buchanan and Kate Steger) and Auckland University (Jarrod Walker). Thanks to staff of the herbaria at Auckland Museum (Ewen Cameron, Mei Nee Lee), Landcare Research (Aaron Wilton, Mary Corver), and Te Papa (Pat Brownsey, Jenn Dalen, Barry Sneddon, Simon Whittaker). Others who provided valuable collecting assistance are Christian Boedeker, Rob Harvey, T. Paenga, Murray Parsons, Mike Stuart, and Willie Waitoa.

Thanks to the committees and Friends of the Kapiti and Te Angi Angi Marine Reserves.

HERBARIUM COLLECTIONS

Coralline algae collections made outside the bounds of this project (i.e. from 1905 onwards) were kindly made available to us by the herbaria at the Auckland Museum (AK), Landcare, Christchurch (CHR), and Te Papa (WELT). The contributions of the following collectors are therefore also recognised: N.M. Adams, A.N. Baker, J.E. Braggins, L. Chadderton, R. Davidson, U.V. Dellow, G. Fenwick, M. Foster, R. Gilpin, V. Hoggard, L.M. Irvine, K.A. Johnson, V.W. Lindauer, J.K. Lowry, R. Mason, L.B. Moore, R. Naylor, W.A. Nelson, W.R.B. Oliver, Mrs H. Parsons, M.J. Parsons, Andy Ritchie, D.B. Rogan, W. Scarfe, J. Thomas, and J.C. Yaldwyn.

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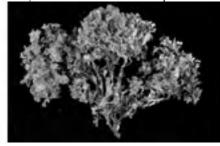
Glossary

Ancillary character: a character useful in helping to identify specimens, but not necessarily diagnostic of a species.

Apical plug: a hard, mucilage-like plug produced at the apex of a tetrasporangium or bisporangium in species of Hapalidiaceae and Sporolithaceae, but not found in the Corallinaceae.



Arborescent: in non-geniculate corallines, a term used to describe the growth form of a plant composed of a distinct holdfast and stipe bearing flattened, ribbon-like to fan-shaped branches.

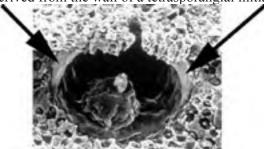


Basionym: the original name under which the coralline was described

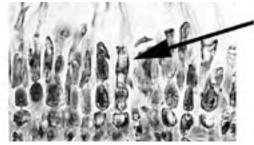
Bisporangium: a sporangium that houses two spores (each termed a bispore).

Bispore: a presumably diploid spore sometimes formed instead of a tetraspore in some species of corallines.

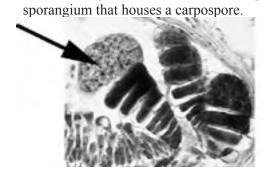
Calcified compartment: a structure housing a tetrasporangium in genera of Sporolithaceae. It is derived from the wall of a tetrasporangial initial.



Carpogonium (carpogonia): a haploid female reproductive cell with which a sperm cell unites during fertilisation. Carpogonia are terminal on filaments arising from the female conceptacle floor.



Carposporangium (carposporangia):

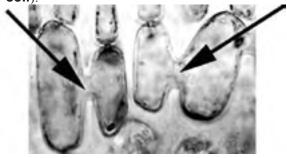


Carpospore: a diploid spore that is produced by a carposporophyte and that gives rise to a diploid tetrasporophyte (see above photo).

Carposporophyte: a small (usually microscopic) diploid, parasitic plant that develops from a fertilised carpogonium, remains attached to the old female haploid parent, and produces carpospores. The carposporophyte is one of three distinct morphological phases in the sexual cycle of corallines; the other two are the haploid gametophyte and the diploid tetrasporophyte.



Cell fusion: a linkage between cells of two adjacent vegetative filaments in which portions of the cell walls break down and the protoplasts then apparently fuse (not to be confused with **fusion cell**).



Collection: for coralline algae, a single 'collection' of a small species may contain thousands of individuals growing on a single host alga or stone; for some larger species, only part of a single individual may make up a collection.

Columella: a structure composed of a group of persistent, but often senescent, sterile cells that arises from the floor of a tetrasporangial or bisporangial conceptacle in some species of corallines.



Conceptacle: a structure consisting of a multicellular chamber housing reproductive bodies. A conceptacle may contain a single pore (uniporate) or a number of pores (multiporate)

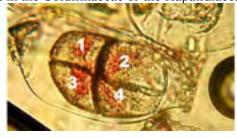
through which spores or gametes are released (see above photo of a conceptacle with a single pore).

Contiguous: adjacent structures that are touching and aligned more or less parallel.

Corona: a ring of filaments that surrounds and protrudes above the surface of a pore in conceptacles of some species of Corallinaceae.

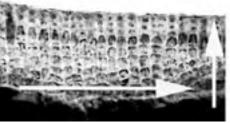


Cruciate: a term used to describe the arrangement of the spores lying side by side in two rows within a tetrasporangium. In corallines, cruciate sporangia characteristically occur in the Sporolithaceae, but not in the Corallinaceae or the Hapalidiaceae.



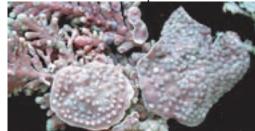
Diagnostic character: any character(s) that unambiguously separates one species from others in the same genus, or more generally any character that unambiguously separates two taxa of the same rank (e.g., two genera within a family).

Dimerous: in non-geniculate corallines, a type of thallus construction involving two distinct groups of filaments that are produced successively and are oriented more or less at right angles to one another.



Diploid: a term used for a nucleus with, or an organism whose nuclei contain, two representatives of each chromosome. Compare with **Haploid**.

Discoid: in non-geniculate corallines, a term used to describe the growth form of a plant composed of an unbranched, largely unattached, flattened, disclike thallus of various shapes.



Encrusting: in non-geniculate corallines, a term used to describe the growth form of a plant that is largely attached, crustose, and flattened or sleevelike, and without protuberances or branches.



Endophytic: growing within another plant.

Epigenous: growing on a substrate.

Epilithic: growing on rock.

Epiphytic: growing upon another plant.

Epithallial cell: in corallines, the terminal cell or cells of a filament at the surface of a plant. Epithallial cells are formed outwardly from the division of a vegetative cell just below the tip of a filament. In some species, more than one epithallial cell may occur at the tip of a filament.



Epizoic: growing on an animal.

Family: in nomenclature, the *principal* taxonomic rank above the rank of genus and below the rank of order. Names of families end in –aceae as, for example, the Corallinaceae.

Filament: in corallines, a row of cells derived from a terminal or subterminal meristematic cell and linked by primary pit connections.



Flat-topped: refers to multiporate conceptacles that are mound-like (raised), or more or less level with the thallus surface, with a flat roof (i.e., lacking a distinct rim and central sunken pore plate).



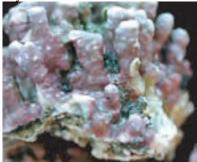
Flared: spread out or broadened above the base as in the distal epithallial cell walls of some corallines (see photo at left of flared **epithallial cells**).

Foliose: in non-geniculate corallines, a term used to describe the growth form of a plant composed of several to many flattened lamellate (plate-like) branches arranged at various angles to one another, but not in horizontal layers.

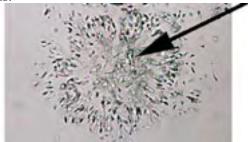


Frond: a term sometimes used to describe a leaf-like part of a plant.

Fruticose: in non-geniculate corallines, a term used to describe the growth form of a plant composed mainly of more or less cylindrical to compressed or knobby branches that are over 3 mm long.



Fusion cell: an enlarged, often irregular, cell formed from the zygote by fusion with other adjacent cells.



Gamete: either of two haploid cells involved in zygote formation; in corallines the female gamete is the carpogonium and the male the spermatium.

Geniculum (genicula): the uncalcified segment found between two calcified segments (intergenicula) in branches of some species of Corallinaceae.



Geniculate: possessing genicula.

Genus: in nomenclature, the *principal* taxonomic rank above the rank of species and below the rank of family.

Gonimoblast filament: in carposporophytes,

a name for a filament that ultimately produces carposporangia.



Growth form: a term used to describe the external appearance of a non-geniculate coralline alga. The array of growth forms in non-geniculate corallines is shown in Figure 7.1.

Haploid: a term used for a nucleus with, or an organism whose nuclei contain, one representative of each chromosome. Compare with **Diploid**.

Heterotypic synonym: in nomenclature, synonyms based on different type specimens.

Holdfast: a small, basal attachment structure that anchors a plant to a substrate.

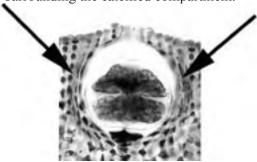
Holotype: the single specimen or sheet upon which an author bases the name of a new taxon.

In: in nomenclature, used to connect the names of two authors the first of which validly published a name in a work by the second author, as for example *Lithocystis* Allman *in* Harvey.

Infraspecific taxon: a taxon below the rank of species (e.g., a form or variety).

Initial: see Subepithallial initial.

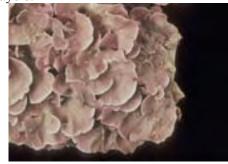
Involucre: in the Sporolithaceae; applies to the vertically flattened sterile filaments surrounding the calcified compartment.



Kina: sea urchins; echinoderms of the class Echinoidea, having a soft body enclosed in a round calcareous shell covered in spines.

Lamella (lamellae): a thin, plate-like structure, as for example, the thallus of *Synarthrophyton patena* (Figure 12.17, D). **Lamellate** (adj.).

Layered: in non-geniculate corallines, a term used to describe the growth form of a plant composed of several to many lamellate branches arranged in horizontally oriented layers.



Lectotype: a specimen selected from the original material used by an author to describe a species when a holotype was not designated or has been lost.

Lumpy: in non-geniculate corallines, a term used to describe the growth form of a plant composed of short, swollen protuberances that are usually unbranched and are often crowded or contiguous.



Maerl: a term used sometimes for the sediment composed principally of irregularly branched, unattached, non-geniculate coralline algae (see **Rhodolith**).

Meiosis: a specialised nuclear division sequence in which the chromosome number is reduced from 2N to N and genetic segregation occurs.

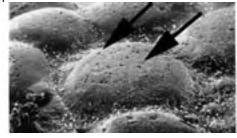
Monographic study: a comprehensive analysis of a taxonomic group occurring in a geographic region, that includes a survey of all past relevant literature and a detailed assessment of all the characters that have been, or potentially could be, used to delimit taxa.

Monomerous: in non-geniculate corallines a type of thallus construction involving a single system

of repeatedly branched filaments forming a core in which filaments are oriented more or less parallel to the thallus surface, and a peripheral region in which portions of filaments become oriented more or less perpendicular to the thallus surface.



Multiporate: in corallines, a term used to describe a conceptacle whose roof is perforated by a number of pores.



Neotype: a specimen selected to serve as the nomenclatural type when all of the original material used by an author to describe a species has been lost or is missing.

Non-geniculate: lacking genicula.

Order: in nomenclature, the *principal* taxonomic rank above family.

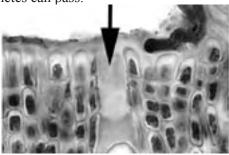
Paua: abalone; any of the various univalve marine molluscs of the genus *Haliotis*.

Pit connection: a link between two cells in which a pit plug occurs in an opening in the contiguous cell walls.

Pit plug: a distinct more or less lens-shaped structure constituting a part of a primary or secondary pit-connection.

Plant: the term 'plant' is used in the guide in the traditional sense (i.e., including photosynthetic protists or algae) as explained in item 7 of the preamble in the International Code of Nomenclature (Grueter et al., 2000).

Pore canal (or pore): in coralline conceptacles, an opening in the roof through which spores or gametes can pass.



Primary pit connection: a pit connection between adjacent cells of the same filament.

Protuberance: in non-geniculate corallines, a cylindrical to compressed or irregularly shaped outgrowth or branch of a plant.

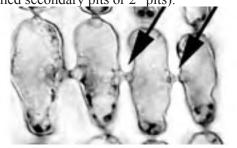
Ribbon-like: in non-geniculate corallines, a term used to describe the growth form of a plant composed of flat, ribbon-like branches and lacking a distinct holdfast.



Rhodolith: a term used to describe an unattached coralline plant that has formed as a result of fragmentation or from envelopment of a looselying stone or other object.

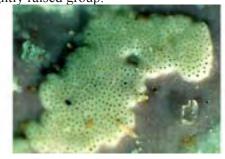


Secondary pit connection: a pit connection between cells of two adjacent filaments (also termed secondary pits or 2° pits).



Semi-endophytic: growing partly within another plant, e.g., *Choreonema*.

Sorus (sori): in the Sporolithaceae, a cluster of calcified compartments (each containing one sporangium) occurring as a surface patch or slightly raised group.



Species: in nomenclature, the lowest *principal* rank that can be accorded to a taxonomic group. Names of species are binomials, consisting of a generic name and a specific epithet, e.g., *Sporolithon durum*.

Spermatium (spermatia): the male gamete.

Spermatangium (spermatangia): a structure that contains spermatia.

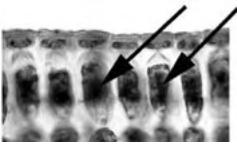
Sporangium (sporangia): spore-producing cells or structures.

Spore: a one-celled reproductive cell derived by mitosis (mitospore) or meiosis (meiospore) and capable of growth directly into another plant.

Stipe: a stalk, which in arborescent corallines

connects the holdfast with the main expanded part of the plant.

Subepithallial initial: in corallines, a meristematic cell situated beneath an epithallial cell in a filament. Subepithallial initials divide to give rise to new epithallial cells outwardly and new ordinary vegetative cells inwardly.



Subfamily: in nomenclature, a subordinate rank between the principal ranks of a family and genus. Names of the subfamilies end in —oideae, for example, Mastophoroideae.

Subtending: immediately below, as, for example, the cells beneath a subepithallial initial in a vegetative filament.

Syntype: any one of the original specimens used by the author to describe a species when no holotype was designated.

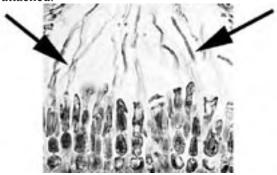
Taxon (taxa): a group of organisms of any taxonomic rank, e.g., family, genus, or species.

Tetrasporangium (tetrasporangia): a sporangium containing four spores, usually in a distinctive arrangement.

Tetraspore: any one of the four spores formed within a tetrasporangium. Tetraspores are generally presumed to be formed as a result of meiosis.

Thallus (thalli): the plant body of a non-vascular plant.

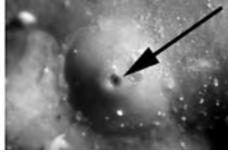
Trichogyne: the slender prolongation from the carpogonium to which spermatia become attached.



Type locality: the geographical site at which the type material of a species or infraspecific taxon was collected.

Unconsolidated: in non-geniculate corallines, a term used to describe the growth form of a plant composed of filaments that do not group together to form a distinct thallus (for example, the thallus of *Choreonema*).

Uniporate: in corallines, a term used to describe a conceptacle with a single pore.



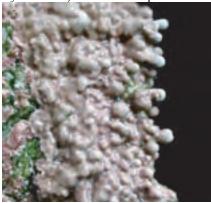
Ventral strut: in non-geniculate corallines, a downward growing branch arising from the ventral side of a lamellate branch.



Volcano-like: refers to multiporate conceptacles that have a distinct rim and central sunken pore plate.



Warty: in non-geniculate corallines, a term used to describe the growth form of a plant with short (usually < 3 mm) unbranched protuberances.



Zonate: a term used to describe the arrangement of four spores lying in a single row within a tetrasporangium. In corallines, zonate tetrasporangia characteristically occur in the Corallinaceae and Hapalidiaceae, but not in the Sporolithaceae.



Zygote: the diploid cell resulting from fusion of two haploid gametes.

Appendix 1. Table A1 – Species found at collection localities

	Appointment in re-	Corallinaceae										Hapalideaceae								Sporolithaceae				
		Lithophylloideae Mastophoroide							ae	Ch.		Melobesioideae												
	T			liopity	ynorac	ac	. Φ	IVIC	Stopii	Orolac	Jac	OH.	l	T		IVICIO	003101	lucac	T	T			1	e G
Locality number	Locality name (# collection sites)	L. carpophylli	L. corallinae	L. johansenii	L. pustulatum	L. stictaeforme	intermediate between L. corallinae & stictaeforme	H. improcerum	P. coronatum	P. fragile	S. yendoi	C. thuretii	M. membranacea	M. engelhartii	M. erubescens	M. macroblastum	M. printzianum	P. repandum	S. patena	S. schielianum	either M. engelhartii or S. patena	H. homalopasta	S. durum	Sterile rhodoliths (either
lorth Islai	nd, west coast					-	-			-		-		ı	-	-	J	-		-	-	-	J	
1	Parahaki Stream Reef																							
2	Radio Mast Boulders														0			0					0	
3	Waiwhakaiho Reef																0	0						
4	Sugar Loaf Islands (2 sites)																0	0				0		
5	Kawaroa Reef						0										0							
6	Hapuka Rock														0		0							
7	Belt Road Reef																							
8	Port Taranaki																							
9	Puketapu Road																							
10	Kapiti Island (3 collection sites)														0			0					0	0
11	Makara (4 collection sites)	0													Ŏ	0	0				0			0
		_																						
North Islai	nd, east coast								,				,				,		,			,	,	
12	Te Rangiharu Bay																							
13	Raukokore																					•		
14	Te Tapuwae O Rongokako																							
15	Tatapouri																							
16	Makorori (2 collection sites)																							
17	Kaiti Beach																							
18	Tuamotu Island (2 collection sites)	•							•						•						0			
19	Mangakuri Beach																							
20	Tuingara Point (3 collection sites)					0							0		0		0	•			0			
21	Pourerere														0									
22	Aramoana (2 collection sites)															0								
23	Mataikona (4 collection sites)				0		0					☆	0		0									
24	Castlepoint									☆														
North Islan	nd, Cook Strait																							
25	Cape Palliser											☆												
26	Scorching Bay																							
27	Breaker Bay																							
28	Palmer Head (6 collection sites)								0		Ō			0	D	Ō	Ō		0	0	D			
29	Island Bay (12 collection sites)	Ŏ				Ŏ			Ŏ	•	Ŏ		0		Ŏ	Ŏ	Ŏ	Ŏ	Ŏ	0	Ŏ			
30	Princess Bay	Ŏ							•															
31	Owhiro Bay								☆						☆									

Table A1 indicates the localities at which each of the 20 species in this guide have been confirmed to occur during the present study. Where only herbarium collections of a species have been identified from a locality (i.e., collected before this study), this is indicated by a star. Otherwise, intertidal, subtidal, or both inter- and subtidal collections at a locality are indicated according to the legend:

- Intertidal
- Subtidal
- Intertidal & subtidal
- ☆ Herbarium specimens only

Details for 14 localities outside central New Zealand are also given.

Notes:

Intermediate between *L. stictaeforme* & corallinae: Seven collections showed conceptacle anatomy intermediate in form between these two species (see Taxonomic Notes, Figures 12.2, 12.5).

Either *M. engelhartii* or *S. patena*: Male plants are required to place collections definitively in either species (see Taxonomic Notes, Figures 12.12, 12.17). Collections in this column lacked males.

Sterile rhodoliths (either *Sporolithon* or *Lithothamnion*): these collections lacked reproductive structures, so are unable to be unequivocally identified. Vegetatively, however, they can be placed in either *Sporolithon* or *Lithothamnion*.

	Table A1 (continued)																							
	(33131)				C	Corallin	пасеа	е							Н	apalid	eacea	ae				Sporoli	thacea	ae
			Lit	thophy	ylloide			Ма	stoph	oroide		Ch.				Melo	besioi	deae						
Locality number	Locality name (# collection sites)	L. carpophylli	L. corallinae	L. johansenii	L. pustulatum	L. stictaeforme	intermediate between L. corallinae & stictaeforme	H. improcerum	P. coronatum	P. fragile	S. yendoi	C. thuretii	M. membranacea	M. engelhartii	M. erubescens	M. macroblastum	M. printzianum	P. repandum	S. patena	S. schielianum	either M. engelhartii or S. patena	H. homalopasta	S. durum	Sterile rhodoliths (either Sporolithon or Lithothamnion)
South Isla	nd, Cook Strait											,										,		
32	Wharariki Beach (2 collection sites)																							
33	Patons Rock (2 collection sites)					Q					0				0			0						
34	Wainui (2 collection sites)					Ó	0									0	0	O						
35	Golden Bay (4 collection sites)					0					0				0		0	0						
36	Abel Head	☆																			A			
37	Pinnacle I																				☆			
38	Nelson Haven														_			0						
39	Boulder Bank														Q							Q		
40	Cable Bay (4 collection sites)					•									0		•					0	0	
41	D'Urville Island (4 collection sites)														Ò		0	0			0			0
42	Maud Island														☆									
	nd, west coast						0		0						\cap	0					0			
43	Steeples (4 collection sites)			0		0					0				0		0	0						
	Charleston																							
45	Seal Rock Island														-									
46	Greigs																							
South Isla	nd, east coast																							
47	Cape Campbell																							
48	Chancet Rocks										Ó		Ŏ								Ŏ			
49	Maungamanu																							
50	Rakautara (4 collection sites)											Ŏ												
51	Kaikoura (3 collection sites)															☆			☆		☆			
52	Oaro Reef																							
53	Sumner (4 collection sites)					0					0				0		0	Ō	0		0			
54	Macintosh Bay														Ŏ									Intertidal
55	Diamond Harbour																							
56	Camp Bay														0		0							O Subtidal
57	Pigeon Bay					0									0		0	0			0			Intertidal & subtidal
58	Decanter Bay														0	0								→ Horborium anasimosas
59	Raupo Bay														0									☆ Herbarium specimens o

Tab	ole A1 (continued)					`oralli	nacea	0								analic	leacea	20				Sporoli	thacea	0	
			1.0	ر ما مر ما ا			Hacea	_	stoph	- u - i - i -		Ch					besioi					Sporon	шасса	.6	
	I		LII	triopri	ylloide	ae	4		IStopni	Jiolue	lae	Ch.		1		ivieio	Desioi	ueae	T	T	1	1	T	1	
Locality number	Locality name (# collection sites)	L. carpophylli	L. corallinae	L. johansenii	L. pustulatum	L. stictaeforme	intermediate between L. corallinae & stictaeforme	H. improcerum	P. coronatum	P. fragile	S. yendoi	C. thuretii	M. membranacea	M. engelhartii	M. erubescens	M. macroblastum	M. printzianum	P. repandum	S. patena	S. schielianum	either M. engelhartii or S. patena	H. homalopasta	S. durum	Sterile rhodoliths (either Sporolithon or Lithothamnion)	
Chatham	Islands						,																		
60	Cape Young														☆					☆	☆				
61	Wharekauri																								
62	Okawa Point (2 collection sites)																								
63	Waitangi West														☆										
64	Whangatete Inlet																								
65	Port Hutt																								
66	Ocean Beach														☆						☆				
67	Waitangi																								
68	Heaphy Shoal (2 collection sites)																								
69	Point Durham (3 collection sites)																			☆	☆				
70	Te One Creek																								
71	Owenga Wharf																								
72	Tommy Solomon (2 sites)																								
73	Pitt Island (2 collection sites)									☆					☆										
North Isla	nd, north of zone																								
74	Russell (2 collection sites)								☆												☆				
75	Poor Knights Is.								M												H			☆	
76	Little Barrier I.	☆																						W	
77	Great Barrier I.	M	☆																						
78	Mangere I.		M												☆										
79	Waiheke Channel														Ö									0	
80	Army Bay																							☆	
81	Little Manly																							Ô	
82	Aldermen Is.																				☆				
83	Slipper Island																				- A			0	
84	Maketu																								Intertidal
85	Te Kaha																						☆		
																				1					O Subtidal
South of z	zone																								Intertidal & subtidal
86	Campbell Island														☆										
87	Antarctica (5 sites, Ross Sea)																				0				☆ Herbarium specimens onl

Appendix 2. Table A2 – Reference codes for specimens pictured in the guide

Species names and herbarium numbers of specimens pictured in the guide. Most specimens are held at WELT (Museum of New Zealand Te Papa Tongarewa), and have an A prefix. One specimen (Figure 7.8, B) from CHR (Landcare, Christchurch) has a CHR prefix.

n/a, not applicable.

Figure		Identification	Herbarium
7.2 Coralline substrates	ABCDEFGH	Corallina sp. Unidentified geniculate coralline Choreonema thuretii Spongites yendoi Hydrolithon improcerum Spongites yendoi Unidentified non-geniculate Sporolithon durum	number n/a n/a A027038 A027039 A026945 A027040 n/a A026998
7.6 Uniporate conceptacles	ABCDEFGH	Pneophyllum fragile Pneophyllum coronatum Spongites yendoi Lithophyllum sp. Lithophyllum carpophylli Unidentified coralline Lithophyllum carpophylli	A026984 A026976 A027041 A027054 A026948 n/a n/a A026948
7.7 Multiporate conceptacles	ABCDEFGH	Mesophyllum printzianum Synarthrophyton schielianum Mesophyllum erubescens « Synarthrophyton patena Melobesia memuranacea	A026972 A027006 A027042 A027042 A026956 A027001 A026966 A027028
7.8 Calcified compartments	ABCDEFG	Sporolithon durum " Heydrichia homalopasta Sporolithon durum Heydrichia homalopasta	A027043 CHR39696 n/a A026942 n/a n/a n/a
7.9 Male, female, and carposporangial conceptacles	ABCDEFGH	Mesophyllum engelhartii Lithophyllum carpophylli Synarthrophyton schielianum S. patena/M. engelhartii "	A026958 A026958 A026948 A027004 n/a n/a n/a

7.10 Vegetative characters	ABCDEFGH	Mesophyllum printzianum Lithophyllum sp. Sporolithon durum Lithophyllum stictaeforme Mesophyllum macroblastum Phymatolithon repandum Synarthrophyton patena Melobesia membranacea	A027044 A027055 A027045 A027020 A027046 A027047 A027002 A027048
9.10 Cell connections	ABCDEFGH	Unidentified non-geniculate Pneophyllum coronatum Unidentified non-geniculate Hydrolithon improcerum Unidentified non-geniculate Lithophyllum pustulatum Unidentified non-geniculate	n/a A026981 n/a A027029 n/a A027017 n/a n/a
9.11 Reproductive features	ABCDEFGH	Lithophyllum stictaeforme Synarthrophyton schielianum Unidentified non-geniculate Synarthrophyton patena	A027024 A027024 A027007 A027007 n/a n/a A027049 A027049
12.1 Lithophyllum carpophylli	ABCDEFG	Lithophyllum carpophylli " " " " " "	A026948 A026948 A026947 A026949 A026948 A026948 A026948
12.2 Lithophyllum corallinae	ABCDEF	Lithophyllum corallinae " " " "	A003210 A003210 A003210 A003210 A003210 A003210
12.3 Lithophyllum johansenii	ABCDEF	Lithophyllum johansenii " " "	A026951 A026952 A026951 A026952 A026950 A026950

12.4 Lithophyllum pustulatum	ABCDEF	Lithophyllum pustulatum " " "	A027017 A027016 A027019 A027018 A027019 A027019
12.5 Lithophyllum stictaeforme	ABCDEFG	Lithophyllum stictaeforme " " " " " " "	A027021 A027023 A027022 A027020 A027022 A027024 A027024
12.6 Hydrolithon improcerum	ABCDEFG	Hydrolithon improcerum " " " " " " "	A026945 A026945 A026945 A026945 A027029 A027029 A026946
12.7 Pneophyllum coronatum	ABCDEFGH-	Pneophyllum coronatum " " " " " " " " " " " " "	A026975 A026978 A026974 A026981 A026976 A026980 A026977 A026982 A026979
12.8 Pneophyllum fragile	ABCDEF G	Pneophyllum fragile " " " " " "	A026984 A026985 A026987 A026985 A026986 A026983 A026985
12.9 Spongites yendoi	ABCDEFG	Spongites yendoi " " " " " "	A027011 A027013 A027012 A027014 A027010 A027015 A027015

12.10 Choreonema thuretii	ABCDEFG	Choreonema thurettii " " " " " " "	A027066 A027038 A027038 A027066 A027066 A027066 A027066
12.11 Melobesia membranacea	ABCDEFGH	Melobesia membranacea " " " " " " " "	A026963 A026967 A026964 A026966 A026966 A027028 A026965 A026965
12.12 Mesophyllum engelhartii	ABCDE	Mesophyllum engelhartii " "	A026958 A026958 A026958 A026958 A026958
12.13 Mesophyllum erubescens	ABCDUFGH	Mesophyllum erubescens " " " " " "	A026953 A026954 A026956 A022625 A026953 A026956 A026955 A026955
12.14 Mesophyllum macroblastum	ABCDEFG	Mesophyllum macroblastum " " " " " "	A026959 A027027 A026957 A026960 A026961 A026961 A026961
12.15 Mesophyllum printzianum	ABCDEFG	Mesophyllum printzianum " " " "	A026972 A026970 A026968 A026969 A026973 A026971 A026968

12.16 Phymatolithon repandum	ABCDEFGH	Phymatolithon repandum " " " " " " " " "	A026993 A026994 A026988 A026989 A026990 A026992 A026995 A026991
12.17 Synarthrophyton patena	ABCDEFG	Synarthrophyton patena " " " " " "	A027003 A027000 A027005 A027001 A027001 A027002 A027002
12.18 Synarthrophyton schielianum	ABCDEFG	Synarthrophyton schielianum " " " " " "	A027008 A027007 A027007 A027006 A027009 A027004 A027004
12.19 Heydrichia homalopasta	ABCDEF	Heydrichia homalopasta " " "	A026941 A026940 A026944 A026943 A026941 A026942
12.20 Sporolithon durum	ABCDEF	Sporolithon durum " " " "	A026997 A026998 A026997 A026996 A026999 A026999