UNIVERSITY OF SOUTHAMPTON

FACULTY OF NATURAL AND ENVIRONMENTAL SCIENCES

Ocean and Earth Science

INTERACTIONS BETWEEN ALGAE AND NEMATODES: HABITAT PROVISION AND DETRITAL DRIVEN PROCESSES

by

Hyeong-Gi Kim

Thesis for the degree of Doctor of Philosophy

June 2017

UNIVERSITY OF SOUTHAMPTON

ABSTRACT

SCHOOL OF OCEAN & EARTH SCIENCE

Doctor of Philosophy

INTERACTIONS BETWEEN ALGAE AND NEMATODES: HABITAT PROVISION AND DETRITAL DRIVEN PROCESSES

by Hyeong-Gi Kim

The overall aim of this thesis was to investigate marine nematode-algae interactions using a twofold approach: habitat provision by algae and the role of algal detritus in structuring nematode assemblages. Nematode assemblages living in algae on the south and east coast of Korea were compared using a hierarchical sampling design with those from the south coast of the British Isles in order to investigate nematode diversity and functional traits at different spatial scales (patch, shore, region and biogeographic realm). The cosmopolitan genus *Corallina* and *Sargassum muticum*, a native of Korea and an invader in the British Isles were investigated as algal habitat providers for nematodes. The species composition and functional traits of nematode assemblage in both species of macroalgae were significantly different at all spatial scales. The alpha, beta and gamma biodiversity of nematodes in each macroalgae were also measured at all spatial scales. The alpha, beta and gamma diversity of nematodes across four spatial scales in two different habitats are an alpha dominant relationship indicating random colonization and high species turnover at small scales.

Nematode species composition and diversity on the wrack-loaded sandy shore were explored using field experiments. Mesh bags filled with three different types of macroalgae (brown, green and red) were used to test the effects of diversity of algal detritus on nematode assemblages involved in decomposition processes. The detrital treatments consisted of 10 combinations of macroalgae (monoculture, two and three species treatments), containing different proportions of algal material in order to test the impacts of detrital diversity on nematode assemblages. The density, diversity and species composition of nematodes in combinations of algae were higher than with single types of macroalgae. This indicated that the combination of two algae mixtures offered more favourable environments than the single species bags. Overall this thesis demonstrated the high diversity of nematodes inhabiting seaweeds and the role of detrital composition in driving nematode assemblages.

Table of Contents

List of Tables

List of figures

DECLARATION OF AUTHORSHIP

I, Hyeong-Gi Kim, declare that this thesis and the work presented in it are my own and has been generated by me as the result of my own original research.

Interaction between algae and nematode: habitat provision and detrital driven processes

I confirm that:

1. This work was done wholly or mainly while in candidature for a research degree at this University;
2. Where any part of this thesis has previously been submitted for a degree or any other qualification at this University or any other institution, this has been clearly stated;
3. Where I have consulted the published work of others, this is always clearly attributed;
4. Where I have quoted from the work of others, the source is always given. With the exception of such quotations, this thesis is entirely my own work;
5. I have acknowledged all main sources of help;
6. Where the thesis is based on work done by myself jointly with others, I have made clear exactly what was done by others and what I have contributed myself;
7. None of this work has been published before submission

Signed: .....

Date: .....

Acknowledgements

Firstly, I thank my supervisors Stephen J Hawkins, Lawrence E. Hawkins and Jasmin A Godbold for all the great support during all phases of my PhD. I would like to extend enormous thank to Stephen J Hawkins for giving me an opportunity to learn how to enjoy “counting things” on the shores and more importantly British beer. I am also very thankful for all the help and support from Moira Fisher and Katrin Bohn. Without their support, I would not have collected my nematode sample and would not have adapted to British life. Many thanks to Hyun Soo Rho and his research group from the Korean Institute of Ocean Science and Technology (KIOST) for nematode identification and taxonomy. I am also vary grateful to Chul-Woong Oh and his students from Pukyong National University for all the help when I stayed and studied in Korea.

Special thanks go to friends for the continuous support and backup and to my officemates Dan and Mat for being with me, talking with me, making me smile all the time, even at the most desperate times in the field, lab or during the write-up.

Finally, I would like to thank all of my family and dedicate this especially to my parents, Taeyong Kim and Jung Ok Lee. My hard work and determination to undertake this study would not have been possible without the investment and sacrifice that they have made over the years to encourage my success. I hope this is the starting point of pay back.

Chapter 1: General Introduction

1.1 Scope of thesis and this chapter

Free-living nematodes are an extremely diverse and numerous group of organisms widely found across marine, freshwater and terrestrial ecosystems (Heip et al. 1985, Holovachov & Schmidt-Rhaesa 2014). Compared with larger taxa they have been relatively understudied despite their prevalence and likely importance in underpinning ecosystem processes. The overall aim of my thesis was to investigate marine nematode-algae interactions using a twofold approach: habitat provision by algae and the role of algal detritus in structuring nematode assemblages.

First, I compared nematode assemblages living in algae on the south coast of Korea (North-East Asia, Indo-pacific biogeographic region) with the south coast of the British Isles (Northern Europe, North-East Atlantic biogeographic region) in order to investigate nematode diversity and functional traits at different spatial scales. The cosmopolitan macroalgal genus *Corallina* is an important habitat forming species on coasts worldwide (Walker et al. 2009). Comparisons were also made of nematode assemblages living in *Sargassum muticum,* a native of Korea and North-East Asia that invaded Northern Europe in the 1970s (Farnham et al. 1973) Assemblage structure composition community and functional traits were compared. Comparisons at different spatial scales in the same habitat structure were performed to understand the role of biogeographical and ecological processes in determining nematode species diversity and distribution patterns. Secondly, the role of diversity of macro-algal detritus in shaping nematode species composition and diversity was explored. Algal detritus is one of the major sources of energy in coastal areas (Raffaelli & Hawkins 1996), influencing community structure and ecosystem functioning (Polis et al. 1997, Barreiro et al. 2012).

In the rest of this general introduction, I first briefly summarise the underlying theories of the relationship between biodiversity and ecosystem functioning. I then briefly review broad-scale studies of biodiversity and assemblage composition made to date in marine ecosystems. I then outline the contribution of nematodes to global biodiversity and their role in ecosystem functioning. This introductory chapter concludes by setting the scene for the rest of the thesis

before outlining the overall aims followed by the rationale of each individual chapter.

1.2 Biodiversity and Functioning of Ecosystems

The relationship between biodiversity and ecosystem functioning (B-EF) has been a topic of interest during the past two decades because of the potential effect of biodiversity loss on ecosystem processes (Naeem et al. 2009, Cardinale et al. 2012, Hooper et al. 2012, Solan et al. 2012, De Laender et al. 2016). With increasing human population, anthropogenic activities have been changing the environment on global and local scales leading to rapid global changes in biodiversity worldwide with consequences for ecosystems (Halpern et al. 2008, Hooper et al. 2012). As a result of rapidly declining biodiversity (Gonzalez et al. 2016), there is a need to know how decreasing biodiversity can affect ecosystem functioning and consequently services. It still remains unclear, however, whether a few or many of the species in an ecosystem are needed to sustain the provisioning of ecosystem services (Isbell et al. 2011, Cardinale et al. 2012). In addition to recent experiments (>600 studies), B-EF studies have led to development of mathematical theory which can explain global patterns in natural ecosystems (Isbell et al. 2011, Cardinale et al. 2012). This theory indicated that increasing the numbers of species enhances the number of functional roles and properties (Isbell et al. 2011). Moreover, reductions in number of species, genera and functional groups leads to decreases in the efficiency by which whole communities capture biologically essential resources and convert those resources into biomass (Cardinale et al. 2012). Therefore, to determine how many species are needed to provide ecosystem functioning requires studies of multiple functional groups replicated in time and place (Isbell et al. 2011, Cardinale et al. 2012, Gamfeldt et al. 2015).

In marine benthic ecosystems, much research has been directed towards this issue, focusing especially on the relationship between biodiversity, species interactions and ecosystem functioning in marine sediments (Ieno et al. 2006, Norling et al. 2007, Godbold et al. 2009, Michaud et al. 2009, Godbold et al. 2011, Byrnes et al. 2014) and also on rocky shores (Crowe et al. 2000, O'Connor & Crowe 2005, Griffin et al. 2009). Most of these previous studies, however, have primarily focused on the influence of macrofauna in marine ecosystems, without any recognition of the contribution of other size-classes of organisms such as meiofauna to ecosystem processes (Piot et al. 2014).

The loss of different size classes of species can affect ecosystem functioning (Coleman & Williams 2002), modify trophic interactions (Thébault et al. 2007) and community dynamics (Vasas & Jordán 2006). Therefore, observations would consider all major relationship between species to explain adequate ecosystem functioning (Thébault et al. 2007). Therefore, understanding B-EF relationships also requires consideration at different size classes in order to draw more realistic conclusions (Vasas & Jordán 2006). A particular gap has been research on patterns of meiofaunal (Hakenkamp & Morin 2000) and microbial (Schimel et al. 2007) diversity and how they influence ecosystem functioning.

1.3 Geographical Patterns of Biodiversity

The biodiversity, structure and dynamics of benthic communities can be affected by environmental factors operating at different scales from microhabitats (~ 1m), through local gradients (10m to 10km) and regional differences (100km) up to biogeographic realms (1000s km) (Levin 1992, Frey 2010). Therefore, many studies have directly dealt with this issue (Mayr 1942, Levin 1992, Frey 2010, Dray et al. 2012). In terrestrial systems, the highest species diversity generally occurs in the tropics, particularly in rain forests with decreases towards the poles (Pianka 1966, Wilson & MacArthur 1967). In the marine environment, patterns seem much less clear (Gray 1997, Tittensor et al. 2010). In terms of latitudinal patterns of diversity, species diversity increases from the Arctic to the tropics have been recorded (Thorson 1957, Kendall & Aschan 1993) that are similar with the species diversity patterns seen in terrestrial systems. In contrast, the gradient from the Antarctic to the tropics is much less clear due to relatively higher diversity for many taxa in Antarctic regions (Clarke 1992). The oceans of the Southern hemisphere are larger than those of the Northern hemisphere ocean, and have been relatively little studied (Poore 1993).

Much less work has been done on longitudinal patterns of diversity. Two major patterns have, however, been observed: coastal species diversity was the highest in Western Pacific; whereas for oceanic species, diversity levels are similar across broad mid-latitudinal bands in all oceans (Tittensor et al. 2010).

Geographical patterns of diversity are strongly dependent on the size of sampling scale (Underwood 1997, Underwood & Chapman 2013). At small scales, species seem to interact with each other and compete for similar limiting resources which are called within-habitat or α diversity (Fisher et al. 1943, Whittaker 1967, Gray

1997). At slightly larger scales, diversity within single or multiple habitats or communities is driven by the surrounding environment – so called between-habitat or β diversity) (Whittaker 1970, Gray 1997). At even broader scales, evolutionary isolation and biogeographic processes have more influence than ecological processes. This scale has been called γ diversity (representing the species pool in a particular geographic location) (Franklin 1993, Tuomisto et al. 1995). Therefore, species diversity needs to be investigated at different spatial scales in order to fully understand geographical patterns of biodiversity.

1.4 Role of Nematodes in Ecosystem Functioning

Nematodes are the most numerous and diverse metazoans in the biosphere (estimates of between 40,000 and 10,000,000 species in the phylum Nematoda) (Holovachov & Schmidt-Rhaesa 2014). They are present in every habitat such as terrestrial plants, soils, algae, sediments of shallow water to the deep sea (Platt et al. 1980). As parasites, they can cause disease in hundreds of millions of people. It is estimated that eight billion nematodes are enjoying food, warmth and shelter in vertebrate and human intestines (Holovachov & Schmidt-Rhaesa 2014). For example, *Ascaris lumbricoides* is one of the best-known parasitic nematodes, as it is the second most infectious organism in the world (Chan 1997). Plant-parasitic nematodes can cause serious damage to crops throughout the world (Ash et al. 1984, Heip et al. 1985). Parasitic nematology has become a vital research topic as a result of their economic and health importance. *Caenorhabditis elegans* (Rhabditina) has become a well-known model species for genome based studies, having been sequenced early on (Fire et al. 1998). Studies aiming to relate gene expression to the development and functioning of organisms using advanced molecular biology are underpinned by the use of *C. elegans* (Fire et al. 1998).

In contrast, free-living nematodes have been little documented and remain relatively unstudied, despite their functional importance in aquatic and soil ecosystems (Platt et al. 1980). They are also the most abundant meiofauna (size between 63 and 500µm) in the marine environment (Platt et al. 1980). They are ubiquitously distributed, with high diversity ranging from very tolerant species to species sensitive to disturbance (Heip et al. 1985). They play an important role in the link between smaller (e.g. bacteria, primary producers) and larger organisms (macrofauna, demersal fauna) in benthic sediments as predators of smaller organisms and as an important food resource for larger organisms (Giere 2009). Thus they play a major energetic role in marine benthic environments (Coull

1999), facilitating the mineralization of organic matter (Coull 1999). Furthermore, nematodes are highly habitat specific (Joint et al. 1982). Changes in the composition of nematode assemblages can provide good indicators of when changes are occurring in the environment, either naturally (Nozais et al. 2005) or as a result of anthropogenic activities (Bongers & Ferris 1999).

Therefore, studies of nematode structure and biodiversity are necessary to understand the whole of benthic ecosystem functioning (Giere 2009, Piot et al. 2014). Nematodes are also a good model system to study the relationship between biodiversity and ecosystem functioning due to their rapid turn-over and short generation times.

1.5 Rationale of thesis

The overall aim of my thesis was to explore the interactions between algae and nematodes focusing on the importance of habitat provisioning by algae for nematode diversity as well as the role of algal detritus in shaping nematode assemblage abundance and composition. My approach to understanding habitat provisioning by seaweeds for nematodes has been a comparative study between the south coast of Korea and south coast of the British Isles. Two macroalgae were chosen as habitat-forming species. The cosmopolitan genus *Corallina* was chosen to investigate broadscale comparisons of nematode biodiversity, assemblage structure and composition. Previously, *Corallina officinalis* was considered as a cosmopolitan species occurring in both the Atlantic and Indo-Pacific (Walker et al. 2009). However, recent work revised its taxonomy (Walker et al. 2009). Despite the taxonomic changes, the very similar morphology of *Corallina* spp. ensures that habitat provision would be similar between the British and Korean coast. I also selected *Sargassum muticum* as a test species in order to investigate nematode diversity, assemblage structure and biological traits at different spatial scales. *S. muticum* is a native species in North-East Asia occurring in China, Japan, Korea and has become a worldwide invasive species (North 1973), reaching Europe (Critchley et al. 1983) in the 1970s. It was first found in the British Isles in the Isle of Wight in 1973 (Farnham et al. 1973). This species provided the opportunity to compare assemblages of nematodes associated with a species of algae within its native home range and when occurring as an invasive species. Moreover, both macroalgae were common species in the British Isles and Korea. Therefore, the south coast of British Isles and Korea were selected to compare geographic differences in nematode diversity

and assemblage composition using a hierarchical nested design. The second section of my thesis was experimental work exploring how biodiversity of algal detritus influenced nematode abundance, diversity and assemblage structure. This gave insights into the important contribution of nematodes to biochemical cycling of nutrients and thus ecosystem functioning.

In chapter 2, the general methods used throughout the thesis are described, along with description of the study sites.

In chapter 3, geographical patterns of the abundance, diversity, composition and biological traits of nematodes living in turf of *Corallina* spp. are compared using a hierarchical survey design between the south coast of Korea and the south coast of the British Isles taking account of the environmental gradients in each area.

In chapter 4, nematode abundance, diversity composition and biological traits in *Sargassum muticum* are compared within the home range of the algae in Korea and where it is an invasive species in the British Isles.

In chapter 5, both hierarchical designs from chapter 3 (habitat provision by *Corallina*) and chapter 4 (habitat provision by *Sargassum*), were used to compare alpha, beta, gamma diversity of seaweed dwelling nematodes in the British Isles and Korea.

In chapter 6, the role of biodiversity in decomposition processes was explored by experimentally investigating how diversity of algal detritus influenced nematode abundance, assemblage composition and diversity.

Chapter 7 provides an overview and synthesis of the thesis, as well as discussing the limitations work and making suggestions for further research.

Chapter 2: General methods and study sites

2.1 Introduction

In this short chapter the study locations used are briefly summarised and key contextual geographic and environmental information summarised. General methods used throughout the thesis are also outlined in more detail. My study used two distinct macroalgae as habitat providers of free-living nematodes to investigate broad-scale comparisons. *Corallina* spp. is one of the most common seaweeds on the south coasts of England and Korea. What was once considered a cosmopolitan species has recently been divided into various species using molecular technics (Walker et al. 2009). *Sargassum muticum* (Yendo) Fensholt is an invasive species that was introduced from Asia into the British Isles in 1973, probably on Japanese Oysters, *Crassostrea gigas* (Farnham et al. 1973). It has become very common on rocky shores on the south coast of England.

2.2 Locations of study

2.2.1 English Channel, British Isles

The English Channel is the body of water that is situated between southern England and northern France; connecting the southern part of the North Sea to the Atlantic Ocean. The southern coast of England on the English Channel is 560 km long and varies in width from 33 to 240km covering an area of some 75,000 km^2; it is known as the smallest of the shallow sea within the continental shelf of Europe average depth between 45 to 120 metres (Organization 1953). To compare spatial variability, two shores in each of three regions were sampled on the south coast of England; in the western English Channel, Looe and Heybrook Bay for both species; in the central English Channel Portland Bill for *Corallina* samples, Osmington for *Sargassum muticum* samples, Swanage for both species, and the eastern English Channel, Brighton and Beachy Head, also for both species. It should be noted that in the central region, different sites were used for each species of macroalgae as there was insufficient *S.muticum* at Portland Bill. Figures 2.1 and 2.2 show the locations and general character of the sampling sites.

The sampling locations on the south coast of England are characterised by various macroalgae and invertebrates (Lewis 1964, Southward 1991, Hawkins & Jones 1992, Raffaelli & Hawkins 1996, Hawkins et al. 2008).

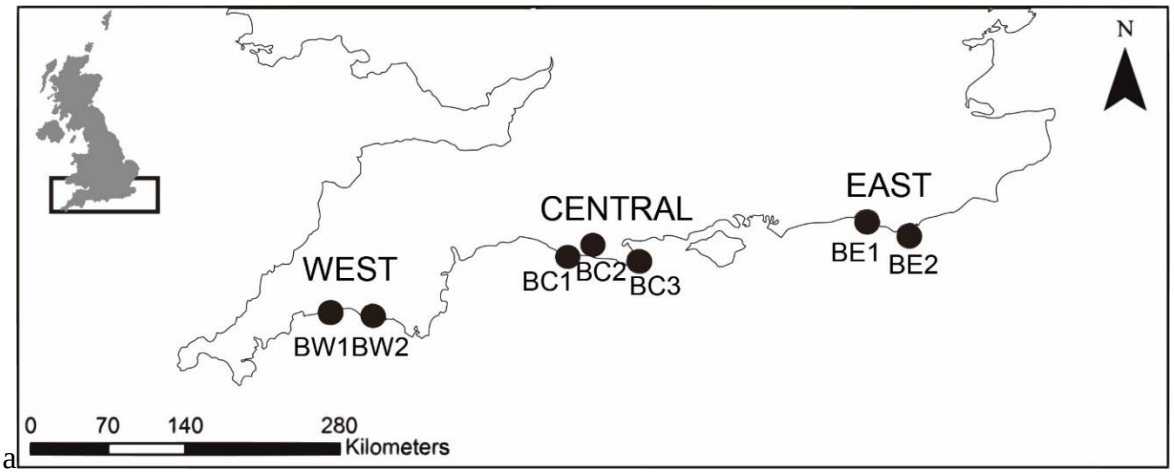
a 

Figure 2.1 The sampling locations in the British Isles; BW1: Looe (S,C), BW2: Heybrook Bay (S,C), BC1: Portland Bill (C), BC2: Osmington (S), BC3: Swanage (S,C), BE1: Brighton (S,C), BE2: Beachy Head (S,C); S: *S. muticum*, C: *Corallina* spp.

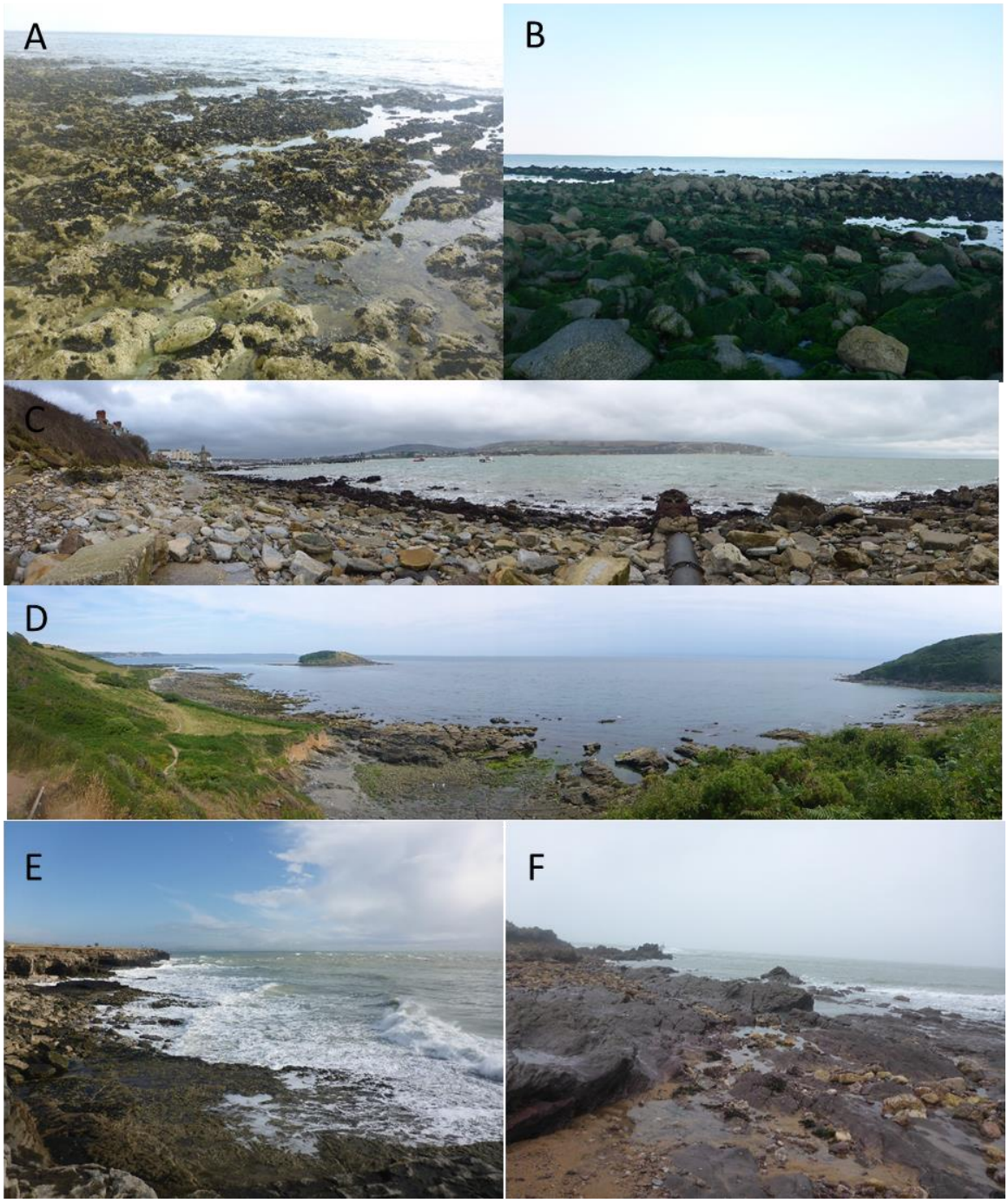


Figure 2.2 Photographs of sampling locations in the British Isles; A: Brighton, B: Beachy Head, C: Swanage, D: Looe, E: Portland Bill, F: Heybrook Bay.

2.2.2 South and East coast of Korea in the North-West Pacific Ocean

The south and east seas of Korea (Sea of Japan) form part of the North-West Pacific Ocean. The south coast of Korea is a ria coast line is a particularly complex; 255 km long in straight line, and 2,251 km in total coast line length covering an area of some 75,000 km^2. Its average depth is 100 meter and maximum depth is 198 metre. In contrast to the south coast of Korea, the east coast of Korea is a relatively simple coastline; it is 1,700 km long and maximum width of about 1,070 km. The East sea of Korea (Sea of Japan) has a surface area of 978,000 km^2, a mean depth of 1,752 meter and maximum depth of 3, 742 metres. The south coast of Korea has a gradient of tidal ranges (decreasing from west to east) while the south east coast is relatively constant (Table 2.1). The sampling locations on the south coast of Korea are characterised by various macroalgae and invertebrates. (Son & Hong 1998, Park et al. 2003, Yoo 2003, Jeong 2011). Figure 2.3, 2.4 and 2.5 show the location and visual information of the sampling sites.

To compare spatial variability, two shores in each of the three regions were surveyed on the south coast of Korea; West-Wando and East-Wando in the west, Yeosu and Namhae in the centre and Gueje and Busan in the east. In terms of *Sargassum muticum* samples, shores on the east cost of Korea were selected due to its limited distribution on the southeast coast of Korea; Jindo and Wando in the west, Yeosu and Busan in the south coast of Korea, and Ulsan and Pohang in the east coast of Korea.

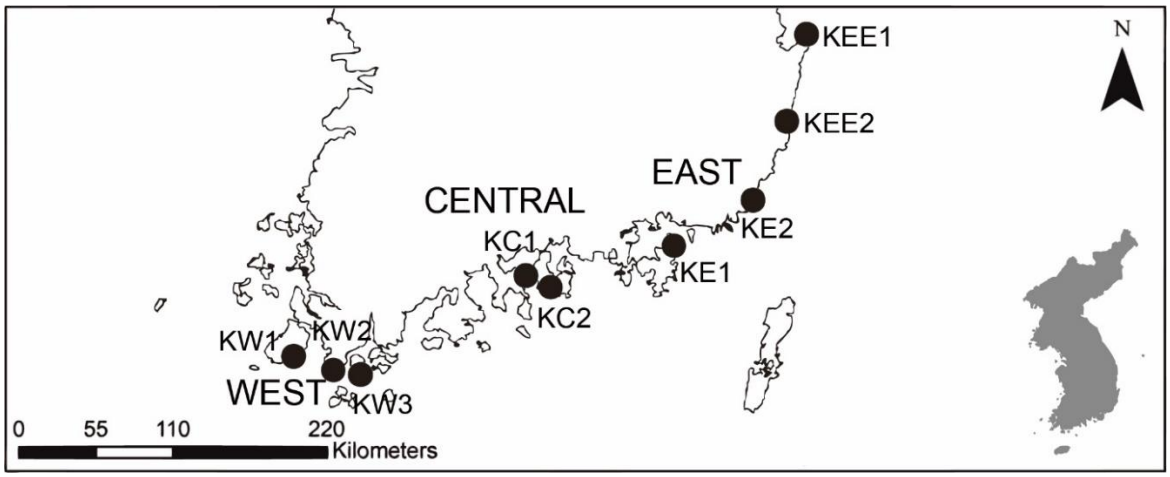


Figure 2.3 The sampling locations in South Korea; KW1: Jindo (S), KW2: west-Wando (C), KW3: east-Wando (S,C), KC1: Yeosu (S,C), KC2: Namhae (C), KE1: Gueje (C), KE2: Busan (S,C), KEE1: Pohang (S), KEE2: Ulsan (S); S: *S. muticum*, C: *Corallina* spp.

Figure 2.4 Photographs of sampling locations in the south coast of Korea; A: west-Wando, B: East-Wando, C: Yeosu, D: Namhae.

Figure 2.5 Photographs of sampling locations in the south and east coast of Korea; A: Jindo, B: Busan, C: Ulsan, D: Gueje, E: Pohang.

2.2.3 Environmental variables

Detailed seawater temperature, geographical position and tidal range in the sampling sites have been described in table 2.1. These broad-scale environmental data were collected from The Centre for Environment, Fisheries and Aquaculture Science (Cefas) in the British Isles and from The National Oceanographic Research Institute in South Korea. *Corallina* samples were collected in summer, 2014 (July in the British Isle and August in Korea). *S. muticum*, however, was collected at the season of maximum growth which differs between its native range in the Northwest Pacific (early spring, 2015), and Europe, invasive range (end of summer, 2014). Each shore in both geographical regions was classified and split into three groups using an Oceanic Index. An Oceanic Index value of three represents areas with largely open ocean water; two indicates mixed oceanic and coastal waters and one an area with mainly coastal water. The oceanic water tends to be clearer, warmer in the winter and have much lower sediment and nutrient.

Table 2.1 Environmental context of sampling locations

NO	Geographical Region	Locality	Latitude N	Longitude W	Tides Range (m)	Annual water temperature range (℃)	Oceanic Index
BW1	Atlantic	Looe	50°20'29.4"N	4°27'39.7"W	5.4	9~17	3
BW2	Atlantic	Heybrook Bay	50°19'09.1"N	4°06'51.7"W	5.53	9~17	3
BC1	Atlantic	Portland Bill	50°30'53.03" N	2°27'19.44"W	2.5	9~17	2
BC2	Atlantic	Osmington	50°38'02.9"N	2°22'34.1"W	2.5	9~17	2
BC3	Atlantic	Swanage	50°36'27.22" N	1°56'39.38"W	2.07	9~17	2
BE1	Atlantic	Brighton Marina	50°48'39.87" N	0°5'28.83"W	6.5	8~17	1
BE2	Atlantic	Beachy Head	50°44'15.0"N	0°15'09.1"E	7.3	9~17	1
KW1	Pacific	Jindo	34°23'45.39" N	126°16'31.5"E	4.03	8~25	2
KW2	Pacific	West-Wando	34°17'47.6"N	126°42'05.8"E	4.04	9~23	2
KW3	Pacific	East-Wando	34°18'13.5"N	126°45'52.3"E	4.04	9~23	2
KC1	Pacific	Yeosu	34°49'08.8"N	127°46'02.9"E	3.69	7~25	1
KC2	Pacific	Namhae	34°44'57.5"N	127°54'37.4"E	3.39	7~24	1
KE1	Pacific	Gueje	34°59'51.4"N	128°42'14.0"E	2.15	12~23	3
KE2	Pacific	Busan	35°09'39.9"N	129°11'38.8"E	1.35	11~23	3

Table 2.1 Environmental context of sampling locations

NO	Geographical Region	Locality	Latitude N	Longitude W	Tides Range (m)	Annual water temperature range (℃)	Oceanic Index
KEE 1	Pacific	Ulsan	35˚37'42.32" N	129˚26'33.66" E	0.56	10~25	3
KEE 2	Pacific	Pohang	36˚4'47.81"N	129˚34'5.13"E	0.3	9~26	3

2.3 Nematodes as model organisms

Nematodes are known as one of the most numerous and diverse taxon in the biosphere and are widely distributed in marine environments (Platt & Warwick 1980). Nematodes are also one of the most abundant meiofauna associated with macroalgae (Rysgaard et al. 2000, Schmid-Araya et al. 2002, De Oliveira et al. 2016). On rocky shores, nematodes enhance nutrition of macroalgae (Bracken et al. 2007) and control the densities of epifaunal species (Warwick et al. 1998). Nematodes are present in every habitat such as plants, soil, shallow water (Platt & Warwick 1980). They have low dispersal activity, are easily collected with low expense. Nematodes can be classified into groups with similar ecological traits (e.g. feeding type and life strategy), and have similar ecological characteristics at the genus level (Wieser 1953, Bongers et al. 1991). These advantages mean that nematode populations can respond rapidly to changes in environmental conditions either naturally (Nozais et al. 2005) or as a result of anthropogenic activities (Bongers & Ferris 1999). Therefore, nematodes are theoretically ideal candidates for use as an indicator for environmental monitoring, and to explore.

2.3.1 General characteristics and classification

Nematodes are generally described as microscopic, non-segmented animals with thread-like bodies (Holovachov & Schmidt-Rhaesa 2014). Over 25,000 species have been described and the majority of nematodes are translucent to allow observation of their internal anatomy without special methods (dissection or sectioning), and this can help to find the number of characters for practical identification (Platt & Warwick 1983). Nematodes have a relatively simple and conserved basic body that consist of an external cylinder (the body wall) and an internal cylinder (the digestive system), which are separated by a pseudocoelomic body cavity containing a number of cells and other organisms (Holovachov & Schmidt-Rhaesa 2014). About 99% of all known nematodes have long and narrow cylindrical body shapes, round in cross section with head and tail regions. Some nematodes rarely have a short and robust body (i.e. genus *Desmoscolex*). Most free-living and plant-parastic nematodes are usually smaller than 1mm but some of the larger forms have been found in kelp holdfasts, and in animal parasitic nematode relationship (i.e. genus *Anisakis*). Nematode growth is varied between parasitic and free-living species, and is influenced by food availability and feeding time (Holovachov & Schmidt-Rhaesa 2014). In free-living nematodes, development consists of four moulting stages, but a large body size is achieved by adult

growth, especially endo-parasitic nematodes (Holovachov & Schmidt-Rhaesa 2014).

Parasitic nematodes and free-living nematodes have been grouped together, although the taxonomic classification has not finished yet. Once it was considered that there were two classes, Adenophorea (free-living) and Secernentea (parasitic) in the phylum (Lorenzen 1981). Secernentea share morphological characteristics, including Phasmidia (excretory system) and a pair of sensory organs (Lorenzen 1981). More recently, however, it has been split into three subgroups (Chromadoria, Dorylaimia and Enoplia) using molecular technics (De Ley & Blaxter 2002).

2.3.2 Taxonomic identification

Nematode species can be difficult to distinguish due to their size and morphological homogeneity at the species level. Many nematode species have been described and 25,000 nematode species have been already reported. Due to a lack of knowledge regarding nematode taxonomy in the North-West Pacific Ocean, identification to the species level was not possible. Instead, nematodes were identified to genus level, and often given arbitrary names (sp. 1, 2, 3, etc.).

Nematodes were identified using the pictorial keys of Platt and Warwick (1983) based on the head and tail regions of nematodes. Each specimen was observed under the light microscope with a × 100 oil immersion lens (Olympus BX53). Total body, oesophagus, tail length, body shape and dimeter were also considered when specimens were difficult to identify. Handbook of Zoology (Holovachov & Schmidt-Rhaesa 2014) and the NeMys Database (Deprez 2007)were also used for identification.

2.4 Methods

2.4.1 Sampling collection and strategy

In order to compare the biodiversity of the coast of the British Isles with Korea, a hierarchical experimental design was required (Figure 2.6). Each set of samples were collected following a fully nested hierarchical design taking into account five spatial scales: geographic setting (British Isles and Korea), regions (five coastal stretches at least 50 kilometres apart), shore (two shores about 1 km long and at least 10 kilometres apart for each habitat), patch (three patches at least 10m

apart), replicates (three replicates of 5 x 5 cm quadrat at least 10cm apart). In *sagassum* samples, I only considered two regions in a formal test due to limited distribution of *S.muticum* on the central south coast of Korea. Two rocky shores in three regions (*Corallina* spp.) and two regions (*S. muticum*) at the low intertidal level (between 1.4 m and 2 m) dominated by chosen macroalgae were selected for sampling. Sampling locations with similar environmental conditions were chosen: moderately exposed, at least 80% turf of *Corallina* spp. and gently scraped bedrock shore. On each shore, three replicate patches were chosen by randomly collecting at least five replicates of *Corallina* spp. or *S. muticum* for morphological identification of nematodes. An additional five replicates were taken for potential future molecular analysis. Each replicate was one holdfast, and completely removed from the substrata by scraping, carefully placed into a labelled plastic bag and immediately moved into an Icebox. They were subsequently frozen to preserve them for further processing in the laboratory (see chapter 3 and 5 for more detail).

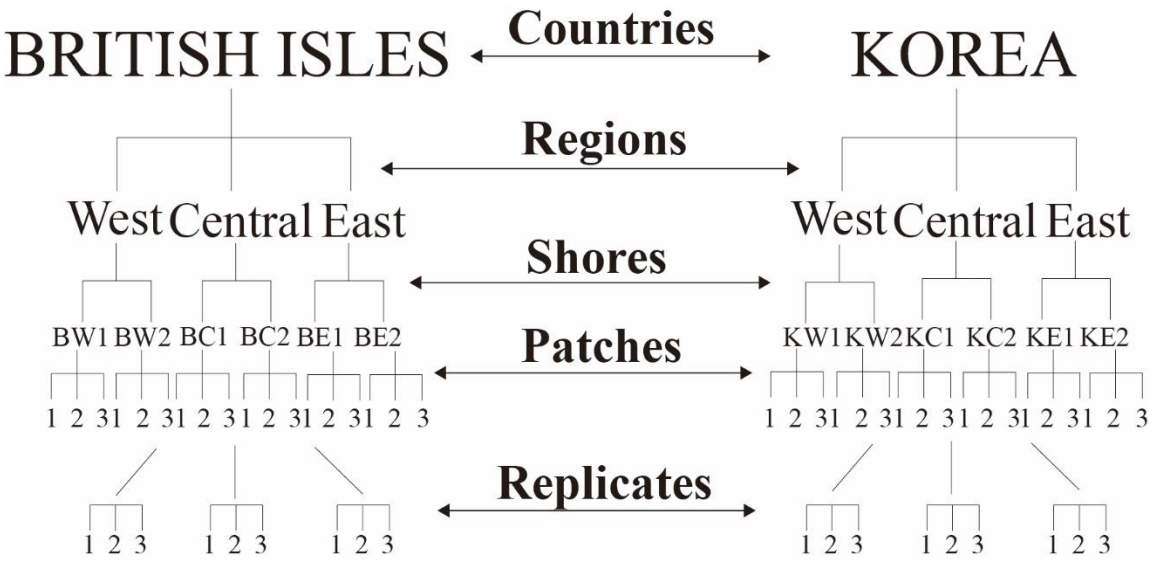


Figure 2.6 The schematic view of nested sampling design

2.4.2 Sampling Analysis

In the laboratory, due to time constraints only three replicates were analysed for morphological identification of nematode species. Total weight of selected macroalgae was measured in order to measure the amount of sediment in each individual seaweed. The plants were then washed with filtered tap water, first decanted through nested sieves of 1-mm to remove the larger fragments of algae and sediments. They were then sieved on 63 µm and 38 µm mesh respectively. Each macroalgal sample was rinsed and decanted three more times to ensure that all the organisms had been removed; then extracted epifauna were preserved in 4% buffered formalin then were washed with distilled water.

After extraction, the seaweeds were blotted and dried with paper to remove the water and kept at room temperature for one hour. Their wet weight was measured to the nearest gram (Taylor & Cole 1994). They were oven-dried 60 ºC for 48 hours to measure their dry weight in order to calculate nematode density per gram of host seaweed. All the nematodes were picked out and counted under a stereo microscope (Leica M125). Nematode samples were divided using a Folsom Plankton Splitter when nematodes were too numerous to count (McEwen et al. 1954). The first 100 nematodes were randomly hand-picked and mounted in anhydrous glycerine on HS slides (Figure 2.7) to observe both sides of the specimens (Shirayama et al. 1993) and identified to genus or species level under a light microscope (Olympus BX53). The pictorial keys of Platt and Warwick (1983), Handbook of Zoology (Holovachov & Schmidt-Rhaesa 2014) and the NeMys Database (Deprez 2007) were used to identify the specimens.

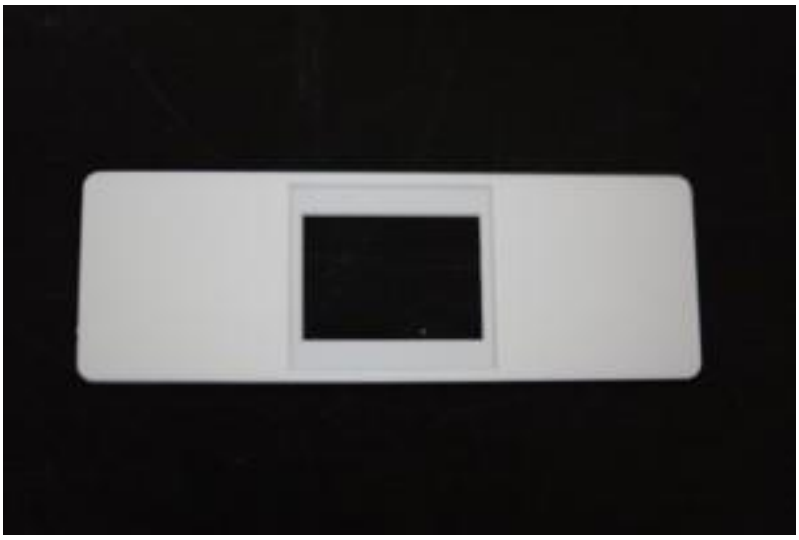


Figure 2.7 Generalised appearance of HS slide. The samples are sealed with two different type of cover glass (circle and rectangular) using a paraffin ring

2.4.3 Statistical Tests

Multivariate data analysis was performed using the software Primer 6.0.2 (Clarke & Gorley 2006) with PERMANOVA add-on. Univariate and bivariate analyses were carried out with Minitab (ver.12). Statistical differences at each different spatial scale were tested by nested analysis of variance (ANOVA) for the quantitative data on the nematode assemblages: abundance, number of genera, Maturity index (MI) and feeding types (FT). The British Isles and Korea were fixed factors with regions, shores and patches as random factors. The central region in both countries was not used in the analyses to preserve a proper nested experimental design. The data were checked with diagnostic graphics to ensure they fulfilled parametric assumptions; when needed, the data were transformed and re-checked to ensure that the transformation improved their frequency distributions. The relationship between relative nematode abundance and both wet and dry macroalgae was tested with Pearson's product-moment correlation.

The data for abundance of each nematode species in each replicate, patch, shore, region and country were standardised by their total abundance and square-root transformed. The means of the transformed abundance for each patch, shore and region were used to construct a Bray-Curtis similarity matrix; this was then subjected to group averaged hierarchical cluster analysis and non-metric multidimen4sional scaling (nMDS) ordination. The samples were coded by regions to visually assess the extent to which the species composition of the samples from the various patches, shores and regions were similar or different. Principal component analysis was performed for the ordination of samples on the basis of physical abiotic factors; the data were normalised due to different units of measurement. Relationships between multivariate structure of nematode assemblages and normalised abiotic factors were compared by BIOENV methods in a BEST test using Spearman's ranked correlation between similarity matrices. BIOENV test allows to find the best match with environmental variable and species compositions based on similarity matrix.

The samples were subjected to fully nested analysis of similarity (ANOSIM) and nested PERMANOVA with Monte Carlo tests to ascertain whether the species compositions of the nematode assemblages differed among patches within shore, among shores within region, and among regions within each country. For this and all subsequent ANOSIM, the null hypothesis, that there were no significant differences among groups, was rejected if the significance level (P) was greater than 0.05 or 5%. The R-statistic values determined with ANOSIM were used to

ascertain the degree to which those groups were significantly different. R-statistic values approaching unity indicate that the compositions of the groups were very different; whereas those close to zero show that they are very similar (Clarke 1993). When ANOSIM detected significant differences among the groups, similarity percentages (SIMPER) were used to determine the species that typified those groups and the species that distinguished each group from each of the other groups (Clarke 1993). PERMANOVA with Monte Carlo tests were used to see at which spatial scale difference in species composition of nematode assemblages occurred. Estimates of components of variation were made in PERMANOVA test to identify variability of nematode composition in each spatial scale.

Regression tests were conducted to measure alpha, beta and gamma diversity at four spatial scale relationships from both macroalgae; country versus region, region versus shore, shore versus patch, patch versus replicate. The total number of genera at beta (regional) scale samples were regressed against the total number of genera in each alpha (local) scale sample in order to compare alpha and beta relationship (Srivastava 1999). In broad scale comparisons, there were only four observations, each of which represented the estimate of alpha genus richness in country versus region, a comparison that provided insufficient data to undertake regression analysis. Therefore, nematode sampling from both macroalgae were pooled and that enabled a regression analyses at geographic scales (country versus region).

Alpha, beta and gamma relationships were conducted using a hierarchical linear model in order to test how regional diversity was partitioned into alpha and beta diversity within a particular spatial scale (Gering & Crist 2002). The total number of taxa in the regional (gamma) diversity sample was plotted against the mean taxa richness among local (alpha) diversity within the region. According to the additive partitioning method, gamma diversity is the sum of alpha and beta diversity (Allan 1975, Lande 1996, Gering & Crist 2002). The additive partitioning method was used to convert each diversity scale into a percentage of total diversity in order to investigate the contribution of each diversity scale into total (gamma) diversity (Lande 1996). Mean genera richness in each spatial scale were measured in order identify alpha diversity. Beta diversity at any scale is determined by subtracting the alpha diversity at that scale from the alpha diversity at the next highest scale (e.g. β = mean of patches – mean of replicates).

2.4.4 Functional Traits Analysis

Each nematode species was classified according to four different biological traits, based on their morphological and functional features. Feeding types based on the morphology of the buccal cavity were developed by Wieser (1953), who classified free-living nematodes into four feeding types such as selective deposit feeders (1A), non-selective deposit feeders (1B), epigrowth feeders (2A), and omnivore/predators (2B). Figure 2.8 shows the shape of the buccal cavity in each four feeding type.

An additional functional group classification can be provided by tail shape of the nematode indicative of mobility, habitat preference and lifestyle (Thistle & Sherman 1985). The diversity of tail shapes together with the features of buccal morphology have proven to be an effective tool for discriminating nematode assemblages (Thistle et al. 1995). Nematodes were classified into four tail shape groups: 1; Short/round tail type with blunt end, 2; Clavate-conicocylindrical tail type, initially conical with an extension to the tip, 3; Conical tail type, with a pointed tip and tail length less than five body widths, 4; Long tail type, with a tail longer than five body widths (Thistle & Sherman 1985). Figure 2.9 indicate four tail shape groups.

The life history strategies of nematodes have been described by Bongers et al. (1991), who proposed a scale (c-p score) to classify the genera of nematodes based on their ability to colonise or persist in a certain habitat. The scale ranges from extreme colonisers (c-p score=1) to extreme persisters (c-p score=5). A maturity index (MI) can also be calculated for each habitat/station based on the c-p scores of inhabiting species according to Bongers et al. (1991).

Biological traits matrix based on the above approaches was used to assess the functional structure of nematode communities at all spatial scales (Schratzberger et al. 2007). The three functional trait categories described above were used: feeding type, life history strategies and tail shape. A biological traits matrix was created by assigning to each nematode taxon to each trait category. The biological trait matrix was then combined with relative species abundance to give abundance-weighted traits matrices at each spatial scale.

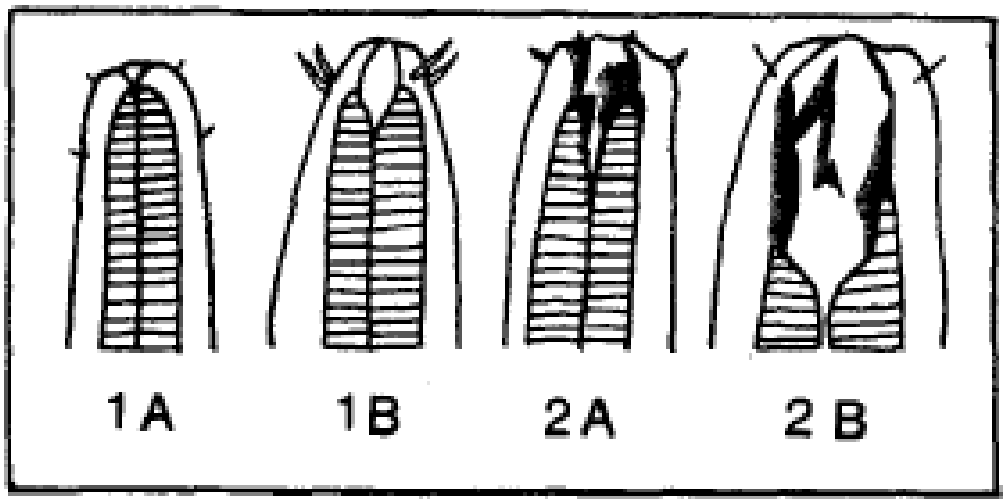


Figure 2.8 Feeding type according to Wieser (1953); the picture was taken from Romeyn and Bouwman (1983). 1A: Selective deposit feeders, 2A: Non-selective deposit feeders, 2A: Epistrate feeders, 2B: Predators/ommnivores.

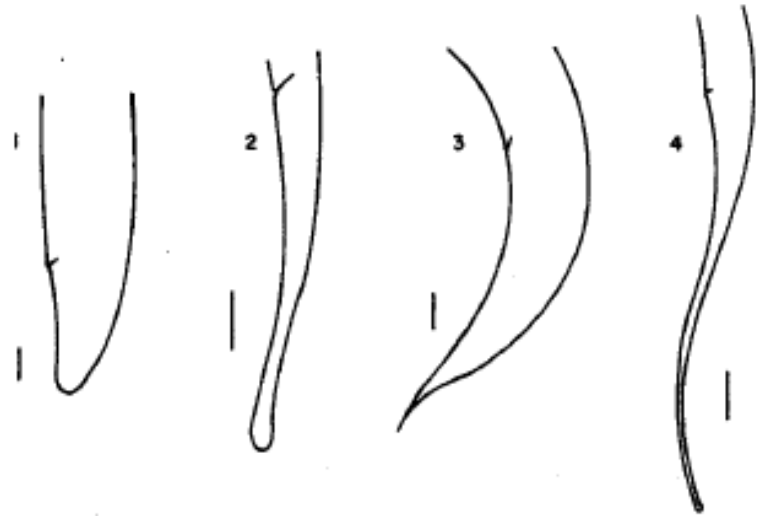


Figure 2.9 The illustration of each tail shape groups, taken from Thistle et al. (1995). Scale bar represents 10µm.

Chapter 3: Comparison of nematode assemblages in *Corallina* spp. between the British Isles and Korea: density, taxonomic versus functional diversity and relationship to environmental factors

3.1 Introduction

The role of biogeographical and ecological processes in determining species diversity and distribution patterns has a long history of study (Mayr 1942, Gobin & Warwick 2006, Frey 2010, Dray et al. 2012). In terrestrial environments, it has been commonly observed that species diversity in tropical areas is greater than at higher latitudes (Pianka 1966, Wilson & MacArthur 1967). In the marine environment, however, the picture is less clear. Some studies have found clear geographical gradients (Rex et al. 1993, Roy et al. 2000, Gobin & Warwick 2006), but many studies have not shown clear nor consistent patterns that are also difficult to explain (Thorson 1957, Warwick 1987, Kendall & Aschan 1993, Gray 1997, 2000, O'Hara & Poore 2000). For example, Boucher (1990) reported that nematode diversity displayed no latitudinal gradient; Mackie et al. (2005) also showed no clear latitudinal difference in polychaete and mollusc assemblages. Moreover, most studies have to date only considered one spatial scale (Gray 2000, O'Hara & Poore 2000, Mackie et al. 2005) or used univariate indices such as species diversity and evenness (Mackie et al. 2005). Often higher levels of taxonomic classification have been used such as family, order, class or phylum (Bracken et al. 2007, Ranjitham et al. 2008, Veiga et al. 2016) in order to demonstrate geographic distribution patterns of diversity.

Hierarchical sampling designs are an appropriate method to evaluate the contribution of each spatial scale to the total variation among samples (Murphy et al. 2009). They are also helpful in determining whether natural and anthropogenic activity has induced change (Chapman et al. 2010, Underwood & Chapman 2013). Community level information such as species diversity, species evenness, density and species composition are strongly dependent on the amount and quality of available data; if insufficient this can lead to erroneous

classification of environmental status (Hewitt et al. 2005, Tataranni & Lardicci 2010). A limited number of studies have looked at the variation of biotic indices using hierarchical sampling approaches (Hewitt et al. 2005, Muniz et al. 2012, Brauko et al. 2015). In addition, there have been broad scale studies of recruitment (Jenkins et al. 2000) and experimentation on a large scale geographic on rocky shores (Jenkins et al. 2005, Coleman et al. 2006). Such replication of sampling at multiple nested scales is an efficient tool for understanding distribution patterns and variation in community structure (Underwood 1997, Kelaher et al. 2001, Kelaher et al. 2004).

Marine nematodes are ubiquitous, found from the littoral to the hadal depths of the ocean (Giere 2008), and the most abundant taxa among meiofauna (Heip et al. 1985). Nematodes, as part of the meiofauna, play an important role in ecosystem functioning (Piot et al. 2014). Nematodes are a major food resource for invertebrates and fish (Huff & Jarett 2007, Giere 2009) as well as being a predator of microbial communities (Jensen 1987, Giere 2009). Additionally, nematodes can control numbers of the bacteria at steady states and influence nutrient cycling (Gibbons & Griffiths 1986). Furthermore, nematodes have been widely used as an ecological indicator in benthic environments (Ekschmitt et al. 2001, Kim et al. 2013). Nematodes display high abundance, wide distribution and diverse traits ranging from tolerant species to sensitive species (Heip et al. 1985, Giere 2008). They have a short generation time and continuous reproduction; they are also restricted to the same habitat throughout their life spans as they have direct development (Tietjen & Lee 1973). These are all valuable characteristics for indicating natural stress or anthropogenic impacts.

Despite their ecological importance, nematode ecology has been little documented, particularly on intertidal rocky shores (Gobin & Warwick 2006, Frame et al. 2007). On rocky shores, abundance of meiofauna is much greater than that of macrofauna, representing 25% of total secondary production (Gibbons & Griffiths 1986). Nematodes are the most abundant taxa among epiphytic species (Heip et al. 1985). Most studies of nematodes on rocky shores have focussed on habitat provided by macroalgae (Gestoso et al. 2010, De Oliveira et al. 2016, Veiga et al. 2016), investigating the relationship between nematode assemblages and macroalgal complexity (Veiga et al. 2016). Nematode density and diversity is dependent on the shapes of macroalgae (Gibbons 1988, Gee & Warwick 1994a, Gee & Warwick 1994b), and modified by a set of abiotic and biotic factors (Gibbons 1988, Giere 2009). To date, no study has investigated the relationship between nematode assemblages

and macroalgae at different spatial scales in order to look at their geographic patterns of species diversity.

Species pooling by similar functional groups can show different relationships between assemblages unlike taxonomic classification (Schratzberger et al. 2007, Armenteros et al. 2009). Traditional taxonomic approaches based on nematode community, diversity and community structure from species abundance data, do not take account of functional diversity based on biological traits (Bremner et al. 2003, Boström et al. 2006). Several previous studies have, however, revealed a comparison between taxonomic and functional diversity can be crucial for inferring ecosystem functioning and the effect of environmental variables and human activity (Bremner et al. 2003).

A broad scale hierarchical approach was adopted to specifically investigate the relationship between a cosmopolitan habitat-forming macroalgae (*Corallina* spp.) and its associated nematode assemblages. Until recently, *Corallina officinalis* was considered a cosmopolitan species, because of its similar morphology worldwide. More recently, it has been split into various species using molecular technique (Walker et al. 2009).

The overall aim of my study was to compare nematode assemblages in *Corallina* along the English Channel coast and the South coast of Korea using a nested design. On both coasts, there are gradations from oceanic to neritic conditions: from west to east in the English Channel and from east to west in Korea, although the current patterns are, however, more complex on the Korean coast (see Chapter 2). The specific objectives were: 1) to compare nematode density, diversity, assemblage composition and functional diversity (feeding type and life histroy strategies) living in *Corallina* spp. in different geographic areas; 2) to determine the relationship between abiotic factors and nematode assemblages associated with macroalgae along the gradation along both coasts; 3) to compare the use of taxonomic and functional measures of diversity in nematodes 4) to synthesize the information from both biogeographic regions to examine whether similarities or differences occur in the species pool and their functional roles.

More formally, the following hypotheses were tested:

1. The nematode diversity would be higher on the Korean coast than in the British Isles due to generally higher diversity in the Pacific than the Atlantic.

2. Taxonomic composition would be different but functional diversity (i.e. feeding type, life history strategy and tail shape) would be similar between British Isles and Korea.
3. Regional and local scale abiotic factors would influence species composition, abundance and diversity.

3.2 Materials and Methods

3.2.1 Sampling Strategy

In order to compare the coast of the British Isles with Korea, a hierarchical experimental design was required (Figure 3.1). Each set of samples was collected following a fully nested hierarchical design across five spatial scales: geographic setting (British Isles and Korea), regions (three coastal stretches at least 50 kilometres apart), shore (two shores about 1 km long and at least 10 kilometres apart for each habitat), patch (three patches at least 10m apart), replicates (five replicates of 5 x 5 cm quadrat at least 10cm apart). Two rocky shores in three regions at the low intertidal level (between 1.4 m and 2 m) dominated by turfs of *Corallina* spp. were selected for sampling. Sampling locations with similar environmental conditions were chosen: moderately exposed, with at least 80% turf of *Corallina* spp. and gently scraped bedrock shore.

3.2.2 Study Areas and Sample Collection

Nematode samples were collected in the British Isles and Korea in 2014 (Figure 3.2 and 3.3). To enable spatial comparisons two shores in each of three regions were surveyed on the south coasts of each country (Table 3.1): in the British Isles in the western English Channel Looe, (BW1) and Heybrook Bay (BW2); in the central English Channel Swanage (BC1), Portland Bill (BC2); and in the eastern English Channel Brighton (BE1), Beachy Head (BE2). In Korea West-Wando (KW1) and East-Wando (KW2) in the west, Yeosu (KC1) and Namhae (KC2) in the centre and Gueje (KE1) and Busan (KE2) in the east were sampled. On each shore, three replicate patches were surveyed by randomly collecting at least five replicates (5 x 5 cm) of *Corallina* spp for morphological identification and an additional five replicates were taken for potential future molecular analysis. Each replicate was completely removed from substrata by scraping, carefully placing into a labelled plastic bag and immediately moving into an Icebox to preserve for further processing in the laboratory.

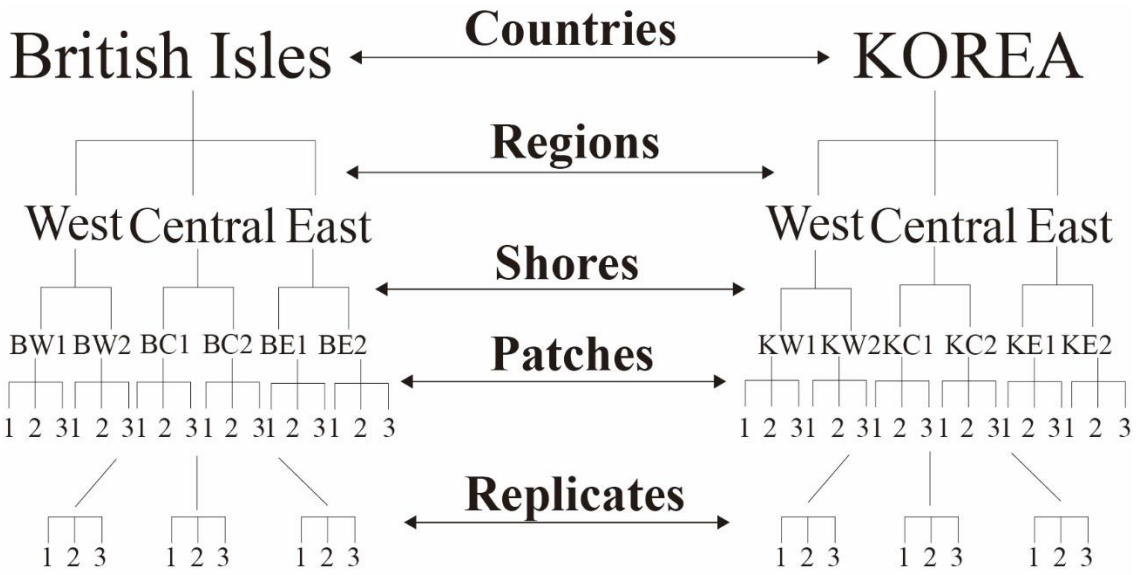


Figure 3.1 Schematic diagram of nested sampling design: BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

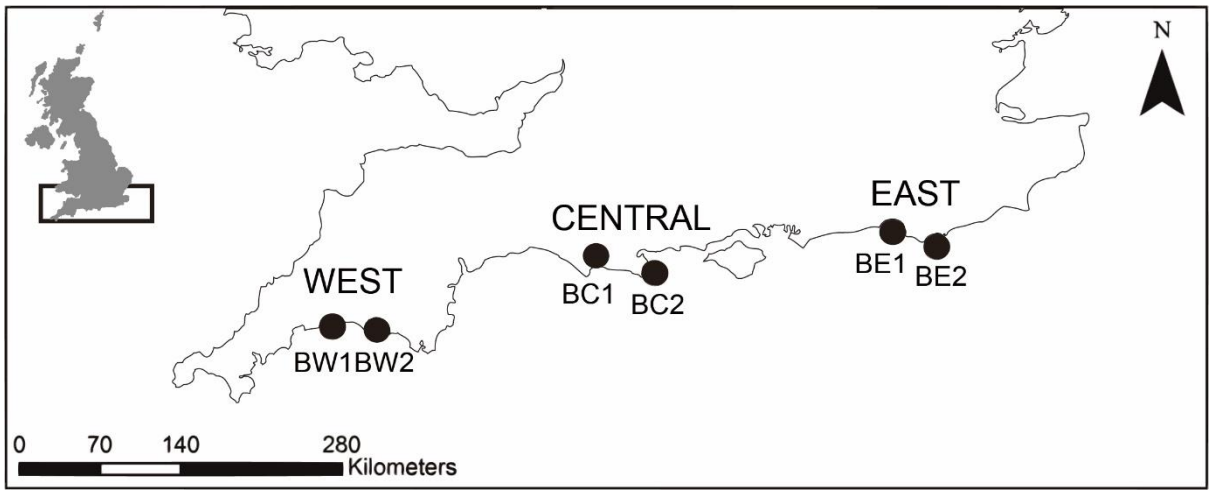


Figure 3.2 The sampling locations in British Isles: BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head.

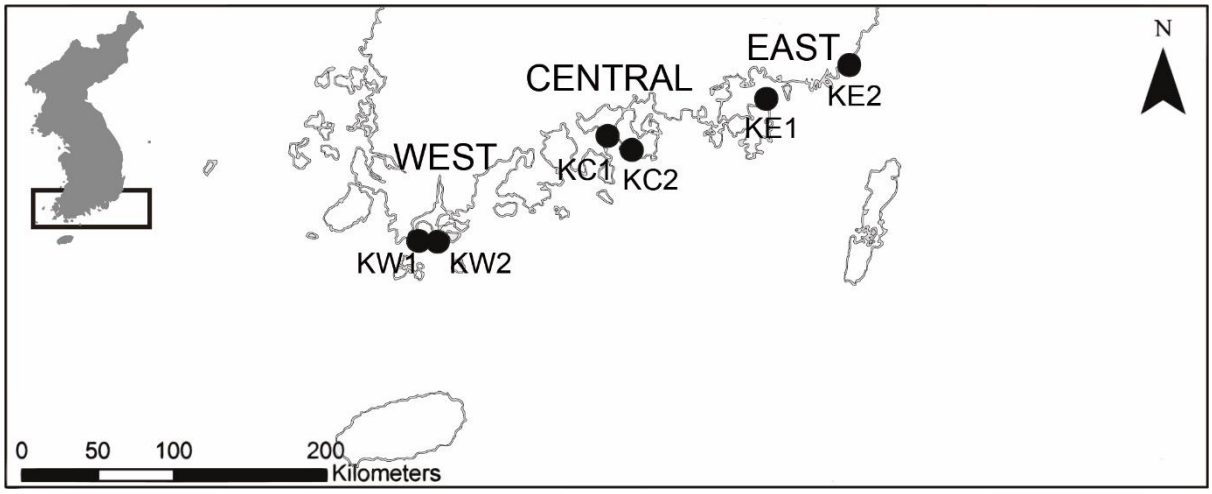


Figure 3.3 The sampling locations in Korea: KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

Table 3.1 Environmental context of sampling locations

NO	Geographical Region	Locality	Latitude N	Longitude W	Tides Range (m)	Annual Water (July) temperature range (℃)	Oceanic Index
BW1	Atlantic	Looe	50°20'29.4"N	4°27'39.7"W	5.4	9~17 (16)	3
BW2	Atlantic	Heybrook Bay	50°19'09.1"N	4°06'51.7"W	5.53	9~17 (16)	3
BC1	Atlantic	Portland Bill	50°30'53.03"N	2°27'19.44"W	2.5	9~17 (15.2)	2
BC2	Atlantic	Swanage	50°36'27.22"N	1°56'39.38"W	2.07	9~17 (16)	2
BE1	Atlantic	Brighton Marina	50°48'39.87"N	0°5'28.83"W	6.5	8~17 (15.4)	1
BE2	Atlantic	Beachy Head	50°44'15.0"N	0°15'09.1"E	7.3	9~17 (16)	1
KW1	Pacific	West-Wando	34°17'47.6"N	126°42'05.8"E	4.04	9~23 (22.7)	2
KW2	Pacific	East-Wando	34°18'13.5"N	126°45'52.3"E	4.04	9~23 (22.7)	2
KC1	Pacific	Yeosu	34°49'08.8"N	127°46'02.9"E	3.69	7~25 (24.5)	1
KC2	Pacific	Namhae	34°44'57.5"N	127°54'37.4"E	3.39	7~24 (24.3)	1
KE1	Pacific	Gueje	34°59'51.4"N	128°42'14.0"E	2.15	12~23 (22.9)	3
KE2	Pacific	Busan	35°09'39.9"N	129°11'38.8"E	1.35	11~23 (23)	3

3.2.3 Sample Analysis

In the laboratory, due to time constraints, three replicates of *Corallina* spp. were processed for morphological identification of nematode species. These were measured as a first total weight (g) of *Corallina* spp. was determined in order to measure approximate sediment weight of each seaweed. Each sample was washed with filtered tap water and decanted through a series of nested sieves of 1-mm to remove the larger fragments of algae and sediments; this was followed by 63 µm and 38 µm mesh. Each *Corallina* spp sample was rinsed and decanted three more times to ensure that all the epifauna had been removed; all extracted epifauna were preserved in 4% buffered formalin then were washed with distilled water.

After washing, the seaweeds were blotted with paper to remove excess water and allowed to dry at room temperature for one hour and their wet weight was measured to the nearest gram (Taylor and Cole, 1994). Samples were oven-dried 60 ºC for 48 hours to measure their dry weight.

After extraction, all nematodes were picked out and counted under a stereo microscope (Leica M125). When nematodes were too numerous to count, nematode samples were divided using a Folsom Plankton Splitter (McEwen et al. 1954). The first 100 nematodes were randomly chosen and mounted in anhydrous glycerine on HS slides to observe both sides of the specimens (Shirayama et al. 1993). They were identified to genus, and where possible species level, under a light microscope (Olympus BX53). The pictorial keys of Platt and Warwick (1983), Handbook of Zoology (Holovachov & Schmidt-Rhaesa 2014) and the NeMys Database (Deprez 2007) were used to identify the specimens.

3.2.4 Statistical Tests

Multivariate data analysis were used in the software Primer 6.0.2 (Clarke & PRIMER 2006) with PERMANOVA addition. Univariate and bivariate analyses were conducted in Minitab (ver.12). Statistical differences among patches within regions were tested by nested analysis of variance (ANOVA) for the quantitative data on the nematode assemblages: abundance, number of genera and Maturity index (MI). For univariate tests, relative nematode abundance were standardized per gram of *Corallina* spp. dry weight. The British Isles and Korea were fixed factors with regions, shores and patches as random factors. The data were checked with diagnostic graphics to ensure they fulfilled the parametric

assumptions; when needed, the data were transformed and re-checked to ensure that the transformation improved their frequency distributions. Whether there was any relationship between relative nematode abundance and both dry macroalgae and sediment weight in each 5 x 5 cm quadrat was tested with Pearson's product-moment correlation.

The data for abundance of each nematode genus in each replicate in each patch, shore and region were standardised by their total abundance and square-root transformed. The means of the transformed abundance for each patch, shore and region were used to construct a Bray-Curtis similarity matrix; this was then subjected to group averaged hierarchical cluster analysis and nonmetric multidimensional scaling (nMDS) ordination. Principal Component Analysis (PCA) was used for the ordination of samples and variables on basis of abiotic factor. The data were normalized due to different units of measurement. The samples were coded by regions to visually assess the extent to which the species composition of the samples from the various patches and regions were similar or different. Relationships between multivariate structure of nematode assemblages and normalised abiotic factors were compared by BEOENV methods in BEST test using Spearman's correlation between similarity matrices.

The samples were subjected to fully nested analysis of similarity (ANOSIM) and nested PERMANOVA with Monte Carlo tests to ascertain whether the species compositions of the nematode assemblages differed among patches within shores within regions in each country. For this and all subsequent ANOSIM, the null hypothesis, that there were no significant differences among groups, was rejected if the significance level (P) was<0.05 or 5%. The R-statistic values determined with ANOSIM for comparisons between those approaching groups that were significantly different were used to ascertain the degree to which those groups were dissimilar. R-statistic values approaching unity indicate that the compositions of the groups are very different, whereas those close to zero show that they are very similar (Clarke 1993). When ANOSIM detected significant differences among the groups, similarity percentages (SIMPER) were used to determine the species that typified those groups and the species that distinguished each group from each of the other groups (Clarke 1993).

3.2.5 Functional Traits Analysis

Each nematode species was classified according to four different biological traits, based on their morphological and functional features. Feeding types based on the morphology of the buccal cavity were described by Wieser (1953), who classified free-living nematodes into four feeding types: selective deposit feeders (1A), non-selective deposit feeders (1B), epigrowth feeders (2A), and omnivore/predators (2B).

An alternative functional group classification can be provided by tail shape of nematode indicative of mobility, habitat preference and lifestyle (Thistle & Sherman 1985). The diversity of tail shapes together with the feature of buccal morphology have proven to be an effective tool to discriminate nematode assemblages (Thistle et al. 1995). Nematodes were classified into four tail shape groups: 1. short/round tail type with blunt end; 2. clavate-conicocylindrical tail type, initially conical with an extension to the tip; 3. conical tail type, with a pointed tip and tail length less than five body widths; 4. long tail type, with a tail longer than five body widths (Thistle & Sherman 1985).

The life history strategies of nematodes have been described by Bongers et al. (1991), who proposed a scale (c-p score) to classify the genera of nematodes based on their ability to colonise or persist in a certain habitat. The scale ranges from extreme colonisers (c-p score=1) to extreme persisters (c-p score=5). A maturity index (MI) can also be calculated for each habitat/station based on the c-p scores of inhabiting species according to Bongers et al. (1991)

A biological traits matrix was assembled by following the above the approaches to assess the functional structure of nematode communities at all spatial scales (Schratzberger et al. 2007). The three functional trait categories described above were used: feeding type, life history strategies and tail shape. Thirteen combined categories were measured in total. A biological traits matrix was created by assigning each nematode taxa to each trait category. The biological trait matrix was then combined with relative species abundance to give abundance-weighted traits matrices for each spatial scale.

3.2.6 Environmental variables

Broadscale environmental data (July SST and tidal range from The Centre for Environment, Fisheries and Aquaculture Science (Cefas) in British Isles and from National Oceanographic Research Institute in South Korea) were combined with sample specific data (sediment load and algal dry weight) to explore abiotic and biotic factors influencing assemblage abundance and composition. An Oceanic Index was derived by the direction of currents coming from open sea (Pingree & Griffiths 1980, Huh 1982) in order to investigate how water type (i.e. oceanic or neritic) affect nematode assemblage. An Oceanic Index value of three represents area exposed to high levels of current action; two indicates moderate current activity and one an area with minimal current activity.

3.3 Results

3.3.1 Abiotic information

ANOVA showed that Sea-Surface Temperature in July (SST) and tidal range were significantly different among shores (P<0.001) in regions (P<0.001) and between countries (P<0.001). It was warmer in Korean waters than in the English Channel. In Korea the warmest shore was Yeosu (23.7℃). In the English Channel, most shores were around 16℃ (Looe, Beachy Head, Heybrook Bay and Swanage). Tidal range was greater in the British Isles than South Korea, but with considerable regional variation. In the English Channel the smallest tidal range was in the central region with greater tidal range in the western and eastern regions. In Korea, the gradient of tidal range was from the higher tidal range western to the eastern region.

Principal component analysis (PCA) showed clear differences between locations in countries in terms of their tidal range, sediment load, SST and the Oceanic index (Table 3.1) (Figure 3.4). The amount of sediment retention in *Corallina* turf was significantly different among patches and among shores (P<0.001), but not among regions and countries. Pearson's product-moment correlation tests indicated that the relative nematode abundance was significantly positively correlated with the amount of sediment (r=0.269, P<0.05) and with the amount of seaweed dry weight (r=0.310, P<0.05).

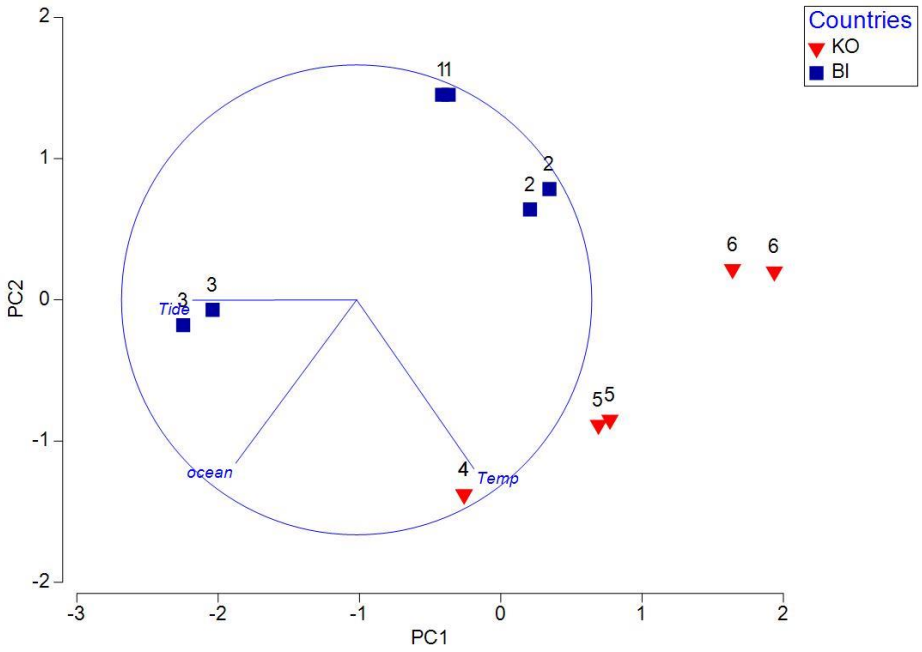


Figure 3.4 Principal component analysis (PCA) based on abiotic factors measured in all geographical regions: KO: south coast of Korea, BI: the British Isles, 1: Southwest of English Channel; 2: South central of English Channel; 3: Southeast of English Channel; 4: Southwest of Korea; 5: South central of Korea; 6: Southeast of Korea.

3.3.2 Nematode assemblages in *Corallina* spp. from the South coast of England

1633 nematodes belonging to 71 taxa (genera and some identified as species) were mounted and identified from samples collected from the shores in the west English Channel coast in July 2014. The most dominant nematode genus across both shores (Looe and Heybrook Bay) in the southwest coast of England was the genus *Euchromadora,* an epigrowth feeder (2A), followed by the omnivore/predator (2B) *Enoplus* and *Cyatholaimus* which was also an epigrowth feeder. The nematode density and number of genus in Heybrook Bay was greater than Looe. SIMPER analysis revealed that *Pontonema,* an omnivore/predator (2B) contributed the most to dissimilarities between the two shores. Epigrowth feeders were the most dominant feeding type across both shores. A Maturity Index of 3 indicating as intermediate between colonizers and persistent with moderate length of life cycle was dominant on both shores. The conical tail type was the most dominant tail type on the Southwest coast of England. In terms of their biological traits, the epigrowth feeders with conical tails and a c-p value 3 were the most common species along the west English Channel coast.

1708 nematodes belonging to 39 taxa (genera and species) were mounted and identified from samples collected from the central English Channel coast in July in 2014. The most dominant nematode genus across both shores (Portland Bill and Swanage) was the omnivore/predator (2B), *Enoplus*. Nematode density at Swanage was slightly larger than the density in Portland Bill; whilst the number of genera at Portland Bill was greater than at Swanage. The SIMPER test showed that the omnivore/predator (2B), *Enoplus* contributed the most to dissimilarities between the assemblages on the two shores. The major feeding type across the central English Channel coast was predator/omnivores. Short/round tail shape was the most dominant tail shape on the South central coast of England. The major common Maturity Index was c-p vale 5 indicating a relatively long life span, delayed maturity and less time spent on reproduction, and persistent species (K-strategists) on both shores.

1420 nematodes belonging to 55 taxa (genera and species) were mounted and identified from samples from the eastern English Channel coast in July in 2014. The most dominant nematode genus across both shores (Brighton Marina and Beachy Head) was *Chromadora,* an epigrowth feeder (2A). The nematode density from the rocky shore to the east of Brighton Marina was greater than the density at Beachy Head; while the number of genera at Beachy Head was more diverse

than Brighton Marina. The SIMPER test demonstrated that *Chromadora* contributed the most to dissimilarities between the two shores. The most abundant tail shape was clavate-conicocylindrical tail type on the Southeast coast of England. The most dominant functional type of nematodes across two shores were epigrowth feeders with clavate-conicocylindrical tail type and c-p value 2 indicating a relatively short life span, early and extensive reproduction leading to rapid colonization (*r*-strategists).

3.3.3 Nematode assemblages in *Corallina* spp. from the South coast of Korea

1058 nematodes belonging to 46 taxa were mounted and identified from samples from the southeast coast of Korea in August in 2014. The most dominant nematode genus across both shores (west-Wando and east-Wando) was *Crenopharynx,* a selective deposit feeder (1A). The density and number of nematode genera in west-Wando was greater than east-Wando. The SIMPER test showed that *Enoplus,* the omnivore/predator (2B) contributed the most to dissimilarities between the two shores. Long tail type was the most dominant shape on the Southwest coast of Korea. The most dominant functional type of nematode across two shores were selective deposit feeders (1A) with c-p value 5 indicating persistent species with relatively long life span, later maturity and less effort spent on reproduction (*k*-strategists).

1114 nematodes belonging 43 taxa (genus and species) were mounted and identified along the South central coast of Korea in August in 2014. The most dominant nematode species across both shores (Yeosu and Namhae) was *Phanoderma wieseri,* an epigrowth feeder (2A). The density of nematodes of Yeosu was greater than the nematode density in Namhae, but the number of genera in Namhae was larger than in Yeosu. Epigrowth feeders were the most common species in Yeosu; whereas predator/omnivores were dominant at Namhae. The conical tail type that is commonly found in epigrowth nematode was the most dominant tail shape in Yeosu whilst short/round tail shape was abundant in Namhae. The Maturity Index of 3 indicating moderate length of life cycle and colonizing ability was the most common component at Yeosu; whilst c-p value 4 indicating a relatively long life span and reproduction time was dominant at Namhae.

1599 nematodes belonging to 66 taxa (mostly genera, some species) were mounted and identified from samples from the Southeast coast of Korea in August in 2014. The most dominant nematode genus across both shores (Gueje and Busan) was *Euchromadora,* an epigrowth feeder (2A). The density and the number of nematode genera in Busan was greater than in Gueje. Epigrowth feeders were the most common species in Gueje whereas predator/omnivores were dominant at Busan. The conical tail shape was the dominant shape in Gueje while conicocylindrical tail type was the most dominant in Busan. A Maturity Index value of 3 was indicative of a relatively moderate life cycle and reproduction time at Gueje. In contrast, a c-p value of 4, indicating a relatively long life span and reproductive effort, was dominant at Busan.

3.3.4 Taxonomic composition of nematode assemblages across geographical scales

From pooled samples 8532 individuals were identified as belonging to 128 taxa along the south coast of England and South Korea combined in July and August in 2014. The top five ranked species, together accounted for 53% of total nematode abundance: *Enoplus* (18%), *Euchromadora* (13%), *Crenopharynx* (10%), *Cyatholaimus* (7%), *Chromadora* (5%). 109 species contributing individually to less than 1% to total relative abundance. The most widely distributed nematode genus in both countries was *Enoplus,* followed by *Cyatholaimus* and *Euchromadora.* Several nematode species and genra were common in particular geographic locations: *Phanoderma wieseri, Phanoderma segmentum* and *Desmodora* were dominant in the UK; whereas *Chromadora* and *Retrotheristus* were dominant in South Korea. Some rare nematode species appeared to be unique to particular locations: *Gomphionema* sp. (Heybrook Bay), *Hypodontolaimus* sp. (Beachy Head), *Prochromadorella* sp. (Beachy Head), *Morlaixia* sp. (Busan), *Ditlevsenella* sp. (Busan), *Ascolaimus* sp. (Geoje) and *Diplolaimelloides* sp. (Namhae).

3.3.5 Density and diversity of nematode assemblages across all geographical scales

The density of nematode assemblages living in *Corallina* turfs were significantly different among patches between shores (P<0.05), but not among regions and between countries (Figure 2.5). The number of genera differed only at patch level (P<0.05), not at other spatial scales (Figure 2.6). The density in Busan was the highest across all shores in both countries. Namhae had the lowest density in all geographical regions. In terms of number of genera, Busan was the most diverse shore across all geographical regions. In contrast, Yeosu was the least diverse among the shores. The average range of nematode density per gram in patches was between 5~30 and the average range of the number of genus in patches was 26~60.

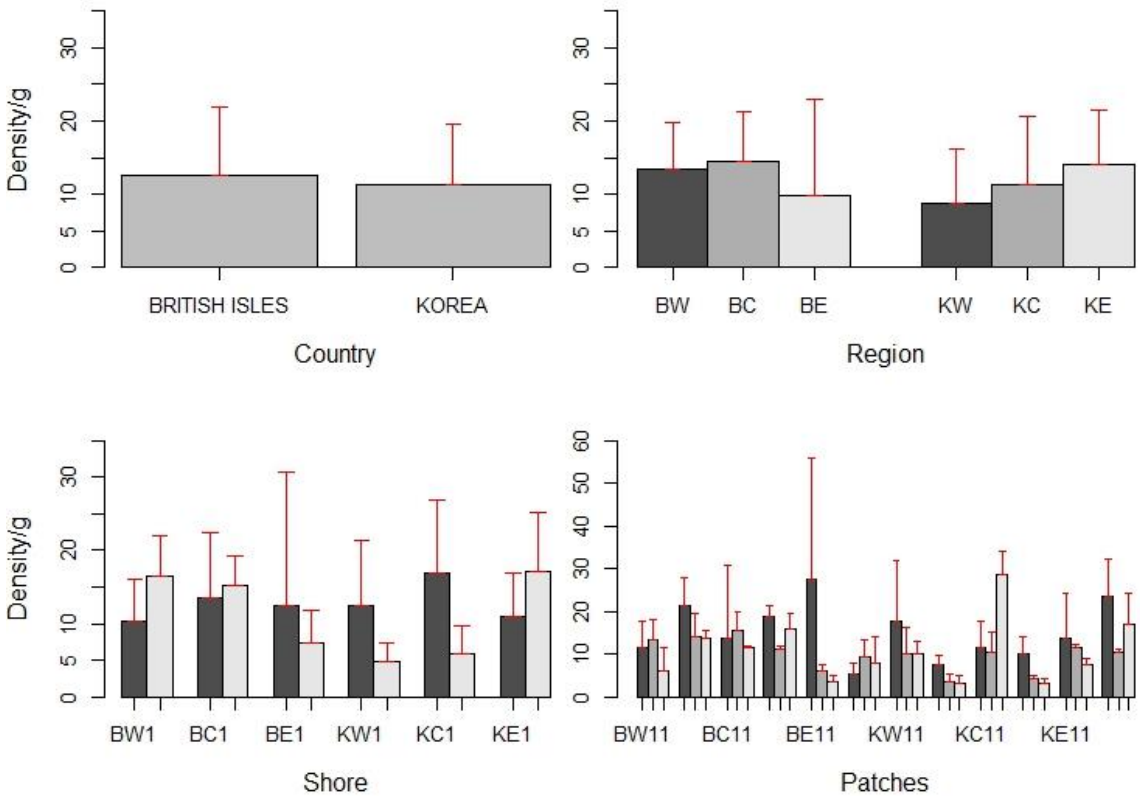


Figure 3.5 Mean density of nematode assemblages with standard deviation at all spatial scales: BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

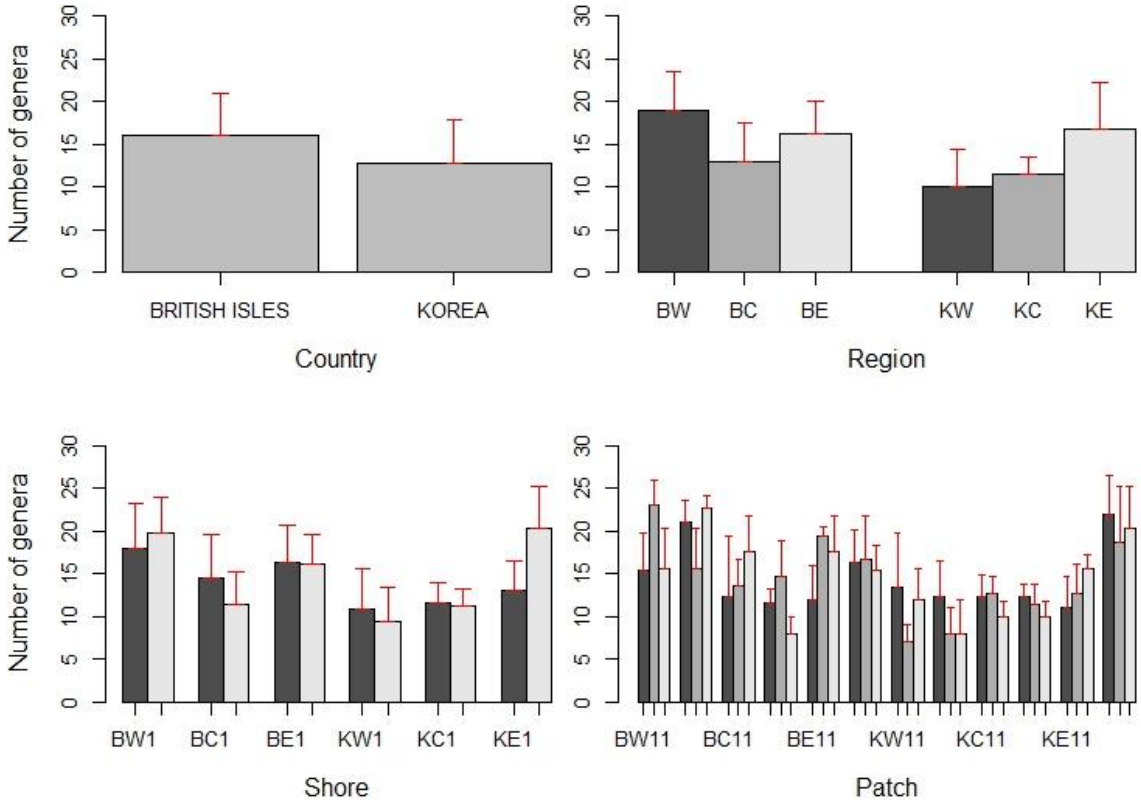


Figure 3.6 Mean number of genera in nematode assemblages with standard deviation in all spatial scales: BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

3.3.6 Comparisons of species compositions across all geographical scales

Nested PERMANOVA with Monte Carlo tests demonstrated that the species composition differed significantly between countries (P<0.05), regions (P<0.05), shores (P<0.001) and patches (P<0.001). The ordination of samples with the nMDS technique showed clear differences at different geographical scales (Figure 3.7). The Estimates of Component Variation test in PERMANOVA indicated that among shore variations in nematode assemblage composition was greater than the other three spatial scales (Table 3.2). Nested ANOSIM tests showed that the species composition was significantly different among regions (R = 0.639, P<0.05), shores (R = 0.919, P<0.001) and patches (R = 0.428, P<0.001) but not between countries (R = 0.852, P>0.05); it is indicated that nematode variation across each shore was greater than at other spatial scales. The ordination of samples with nMDS showed clear differences between countries and among regions in the British isles, while no clear differences were apparent among regions in Korea.

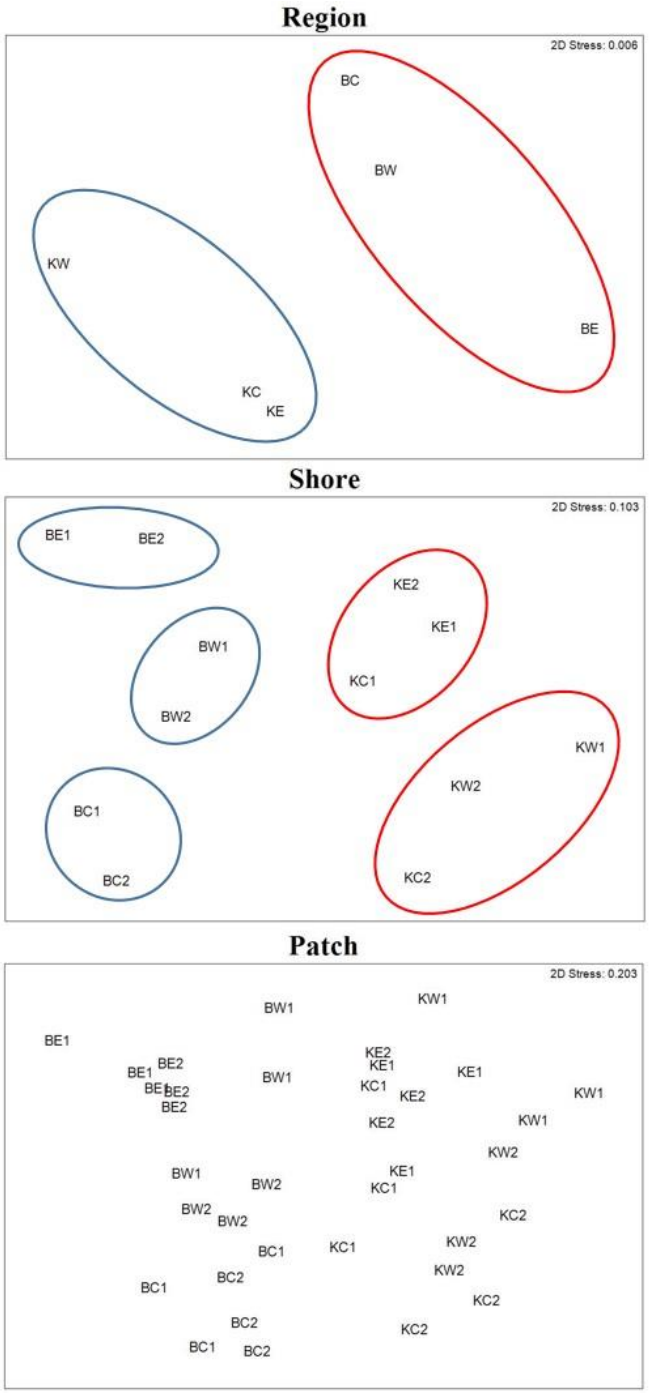


Figure 3.7 Non-parametric multi-dimensional scaling (MDS) ordination based on the standardised abundance of nematode genera at three spatial scales: BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

Table 3.2 Estimates of Components of Variation in PERMANOVA; Co: Countries, Re: Regions, Sh: Shores, Pa: Patches.

Estimates of components of variation		
Source	Estimate	Sq.root
S (Co)	454.11	21.31
V (Re(Co))	456.12	21.357
V (Sh(Re(Co)))	502.08	22.407
V (Pa(Sh(Re(Co))))	339.05	18.413
V (Res)	970.89	31.159

3.3.7 Biological traits across all geographical scales

nMDS plots showed that the ordination of taxonomic based classification did not match the ordination based on functional groups and traits (Figure 3.9). Feeding-type composition was characterised by epigrowth feeders (43%), predator/omnivores (33%), selective deposit feeders (13%) and non-selective deposit feeders (11%) (Figure 3.8). Similar feeding type compositions were present in both countries, but PERMANOVA test showed clear differences among regions (P<0.05), among shores (P<0.05), among patches (P<0.05) and among replicates (P<0.001). The Maturity Index (MI) was calculated for each different spatial scale and did not differ at the country and at shore level. Significant differences were, however, found among patches and regions. Nematode species with a c-p value of 3 indicating a moderate length of life and reproduction investment formed the most dominant component (35%) across both geographical scales. The most dominant tail shape was conical tail type (34%) followed by short/round (28%), conicocylindrical (24%) and long tail types (13%). PERMANOVA test showed significant difference among patches (P<0.001), shores (P<0.001) and regions (P<0.05), but not between countries.

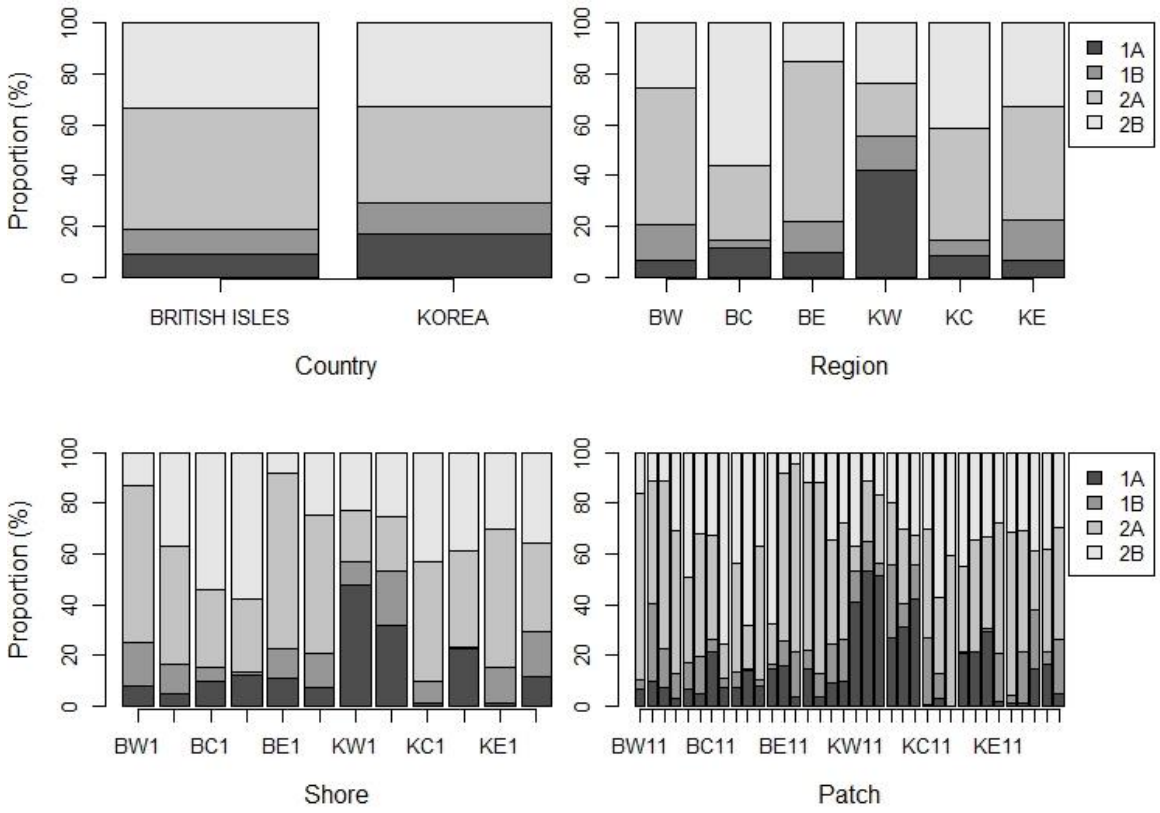


Figure 3.8 Trophic composition of nematode assemblages on the basis of average percentages at all spatial scales. The feeding types defined after Wieser (1953); 1A=selective deposit feeders, 1B=non-selective deposit feeders, 2A=epigrowth feeders, 2B=predators/omnivores: BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

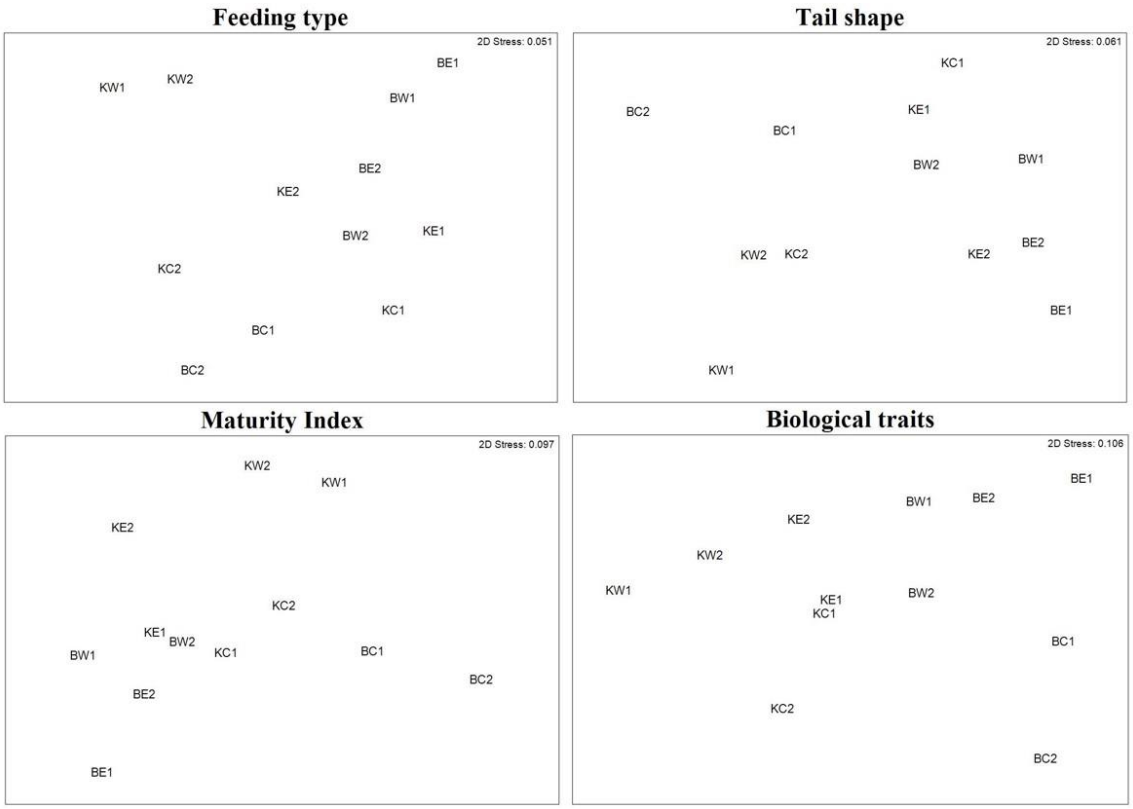


Figure 3.9 Non-parametric multi-dimensional scaling (MDS) ordination based on the standardised abundance of nematode feeding type, tail shape, maturity index (life strategies) and Biological traits in shore level: BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

3.3.8 Relationships between abiotic factors and nematode assemblages

The BIOENV method in BEST test using similarity matrices derived from relative nematode abundance and biological traits (feeding type, Maturity Index and tail shape) indicated that nematode composition and combined biological traits were significantly positively correlated with the combination of Sea-Surface Temperature (SST) and tidal range (Table 3.4). The feeding type of nematode was significantly correlated with the Oceanic index (R = 0.136, P<0.001). Maturity Index was, however, significantly correlated with the combination of sea-surface temperature (SST), tidal range and the amount of retention sediment (R = 0.130, P<0.001). Tail shape of nematode was positively correlated with the combination of tidal range and Oceanic Index (Table 3.4).

Table 3.3 P-value of nested PERMANOVA in each measurement; Co: Countries, Re: Regions, Sh: Shores, Pa: Patches, Res: Replicates

Source	Species abundance	Feeding type	Maturity index	Tail shape	Biological traits
S (Co)	**0.02**	0.631	0.293	0.823	**0.048**
V (Re(Co))	**0.004**	**0.02**	**0.006**	**0.004**	**0.001**
V (Sh(Re(Co)))	**0.001**	**0.011**	0.266	**0.001**	**0.001**
V (Pa(Sh(Re(Co))))	**0.001**	**0.001**	**0.001**	**0.002**	**0.001**

Table 3.4 Results of BIOENV analysis matrices of similarity derived data of species abundance and biological traits. The combination of abiotic factors with the highest value of Spearman rank correlation (ρ) and significance values (*p*) in each matrix were shown; Te: sea-surface temperature, Ti: tidal range, Oc: Oceanic index, Sd: the amount of retention sediment.

Matrix	ρ (rho)	*p*	Abiotic factors
Species	0.401	0.001	Te, ti
Feeding type	0.136	0.001	Oc
life strategy	0.13	0.001	Te, ti, sd
Tail shape	0.161	0.001	Ti, oc
Combined biological traits	0.331	0.001	Te, ti

3.4 Discussion

The density, number of genera, biological traits and composition of nematode assemblages living in *Corallina* spp. were investigated at different spatial scale (patches, shores, regions and countries). Most previous studies of nematode living in macroalgae have been made in the temperate North-East Atlantic (Wernberg et al. 2004, Gobin & Warwick 2006, Harries et al. 2007, Gestoso et al. 2010, Veiga et al. 2016), or in Atlantic tropical areas (Boucher 1990, Da Rocha et al. 2006, Armenteros et al. 2009, De Oliveira et al. 2016). In contrast, relatively few studies have been reported from the Pacific area (Andrews 1977, Boucher 1997). Most of these studies have focussed on the relationship between nematode assemblages and macroalgal complexity (Veiga et al. 2016). Several investigations have shown differences in composition and structure of epiphytic assemblages associated with different types of macroalgae (Wernberg et al. 2004, Wikström & Kautsky 2004, Harries et al. 2007, Vázquez-Luis et al. 2008). In contrast, other studies have found no difference among different species of macroalgae in a particular study area (Parker et al. 2001, Schreider et al. 2003, Cacabelos et al. 2010b).

Previous studies on the distribution of free-living nematodes living among/on macroalgae found common nematode species on different types of macroalgae although the dominant species on each algae seemed to vary (Wieser 1959, Heip et al. 1985, Da Rocha et al. 2006, De Oliveira et al. 2016). The density and size of nematodes was correlated with the morphological shape of macroalgae and the amount of detritus caught in the rhizoids (Heip et al. 1985). For example, in foliaceous algae from exposed coasts, small Chromadoridae were dominant (approximately 70% of total abundance). In contrast, tuft-like algae, including *Corallina,* from exposed coasts have large number of Enoploidea (Wieser 1959). Moreover, De Oliveira et al. (2016) found dissimilarity of nematode composition with few same abundant genera such as *Euchromadora* and *Draconema* in *Halimeda opuntia*. My work also showed similar dominant nematode genera such as *Euchromadora*, *Enoplus*, *Chromadora* and *Eurystomina* consisting of more than 50% of total abundance in both countries. This suggests that several common nematode species are appearing in macroalgae of similar morphology (i.e. turf forming or foliaceous species)

In my study, nematode density and diversity were only significantly different at small spatial scale (patches and shores) and not on larger scales (regions and countries). This suggests that nematode species diversity at a small scale seems to

vary due to available resources and local environmental conditions. The overall density of nematodes at small scales depends on the amount of dominant nematode genera colonizing a patch of habitat. At larger scales, however, the number of nematode taxa are similar in both countries, suggesting similar evolutionary and biogeographic processes operating in different oceans even if the nematode diversity in British Isles was slightly higher than in Korea. This mirrors work by De Oliveira et al. (2016) who found similar results working with *Halimeda* at the northeast of the Brazilian coast showing no difference in density and species diversity at two different spatial scales.

Despite nematode species diversity being similar within regions and countries, the nematode species composition was significantly different at all spatial scales. At the patch level, SIMPER tests showed that the characteristics of patches on each shore were determined by relative abundance of a few dominant genera (Table A1.1). This small-scale heterogeneity was also found in previous studies indicated by aggregation of dominant nematode species (Blome et al. 1999). At the shore level, nested PERMANOVA with Monte Carlo test showed higher Estimate Component Variation among other three spatial scales (Table 3.2). The most dominant genus on each shore was different. This suggests that nematode compositions seem to vary in response to the surrounding environment. The relationship between environmental factors and intertidal nematode communities have been reported in several previous studies (Warwick 1977, Gibbons 1988, Armenteros et al. 2009). The composition and abundance of epifaunal nematode communities were affected by wave exposure, water temperature, changes in nutrient availability and sediment accumulation or the combination of these environmental factors (Warwick 1977, Gibbons 1988, Holovachov & Schmidt-Rhaesa 2014).

De Oliveira et al. (2016) reported sediment retention in *Halimeda opuntia* increased nematode richness and was positively correlated with *Euchromadora* and *Draconema*. They showed that sediment accumulation by seaweeds led to higher nematode overall diversity and density. My results also showed a positive correlation between both number of nematode genera and amount of sediment retention by *Corallina* spp. ($R = 0.319$, $P < 0.001$), and between nematode density and amount of sediment retention by *Corallina* spp. ($R = 0.269$, $P < 0.005$). Nematode assemblage compositions on each shore seemed to be affected by the sediment accumulation by *Corallina* spp. For example, the shores on the southwest of Korea (KW1 and KW2) were characterised as having high amounts of

sediment retention in *Corallina* spp. leading to dominance by *Crenopharynx* that is commonly found in sediments. Other shores showed several dominant epifaunal nematode genera such as *Euchroamdora*, *Cyatholaimus* and *Eurystomina*. This contrasts with species composition in seaweeds reported previously (Wieser 1952, Hopper & Meyers 1967, Hopper 1967, Warwick 1977, De Oliveira et al. 2016). Moreover, BIOENV tests showed that tidal range and sea-surface temperature were the best matches with nematode abundance ($R = 0.401$, $P < 0.001$). This suggests that the variation of nematode compositions on shores were determined by a combination of environmental factors (Table 3.4).

At even larger scales, at the regional and country level, nMDS plot showed clear differences among regions in both countries, and between countries (Fig 3.7). The nematode compositions were clearly divided by three regions (west, centre and east) in the British Isles while only two regional groupings (west and east) were found in Korea. This probably reflect the geography and oceanography of both countries. Strong current flow from southwest to southeast coasts of the British Isles including up the English Channel (Pingree & Griffiths 1980). The Tsushima Warm Current to southeast, and Yellow Sea Current from west to east converge on the South coast of Korea (Huh 1982). These different current systems might cause the differences in regional patterns of nematode composition between each country. At the countries level, biogeographic and evolutionary processes might play an important role in the taxonomic variation of nematode composition between biogeographic realms (Witman et al. 2004). Although the dominant group of nematode genera were similar, more nematode taxonomic or molecular studies are needed to investigate biogeographic differences between the two countries, preferably working at the species rather than generic level of taxonomic resolution.

In terms of biological traits, increasing species richness potentially enhances functional diversity (Petchey & Gaston 2006). To date, many studies have investigated the relationship between functional diversity and taxonomically based species diversity (Walker et al. 1999, Petchey & Gaston 2002, Mouillot et al. 2005); but only a few studies have used a functional concept in considering free-living nematode communities (De Mesel et al. 2003, Schratzberger et al. 2007, Armenteros et al. 2009). Armenteros et al. (2009) showed no significant temporal and spatial variation of functional nematode diversity in a semi-closed tropical bay in contrast to differences in taxonomic diversity. Whilst Schratzberger et al. (2006) found clear functional differences spatially in nematode communities in the southwestern North Sea. My results also showed significant differences in

combined biological traits matrix at all spatial scales; but this was not clear when considering single functional traits (Table A1.1).

Composition of community structure comparison based on a taxonomic approach is more powerful than using biological traits to detect spatial patterns (Schratzberger et al. 2006, Armenteros et al. 2009). My results indicated that common morphological type of nematodes appeared across all spatial scales. There was strong dominance of two morphotypes (epigrowth feeder/c-p value 3/ tail conical, and predator/omnivores/ c-p value 5/ tail round/short). They appeared across all spatial scales indicating a few cosmopolitan genera. These two morphotypes appeared at all spatial scales, and probably perform similar ecological processes contributing to ecosystem functioning. Several shores were distinguished from other shores indicating local prevalence of a particular dominant biological trait. For example, feeding type and tail shapes were strongly influenced by the amount of sediment in *Corallina* spp. (KW1 and KW2). Long life span and short/round tail shapes were dominant at relatively exposed shores (BC2). This indicates that the role of nematodes in ecosystem functioning can be changed by characteristics of the surrounding environment such as sediment retention, wave action and nutrient resources.

Although the cosmopolitan morphotypes were found in nematode assemblages, combined biological traits significantly differed at all spatial scales. Species abundance data were strongly correlated with number of functional traits (RELATE test, $R = 0.8$, $P < 0.001$). This indicates that increasing species diversity probably lead to increasesd functional diversity. The relationship between species richness and functional diversity, and whether some species can be lost from some ecosystems is still controversial (Petchey & Gaston 2002, Schratzberger et al. 2007). Knowing how different size class organisms contribute to ecosystem processes is essential for a more integrated and complete understanding of biological influences in ecosystem process (Horner-Devine et al. 2004). Therefore, combined trait approach in nematode assemblages can be a useful tool to evaluate ecosystem processes, properties and diversity loss in ecosystems.

Summary

Returning to the hypotheses, hypothesis 1 was rejected; the converse occurred with species diversity being higher in the British Isles. This may be due greater variation among regions in the British Isles (For further discussion see chapter 4).

Hypothesis 2 can also be rejected as both taxonomic and functional diversity differed between Korea and the British Isles. Both classifications were affected by the environmental characteristics of the shore. This indicates that the role of nematode in *Corallina* spp. would be different by surrounding environment

Hypothesis 3 was, however, accepted, Abiotic factors clearly influenced species composition, abundance and diversity at the regional scale, in relation to tidal range, SST and sedimentary characteristics. Biotic factors such as weight of algae also influenced abundance, composition and diversity of nematode. The amount of sediment trapped in *Corallina* spp. was a predominant factor shaping nematode abundance and composition assemblages.

Further experimental studies are required to further understand the abiotic and biotic factors determining nematode abundance, assemblage and composition.

Chapter 4: A comparative study of nematode assemblages associated with *Sargassum muticum* in its native range in South Korea and as an invasive species in the English Channel

4.1 Introduction

Invasive species have become one of the greatest threats to biodiversity and ecosystem functioning (Pejchar & Mooney 2009, Salvaterra et al. 2013, Veiga et al. 2016). Invasive species can cause strong effects on receiving communities such as decreasing abundance of native species abundance (Race 1982, Delibes et al. 2004), altering diversity (Meiners et al. 2001, Hejda et al. 2009) and changing community structure (Posey 1988, Britton-Simmons 2004). Invasive species can also hybridize with native species (Nehring & Hesse 2008), alter habitat quality (Levin et al. 2002), and can lead to local or regional scale species extinction (Gurevitch & Padilla 2004). Marine systems are particularly sensitive and vulnerable to biological invasions due to their open nature, geographical connectivity and the great dispersal potential of marine species (Rapoport 1994). With anthropogenic vectors such as transoceanic shipping (Holeck et al. 2004), recreational activity (Cameron et al. 2007), introduction of species in aquaculture (De Schryver & Vadstein 2014) and use of ballast water (Hallegraeff & Bolch 1992), the transfer and dispersal of non-indigenous species outside their native habitat has greatly increased in recent decades (Salvaterra et al. 2013).

As marine invasive species are considered a major problem for biodiversity and ecosystem functioning especially in coastal systems, many studies have been directed towards this issue (Stachowicz et al. 2002, Gurevitch & Padilla 2004, Fridley et al. 2007, Pejchar & Mooney 2009). To date, most studies have, however, been limited to notorious invasive species (Smith et al. 2014). It has been estimated only 6% of exotic macroalgae have been studied concerning their ecological impact (Smith et al. 2014). Invasive meiofauna have not been studied at all. This indicates that many unknown invasive species may have been introduced to recipient communities but yet remain undetected. The impacts of

invasive species are not only invader specific but also dependent on the characteristics of the recipient communities (Cacabelos et al. 2010a, Salvaterra et al. 2013). Therefore, it is important to demonstrate whether invasive species add to the overall density of organisms in natural communities, or replace some of the native organisms so that the overall density in the community remains unchanged (Salvaterra et al. 2013). Moreover, determining which invader replaces what native species is needed to understand the influence of biological invasion in recipient communities (Stachowicz et al. 2002, Fridley et al. 2007).

On rocky shores, macroalgae are the major primary producer and also provide habitats for macro and meiofaunal invertebrates (Raffaelli & Hawkins 1996, Thompson et al. 1996). Many studies have indicated that the number of species and their abundance in epifaunal assemblages are related to the morphological and structural complexity of habitat-providing macroalgae (Bell & Coen 1982, Heip et al. 1985, Gee & Warwick 1994a, McDonald & Bingham 2010, De Oliveira et al. 2016). These studies have reported a common relationship between macroalgae and epifaunal species. For example, epifaunal species can feed on epiphytes or detritus from seagrasses and algae which increase the growth rates of their host (Stachowicz & Whitlatch 2005). Furthermore, the growth rate of seaweeds tend to be affected by their nitrogen resources and are enhanced by the nitrogen excretion from meio-epiphytal species living on their host seaweed (Bracken et al. 2007).

Nematoda comprise one of the most numerous and diverse taxon in the biosphere and are widely distributed in the marine environment (Platt et al. 1980). They live in every habitat such as macroalgae, sediments from shallow water to deep sea (Platt & Warwick 1980) and are parasites in most classes of Metazoa (Heip et al. 1985). Nematodes are also one of the most abundant meiofauna associated with macroalgae (De Oliveira et al. 2016). They play a key role in coastal ecosystem functioning in processes such as detrital breakdown and biomineralization, They serve as a food source for higher trophic levels and are predators regulating bacterial populations and feeding on other meiofauna (Rysgaard et al. 2000, Schmid-Araya et al. 2002, De Oliveira et al. 2016). On rocky shores, nematodes enhance nutrition of macroalgae (Bracken et al. 2007) and control the densities of epifaunal species (Warwick et al. 1998).

Sargassuum muticum (Yendo) Fensholt is a one of the most well-known invasive species in the world. This macroalga was first introduced to Europe in the early 1970s (Rueness 1989). It reached the British Isles in 1973, probably on Japanese

Oyster, *Crassostrea gigas* (Farnham et al. 1973). It has become a common species on the south coast of England (Farnham et al. 1973). Several studies have compared the associated fauna on invasive macroalgae with those living on native species (Norton & Benson 1983, Britton-Simmons 2004, Wernberg et al. 2004, Harries et al. 2007, Veiga et al. 2016). Some studies found a negative effect on related species by reducing abundances of native species which are preferred food (Britton-Simmons 2004), or decreasing abundance and changing species composition (Salvaterra et al. 2013).

There are various limitations to the previous work. In many cases, higher level taxonomic classification (e.g. phylum, class or family) of associated fauna was used (Curvelo & Corbisier 2000, Wikström & Kautsky 2004, Gestoso et al. 2010). Comparisons were also only made with other native macroalgae of similar morphology, and tended to be restricted to a single locality (Stæhr et al. 2000, Wernberg et al. 2004, Veiga et al. 2016). To date, no studies have compared epifaunal assemblages associated with *S. muticum* in its native range with its invaded regions. Therefore, my overall aim was to compare the assemblages of nematodes associated with *Sargassum muticum*, a native of Korea, both in its home range and on the coast of England, where it is a relatively recently-arrived invader. This enabled an evaluation of the role of *Sargassum muticum* as a habitat provider at both home and abroad and whether there are differences in nematode assemblage composition and structure in its invasive range compared to its native range. This provides a good model to test hypotheses concerning the functional role of habitat providing non-natives species in areas that have been invaded. The following specific hypotheses were tested, using a hierarchical design with sampling at the scale of biogeographic realms (South coast of Korea in North-West Pacific versus the English Channel in the North-East Atlantic), regional (along the coast of each country), shore and patch level.

1. Species diversity, density and composition of nematodes would be higher in Korea than in its new home in the British Isles.
2. Despite expected differences in taxonomic composition, functional diversity measured by traits would be similar between the two countries as a result of the host species being the same.
3. There would be some “cosmopolitan species’ which might possibly have ‘hitchhiked’ with *Sargassum muticum*. This hypothesis can be only confirmed by molecular genetic approaches but evidence of very similar species morphologically would prompt further work.

4.2 Material and Methods

4.2.1 Sampling Strategy

To compare the coast of the British Isles with Korea, a hierarchical experimental design was planned (Figure 4.1). Each set of samples were to be collected following a fully nested hierarchical design taking into account five spatial scales: geographic setting (British Isles and Korea), regions (three coastal stretches at least 50 kilometres apart), shores (two shores about 1 km long and at least 10 kilometres apart for each habitat), patches (three patches at least 10m apart), replicates (three replicates of each patch at least one metre apart). Two rocky shores in three regions at the low intertidal level (between 1.4 m and 2 m above chart datum), on moderately exposed shores with dense cover of *Sargassum muticum* were selected for sampling. Sampling locations with similar environmental conditions were chosen, mainly open rock avoiding pools. The sampling unit was one holdfast with a plant of approximately 60cm height. Unfortunately, the planned experimental design was compromised by lack of enough *Sargassum muticum* at one of the selected shores in the central region of the English Channel. In Korea, there were problems in finding sufficient *Sargassum muticum* on the south coast. Thus the central regions on both coast shave been removed from formal statistical analyses.

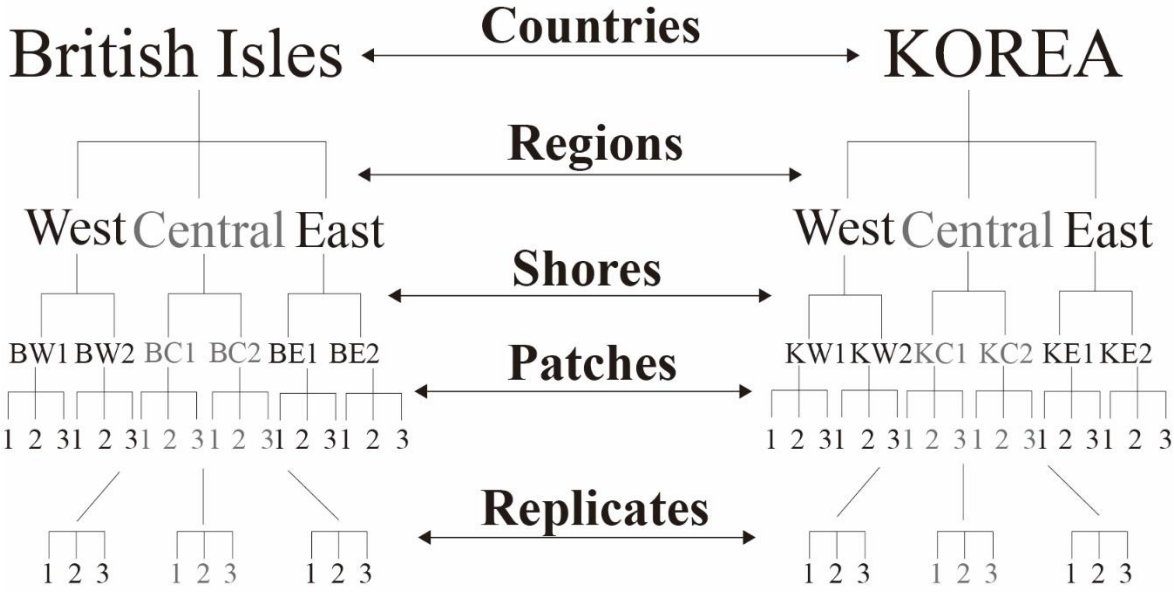


Figure 4.1 The schematic view of planned nested sampling design; BW1: Looe, BW2: Heybrook Bay, BC1: Osmington (in July 2016), BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang. In Korea, there were problems in finding sufficient *Sargassum muticum* on the central south coast. Thus the central regions on both coast have been removed from formal statistical analyses.

4.2.2 Study Areas and Sample Collection

Samples of *Sargassum muticum* hosting nematodes were collected in the British Isles in July in 2014 and Korea in March in 2015 (Figure 4.2 and 4.3). *S. muticum* was collected at the season of maximum growth which differs between its native range in the Northwest Pacific (early spring), and Europe, invasive range (end of summer). To compare spatial differences two shores in each of three regions were surveyed on the coasts of each country (Table 4.1): in the British Isles in the western English Channel, Looe (BW1) and Heybrook Bay (BW2), in the central English Channel, Osmington (BC1 in July, 2016), Swanage (BC2), and the eastern English Channel Brighton (BE1), Beachy Head (BE2); in Korea Jindo (KW1) and Wando (KW2) in the west, Yeosu (KC1) and Busan (KC2) in the south coast of Korea, and Ulsan (KE1) and Pohang (KE2) in the eastern coast of Korea. Locations selected in Korea were limited due to gaps in distribution of *S. muticum*. In July 2014, there was insufficient *S.muticum* at one of the selected shores (Portland Bill). Therefore, I recollected samples from a nearby replacement shore (Osmington) in the central English Channel (BC1) in July 2016 to provide a balanced sampling design. On each shore, three replicate patches were chosen within which at least five replicates of *S. muticum* were collected for morphological identification of nematodes. An additional five replicates were taken for potential future molecular analysis. Each replicate consisted of one plant with a single holdfast; this was completely removed from the substrata by scraping, carefully placed into a labelled plastic bag and immediately moved into an Icebox. They were subsequently frozen to preserve them for further processing in the laboratory.

Table 4.1 Environmental context of sampling locations

NO	Geographical Region	Locality	Latitude N	Longitude W	Tides Range (m)	Annual water temperature range (℃)	Oceanic Index
BW1	Atlantic	Looe	50°20'29.4"N	4°27'39.7"W	5.4	9~17	3
BW2	Atlantic	Heybrook Bay	50°19'09.1"N	4°06'51.7"W	5.53	9~17	3
BC1	Atlantic	Osmington	50°38'02.9"N	2°22'34.1"W	2.5	9~17	2
BC2	Atlantic	Swanage	50°36'27.22"N	1°56'39.38"W	2.07	9~17	2
BE1	Atlantic	Brighton Marina	50°48'39.87"N	0°5'28.83"W	6.5	8~17	1
BE2	Atlantic	Beachy Head	50°44'15.0"N	0°15'09.1"E	7.3	9~17	1
KW1	Pacific	Jindo	34°23'45.39"N	126°16'31.5"E	4.03	8~25	2
KW2	Pacific	Wando	34°17'47.6"N	126°42'05.8"E	4.04	9~23	2
KC1	Pacific	Yeosu	34°49'08.8"N	127°46'02.9"E	3.69	8~27	1
KC2	Pacific	Busan	35°09'39.9"N	129°11'38.8"E	1.35	11~27	1
KE1	Pacific	Ulsan	35°37'42.32"N	129°26'33.66"E	0.56	10~25	3
KE2	Pacific	Pohang	36°4'47.81"N	129°34'5.13"E	0.3	9~26	3

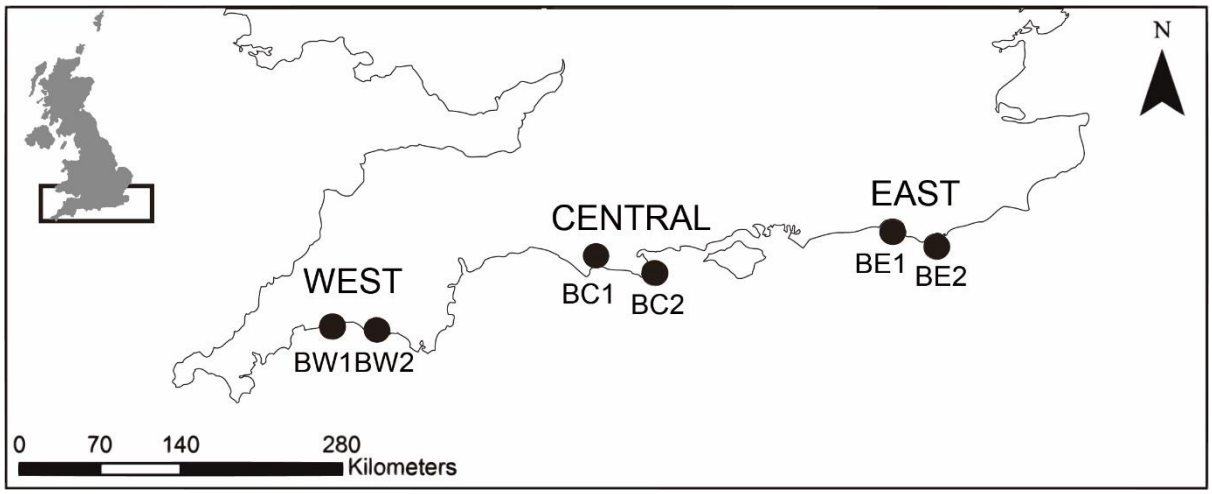


Figure 4.2 The sampling locations in British Isles; BW1: Looe, BW2: Heybrook Bay, BC1: Osmington, BC2: Swanage, BE1: Brighton, BE2: Beachy Head.

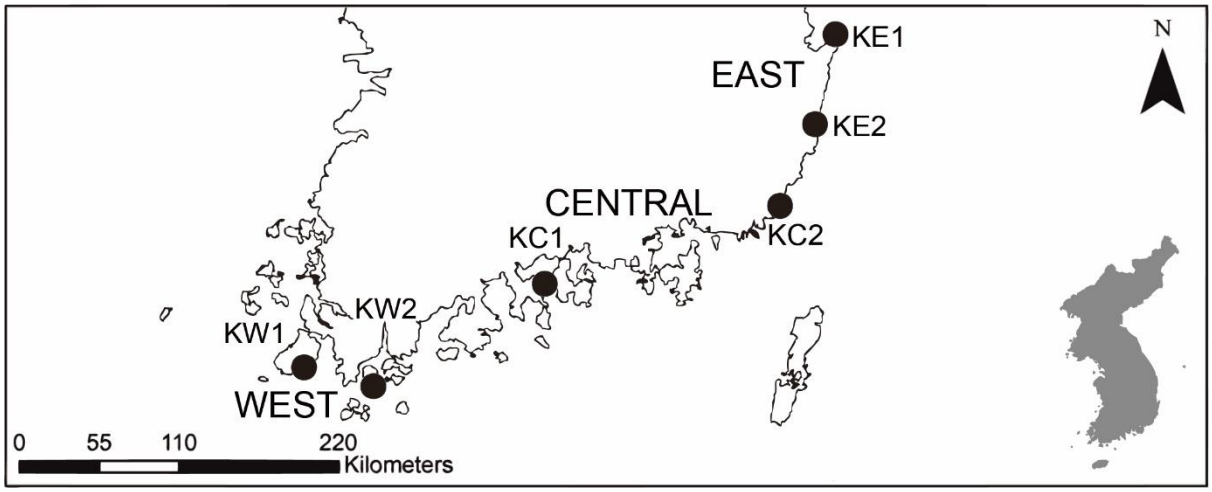


Figure 4.3 The sampling locations in Korea; KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang.

4.2.3 Sample Analysis

In the laboratory, three replicates were analysed for morphological identification of nematodes to species level. The plants were then washed with filtered tap water, firstly being decanted through nested sieves of 1mm to remove the larger fragments of algae and sediments and then sieved on 63 μm and 38 μm mesh respectively. Each *S. muticum* sample was rinsed and decanted three more times to ensure that all the organisms had been removed; then extracted epifauna were preserved in 4% buffered formalin then were washed with distilled water.

After extraction, the *S. muticum* were blotted and dried with paper to remove water, kept at room temperature for one hour, before their wet weight was measured to the nearest gram (Taylor & Cole 1994). They were oven-dried 60 ºC for 48 hours to measure their dry weight in order to calculate nematode density per gram of host seaweed. All the nematodes were picked out and counted under a stereo microscope (Leica M125). Nematode samples were divided using a Folsom Plankton Splitter when nematodes were too numerous to count (McEwen et al. 1954). The first 100 nematodes were randomly chosen and mounted in anhydrous glycerine on HS slides to observe both sides of the specimens (Shirayama et al. 1993) and identified to genus or species level under a light microscope (Olympus BX53). The pictorial keys of Platt and Warwick (1983), Handbook of Zoology (Holovachov & Schmidt-Rhaesa 2014) and the NeMys Database (Deprez 2007) were used to identify the specimens.

4.2.4 Statistical Tests

Multivariate data analysis was performed using the software Primer 6.0.2 (Clarke & Gorley 2006) with PERMANOVA add-on. Univariate and bivariate analyses were carried out with Minitab (ver.12). Statistical differences at each different spatial scale were tested by nested analysis of variance (ANOVA) for the quantitative data on the nematode assemblages: abundance and number of genera standardised by dry weight biomass of seaweed. The British Isles and Korea were fixed factors with regions, shores and patches as random factors. The central region in both countries was not used in the analyses to preserve a proper nested experiment design. The data were checked with diagnostic graphics to ensure they fulfilled parametric assumptions; when needed, the data were transformed and re-checked to ensure that the transformation improved frequency distributions.

The data for abundance of each nematode species in each replicate, patch, shore, region and country were standardised by their total abundance and square-root transformed. The means of the transformed abundance for each patch, shore and region were used to construct a Bray-Curtis similarity matrix; this was then subjected to group averaged hierarchical cluster analysis and non-metric multidimensional scaling (nMDS) ordination. The samples were coded by regions to visually assess the extent to which the species composition of the samples from the various patches, shores and regions were similar or different. Relationships between multivariate structure of nematode assemblages and normalised abiotic factors were compared by BEOENV methods in BEST test using Spearman's correlation between similarity matrices.

The samples were subjected to fully nested analysis of similarity (ANOSIM) and nested PERMANOVA with Monte Carlo tests to ascertain whether the species compositions of the nematode assemblages differed among patches within shore, among shores within region, and among regions within each country. For this and all subsequent ANOSIM, the null hypothesis, that there were no significant differences among groups, was rejected if the significance level (P) was greater than 0.05 or 5%. The R-statistic values determined with ANOSIM were used to ascertain the degree to which those groups were significantly different. R-statistic values approaching unity indicate that the compositions of the groups were very different; whereas those close to zero show that they are very similar (Clarke 1993). When ANOSIM detected significant differences among the groups, similarity percentages (SIMPER) were used to determine the species that typified those groups and the species that distinguished each group from each of the other groups (Clarke 1993). PERMANOVA with Monte Carlo tests were used to see at which spatial scale difference in species composition of nematode assemblages occurred. Estimates of components of variation were made in a PERMANOVA test to identify variability of nematode composition in each spatial scale.

4.2.5 Functional Traits Analysis

Each nematode species was classified according to four different biological traits, based on their morphological and functional features. Feeding types based on the morphology of the buccal cavity were developed by Wieser (1953), who classified free-living nematodes into four feeding types such as selective deposit feeders (1A), non-selective deposit feeders (1B), epigrowth feeders (2A), and omnivore/predators (2B).

An additional functional group classification can be provided by tail shape of nematode indicative of mobility, habitat preference and lifestyle (Thistle & Sherman 1985). The diversity of tail shapes together with the features of buccal morphology have proven to be an effective tool for discriminating nematode assemblages (Thistle et al. 1995). Nematodes were classified into four tail shape groups: 1; Short/round tail type with blunt end, 2; Clavate-conicocylindrical tail type, initially conical with an extension to the tip, 3; Conical tail type, with a pointed tip and tail length less than five body widths, 4; Long tail type, with a tail longer than five body widths (Thistle & Sherman 1985).

The life history strategies of nematodes have been described by Bongers et al. (1991), who proposed a scale (c-p score) to classify the genera of nematodes based on their ability to colonise or persist in a certain habitat. The scale ranges from extreme colonisers (c-p score=1) to extreme persisters (c-p score=5).

Biological traits matrix based on the above the approaches was used to assess the functional structure of nematode communities at all spatial scales (Schratzberger et al. 2007). The three following functional trait categories were used: feeding type, life history strategies and tail shape, giving thirteen combined categories in total. A biological traits matrix was created by assigning to each nematode taxon to each trait category. The biological trait matrix was then combined with relative species abundance to give abundance-weighted traits matrices at each spatial scale.

4.2.6 Environmental variables

Broadscale environmental data (maximum SST and average tidal range in British Isles and Korea) was collected from The Centre for Environment, Fisheries and Aquaculture Science (Cefas) in British Isles and from National Oceanographic Research Institute in South Korea. These data were combined with algal dry and

wet weight to investigate abiotic and biotic factors influencing assemblage abundance and composition. An Oceanic Index was derived by the direction of currents coming from open sea (Pingree & Griffiths 1980, Huh 1982) in order to investigate how currents affect nematode assemblage. Therefore, Oceanic Index 3 indicates near to the open ocean; 2 is intermediate and 1 is more coastal neritic waters.

4.3 Results

4.3.1 Abiotic information

ANOVA tests indicated that maximum, minimum Sea-Surface Temperature (SST) and tidal range were significantly different among shores (P<0.001), among regions (P<0.001) and between countries (P<0.001). Annual SST in Korea is generally warmer in summer and colder in winter than in the British Isles. The samples in Korea were collected in early spring while the samples in British Isles were collected in summer reflecting the time of maximum growth of *Sargassum*. Tidal range was greater in the British Isles than Korea. In the English Channel the smallest tidal range was in the central region (BC) with greater tidal range in the western (BW) and eastern (BE) regions. In Korea, the gradient of tidal range was from the western higher tidal range region (KW) to the eastern region (KC and KE).

4.3.2 Nematode assemblages in *S. muticum* from the South coast of the British Isles

2706 nematode specimens belonging to 69 taxa were mounted and identified from *S. muticum* samples taken from the south coast of the British Isles in July 2014. The density and the number of nematodes from the southeast (1621 individuals, 49 taxa) of British Isles were greater than other two regions of British Isles (the southwest, 576 individuals, 41 taxa; central coast, 509 individuals, 43 taxa) (Figure 4.4 and 4.5). The most dominant nematode species across all British sites was *Euchromadora* sp. 1, followed by *Chromadora* sp. 1 and *Cyatholaimus* sp. 1, together contributing more than 50% of total abundance; whilst 52 species contributed less than 1% of total abundance. *Euchromadora* sp. 1, an epigrowth feeder (2A), was the most dominant nematode species on the southeast and the central coast of British Isles; whereas *Viscosia* sp. 1 was the most dominant nematode on the southwest. The most common functional type of nematodes

across British shores was epigrowth feeders (2A), with conical tail types, and a c-p value 3 being intermediate between colonizers and persisters with moderate length of life.

On the southwest coast of the English Channel (Looe, BW1, and Heybrook Bay, BW2), 576 nematode specimens belonging to 41 taxa were mounted and identified from samples collected in July 2014. The density and the number of nematode species in Heybrook Bay was greater than Looe. SIMPER tests showed that *Viscosia* sp. 1, an omnivore/predator (2B), contributed the most to dissimilarity between the two shores (Table 4.4). The functional traits were different between the two shores: epigrowth feeders (2A) with c-p value 3, and clavate-conicocylindrical tail type were most common at Looe; the omnivore/predators (2B), with c-p value 3 and conical tail type at Heybrook Bay.

509 nematode specimens belonging to 43 taxa were mounted and identified from samples collected from the central coast of the English Channel (Osmington, BC1 in July 2016, Swanage, BC2 in July 2014). The density and the number of nematode species in Swanage was greater than Osmington. A SIMPER test showed that *Euchromadora* sp 1. an epigrowth feeder (2A), contributed the most to dissimilarity between two shores (Table 4.4). The most dominant feeding type of nematodes on both two shores was epigrowth feeders (2A), with c-p value 3 and a conical tail type.

1621 nematode specimens belonging to 49 taxa were mounted and identified from samples from the southeast of the English Channel (Brighton, BE1, Beachy Head, BE2) in July 2014. The number of nematode species found in Brighton was greater than at Beachy Head; while the density of nematodes were similar between the two shores. Epigrowth feeders (2A) were the most dominant feeding type on both shores. A Maturity Index of 3, intermediate between colonizers and persisters indicating moderate length of life, was dominant on both shores.

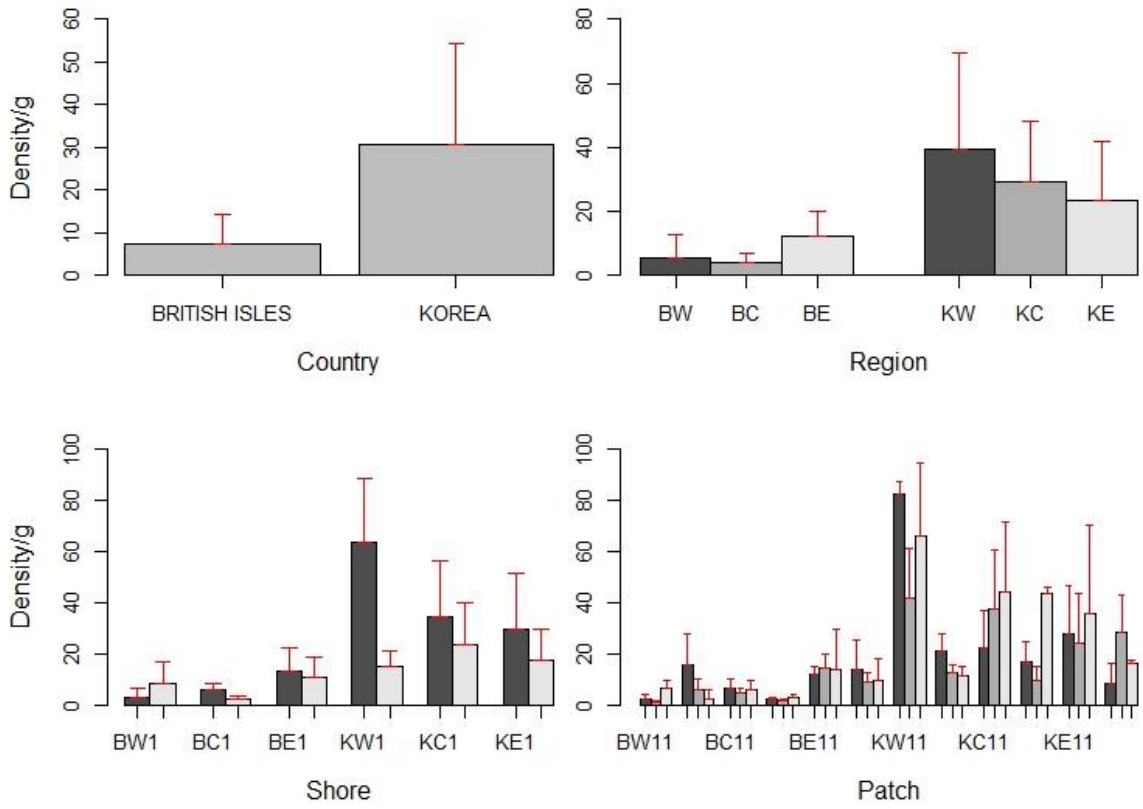


Figure 4.4 Mean density of nematode assemblages with standard deviation at all spatial scales; BW1: Looe, BW2: Heybrook Bay, BC1: Osmington, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang.

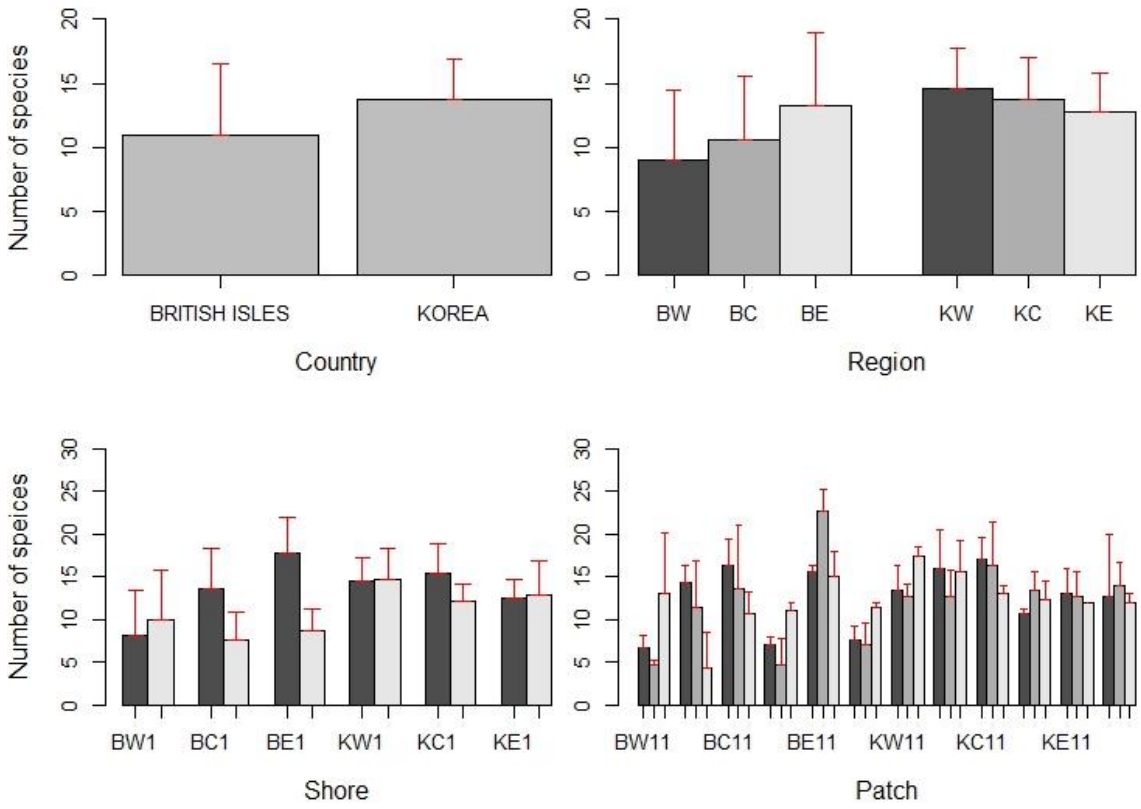


Figure 4.5 Mean number of nematode species with standard deviation at all spatial scales; BW1: Looe, BW2: Heybrook Bay, BC1: Osmington, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang.

4.3.3 Nematode assemblages in *S. muticum* from the south and east coast of Korea

5224 nematode specimens belonging to 62 taxa were mounted and identified from *S.muticum* samples collected from the south and east coast of Korea in March in 2015. The density of nematodes on the southwest coast of Korea (1885 individuals, 42 taxa) was greater than the other two regions (the southeast, 1666 individuals, 45 taxa, and the east of Korea, 1673 individuals, 39 taxa) (Figure 4.4 and 4.5). The most dominant nematode species across all shores in Korea were *Pontonema* sp. 1 followed by *Eurystomina* sp. 1, *Theristus* sp. 1, *Acanthonchus* sp. 1 together contributing more than 50% of total abundance. Less than 1% of total abundance were contributed by 43 species. The most dominant nematode species were different in each region: *Pontonema* sp. 1 on the southwest coast, *Eurystomina* sp. 1 on the southeast coast and *Theristus* sp. 1 on the east coast of Korea. The most dominant functional type of nematode across Korean shores was the omnivore/predator (2B), with clavate-conicocylindrical tail type, and c-p value 4 indicating a persistent species with relatively long life span, later maturity and less effort spent on reproduction (k-strategists).

From samples collected from the southwest coast of Korea in March 2015 (Jindo, KW1, and Wando, KW2), 1885 nematode specimens belonging to 42 species were mounted and identified. The density of nematodes in Wando was greater than Jindo, but the number of nematode species was similar on both shores. *Pontonema* sp. 1, the omnivore/predator (2B), was the most dominant nematode species. The SIMPER test also showed that *Pontonema* sp. 1 contributed the most to dissimilarity between Jindo and Wando (Table 4.4). Omnivore/predator 2B) was the most dominant feeding type across both shores. The Maturity Index of c-p 4 indicated dominance by persistent, relatively long-lived, delayed maturity species spending less effort on reproduction (K-strategists) on both shores. The short/round tail type was the most dominant tail type in *S. muticum* on the Southwest coast of Korean. In terms of their biological traits, the omnivore/ predators with short/round tails and c-p value 4 were the most common nematode species living in *S. muticum* along the southwest of Korea.

1666 nematode specimens belonging to 45 taxa living in *S. muticum* were mounted and identified from samples collected on the southeast coast of Korea (Yeosu, KC1, and Busan, KC2) in March 2015. The density and number of nematode species in Yeosu were significantly higher than in Busan. The dominant

species were different between two shores: the non-selective deposit feeders *Theristus* sp 1 (1B), in Yeosu (KC1) and *Eurystomina* Sp. 1, an omnivore/predator (2B) in Busan (KC2). The SIMPER test showed that *Theristus* sp. 1 contributed the most to dissimilarity between two shores (Table 4.4). The major feeding type across the southeast coast of Korea was predator/omnivores (2B). However, non-selective deposit feeders were also abundant in Yeosu; whereas only predator/omnivores were dominant in Busan. Nematode species with a c-p value 2 were dominant in Yeosu; in contrast nematodes with a c-p value 4 were abundant in Busan. The clavate-conicocylindrical tail type was the most dominant tail shape on both shores.

1673 nematode specimens belonging to 39 taxa living in *S. muticum* were mounted and identified from samples collected on the east coast of Korea (Ulsan, KE1, and Pohang, KE2) in March 2015. The nematode density in Ulsan was significantly higher than Pohang; whereas the number of nematode species from the two shores was similar. The dominant nematode species in the east coast of Korea were also different between the shores with *Theristus* sp. 1, the non-selective deposit feeders (1B), dominant at Ulsan and *Euchromadora* sp. 1, on epigrowth feeder (2A) at Pohang. The SIMPER test indicated that *Theristus* sp. 1 contributed the most to dissimilarity between the two shores. Predator/omnivores (2B) were the most dominant feeding type on the east coast of Korea. A Maturity Index of 3, intermediate between colonizers and persisters, indicating a moderate life span, was dominant on both shores. The clavate-conicocylindrical tail type was the most dominant tail type in Ulsan; while the short/round tail shape was the most prevalent in Pohang.

4.3.4 Taxonomic composition of nematode assemblages across geographical scales

In total, 7930 individuals were identified as belonging to 90 taxa when pooling samples from all sites in both the British Isles and Korea. The top five ranked species, accounted for 52% of total nematode abundance: *Euchromadora* sp 1. (15%), *Pontonema* sp. 1 (13%), *Eurystomina* sp. 1 (9%), *Theristus* sp. 1 (8%), *Acanthonchus* sp. 1 (6%); whereas 73 species contributed individually to less than 1% to total relative abundance. The most widely distributed nematode species in British Isles was *Euchromadora* sp. 1, whereas *Pontonema* sp. 1 was the most common nematode species in Korea. Several nematode species were commonly

found in both countries: *Euchromadora* sp. 1, *Enoplus* sp. 1, *Oncholaimus* sp. 1 and *Phaenoncholaimus* sp. 1. However, the dominant nematode species in each country were different: *Pontonem*a sp. 1 and *Euchromadora* Sp. 1 in Korea and *Chromadora* Sp. 1 and *Eurystomina* Sp. 1 in the British Isles.

4.3.5 Density and number of nematode assemblages across all geographical scales

The density of nematode assemblages in *S. muticum* were significantly different among patches (P <0.05), between countries (P<0.05), but not between shores among regions (Figure 4.4). The number of species differed among patches (P<0.05), between shores (P<0.05), but not at other spatial scales (Figure 4.5). The density in Wando was the highest across all shores in both countries; the lowest density recorded in all geographical regions was at Osmington. In terms of number of species, Brighton was the most diverse shore across all geographical regions. In contrast, Ulsan and Osmington were the least diverse among all the shores. The average range of nematode density per gram in samples was between 80~100 in Korea, and 20~90 in British Isles. The average range of the number of nematode species was 22~40 across both geographical scales.

4.3.6 Comparisons of species compositions across all geographical scales

Nested PERMANOVA with Monte Carlo tests demonstrated that the species composition differed significantly between countries (P<0.001), between shores *(*P<0.001) and among patches (P<0.001), but not among regions (Table 4.3). The ordination of samples with the nMDS technique showed clear differences between countries and among shores (Figure 4.7). The Estimates of Component Variation test in PERMANOVA indicated that between countries variations in nematode assemblage composition was greater than at the other three spatial scales (Table 4.2). Nested ANOSIM test showed that the species composition was significantly different among shores (R = 0.628, P<0.001) and patches (R = 0.368, P<0.001), but not among regions and between countries. The ordination of samples with nMDS showed clear differences between countries; but no clear differences were apparent among regions in both countries.

Table 4.2 Estimates of components of variation in PERMANOVA; Co: Countries, Re: Regions, Sh: Shores, Pa: Patches, Res: Replicates.

Estimates of components of variation		
Source	Estimate	Sq.root
S (Co)	835.91	28.912
V (Re(Co))	354.84	18.837
V (Sh(Re(Co)))	505.8	22.49
V (Pa(Sh(Re(Co))))	295.98	17.204
V (Res)	1147.2	33.87

Table 4.3 P-value of nested PERMANOVA with Monte Carlo tests in each measurement; Co: Countries, Re: Regions, Sh: Shores, Pa: Patches. Significant values were marked as "bold".

Source	Species abundance	Feeding type	Maturity index	Tail shape	Biological traits
S (Co)	**0.032**	0.066	0.155	0.194	**0.031**
V (Re(Co))	0.44	0.416	0.218	0.081	0.141
V (Sh(Re(Co)))	**0.001**	**0.001**	**0.004**	0.077	**0.001**
V (Pa(Sh(Re(Co))))	**0.001**	0.316	0.091	**0.017**	**0.002**

4.3.7 Biological traits across all geographical scales

Nested PERMANOVA with Monte Carlo tests showed that the feeding type of nematode assemblages differed significantly between shores (P<0.05) but not other spatial scales. The most dominant feeding types was the epigrowth feeders (2A) in the British Isles while the predator/omnivores (2B) were most abundant in Korea (Figure 4.6). The dominant life history strategy of nematode assemblages (MI) were significantly different between shores (P<0.001); but were not different between countries, among regions and patches. Nematode species with c-p value 4 indicating persistent species with relatively long life spans, later maturity and less effort spent on reproduction (k-strategists) formed the most dominant component (42%) across both geographical scales. Conical tail type was the most common tail type at the majority of shores in both countries. nMDS plots based on the relative abundance of nematode species, feeding type, life history strategies and tail shape showed clear differences between countries; but there were not any clear differences at other spatial scales (Figure 4.8). Biological traits of nematode assemblages in *S. muticum* were significantly different between countries (P<0.05), between shores (P<0.001) and among patches (P<0.001), but not among regions. nMDS plots of biological traits indicated clear differences similar to those nMDS plots based on relative nematode abundance data (Figure 4.9).

4.3.8 Relationship between abiotic conditions and nematode assemblage structure

The BIOENV method in the Best test using similarity matrices based on relative nematode abundance and biological traits (feeding type, Maturity Index, tail shape and combined biological traits) showed that nematode species composition was significantly positively correlated with the combination of Sea-Surface Temperature (SST) and tidal range (R = 0.497, P<0.001). The feeding type of nematode was positively correlated with the combination of Sea-Surface Temperature (SST) and tidal range (R = 0.42, P<0.001). Life strategy, tail shape of nematode and combined biological traits were also positively correlated with the combination of Sea-Surface Temperature (SST) and tidal range (Table 4.5).

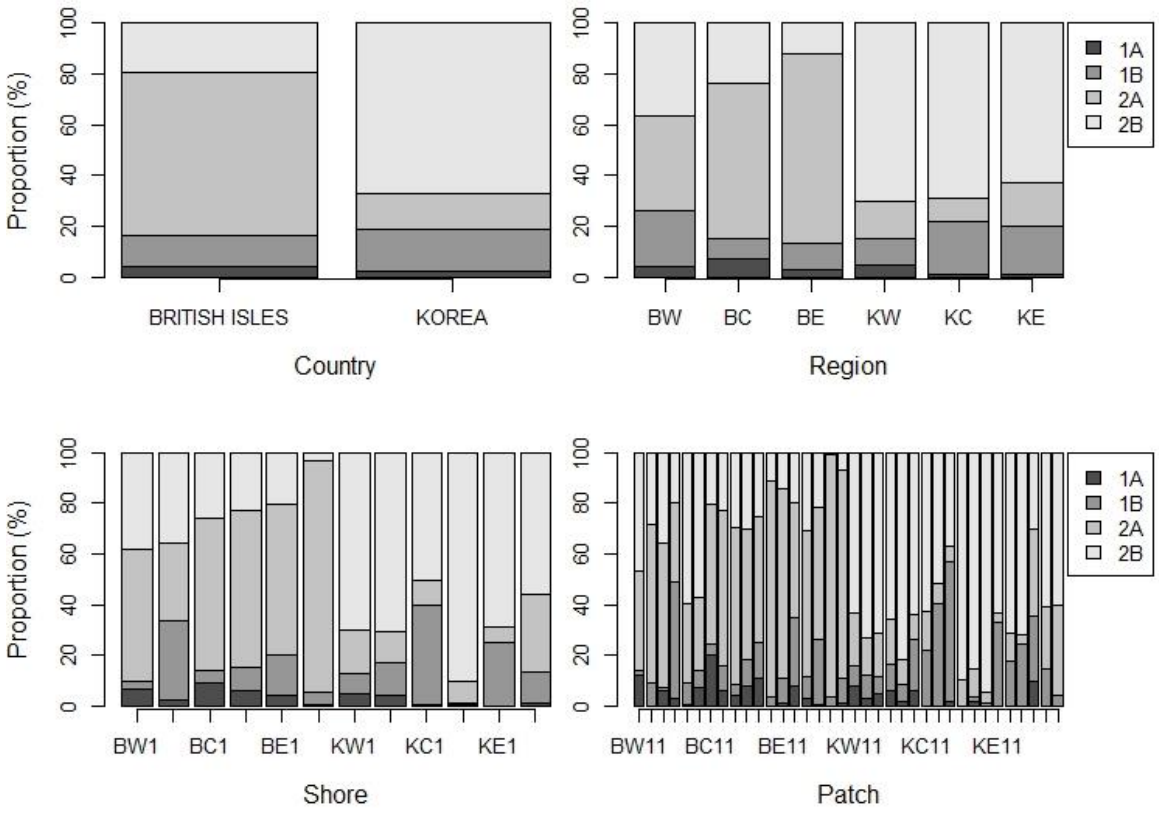


Figure 4.6 Trophic composition of nematode assemblages on the basis of average percentages in all spatial scales. The feeding types defined after Wieser (1953); 1A=selective deposit feeders, 1B=non-selective deposit feeders, 2A=epigrowth feeders, 2B=predators/omnivores, BW1: Looe, BW2: Heybrook Bay, BC1: Osmington, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang.

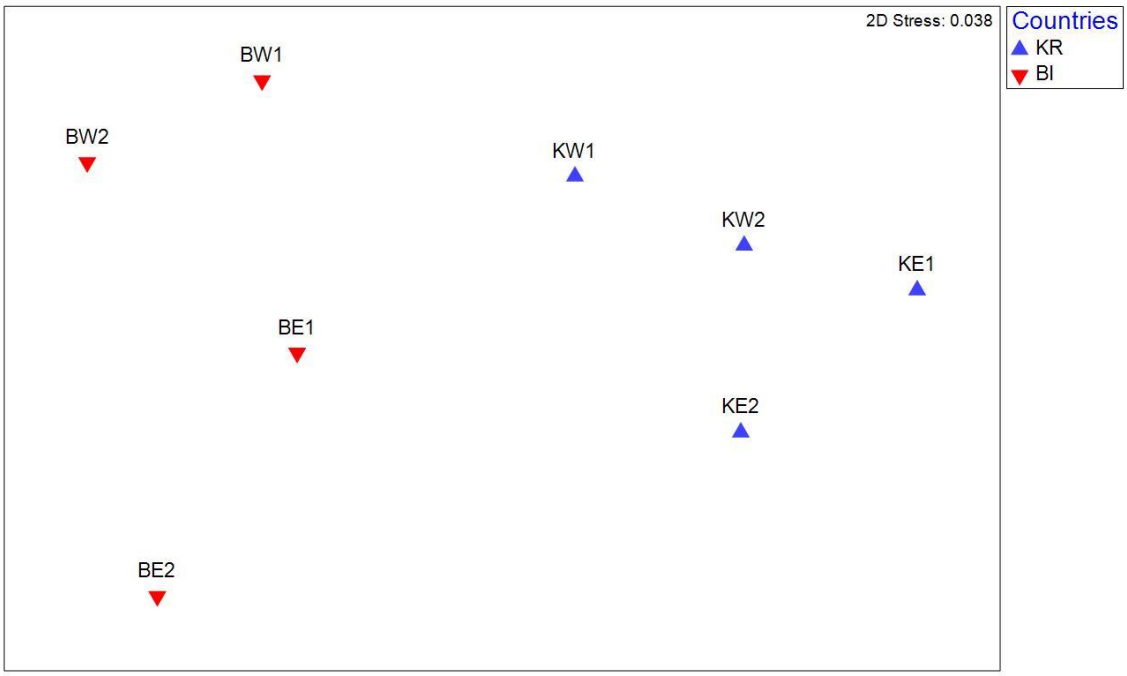


Figure 4.7 Non-parametric multi-dimensional scaling (MDS) ordination based on the standardised abundance of nematode species by shores. BW1: Looe, BW2: Heybrook Bay, BC1: Osmington, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang, BI: British Isles, KR: Korea.

Figure 4.8 Non-parametric multi-dimensional scaling (MDS) ordination based on the standardised abundance of nematode species, feeding type, tail shape and life history strategy (Maturity index) in shore level; BW1: Looe, BW2: Heybrook Bay, BC1: Osmington, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang, BI: British Isles, KR: Korea.

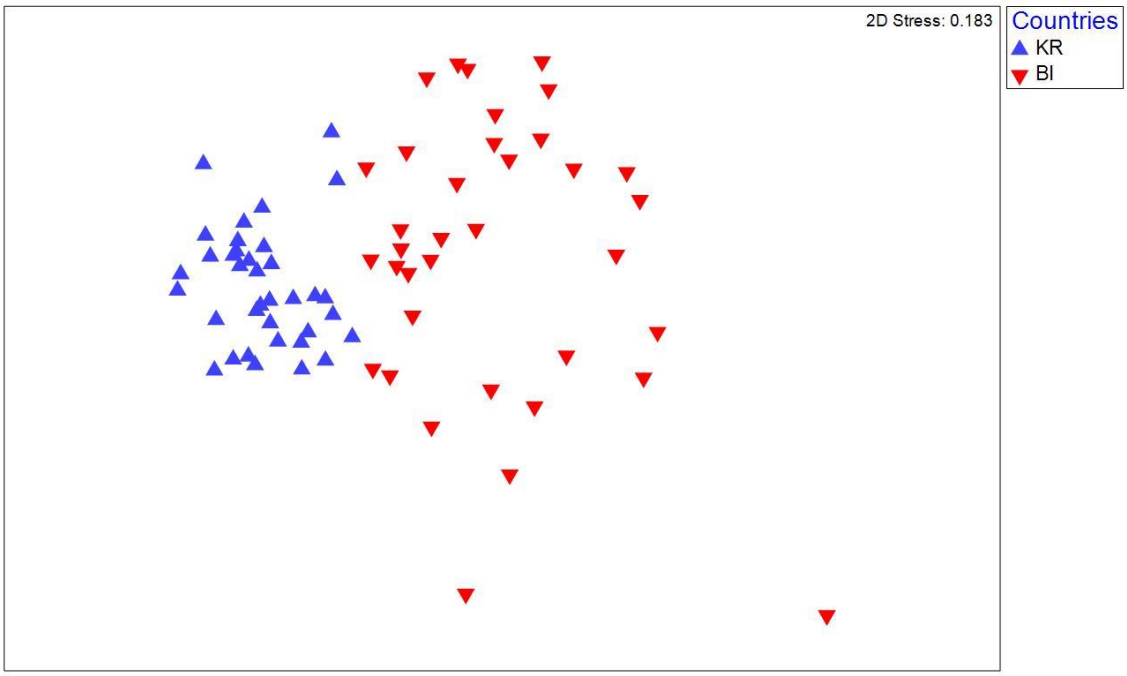


Figure 4.9 Non-parametric multi-dimensional scaling (MDS) ordination based on the standardised nematode biological traits data at all spatial scales; BI: British Isles and KR: Korea.

Table 4.4 Result of one way SIMPER analysis of nematode abundance data at the level of shore listing the main three discriminating species, their average abundance on each shore (Av. Abund), average of dissimilarity (Av. Diss), standard deviation of dissimilarity (Diss/SD), contribution (Contrib%), accumulation (Cum%) and average dissimilarity (AD); BW1: Looe, BW2: Heybrook Bay, BC1: Osmington, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: Jindo, KW2: Wando, KC1: Yeosu, KC2: Busan, KE1: Ulsan, KE2: Pohang.

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
BW1 & BW2						
AD= 52.89	BW1	BW2				
Pontonema Sp. 1	40.11	43.33	9.74	1.45	18.42	18.42
Eurystomina Sp. 1	13.89	5.33	4.66	1.33	8.82	27.24
BC1 & BC2						
AD=72.44	BC1	BC2				
Theristus Sp. 1	36.11	0.11	19.82	2.13	27.36	27.36
Eurystomina Sp. 1	6.11	28.33	12.62	1.81	17.42	44.78
BE1 & BE2						
AD= 63.32	BE1	BE2				
Theristus Sp. 1	23.44	8.89	8.97	1.27	14.16	14.16
Euchromadora Sp. 1	3.89	18.67	8.48	1.25	13.39	27.55
KW1 & KW2						
AD= 86.10	KW1	KW2				
Viscosia Sp. 1	3.33	6.11	11.71	0.95	13.6	13.6
Sabatieria Sp. 1	0	9.33	11.21	0.85	13.02	26.62
KC1 & KC2						
AD= 77.44	KC1	KC2				
Euchromadora Sp. 1	3.22	8.67	13.19	1.32	17.03	17.03
Neochromadora Sp.1	5	0.56	8.27	1.18	10.68	27.71

Table 4. 4 Result of one way SIMPER analysis of nematode abundance data at the level of shore

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
KE1 & KE2						
AD= 64.18	KE1	KE2				
Euchromadora Sp. 1	31.33	49	17.81	1.14	27.75	27.75
Chromadora Sp. 1	2.33	28.11	14.56	1	22.68	50.43

Table 4.5 Results of BIOENV analysis matrices of similarity derived data of species abundance and biological traits. The combination of abiotic factors with the highest value of Spearman rank correlation (ρ) and significance values (p) in each matrix were shown; Te: sea-surface temperature, Ti: tidal range, Oc: Oceanic index, Sd: the amount of retention sediment.

Matrix	P (rho)	*p*	Abiotic factors
Species	0.497	0.001	Te, Ti
Feeding type	0.42	0.001	Te, Ti
Life Strategy	0.25	0.001	Te, Ti
Tail shape	0.117	0.001	Te, Ti
Combined biological traits	0.424	0.001	Te, Ti

4.4 Discussion

The results of my study demonstrated that the density, number of species, assemblage composition and biological traits of nematode assemblages living in *S. muticum* were significantly different between the English Channel and South coast of Korea. The density and number of nematode species in Korea was higher than the density and number of nematodes in the British Isles, thus supporting the first hypothesis (in section 4.1). Several common nematode genera associated with *S. muticum* appeared in both countries. These genera belong to the families Chromadoridae and Oncholaimidae which are commonly found in macroalgae (Holovachov & Schmidt-Rhaesa 2014). There were also more nematode species per common genus in *S.muticum* in Korea than in the British Isles. This result might reflect different evolutionary histories and processes between the two oceans (Witman et al. 2004). Thus *S. muticum* hosts greater diversity within its native area then in its invasive range. This probably reflects a more diverse regional species pool and a larger time for species to colonize and adapt to the habitat provided by *S. muticum*.

Nematode species compositions were different among patches, between shores and countries. They were not, however, different among regions on the same coast. Nevertheless, this pattern varied across patches and shores (Table 4.2). This small scale variability in composition could be due to the interaction of local physical and biological factors. For examples, aggregation of dominant nematode species causes small scale heterogeneity (Blome et al. 1999); predation can have a major influence on epifaunal communities (Edgar & Klumpp 2003). Nutrient loading might also affect the epiphytic load of algae that lead to changes in epifaunal assemblages (Viejo 1999, Wikström & Kautsky 2004).

At larger scales, several nematode species were commonly found across regions in each country, although the most dominant nematode species varied on each shore. Several studies have shown little specialization in epifaunal assemblages to particular host macroalgae because most epifaunal species feed directly on host tissues irrespective of host species (Arrontes 1999, Wikström & Kautsky 2004, Prado & Thibaut 2008). In contrast, others have reported a strong relationship between epifaunal assemblages and host-macroalgae, probably determined by specific chemical, structural or morphological characteristics of the algal species (Wieser 1959, Heip et al. 1985, Edgar & Klumpp 2003, Schmidt & Scheibling

2006). My results showed considerable variation in the dominant species at local patch and shore scales in both countries indicating a suite of specialist species.

Previous studies of epifauna associated with *S. muticum* have focused on the comparison between epifaunal community structure in *S. muticum* as an invasive species when compared to native macroalgae (Arrontes 1999, Wikström & Kautsky 2004, Prado & Thibaut 2008, Engelen et al. 2013, Veiga et al. 2016). These studies have reported the result of comparisons between invertebrate communities between native algae and *S.muticum* are dependent on the morphological complexity of native algae. For example, some studies of morphologically complex native macroalgae (e.g. *Halidrys siliquosa*, *B. bifurcata*, *Cystoseira* spp., *Stypocaulon scoparium*) concluded there were no substantial changes or loss in the composition of epifaunal assemblages (Wernberg et al. 2004, Buschbaum et al. 2006, Engelen et al. 2013, Veiga et al. 2014). In contrast, other studies with less morphologically complex native macroalgae (i.e. *Fucus* sp. and *C. crispus*) indicated that epifaunal assemblages were relatively more diverse in invasive macroalgae, thereby potentially increasing biodiversity (Viejo 1999, Buschbaum et al. 2006, Veiga et al. 2014).

Moreover, some previous studies have only used one spatial scale or higher taxonomic level (Stæhr et al. 2000, Wernberg et al. 2004, Gestoso et al. 2010) or focused on epifaunal macrofauna assemblages. Therefore, comparative study of the epifaunal assemblages associated with *S.muticum* in its native regions and invasive regions are needed to investigate the role of invasive macroalgae on their associated fauna. My study showed that the host-specialist nematode in each country were different. Estimates of components of variation in PERMANOVA also showed the species variations between countries were higher than other spatial scales indicated as two different host-specialist groups; species belonging to Chromadoridae in the British Isles and species belonging to Oncholaimidae in Korea (Table 4.2). nMDS test also showed clear differences between countries (Fig 4.7). This contrasts with species composition between two countries that were correlated with physical and biological factors. BIOENV tests showed that the combination of tidal range and sea-surface temperature were correlated with nematode abundance ($R = 0.631$, $P < 0.001$). This may, however, just be coincident with differences in the physical setting between the Korean coast and the English Channel.

The food resource that is available from the seaweed host might be an important variable in determining the structure of epifaunal assemblages (Gestoso et al.

2010). In my results, feeding type and life history strategy traits were different among patches, between shores and countries but not among regions. Thus I rejected my third hypothesis. In terms of feeding type, two contrasting dominant feeding types were apparent between countries (epigrowth feeders in British Isles, and omnivore/predators in Korea). This distinctive difference is also shown in the data based on taxonomic species composition. A RELATE test showed that biological traits composition was strongly correlated with nematode taxonomic species composition (R = 0.8, P< 0.001). Earlier studies have reported the positive correlation between epiphytes supported by seaweed and free-living epifaunal assemblages (Worm & Sommer 2000, Parker et al. 2001, Wikström & Kautsky 2004). Peak biomass of epiphytes occurred at the season of maximum size of the *S. muticum* plants (Gestoso et al. 2010). *S. muticum* has different life cycles between the two countries. For instance, *S.muticum* living on the European coast reach maximum length of growth in late summer (Plouguerné et al. 2006), while *S. muticum* display maximise size in early Spring in Korea (Mukai 1971). I sampled at around peak biomass in both countries. Therefore, alternation in the food resource provided by *S.muticum* could be one of the important factors that determine assemblage composition and functional traits of nematodes.

The density and diversity of nematodes in Korea were higher than in the British Isles in *S. muticum*. Nematode species diversity per genus was also higher in samples collected in Korea. Previous study also showed the species reduction in invasive species. For instance, the red turpentine beetle, *Dendroctonus valens,* showed less genetic diversity in the invasive population in China than in its native region of North America (Cai et al. 2008). My results might also reflect the different evolutionary history and processes between the two oceans. The longer association over evolutionary time of nematodes with *S. muticum* may lead to greater niche differentiation and speciation in seaweed dwelling nematodes. The higher number of species of nematodes and biological traits in Korea could also lead to a greater contribution to ecosystem functioning (Ieno et al. 2006, Godbold et al. 2011).

Despite different assemblage structures between countries, some cosmopolitan species appeared in both countries. *S.muticum* was accidentally introduced in Europe due to importation of Japanese Oyster (*Crassostrea gigas*) from 1966 (Farnham et al. 1973). As a consequence, some meiofauna and microfauna could have come to Britain with *S.muticum* growing on oysters. In my study, I found several cosmopolitan nematode species belonging to specific genera:

Euchromadora, *Eurystomina* and *Oncholaimus*. These nematode species might potentially come from Korea to the British Isles. The most likely candidates to be an invasive species are belong to *Euchromadora*, *Eurystomina*, and *Enoplus*. These genera were also found in *Corallina* spp.. Therefore, molecular genetic approaches with those cosmopolitan nematode species or genera are needed to identify whether those species have 'hitchhiked' with *S. muticum* from Korea to British Isles.

Summary

Returning to the hypotheses, hypothesis 1 was accepted; the species diversity and density was higher in Korea than British Isles. This might due to different evolutionary history and processes between two oceans including the higher diversity in Pacific Ocean and the longer association between nematodes and host plants.

Hypothesis 2 was rejected as both taxonomic and functional diversity differed between Korea and the British Isles. Although functional diversity was similar in smaller geographic scales (within country), both classifications were clearly divided by countries. This may be related with the different life strategy of *S.muticum* in each country.

Hypothesis 3 was accepted; I found several cosmopolitan species which might possibly have hitchhiked with *S.muticum.* However, some of these nematode species were also commonly found on other macroalgae (see Chapter 3). Further molecular studies are required to understand whether those species have invaded with *S. muticum* from Korea to British Isles.

Chapter 5: Alpha, beta and gamma diversity of nematode assemblages inhabiting seaweeds in the English Channel and Korea

5.1 Introduction

Comparing spatial variability in patterns of community structure has had a long history of study for ecologists (Whittaker 1970, Cornell 1985, Ricklefs 1987). Whittaker (1960) first introduced the terms of alpha, beta and gamma diversity based on compositional heterogeneity among places. Subsequently the terms of alpha, beta and gamma diversity have become broadly used in ecology. Following Tuomista (2010a), I have used the following definition: alpha (α) diversity is the mean species diversity at the local or within habitat scale; beta (β) diversity represents between habitat patch diversity; gamma (γ) diversity is the total species pool at the regional or landscape scale (Tuomisto 2010a).

Species diversity at different spatial scales can be measured as a ratio in many different ways: species richness/species richness (e.g. 1), percentage of abundance/percentage of abundance (e.g. 2), unit of diversity index/unit of diversity index (e.g. 3) and unit of external gradation/unit of external gradation (e.g. 4) (Tuomisto 2010a). In term of these definitions, diversity can be measured as sampling at different habitat classes (e.g. 1 and 2), the same habitat classes (e.g. 3) or along an environmental gradient that has not been divided into habitat classes (e.g. 4) (Tuomisto 2010a). It is, therefore, important to distinguish which quantitative interpretation of diversity is appropriate and how diversity should be calculated, and at what spatial scale. Several researchers have attempted to determine specific spatial scales of alpha, beta and gamma diversity (Whittaker 1970, Lande 1996, Tuomisto 2010a), but it still remain unclear. Srivastava (1999) reported that local and regional species richness relationships were best suited for comparing similar habitats between different regions, not different habitats in same region.

Although the definition of alpha, beta and gamma diversity remains controversial, many studies have adopted this approach to understand how diversity at different

spatial scales varies in communities (Ricklefs 1987, Srivastava 1999, Whittaker et al. 2001, Gering & Crist 2002, Tuomisto 2010a). Most previous studies have reported alpha – beta (local and regional, LR) relationships in bivariate plots suggesting a curvilinear or a linear relationship between alpha and beta (local and regional) species richness (Lande 1996, Griffiths 1999, Srivastava 1999, Gering & Crist 2002). A curvilinear model is a saturated relationship and suggests that alpha richness becomes increasingly independent of beta diversity or the regional species pool. Such a relationship indicates that alpha diversity is regulated by interactive processes such as competition and predation (Srivastava 1999). In contrast a linear relationship is indicative of alpha species diversity depending upon the beta species pool and biological processes such as evolution, colonization, extinction, and speciation in the local community (Srivastava 1999). Other studies, however, revealed that interactive processes were also found in linear models when habitat disturbance occurred.

Previous work developed mathematical properties in order to define diversity at different spatial scales (Whittaker 1960, Allan 1975, Lande 1996, Srivastava 1999, Tuomisto 2010a, b). For example, Whittaker (1960) suggested a multiplicative formula to measure the relationship between alpha, beta and gamma diversity (gamma = alpha × beta), while an additive partitioning formula (gamma = alpha + beta) was subsequently proposed by Allan (1975), and has been adopted in this study. In addition, multivariate methods have also frequently been adopted and focus on similarity of community composition within and between regions (Soininen et al. 2007, Leduc et al. 2012). These approaches have all been applied to assess ecological phenomena in empirical and experimental work (Whittaker 1960, Ricklefs 1987, Griffiths 1999, Srivastava 1999, Gering & Crist 2002, Leduc et al. 2012). However, only a few studies have been conducted in marine ecosystems (Soininen et al. 2007, Zinger et al. 2011, Leduc et al. 2012) due to their open nature, geographical connectivity and difficulty of sample collection with appropriate experimental designs.

Achieving a full inventory of species in habitats and regions is practically impossible. So collecting species composition data with increasing sampling effort to construct species-area relationships for a region has been widely adopted (Tuomisto 2010a). Nematodes are a good model for understanding species-area relationships in marine ecosystems. Nematodes are one of the most numerous and diverse taxa in the biosphere and are widely distributed in the marine environment (Platt & Warwick 1980). They are present in every habitat

such as in coastal macroalgae and sediments from shallow water to the deep sea (Platt & Warwick 1980).

The overall aim of this chapter was to determine changes in alpha, beta and gamma diversity of nematodes inhabiting two macroalgae species (*Corallina officinalis* and *Sargassum muticum*). I adopted a hierarchical sampling design: at the largest scale, biogeographic realms (South coast of Korea in North-West Pacific and the English Channel in the North-East Atlantic), in which regional (along the coast of each country), shore and patch level sampling was nested, in order to address the following questions:

1. Are relationships between alpha, beta and gamma nematode diversity similar between two different types of habitat (*Corallina* spp.and *Sargassum muticum*)?
2. Which spatial scale has the most influence on nematode diversity across two geographical realms?

5.2 Materials and Methods

5.2.1 Sampling Strategy

In order to compare the biodiversity of the coast of the British Isles with Korea, a hierarchical experimental design was used (see chapter 2) combining samples collected for chapter 3 and 4. My study used two distinct macroalgae as habitat providers of free-living nematode to enable broad-scale comparisons. *Corallina* spp. is one of the most common seaweeds on the south coasts of England and Korea. What was once considered a cosmopolitan species has recently been divided into various species using molecular techniques (Walker et al. 2009). *Sargassum muticum* (Yendo) Fensholt is a native species in Korea, and is an invasive species that was introduced in the British Isles in 1973, probably on Japanese Oysters, *Crassostrea gigas* (Farnham et al. 1973). It has become very common on rocky shores on the south coast of England.

Each set of samples were collected following a fully nested hierarchical design taking into account five spatial scales: geographic setting (British Isles and Korea), regions (three coastal stretches at least 50 kilometres apart), shore (two shores about 1 km long and at least 10 kilometres apart for each habitat), patch (three patches at least 10m apart), replicates (three replicates of 5 x 5 cm quadrat at least 10cm apart). Two rocky shores in three regions at the low intertidal level (between 1.4 m and 2 m) dominated by the chosen macroalgae were selected for sampling. Sampling locations with similar environmental conditions were chosen: moderately exposed, at least 80% turf of *Corallina* spp. and *S. muticum*. Macroalgae were gently cut and the scraped bedrock shore.

5.2.2 Study areas and Sampling Collections

Nematodes were collected from *Corallina* spp. in the British Isles and Korea in July 2014, and from *S. muticum* in the British Isles in July 2014, and in Korea in March 2015. *S. muticum* were collected at the season of maximum growth which differs between its native range in the Northwest Pacific (early spring in March), and Europe, invasive range (summer in July. To compare spatial differences two shores in each of three regions were surveyed on the coasts of each country on each macroalgae (see chapter 2): in the British Isles in the western English Channel, Looe (BW1), Heybrook Bay (BW2), in the central English Channel,

Osmington (BC1 in July in only *S.muticum* sample, 2016), Portland Bill (BC2, in July 2014 in only *Corallina* sample), Swanage (BC3), and the eastern English Channel Brighton (BE1), Beachy Head (BE2); in Korea, West-Wando (KW1), East-Wando (KW2), Jindo (KW3 in only *S. muticum* sample) in the west, Yeosu (KC1) and Namhae (KC2) in the centre and Gueje (KE1), Busan (KE2), Pohang (KEE1, in only *S. muticum* sample), Ulsan (KEE2 in only *S. muticum* sample) in the east (see chapter 2, 3, 4 for further details). Locations selected for collection of *Corallina* spp. and *S.muticum* were slightly different due to their limited distributions. As in chapter 4, nematodes living in *S. muticum* from the central regions in both countries were excluded from the formal analyses. I was not interested in the comparison of nematode assemblage between the two macroalgae. In this chapter, I focussed on alpha, beta and gamma diversity of nematodes.

On each shore, three replicate patches were chosen by randomly collecting at least five replicates of *Corallina* spp. or *S. muticum* for morphological identification of nematodes. An additional five replicates were taken for potential future molecular analysis. Each replicate was one holdfast, and completely removed from the substrata by scraping, carefully placing into a labelled plastic bag and immediately moved into an Icebox. They were subsequently frozen to preserve them for further processing in the laboratory (see Chapters 3 and 4 for further details).

5.2.3 Sample analysis

In the laboratory, only three replicates of each macroalgae were analysed for morphological identification of nematode species. The plants were then washed with filtered tap water, first decanted through nested sieves of 1mm to remove the larger fragments of algae and sediments. They were then sieved on 63 µm and 38 µm mesh respectively. Each seaweed sample was rinsed and decanted three more times to ensure that all the organisms had been removed; then extracted epifauna were preserved in 4% buffered formalin then were washed with distilled water.

After extraction, nematodes were picked out and counted under a stereo microscope (Leica M125). Nematode samples were divided using a Folsom Plankton Splitter when nematodes were too numerous to count (McEwen et al. 1954). The first 100 nematodes were randomly chosen and mounted in anhydrous glycerine on HS slides to observe both sides of the specimens

(Shirayama et al. 1993) and identified to genus level under a light microscope (Olympus BX53). The pictorial keys of Platt and Warwick (1983), Handbook of Zoology (Holovachov & Schmidt-Rhaesa 2014) and the NeMys Database (Deprez 2007) were used to identify the specimens.

5.2.4 Statistical Tests

Alpha and beta (local – regional) relationships of nematodes on both macroaglae were calculated at four spatial scales: country versus region, region versus shore, shore versus patch, and patch versus replicate. The total number of genera at beta (regional) scales samples were regressed against the total number of genera in each alpha (local) scale sample. Previous studies showed that this approach had spatial pseudoreplication (Srivastava 1999, Gering & Crist 2002). However, this analysis approach has still been found to be useful and appropriate when an alpha-beta relationship is compared with an alpha, beta and gamma (alpha, beta and regional) relationship (Gering & Crist 2002).

At the broad scale comparisons, there were only four observations, representing the estimate of alpha genus richness in a country vs. region comparison that provided insufficient data to be analysed using a regression approach. Therefore, in order to be able to conduct a regression analyse for comparison of geographic scales (country versus region) I compared nematode assemblages between the two macroalgae species.

Simple linear regression tests were used to determine each alpha and beta (local and regional, LR) relationship. Model validation confirmed that all LR relationship were linear and positive, therefore curvilinear models were not fitted to the data. Although local-regional regressions are typically constrained (Caley & Schluter 1997, Srivastava 1999), the relationship between alpha and beta diversity can be tested as a unconstrained relationship when it was only used for comparison with alpha, beta and gamma diversity (Gering & Crist 2002). Moreover, a constrained approach can be misleading due to incorrect assumptions about the position of the intercept (Gering & Crist 2002).

Alpha, beta and gamma (alpha and beta and regional, ABR) relationships were analysed using a hierarchical experimental design in order to test how regional diversity was partitioned into alpha and beta within a particular spatial scale (Gering & Crist 2002). The total number of taxa in the regional (gamma) diversity

sample was plotted against the mean taxa richness among local (alpha) diversity within the region. According to the additive partitioning method, gamma diversity is the sum of alpha and beta diversity (Allan 1975, Lande 1996, Gering & Crist 2002). Therefore, only alpha diversity was used in the regression test to avoid spatial pseuodreplication (Srivastava 1999). The additive partitioning method was used to convert each diversity scale into a percentage of total diversity in order to investigate the contribution of each diversity scale into total (gamma) diversity (Lande 1996). The additive partitioning of gamma diversity into alpha and beta components at four nested spatial scales were measured in order to look at the contribution of alpha and beta diversity to gamma diversity. Mean genera richness in each spatial scale was calculated in order to identify alpha diversity. Beta diversity at any scale is determined by subtracting the alpha diversity at that scale from the alpha diversity at the next highest scale (e.g. β = mean of patches–mean of replicates).

PERMANOVA tests with Monte Carlo tests were used to see at which spatial scale difference in species composition of nematode assemblages occurred. Estimates of components of variation were made in the PERMANOVA tests to identify variability of nematode composition of each spatial scale.

5.3 Results

5.3.1 General community patterns

A total of 16,462 individual nematodes were collected in both macroalgae. More individual nematodes were found in *Corallina* spp. (8532) than in *S. muticum* (7930), and more genera in *Corallina* spp. (128) than in *S. muticum* (78). Nematodes in British Isles in *Corallina* samples were more diverse and abundant than in Korea. In contrast the diversity and abundance of nematodes in *S. muticum* in Korea were higher than in the British Isles. The dominant nematode genera were different between the two macroalgae: *Enoplus* (18%), *Euchromadora* (13%), *Crenopharynx* (10%), *Cyatholaimus* (7%), *Chromadora* (5%) in *Corallina* spp. whereas *Euchromadora* sp. 1 (15%), *Pontonema* sp. 1 (13%), *Eurystomina* sp. 1 (9%), *Theristus* sp. 1 (8%), *Acanthonchus* sp. 1 (6%) in *S. muticum.*

5.3.2 Alpha and beta (local and regional, LR) relationships

All LR relationships were linear and positive in each macroalgae (Table 5.1). The slope coefficients of each alpha and beta relationship departed significantly from zero. Relationships at smaller scales were partitioned into larger scale relationships that exhibited patterns of proportional sampling. According to the slope from each regression test, the shore vs. region relationships had higher genera variability than the other alpha and beta relationships in both macroalgae. Region vs. country relationships were not significant in both habitats. This result might be due to only six observations in each comparison. The LR relationship including both macroalgae showed that genera variability in shore versus region comparisons were the highest among all spatial scale relationships.

Table 5.1 Regression statistics and significance tests for departure of slopes from zero (H_o: slope = 0) for alpha and beta (LR) relationships of nematode genus richness across four spatial scales in *Corallina* spp., *S. muticum* and including both macroalgae. Data collected in summer 2014 and 2015 from the south coast of British Isles and south and east coast of Korea using a hierarchically nested sampling design. I conducted separate regression analyses for each spatial scale; Co: Countries, Re: Regions, Sh: Shores, Pa: Patches, Res: Replicates of each macroalga.

Habitat	Spatial scale	Comparison	Y-int	Slope	R^2	d.f	F
Corallina spp.	Broadest	Re vs. Co	-84	1.5	0.07	4	0.31
		Sh vs. Re	0.06	0.72	0.59	10	14.39**
		Pa vs. Sh	1.4	0.57	0.71	34	84.5***.
	Finest	Res vs. Pa	0.71	0.55	0.58	106	147***
S. muticum	Broadest	Re vs. Co	13.2	0.45	0.65	4	7.67
		Sh vs. Re	0.03	0.88	0.23	10	3.13
		Pa vs. Sh	1.18	0.61	0.53	34	39***.
	Finest	Res vs. Pa	0.56	0.58	0.56	106	138.5***
All	Broadest	Re vs. Co	13	0.45	0.52	10	10.9**
		Sh vs. Re	1.1	0.7	0.62	22	35***
		Pa vs. Sh	2.2	0.56	0.69	70	156***.
	Finest	Res vs. Pa	0.88	0.55	0.6	214	325***

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

5.3.3 Alpha, beta and regional (gamma) relationships (ABR)

Regression analyses of the ABR relationship for each spatial scale indicated that gamma richness explained a high percentage of alpha richness (Table 5.2). All ABR relationships showed that shore versus region relationships had higher genera variability than other spatial scale comparisons (Figure 5.1). The slope coefficients of each alpha and beta relationship departed significantly from zero. The alpha, beta and gamma relationship showed that gamma richness is comprised of alpha and beta richness in varying percentages depending upon the alpha and beta relationship (Table 5.2).

The Estimates of Component Variation test using a nested PERMANOVA test indicated that the estimates of component variation among replicates was the highest across all spatial scales in both macroalgae. The Estimates of Component Variation among the shore variations in nematode assemblage composition in *Corallina* spp. were greater than at the other three spatial scales (see chapter 3). In contrast to *Corallina*, the nematode assemblage composition in *S. muticum* showed the higher estimates component variation at country level compared to the other three spatial scales.

Table 5.2 Results from the analysis of alpha–beta–gamma (ABR) relationships indicating the percentage into which gamma genus richness was partitioned into alpha and beta components on four spatial scales *Corallina* spp., *S. muticum* and including both macroalgae. The percentages of alpha and beta were determined by applying additive partitioning to the gamma nematode genus richness within an individual spatial scale. Data collected in summer 2014 and 2015 from the south coast of British Isles and south and east coast of Korea using a hierarchically nested sampling design. Separate regression analyse was conducted for each spatial scale; Co: Countries, Re: Regions, Sh: Shores, Pa: Patches, Res: Replicates of each macroalga.

Habitat	Spatial scale	Comparison	Y-int	Slope	R^2	d.f	F	ABR	
								Alpha (%)	Beta (%)
Corallia spp	Broadest	Re vs. Co						59	41
		Sh vs. Re	0.06	0.72	0.97	4	194***	73	27
		Pa vs. Sh	1.49	0.57	0.94	10	184***.	61	39
	Finest	Res vs. Pa	0.7	0.56	0.89	34	276***	59	41
S.muticum	Broadest	Re vs. Co						68	32
		Sh vs. Re	4.41	0.62	0.83	4	19.8*	73	27
		Pa vs. Sh	1.18	0.61	0.86	10	63***	65	35
	Finest	Res vs. Pa	0.56	0.58	0.83	34	158***	59	41
All	Broadest	Re vs. Co	13	0.45	0.97	2	63*	62	38
		Sh vs. Re	1.1	0.7	0.97	10	334***	73	27
		Pa vs. Sh	2.2	0.56	0.93	22	292***.	63	37
	Finest	Res vs. Pa	0.88	0.55	0.87	70	477***	60	40

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

Table 5.3 Estimates of components of variation in PERMANOVA in *Corallina* spp. and *S. muticum*. Data collected in summer 2014 and 2015 from the south coast of British Isles and south and east coast of Korea using a hierarchically nested sampling design; Co: Countries, Re: Regions, Sh: Shores, Pa: Patches, Res: Replicates of *Corallina* spp. and *S. muticum.*

Habitat	Estimates of components of variation		
	Source	Estimate	Sq.root
Corallina spp.	S (Co)	454.11	21.31
	V (Re(Co))	456.12	21.357
	V (Sh(Re(Co)))	502.08	22.407
	V (Pa(Sh(Re(Co))))	339.05	18.413
	V (Res)	970.89	31.159
S. muticum	S (Co)	835.91	28.912
	V (Re(Co))	354.84	18.837
	V (Sh(Re(Co)))	505.8	22.49
	V (Pa(Sh(Re(Co))))	295.98	17.204
	V (Res)	1147.2	33.87

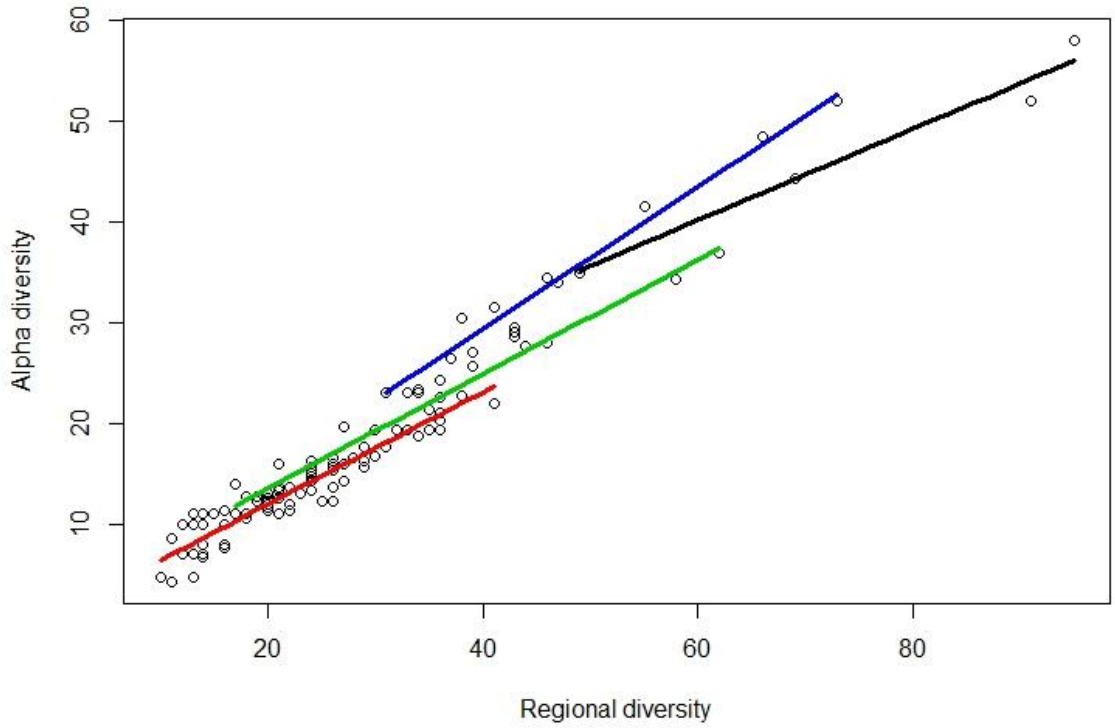


Figure 5.1 Regression plots of the relationship between alpha and regional (gamma) richness for the four spatial scales in the study. Each regression line indicate each ABR relationship; red line: replicate versus patch, green line: patch versus shore, blue line: shore versus region, black line: region versus country.

5.4 Discussion

The results of this chapter described the alpha, beta and gamma diversity relationship of nematode in two different habitats (*Corallina* spp. and *S. muticum*). All alpha and beta (local and regional, LR) regression tests in my studies were linear and positive indicating alpha species richness is dependent upon beta species richness. Non-saturating (linear) relationship have been commonly reported in many studies comparing alpha and beta relationships particularly in proportional sampling studies (Srivastava 1999). Non-saturated relationships were often found in non-interactive communities that indicate density independent fluctuations, limited disposal abilities, and random colonization in open niche space (Gering & Crist 2002). For example, Cornell (1985) found local and regional species richness of cynipine wasp communities on California oaks were linear and regulated by dispersal activity, host specificity and distribution. Moreover, ecological studies of beetles on temperate and tropical trees showed that high host selectivity, density-dependent population fluctuation and random colonization, indicated typical unsaturated patterns (Cornell 1993, Gering & Crist 2002). Nematodes are also known to have limited dispersal capability, high local colonisation ability, and large population differences at small scales (Hopper & Meyers 1967, Platt & Warwick 1980, Bongers et al. 1991). Although my study cannot explain what processes drive the unsaturated relationship between local and regional comparisons, all alpha and beta relationships were not strong enough to limited local species richness in nematode assemblages. The processes that underpin the observed linear relationships might be different in each four LR relationship.

The comparison between LR and ABR relationships indicate how alpha and beta diversity contributes to gamma (total) diversity in each four spatial scale comparisons. If only LR relationships were considered to determine alpha and beta relationships, the results might indicate that the relationships between alpha and beta species richness are caused by same ecological scenarios (Gering & Crist 2002). ABR relationship showed different combinations of alpha and beta genus richness at each spatial scale comparison (Table 5.2). Although the broad scale comparison was not significant in both macroalgae due to their small sample size, the alpha value was continually higher than the beta diversity in both macroalgae. Alpha richness varied between 59-73%, whereas beta richness accounted for 27-41% of total richness. To date, several previous studies have focused on how alpha and beta diversity change across all spatial scales (Wiens

1989, Huston 1999, Loreau 2000, Gering & Crist 2002). There are three possible scenarios; “alpha dominant system”, “beta dominant system” and “irregular dominant system” with shifted dominance in alpha and beta components across a range of spatial scales (Gering & Crist 2002). For instance, Loreau and Mouquet (1999) revealed that immigration of species can increases alpha values and decrease the contribution of beta diversity in regional diversity – an alpha dominant system. In contrast, beta richness increase when strong species interaction dominates local community, thereby decreasing alpha diversity – a beta dominant system (Loreau 2000). Gering and Crist (2002) revealed that interspecific interactions (e.g. competition, facilitation, and resource sharing) and dispersal and colonization–extinction dynamics can shift the contribution of alpha and beta species richness in different spatial scales- irregular dominant system.

In my study, the proportion of alpha richness in each ABR relationship was clearly higher than beta richness. This result is indicative of a higher species variability within habitat rather than between habitats influences regional diversity at each spatial scale. Nematodes are one of the most diverse and abundant metazoan in marine system (Platt & Warwick 1980, Platt & Warwick 1983, Holovachov & Schmidt-Rhaesa 2014). They have wide variety of feeding strategies, life history strategy and limited active dispersal ability (Wieser 1953, Bongers et al. 1991, Holovachov & Schmidt-Rhaesa 2014). These characteristics affect species and genetic composition of local patches, causing strong differentiation among nearby patches (Derycke et al. 2013). Therefore, the alpha dominant system might be realistic for nematode assemblages due to their random colonization and high species turnover (Derycke et al. 2013).

Although alpha diversity is predominant in nematode assemblages, the ecological processes that can influence the alpha and beta relationship might be different at each spatial scale and in each habitat (i.e. species competition, physical and chemical variation and food resource). In my study, a nested PERMANOVA test showed clear differences of estimates of components of variation in each scales and in each macroalgal host (Table 5.3). For example, the estimate of variation in nematode assemblage composition between shores in *Corallina* spp. was higher than between countries. In contrast the estimate of variation of nematode assemblage composition between countries was higher than between shores in *S. muticum*. Moreover, the slope of ABR relationship also showed clear differences at each spatial scale (Table 5.2 and figure 5.1). The estimated component at the finest spatial scale (replicates) was the highest in both macroalgal hosts, but

increasing diversity is strongly restricted at larger geographic scales (in shore level, within 100km in *Corallina*, and at country scale, several 1000's km in *S.muticum*). Thus the scale is variable probably depending on a variety of environmental factors and habitats (Derycke et al. 2013).

My data suggest that alpha, beta and gamma diversity of nematodes across four spatial scales in two different habitats are indicative of an alpha dominant relationship. However, the contribution of the diversity component could switch dominance over the different range of sampling scales (Gering & Crist 2002). The pattern of diversity might be related to change in ecological process either naturally or as a result of anthropogenic activities. However, only limited alpha, beta and gamma diversity studies have been reported using a hierarchical nested design. Therefore, further studies are needed to investigate to draw more realistic conclusion of alpha, beta and gamma diversity. Nematodes living in seaweeds make an ideal test system for such future work.

Summary

Returning to question 1, the contribution of alpha and beta nematode diversity to gamma diversity in each habitat (*Corallina* and *S. muticum*) were similar. The proportion of alpha richness on both macroalgae were clearly higher than beta richness. However, the percentages of alpha and beta richness were different at each spatial scale on both macroalgae. This might indicate that the ecological processes that determine the alpha and beta relationships might be different in each spatial scales.

For question 2, the nematode assemblages were the highest at the smallest scale (replicates) in both macroalgae, but the Variations of Estimate Component at each scales was different between macroalgae. This might be due to their random colonization, high species turnover, limited dispersal ability and a variety of environmental factors and habitats. It might also reflect the shorter term relationship between nematodes and *Sargassum* in Europe than in Asia (see Chapter 4).

Chapter 6: The influence of composition of algal detritus on nematode assemblages

6.1 Introduction

Species diversity has been known to be strongly related to habitat characteristics and nutrient resources (Godbold et al. 2011). Habitat heterogeneity leads to different assemblage composition and community structure (Guégan et al. 1998, Loreau et al. 2003, Levinton & Kelaher 2004, Cadotte & Fukami 2005, McClain & Barry 2010). Environmental heterogeneity leads to more complex and diverse biological assemblages (Guégan et al. 1998, Loreau et al. 2003, McClain & Barry 2010). For example, in terrestrial ecosystems, topographic complexity such as canyons or hills leads to increased diversity by providing multiple habitats and leads to geographic population differentiation (Nevo 1995, Fleishman et al. 2000). In the deep sea, submarine canyons are also an important feature as potential sinks of carbon resource that increase diversity of community (Vetter & Dayton 1999, Puig et al. 2008). Organisms that create habitat, referred as ecosystem engineer species, also modify the physical and biological features of living space that available to other organisms as a resource (Wright et al. 2002).

The intertidal area is a dynamic interface that tightly connects marine and terrestrial ecosystems (Raffaelli & Hawkins 1996, Barreiro et al. 2012). The transfer of detrital nutrient and energy passing through the coastline is a common process affecting ecosystem functioning (Polis et al. 1997, Barreiro et al. 2012). In coastal marine ecosystems, most primary production from macroalgae and seagrasses enters the food web as detritus.

High concentrations of carbon, nitrogen, phosphorus and other nutrients in the coastal waters have been identified as a major component of global biogeochemical cycles (Bauer et al. 2013). Input of macroalgae detritus is one of the major sources of carbon in coastal areas (Griffiths et al. 1983, Dugan et al. 2003). Such fluxes of nutrients and energy influence species abundance and diversity plus community structure. Drifting algae and seagrasses can alter community structure (Holmquist 1997, Norkko et al. 2000), by providing habitat

and food to invertebrates; such detritus also has a strong influence on sediment chemistry (Chown 1996, Pennings et al. 2000).

Wrack-loaded sandy beaches are a biogeochemical hot-spot compared to other coastal regions (Coupland et al. 2007). It has been found that a square metre of wrack deposited on Australian coasts has a metabolic rate three times higher than an adjacent living macroalgal habitat (Coupland et al. 2007). These high metabolic rates, are associated with highly diverse and abundant macro and meiofaunal communities (Griffiths & Stenton-Dozey 1981, Colombini et al. 2000, Jaramillo et al. 2006, Ince et al. 2007, Olabarria et al. 2007, Lastra et al. 2008, Rodil et al. 2008) that colonise freshly deposited algal material. All organic matter from detritus is decomposed and mineralised by the macrofauna, meiofauna and bacteria (Koop & Lucas 1983, Mews et al. 2006, Coupland & McDonald 2008, Lastra et al. 2008, Urban-Malinga & Burska 2009). This results in the rapid breakdown of detrital material releasing macronutrients and carbon, further affecting spatial habitat and community variability (Buchsbaum et al. 1991, Garcia-Robledo et al. 2008, Hardison et al. 2010, Chapin III et al. 2011).

Although the role of meiofauna and, particularly that of nematodes in the decay process has been well documented (Findlay & Tenore 1982, Alkemade et al. 1992a, Alkemade et al. 1992b, De Mesel et al. 2004, Moens et al. 2005), the environmental factors that determine the diversity of nematode communities are still unclear. Nematodes are the one of the most abundant taxa involved in decomposition processes (De Mesel et al. 2004). They are bacteria-grazers particularly linked to nutrient mineralisation (Valiela 2013), having been shown to influence bacterial activity and abundance both positively and negatively (Findlay & Tenore 1982). Furthermore, nematodes are a major food resource for macrofauna (Huff & Jarett 2007, Giere 2009). As a consequence, there have been a wealth of studies focussing on the importance of nematode community structure, functional traits and interactions between macrofauna, meiofauna and microbiota in decomposition processes (Freckman 1988, Alkemade et al. 1992b, De Mesel et al. 2003, Urban-Malinga et al. 2008).

Using litter-bag experiments, Urban-Malinga et al. (2008) found that nematode biodiversity and abundance patterns differed along tidal elevation gradients on wrack-loaded sandy beaches. For example, the upper shore was dominated with Rhabditidae, was recorded the lowest density along the shore whereas middle and bottom of the shore Dorylaimoidea were dominant (Urban-Malinga et al. 2008). The structure, composition and diversity of macrofauna on algal detritus-

dominated beaches are closely linked to the input, size and type of macrophyte detritus (Griffiths et al. 1983, Stenton-Dozey & Griffiths 1983, McLachlan 1985, Dugan et al. 2003, Olabarria et al. 2007). The relationship between type of macrophyte detritus and meiofauna assemblages remain unclear.

To date, research has only considered single, locally important macrophyte detritus species when investigating the importance of resource availability on meiofaunal biodiversity and assemblage structure (Valiela et al. 1985, Alkemade et al. 1993, Urban-Malinga et al. 2008). In the natural environment, however, different types of detritus co-occur, rarely occurring as individual species (Gartner & Cardon 2004, Hättenschwiler et al. 2005, Godbold et al. 2009). The impact of a single type of macrophyte detritus in associated assemblages may differ from algal mixtures. Moreover, decomposition rates vary between species of each macroalgae due to their morphological and chemical characteristics (Smith & Foreman 1984). Therefore, biodiversity of macroalgal detritus in terms of species composition and relative proportions would be expected to affect diversity and density of associated fauna. Therefore, here I focused on the effects of various combinations of three different types of macroalgae (red, Rhodophyta, brown, Phaeophyceae, and green, Chlorophyta) on nematode assemblages involved in decomposition processes. Mesh bags filled with shredded macroalgae were used to test the following specific hypotheses.

1. The nematode assemblages on the detritus would be different to the nematode assemblages in associated sediment.
2. The nematode density and diversity would be higher in algae mixtures in comparison to a single type of macroalgae.
3. Decomposition as indicated by weight loss would be different between combinations of three (red, brown and green) macroalgae and that would be correlated with nematode biodiversity and density.

6.2 Material and methods

6.2.1 Sampling site

Poole Harbour is a large natural harbour in Dorset, southern England, with an area of approximately 36 square kilometres, one of the largest natural harbours in Europe. The tides are regular, semi-diurnal of amplitude (2m). The foreshore can extend up to 1.5 km (Figure 6.1).

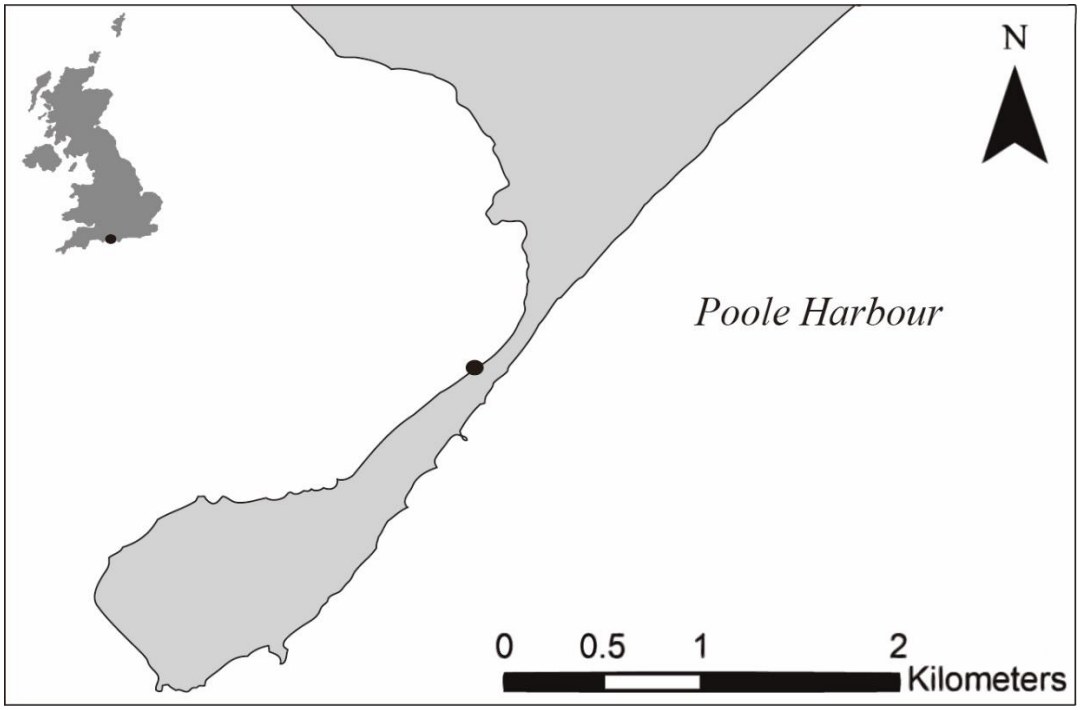


Figure 6.1 the sampling location of algal detritus experiment

6.2.2 Algal detritus experiment

Mesh bags were used that had a pore diameter of 1 mm measured approximately 8 x 12 cm (Fig 6.2). Three different types of macroalgae (red, brown and green) were selected to investigate the effect on nematode assemblages. The species *Corallina officinalis*, *Fucus vesiculosus* and *Ulva intestinalis* were freshly collected at the study site and washed to remove any organisms, then oven-dried 60 ºC for 48 hours. These species were chosen, as they are common on rocky shores in the surrounding areas.

In order to test the impacts of detrital diversity on nematode assemblages, The detrital treatments consisted of 10 combinations of macroalgae (monoculture, two and three species treatments), containing different proportions of algal material (two-species mixtures: 1 species dominant, three-species mixture: all species in equal proportion) whilst maintaining a total dry weight 9g mesh bag^{-1}.A control treatment containing no detrital material was also incorporated (Table. 6.1).

Mesh bags (6 replicates $treatment^{-1}$) were placed on the mid shore (close to natural wrack line and high water level) where the most intensive accumulation of fresh algal material was observed. Mesh bags were placed with treatments randomly interspersed in blocks parallel to shoreline 50 cm apart and fixed to the sediment surface with metal pegs (Figure.6.2). This experiment was run for one month (between 15th September~ 15th October, 2014). After one month, the mesh bags were carefully placed into separate labelled plastic bags and were transported to the laboratory.

Table 6.1 Macroalgae combination in mesh bags. Each component in each combinations has 3g; R: red algae, *Corallina officinalis*, G: green algae, *Ulva intestinalis*, and B: brown algae, *Fucus vesiculosus*.

GGG	BBB	RRR
GBB	BRR	RGG
GRR	BGG	RBB
GRB	Empty (Control)	

Figure 6.2 Macroalgal detritus bag *in situ*

6.2.3 Background sediment nematode community compositions

A sediment sample from under each of the mesh bags and away from the bag bags (at least 1 metre) were collected in order to study the nematode community associated with wrack. Three replicate samples were collected using a plastic corer, with a surface area of 10 cm^2 and down to a depth of 0-2 cm. Each core was put into separate plastic bags and transported to the laboratory for further analysis. Only three sediment samples, however, was analysed due time to constraints in order to compare with mesh bag samples. The sediment sample were treated with LudoxHS-40 to increase the efficiency of extraction of nematodes from sediment (Burgess 2001), and each sample was then sieved on 63 µm and 38 µm mesh respectively. All nematodes were preserved in 4% buffered fomaldehyde then were washed with distilled water.

6.2.4 Analysis of nematodes in the mesh bags

In the laboratory, three mesh bag replicates were analysed for morphological identification of nematodes. The content of each bag was washed with filtered tap water, decanted through 1-mm sieve to remove large fragments and sediment, then washed through 63 µm and 38 µm mesh respectively. Algal fragments remaining on the sieve were carefully collected, oven-dried at 60 ºC for 48 hour and to determine dry weight. The difference between initial and final dry weight was used as a proxy for wrack decomposition.

Nematodes retained on the sieve were preserved with 4% fomaldehyde solution. All nematode samples were picked and counted under a stereo microscope (Leica M125). Nematode samples were divided using a Folsom Plankton Splitter when nematodes were too numerous to count (McEwen et al. 1954). The first 100 nematodes were randomly chosen and mounted in anhydrous glycerine on HS slides to observe both sides of the specimens (Shirayama et al. 1993) and identified to genus or species level under a light microscope (Olympus BX53). The pictorial keys of Platt and Warwick (1983), Handbook of Zoology (Holovachov & Schmidt-Rhaesa 2014) and the NeMys Database (Deprez 2007) were used to identify the specimens.

6.2.5 Statistical analysis

Multivariate data analyses were performed using the software Primer 6.0.2 (Clarke & PRIMER 2006) with PERMANOVA addition. Univariate and bivariate analyses were made using Minitab (ver.12) and the R program to test the effect of detrital diversity (n = 11) on nematode density, genera and Maturity Index (MI). The data were checked with diagnostic graphics to ensure they fulfilled the parametric assumptions; when needed, the data were transformed and re-checked to ensure that the transformation improved their frequency distributions. The relationships between the measurements of the nematode assemblage metrics (number of species and density of nematode) and the seaweed weight loss were tested with Pearson's product moment correlation with untransformed data.

The data for abundance of each nematode genus in each replicate in combinations were standardised by their total abundance and square-root transformed. The means of the transformed abundance for each combinations were used to construct a Bray-Curtis similarity matrix; this was then subjected to nonmetric multidimensional scaling (nMDS) ordination to visualize the similarity between combination and replicates.

The samples were subjected to analysis of similarity (ANOSIM) and PERMANOVA with Monte Carlo tests to ascertain whether the species compositions of the nematode assemblages differed among combinations of algae mixture. For this and all subsequent ANOSIM, the null hypothesis, that there were no significant differences among groups, was rejected if the significance level (P) was<0.05 or 5%. The R-statistic values determined with ANOSIM for comparisons between those approaching groups that were significantly different were used to ascertain the degree to which those groups were dissimilar. R-statistic values approaching unity indicate that the compositions of the groups are very different, whereas those close to zero show that they are very similar (R 1993). When ANOSIM detected significant differences among the groups, similarity percentages (SIMPER) were used to determine the species that typified these groups and the species that distinguished each group from each of the other groups (Clarke 1993).

6.2.6 Functional Traits Analysis

Each nematode species was classified according to four different biological traits, based on their morphological and functional features. Feeding types based on the morphology of the buccal cavity were developed by Wieser (1953), who classified free-living nematodes into four feeding types such as selective deposit feeders (1A), non-selective deposit feeders (1B), epigrowth feeders (2A), and omnivore/predators (2B).

An additional functional group classification can be provided by tail shape of nematode indicative of mobility, habitat preference and lifestyle (Thistle & Sherman 1985). The diversity of tail shapes together with the features of buccal morphology have proven to be an effective tool for discriminating nematode assemblages (Thistle et al. 1995). Nematodes were classified into four tail shape groups: 1: Short/round tail type with blunt end, 2: Clavate-conico-cylindrical tail type, initially conical with an extension to the tip, 3: Conical tail type, with a pointed tip and tail length less than five body widths, 4: Long tail type, with a tail longer than five body widths (Thistle & Sherman 1985).

The life history strategies of nematodes have been described by Bongers et al. (1991), who proposed a scale (c-p score) to classify the genera of nematodes based on their ability to colonise or persist in a certain habitat. The scale ranges from extreme colonisers (c-p score=1) to extreme persisters (c-p score=5). A maturity index (MI) can also be calculated for each habitat/station based on the c-p scores of inhabiting species according to Bongers et al. (1991).

A biological traits matrix based on the above approaches was used to assess the functional structure of nematode communities at all spatial scales (Schratzberger et al. 2007). The three functional trait categories described above were used: feeding type, life history strategies and tail shape. Thirteen combined categories were measured in total. A biological traits matrix was created by assigning to each nematode taxon to each trait category. The biological trait matrix was then combined with relative species abundance to give abundance-weighted traits matrices at each spatial scale.

6.3 Results

6.3.1 Dry weight loss

Seaweed dry weight loss was significantly different in each treatment (n = 11, F = 4.106, $P < 0.001$). The seaweed weight loss in each combination varied from 6.02g to 8.88g in total 9g. The combinations that had brown algae were relatively lower weight loss than other combinations. Seaweed dry weight loss was positively correlated with nematode genera richness ($R = 0.511$, $P < 0.05$), but not between loss of dry weight of seaweed and total density of nematodes ($R = 0.119$, $P = 0.53$) (Figure 6.3 and 6.4)

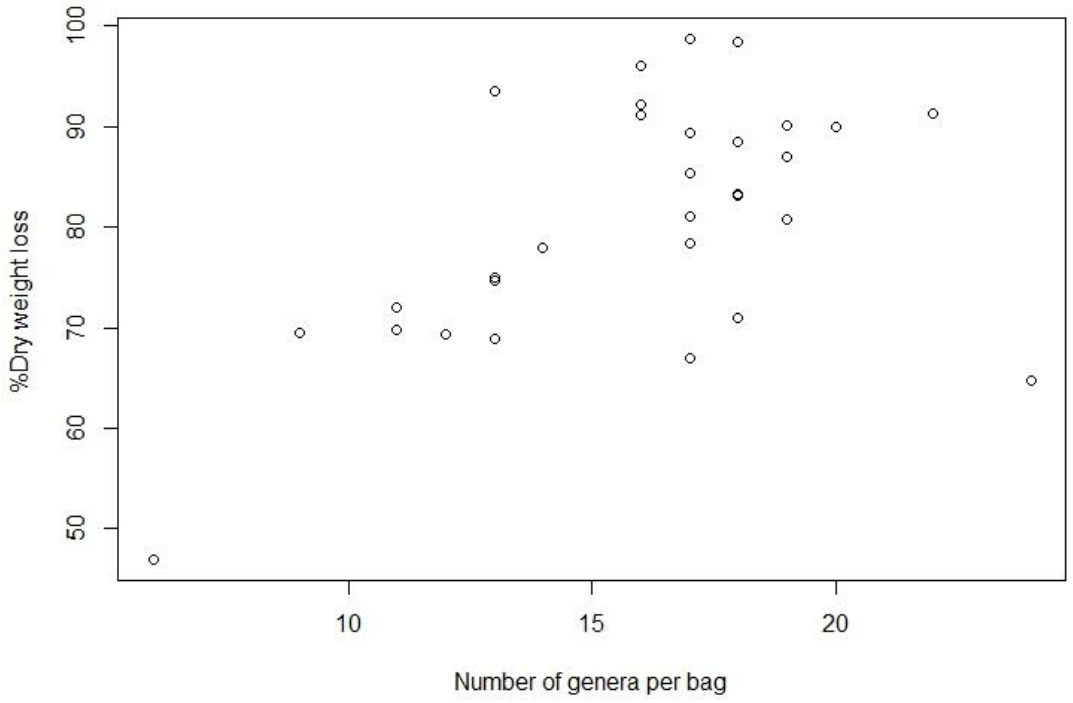


Figure 6.3 Relationship between number of nematode genera and the dry weight loss of seaweed as an indicator of decomposition in each bag ($R = 0.511$, $P < 0.05$).

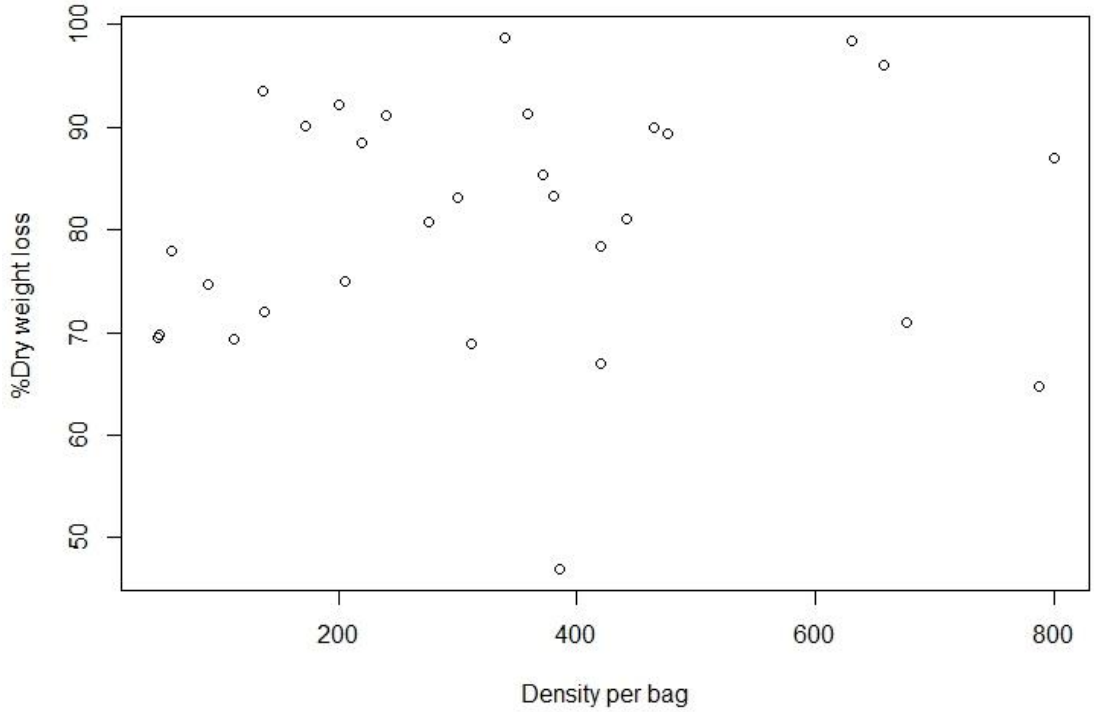


Figure 6.4 Relationship between nematode density and the dry weight loss of seaweed as an indicator of decomposition in each bag (R = 0. 119, P = 0.53).

6.3.2 Nematode density, diversity and composition

The densities of nematodes in the mesh bags were extremely variable and relatively low, ranging from 48 to 800 with an average of 325±28 individuals per bag. The density and diversity were significantly higher than those nematodes living on the sediment surrounding the mesh bags (Figure 6.5 and 6.6). Habitat were provided by mesh bags themselves, with controls including more nematodes than sediment alone. Large variance in density was shown among replicates among treatments. The number of individuals found in single algae treatments were relatively lower than mixed algae treatments (Figure 6.5). ANOVA tests showed that the density of nematode assemblages among treatments were significantly different (n = 11, F = 10.192, $P < 0.001$).

The number of nematode genera also significantly differed among treatments (n = 11, F = 4.4164, $P < 0.001$). The nematode density and diversity were the highest in the combination of red and brown algae (BRR), and the lowest in the single brown combination (BBB). The composition with three different types of algae (GRB) had relatively low density and low diversity compared to the combinations of two types of algae. The dominant nematode genera were *Euchromadora* (23%), *Oncholaimus* (9%), *Vicosia* (8%), *Crenopharynx* (8%), and *Enoplus* (8%) in all 10 treatment combinations.

PERMANOVA tests showed that the nematode assemblage composition in different mixtures of algal combinations was significantly different ($P < 0.001$). The species compositions in the sediment were similar with the nematode composition in the mesh bags although the relative abundance was different ($P < 0.05$). A cluster test and nMDS plot showed clear difference among the nematode composition in single and mixture algae combination and sediment (Figure 6.7 and 6.8).

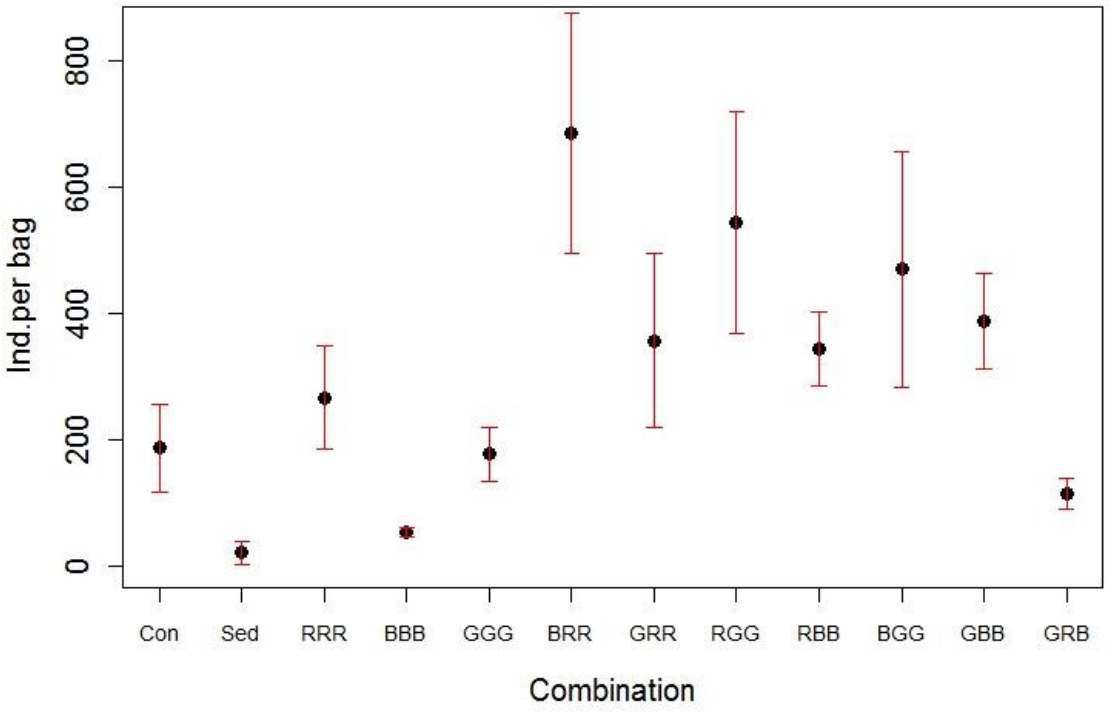


Figure 6.5 Mean abundance of the nematode assemblage with standard deviation in each algal treatment combination; Con: control, R: red algae, *Corallina officinalis*, G: green algae, *Ulva intestinalis*, B: brown algae, *Fucus vesiculosus*, and Sed: sediment.

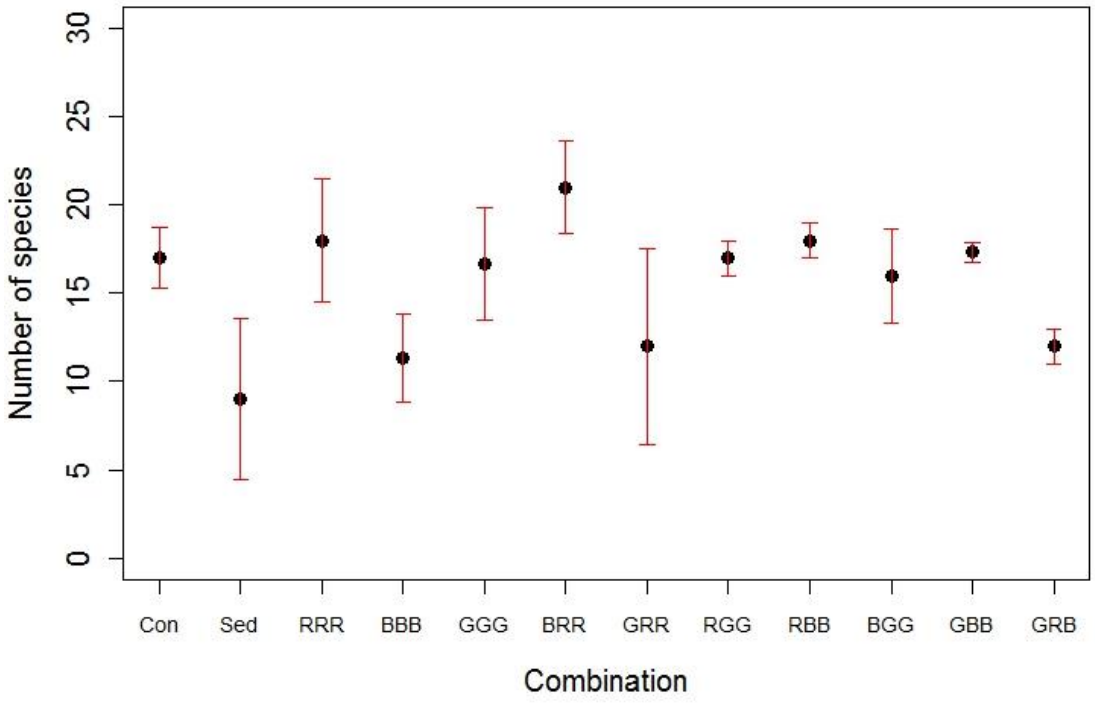


Figure 6.6 Taxa richness (Mean number of nematode genera) with standard deviation in each combination; Con: control, R: red algae, *Corallina officinalis*, G: green algae, *Ulva intestinalis*, B: brown algae, *Fucus vesiculosus*, and Sed: sediment.

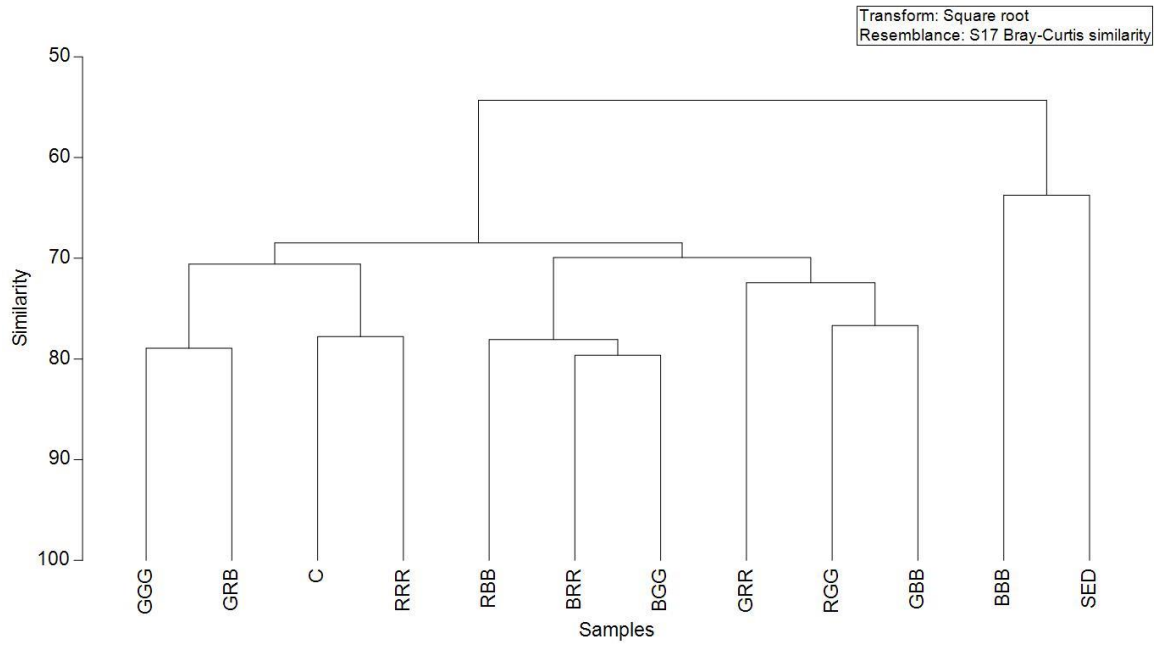


Figure 6.7 Cluster analysis of nematode assemblages based on species abundance data; C: control, R: red algae, *Corallina officinalis*, G: green algae, *Ulva intestinalis*, B: brown algae, *Fucus vesiculosus*, and Sed: sediment.

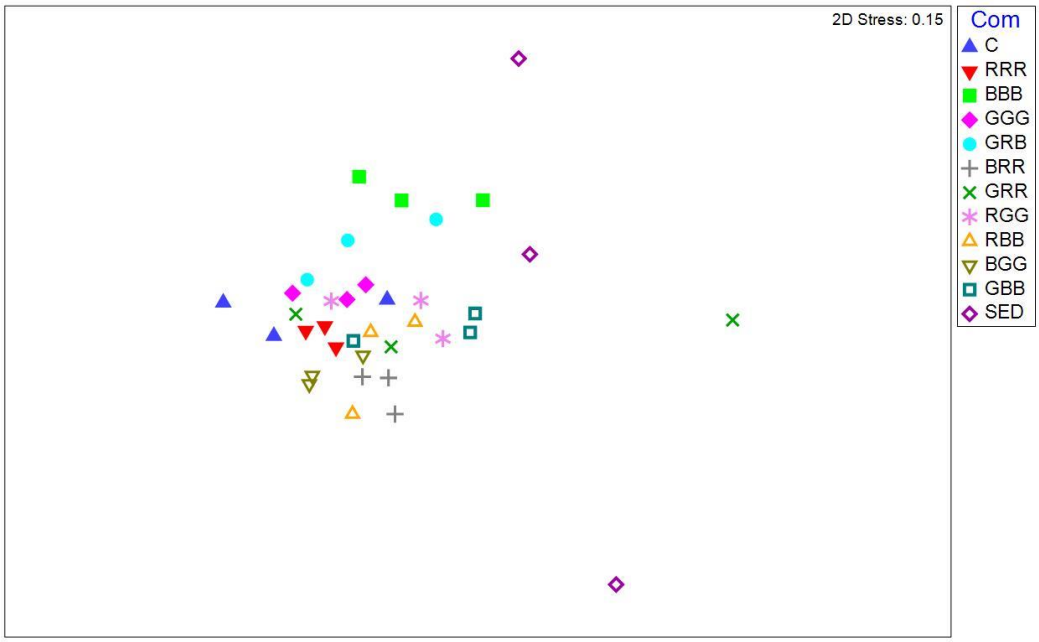


Figure 6.8 Non-parametric multi-dimensional scaling (nMDS) ordination based on the assemblage of nematode species in each bag; C: control, R: red algae, *Corallina officinalis*, G: green algae, *Ulva intestinalis*, B: brown algae, *Fucus vesiculosus*, and Sed: sediment.

6.3.3 Functional traits

PERMANOVA test showed that the feeding type, life strategy, tail shape and biological traits were significantly different among combinations ($P < 0.001$). nMDS plot of each functional traits also indicated clear differences among the nematode assemblages in single, mixture algae combinations and sediments (Fig 6.8). Epigrowth feeders were the most dominant feeding group in the mesh bags. The combination including brown algae had relatively more predatory nematodes than other combinations. The dominant feeding type of nematode in the sediment (epigrowth feeders) was the same as in the nematodes in the mesh bags. The life strategy was similar among combinations indicating as c-p value 3 that has moderate length of life and reproductive effort. The most common functional type was the epigrowth feeder/ c-p value 3 with conical tail following by the omnivore/predators/ c-p value 4 with clavate-conico-cylindrical tail, non-selective deposit feeder / c-p value 2 with clavate-conic-ocylindrical tail.

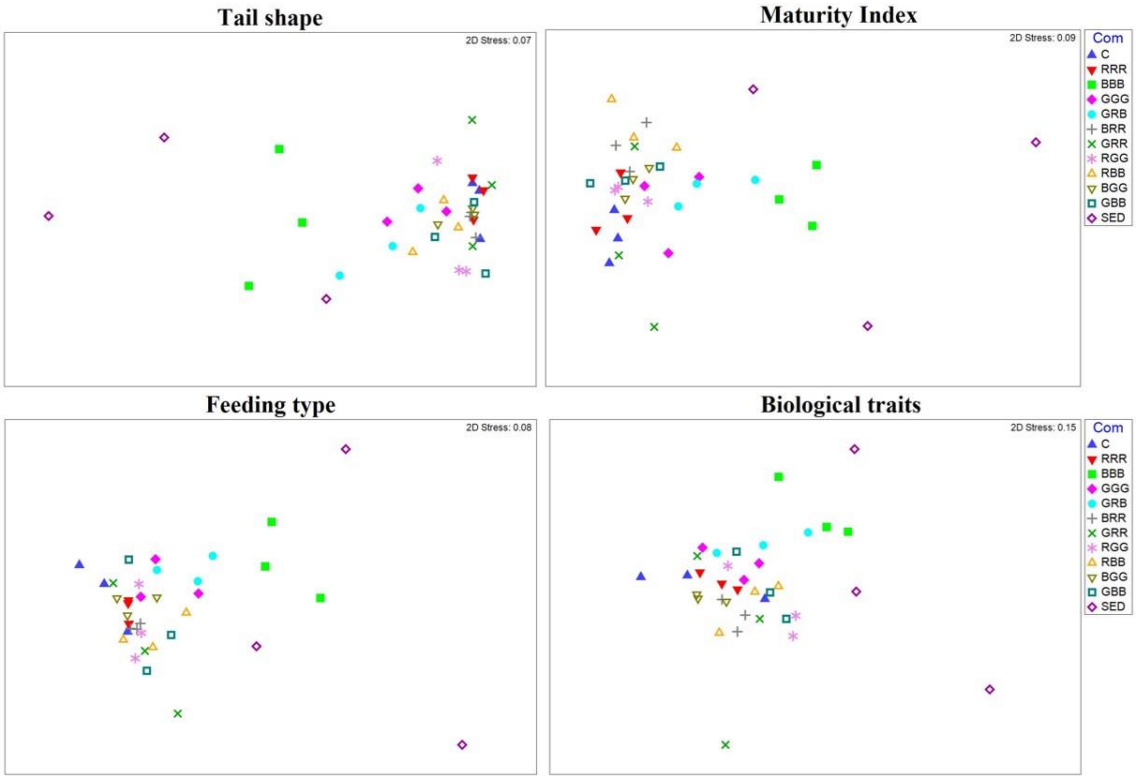


Figure 6.9 Non-parametric multi-dimensional scaling (nMDS) ordination based on the nematode tail shape, maturity index (life strategies), feeding type and biological traits in each bag; C: control, R: red algae, *Corallina officinalis*, G: green algae, *Ulva intestinalis*, B: brown algae, *Fucus vesiculosus*, and Sed: sediment.

6.4 Discussion

My study showed the diversity of algal detritus in a decomposition environment (i.e., two types of algae) leads to increased density and diversity of nematode assemblages, the exception being the bags containing three types of algae in combination. The lower numbers and diversity in the three combination of algae was not the expected result. The decomposition rates of seaweed differed in each combination, and weight loss of seaweed was positively correlated with nematode diversity. The importance of nematode assemblages in decomposition process has been well-documented (Gerlach 1978, Findlay & Tenore 1982, Alkemade et al. 1992a, Alkemade et al. 1992b, De Mesel et al. 2003, Urban-Malinga et al. 2008). Nematodes enhance mineralization of organic matter due to their bioturbation which improves vertical flux of oxygen and nutrient (Alkemade et al. 1992a, Alkemade et al. 1992b). Nematodes also affect the activity and composition of macrofauna, and control bacteria communities as predators (De Mesel et al. 2004, Moens et al. 2005).

Most previous studies have focused on the relationship between decomposition rate and nematode density (Findlay & Tenore 1982, Alkemade et al. 1992b, Alkemade et al. 1993), while only a few studies have investigated nematode species diversity with their effect on the decomposition process (De Mesel et al. 2006, Urban-Malinga et al. 2008). These studies showed the decomposition rates were positively correlated with nematode diversity, but the results did not consider that how the type of macroalgae might affect nematode biodiversity. My study showed relatively more nematode individuals in the two species of algae combinations in comparison to the algal monoculture treatments, although there were large density variations between replicates. This result indicates diversity of detritus components increases food resources for nematode assemblages. The different morphologies of detrital seaweed may have also influenced habitat provision.

Euchromadora was the most dominant genus across all combinations although the diversity and the composition of nematode assemblages were significantly different. Previous studies reported different dominant nematode species in decomposition environment. For example, *Rhabditis* and *Halomonhystera* were dominant in *Fucus distichus* from Hornsund while *Pellioditis* and *Geomonhystera* in *Fucus* the Westerschelde Estuary (Derycke et al. 2007, Urban-Malinga & Burska 2009). However, most dominant nematode species from wrack were

bacterivorous nematode that are well known opportunistic colonisers of algal deposits in littoral environments (Inglis & Coles 1961, Bongers et al. 1991, Alkemade et al. 1993). My result also showed that epigrowth feeders were the most dominant group in all combinations.

Nevertheless, the compositions of nematode species were significantly different between the combinations of algae mixture (Table A2.1). nMDS plots show clear differences between the combinations including brown algae and others. SIMPER tests showed that the combinations including brown algae have relatively more predatory nematodes than other combinations. Moreover, the single species of detritus has relatively low density and diversity when compared to mixed two species combinations of detritus. Therefore, the diversity of detritus might affect growth and diversity of nematode assemblages and associated fauna.

The presence of marine-derived detritus on the shore not only increases nutrients in intertidal ecosystems but also affects the abundance of the intertidal meiofaunal community (Urban-Malinga & Burska 2009). Higher numbers and strong dominance of nematodes were observed on wrack deposit on New Zealand beach (Inglis 1989), on the Antarctic coast (Alkemade & Van Rijswijk 1993), on Netherlands estuary (Derycke et al. 2007) and on Arctic beaches (Urban-Malinga & Burska 2009). The component of wrack associated macrofauna were mainly terrestrial species indicating as oligochaetes, acarids and collembola (Inglis 1989, Colombini et al. 2000, Orr et al. 2005, Coupland & McDonald 2008, Brown & McLachlan 2010). The component of wrack associated meiofauna were similar with meiofauna in sediment but have relatively low density (Urban-Malinga & Burska 2009). My study also showed that nematode assemblages in background sediment had relatively lower abundance and diversity compare to the nematode assemblage in the mesh bags.

Breakdown rates of macroalgae in previous mesh bag experiments were varied with macroalgae species, temperature and other environmental factors (Valiela et al. 1985, Inglis 1989, Alkemade & Van Rijswijk 1993, Jedrzejczak 2002). The differential mass loss of different macroalgae are common phenomenon (Mews et al. 2006). In my study, the combination containing brown algae had relatively less weight loss compared to others. *Fucoids* are heavily defended by phlorotannins (Wikström et al. 2006) which may inhibit bacteria and provide less food for nematodes. Moreover, the nematode diversity was positively correlated with the weight loss of wrack. The breakdown rate vary depending on macroalgal species and other environmental stress (Urban-Malinga & Burska 2009). Therefore,

diversity and identity of wrack species on the beach might affect nematode composition and decay processes.

My study only focused on how wrack diversity affects nematode assemblages and diversity and associated nematode fauna. The time scale of my experiment was limited to determine decomposition pathways involving nematode assemblages. Therefore, serial sampling during the time course of an experiment is needed to understand the relationship between nematode assemblages and break down rate (i.e. collect 3 days, 7 days, 2 weeks period). Moreover, nematode assemblage and bacterial community interaction in decomposition process are also required to investigate whether nematode assemblages boost breakdown rates or not.

Summary

Returning to the hypotheses, hypothesis 1 was rejected. The nematode compositions in the background sediment were similar to the nematode composition in the mesh bags. However, the nematode density and diversity in associated sediment were significantly lower than in mesh bags. This result showed that wrack input is major food resource on the beach and shapes the meiofauna community.

Hypothesis 2 was accepted. The diversity and density of combination algal mesh bags were higher than a single type of macroalgae mesh bags. This might indicate that the combination of two algae mixtures offered more favourable environments than the single species bags. The three species combination did not, however, show greater density nor diversity of nematodes.

Hypothesis 3 was accepted, the combinations including brown algae were relatively smaller weight loss than others, positive relationship was found between nematode diversity and wrack weight loss. Therefore, break down rate depending on the specific identity of macroalgae. Different species might offer different nutrient input and that encourage nematode growth and diversity. Brown algae are packed with tannins that might inhibit bacteria and thus provide less food for nematode.

Chapter 7: General Discussion

In this chapter, I summarise the main findings of my thesis and put them into a broader context. I then briefly outline the advantages of using nematode assemblages as ecological models before considering the limitations of the study and making suggestions for further studies.

7.1 Summary of main findings

The broad scale studies of nematode assemblages living in *Corallina* and *Sargassum muticum* on the south and east coast of Korea (North-East Asia, Indo-pacific biogeographic region) compared with the south coast of the British Isles (Northern Europe, North-East Atlantic biogeographical region) showed clear differences at all spatial scales.

In *Corallina*, the density and diversity of nematodes in the British Isles were significantly higher than in Korea. The regional variations in species composition along the English Channel coast were greater than in the south coast of Korea leading to an increase in total diversity of nematode assemblage composition in the British Isles. The regional variation of nematode assemblages among regions might be affected by meso-scale environmental factors such as the oceanic nature of water (Dugan et al. 2003), sea water temperature (Kim et al. 2013) and tidal range (Urban-Malinga et al. 2008). There may be a sharper environmental gradient along the English Channel coast, which has long been known to be a biogeographic boundary zone for macroflora and fauna (Crisp & Southward 1958, Southward et al. 1995, Herbert et al. 2009).

The functional traits comparison in *Corallina* spp. showed relatively more deposit feeding nematodes in Korea than in British Isles. This probably reflects the characteristics of the shores in Korea. *Corallina* turves collected from southwest coast of Korea had more sediment trapped than all other shores. The amount of sediment trapped by macroalgae would be expected to lead to an increase in abundance of deposit feeders such as *Crenopharynx* and *Phanoderma* (Holovachov & Schmidt-Rhaesa 2014). Therefore, the characteristics of shores and host plants are probably determining the composition of functional traits of nematode assemblages (Warwick 1977, Bell & Coen 1982, Heip et al. 1985, Gee & Warwick 1994a, De Oliveira et al. 2016).

In contrast, the density, diversity and species composition of nematodes from *S. muticum* in its home range in Korea were higher than in the British Isles in *S. muticum*. There were several common nematode genera in both countries. Nematode species diversity per genus was also higher in samples collected in Korea (see chapter 4). This reduction is seen in other invasive species, for example the red turpentine beetle, *Dendroctonus valens,* showed less genetic diversity in the invasive population in China than in its native region of North America (Cai et al. 2008). My results might also reflect the different evolutionary history and processes between the two oceans. *S. muticum* is an invasive species in the British Isles and a native species in Korea. The longer association over evolutionary time of nematodes with *S. muticum* may lead to greater niche differentiation and speciation in seaweed dwelling nematodes. The higher number of species of nematodes in Korea could also lead to a greater contribution to ecosystem functioning (Ieno et al. 2006, Godbold et al. 2011).

The functional traits of nematode assemblages were also significantly different between countries. Epigrowth feeders were dominant in the British Isles; in contrast predator/omnivores were the most abundant in Korea. This might indicate that the role of nematodes in *S. muticum* may differ between the biogeographic realms. Although the major feeding types of nematodes were different between countries, some cosmopolitan nematode genera were found. These might possibly have "hitchhiked" with *S. muticum* (i.e. species in the genera *Euchromadora*, *Enoplus* and *Eurystomina*). The most likely species that could have come to Europe associated with *S. muticum* was *Enoplus* sp. 1. This predatory species has relatively large body size and was dominant in Korea. Moreover, only 53 species were identified, facilitating comparison between Korea and the British Isles.

Several cosmopolitan nematode genera were found in both species of macroalgae (genus *Euchromadora*, *Oncholaimus* and *Eurystomina*). These genera belong to the families Chromadoridae and Oncholaimidae which are commonly found in macroalgae (Hillebrand 2004). This might indicate that nematodes living in seaweed have less host selectivity. The pattern of dominant nematode species, however, differed between the macroalgae (more predatory species in *S. muticum* and relatively more deposit feeder in *Corallina*). The dominant group of nematodes living in most seaweeds are epigrowth feeders (i.e. family Chromadoridae); these feed on epiphytes or detritus from the host plant (Hopper & Meyers 1967, Heip et al. 1985, Holovachov & Schmidt-Rhaesa 2014). In my

observations, however, the most dominant nematode group varied among shores within regions. This suggests that nematode compositions were highly affected by the surrounding environment (Warwick 1977, Heip et al. 1985, Holovachov & Schmidt-Rhaesa 2014).

Alpha, beta and gamma diversity of nematodes across all spatial scales in the two different macroalgae showed clear alpha-dominant relationships. This is different to work on other species and systems: such as the irregular dominant relationship in beetles (Gering & Crist 2002). An alpha-dominant relationship indicates that nematode species richness within habitat contributed more than nematode richness between habitats to the regional nematode species pool. High species variability of nematode at small scales indicating patchiness, random colonization and high species turn-over might lead to increased nematode species richness within habitat (Derycke et al. 2013). Increasing alpha species richness leads to increased regional species richness, but increasing diversity is strongly restricted at larger geographic scales (see chapter 5), perhaps constrained by the species pool present in the area due to phylogeographic processes. There are also various other *Sargassum* species in Asia that might provide sources of colonization of nematode (Koh et al. 1993, Phillips 1995); only *Sargassum muticum* is present in the British Isles.

The density and diversity of nematode assemblages in the mixed algal detritus (two species mixtures) were greater than nematode assemblages in single types of algal detritus. The species composition was also different. Combinations of algal detritus clearly offer more favourable environments than single species. The bags with all three species surprisingly did not show greater diversity and density of nematodes. The density and diversity of nematode were positively correlated with dry seaweed weight loss. This suggests that nematode assemblages are directly or indirectly related with the decomposition pathway of algal detritus (Freckman 1988, Alkemade et al. 1993, De Mesel et al. 2004, De Mesel et al. 2006). More species could drive decomposition processes as predicted by theory on biodiversity-ecosystem functioning relationships. The alternative hypothesis is that more diverse nematodes colonize faster rotting seaweed (Findlay & Tenore 1982, Valiela 2013).

7.2 The advantages of using nematode assemblage as ecological models

My work showed that nematodes can be good model species for ecological study. Nematodes are the most numerous and diverse metazoans in marine systems (Platt & Warwick 1983). They are ubiquitously distributed with high diversity ranging from very tolerant species to species sensitive to disturbance (Platt & Warwick 1983, Hillebrand 2004). Nematodes are highly habitat-specific and respond quickly when changes occur. In my experiment, the nematode assemblages clearly colonized mesh bags within one month. Moreover, they are present in every habitat at high abundance which enables setting up ecological experiments on broad scales. Although the taxonomic identification of nematodes is still difficult, my results show that classification based on functional traits of nematode (e.g. feeding type, life strategy) were significantly correlated with species abundance data (Schratzberger et al. 2007, Armenteros et al. 2009). This approach can be a useful tool to evaluate ecosystem processes, properties and diversity loss in ecosystems (Piot et al. 2014).

7.3 Limitations of my work and suggestions for further studies

Nematodes living in two different macroalgae were studied using a fully nested design. However, nematodes were only identified to genus level due to lack of taxonomic nematode studies in Korea. Therefore, morphological studies of nematodes are needed to enable accurate broad scale comparisons. Metagenomics, as has been used in sediment meiofauna (Creer et al. 2010), whilst expensive and beyond the scope of my study could be used to get better insights into diversity using Operational Taxonomic Units (OTUS). OTUs still need to be matched to morphologically defined species, so whilst a good start they are not the complete solution (Bhadury et al. 2006). Moreover, there were several common nematode genera in both macroalgae in both countries in the genera *Euchromadora*, *Enoplus* and *Eurystomina*; these nematode species were also commonly found in other macroalgae (see Chapter 3). Therefore, further molecular studies are required to understand whether those species, particularly

Enoplus sp. 1 and genus *Eurystomina*, may have invaded Europe with *S. muticum* from Asia,

The nested design was compromised by difficulties finding enough *S. muticum* in the central regions on both coasts. This could have been overcome by greater exploration of suitable study sites at the time.

In the alpha, beta and gamma study, I found the highest species variability of nematodes at small scales (replicates versus patches) but observed irregular species variability of nematodes along the range of spatial scales. Therefore, further studies are needed to investigate how alpha and beta diversity contribute to regional diversity over different ranges of spatial scale in order to examine patterns of nematode diversity. More broad scale comparisons are needed in order to understand how alpha and beta diversity relationship is restricted at larger geographical scales. Nematodes are present in almost every habitat and are easy to sample making nematodes a good ecological model for this kind of study.

In the experiment on decomposition processes, I found nematode assemblages were either directly driving or indirectly related to the decomposition processes. Therefore, further experimental study should be considered for the comparison between the decomposition processes with nematode assemblages present and without nematode assemblages. A pilot experiment that was not written up for this thesis used nematocides to knock out the nematodes. This approach was promising; with proper controls for the domestic pet tablets (blank chalk tablets without the nematocide – which proved difficult to source) this might be a good way forward to test the role of nematodes in decomposition processes in coastal ecosystems.

7.4 Concluding remarks

Overall my thesis demonstrated the high diversity of nematodes inhabiting seaweeds and the role of detrital composition in changing nematode assemblages. The high diversity of nematodes clearly plays an important role in ecosystem functioning in coastal areas. Therefore, studies of different-size class organisms such as nematodes are essential to fully understand ecosystem functioning and ecological impact of biodiversity change.

Appendices

Appendix 1

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level listing the main three discriminating species, their average abundance in each patch (Av. Abund), average of dissimilarity (Av. Diss), standard deviation of dissimilarity (Diss/SD), contribution (Contrib%), accumulation (Cum%) and average dissimilarity (AD); BW1: Looe, BW2: Heybrook Bay, BC1: Portland Bill, BC2: Swanage, BE1: Brighton, BE2: Beachy Head, KW1: west-Wando, KW2: east-Wando, KC1: Yeosu, KC2: Namhae, KE1: Gueje, KE2: Busan.

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
AD = 73.02						
BW1 P1 & P2	Group 1	Group 2				
Euchromadora	43.67	4.67	19.14	1.37	26.21	26.21
Sabatieria	0	14	7.08	1.71	9.69	35.9
Metalinhomoeus	0.33	10.67	5.36	1.49	7.34	43.24
AD = 79.57						
BW1 P1 & P3	Group 1	Group 3				
Euchromadora	43.67	4	25.64	1.27	32.22	32.22
Cyatholaimus	8	9	6.68	2.97	8.39	40.62
Desmodora	7	0.33	5.69	0.68	7.15	47.77
AD = 60.05						
BW1 P2 & P3	Group 2	Group 3				
Cyatholaimus	16.67	9	9.61	2.18	16.01	16.01
Sabatieria	14	2	8.3	1.4	13.83	29.83
Metalinhomoeus	10.67	2.33	6.52	1.12	10.86	40.69
AD = 68.43						

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
BW2 P1 & P2	Group 1	Group 2				
Euchromadora	28.33	2.33	13.71	2.63	20.03	20.03
Pontonema	2	15.67	7.2	1.62	10.52	30.55
Enoplus	18.67	9.67	6.7	1.65	9.78	40.33
AD = 55.01						
BW2 P1 & P3	Group 1	Group 3				
Euchromadora	28.33	10.67	8.6	1.62	15.62	15.62
Enoplus	18.67	8	6.3	1.57	11.46	27.08
Cyatholaimus	7	15	3.85	1.74	7	34.08
AD = 53.81						
BW2 P2 & P3	Group 2	Group 3				
Euchromadora	2.33	10.67	4.41	1.3	8.2	8.2
Pontonema	15.67	8.67	4.32	1.04	8.03	16.24
Enoplus	9.67	8	3.8	1.28	7.06	23.3
AD = 69.72						
BC1 P1 & P2	Group 1	Group 2				
Enoplus	13	59.67	35.24	2.04	50.55	50.55
Dolicholaimus	0	8	5.3	0.81	7.61	58.16
Spilophorella	5.67	1.33	3.15	1.66	4.52	62.68
AD = 71.01						
BC1 P1 & P3	Group 1	Group 3				

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Enoplus	13	35	18.26	1.47	25.72	25.72
Monoposthia	0.33	21.67	14.55	3.19	20.49	46.21
Cyatholaimus	3	11.67	5.61	2.54	7.9	54.11
AD = 53.53						
BC1 P2 & P3	Group 2	Group 3				
Enoplus	59.67	35	11.68	2.21	25.46	25.46
Monoposthia	3.33	21.67	8.84	3.62	19.25	44.71
Cyatholaimus	2.33	11.67	4.5	2.72	9.81	54.52
AD = 57.41						
BC2 P1 & P2	Group 1	Group 2				
Enoplus	68	28	19.39	4.45	33.78	33.78
Cyatholaimus	1.67	27.33	12.35	8.02	21.52	55.3
Metacyatholaimus	2.33	13	5.06	1.88	8.82	64.12
AD = 25.89						
BC2 P1 & P3	Group 1	Group 3				
Cyatholaimus	1.67	13	5.3	4	20.45	20.45
Enoplus	68	71.33	5.29	2.17	20.44	40.89
Crenopharynx	14	15.33	5.2	1.59	20.08	60.97
AD = 53.53						
BC2 P2 & P3	Group 2	Group 3				
Enoplus	28	71.33	20.7	2.99	38.66	38.66

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Cyatholaimus	27.33	13	6.81	3.41	12.73	51.39
Crenopharynx	4.67	15.33	6.62	1.32	12.36	63.75
AD = 49.63						
BE1 P1 & P2	Group 1	Group 2				
Chromadora	29.33	52.67	18.35	1.15	36.98	36.98
Retrotheristus	4.33	13.33	5.59	1.49	11.26	48.25
Desmoscolex	5	0.67	2.44	2.17	4.92	53.16
AD = 49.28						
BE1 P1 & P3	Group 1	Group 3				
Chromadora	29.33	25	13.21	1.66	26.8	26.8
Euchromadora	9.67	15.33	6.1	1.02	12.38	39.18
Cyatholaimus	4	8.33	3.49	1.04	7.09	46.27
AD = 47.34						
BE1 P2 & P3	Group 2	Group 3				
Chromadora	52.67	25	16.37	1.45	34.59	34.59
Euchromadora	10.33	15.33	5.25	1.21	11.1	45.69
Retrotheristus	13.33	4.33	4.73	2.6	9.99	55.68
AD = 66.36						
BE2 P1 & P2	Group 1	Group 2				
Chromadora	35.67	1.67	19.6	0.89	29.54	29.54
Enoplus	4	12.67	7	1.03	10.54	40.08

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Euchromadora	14.33	17.67	5.58	1.35	8.4	48.49
AD = 62.75						
BE2 P1 & P3	Group 1	Group 3				
Chromadora	35.67	3	22.13	0.92	35.27	35.27
Euchromadora	14.33	11	6.3	1.19	10.04	45.3
Enoplus	4	6	3.61	2.54	5.76	51.06
AD = 53.96						
BE2 P2 & P3	Group 2	Group 3				
Enoplus	12.67	6	6.98	1	12.94	12.94
Euchromadora	17.67	11	6.95	1.37	12.88	25.82
Oncholaimus	10.33	4.67	5.52	1.73	10.23	36.05
AD = 67.33						
KW1 P1 & P2	Group 1	Group 2				
Crenopharynx	33	25.67	23.77	1.74	35.31	35.31
Phanoderma segmentum	2.33	9.33	6.43	1.07	9.55	44.86
Eurystomina	8	0.67	5.65	2.37	8.39	53.25
AD = 50.87						
KW1 P1 & P3	Group 1	Group 3				
Phanoderma segmentum	2.33	23.33	13.22	1.55	25.99	25.99
Crenopharynx	33	47.33	12.64	1.21	24.84	50.83

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Calyptonema	7.67	2.67	3.49	1.02	6.87	57.7
AD = 59.50						
KW1 P2 & P3	Group 2	Group 3				
Crenopharynx	25.67	47.33	25.99	1.7	43.68	43.68
Phanoderma segmentum	9.33	23.33	13.43	1.29	22.56	66.25
Daptonema	0	4	2.63	1.03	4.42	70.66
AD = 66.36						
KW2 P1 & P2	Group 1	Group 2				
Metalinhomoeus	16.33	0	15.67	0.66	23.61	23.61
Crenopharynx	18	7.33	14.88	1.36	22.42	46.03
Phanoderma	7.67	3.33	5.87	0.78	8.84	54.87
AD = 61.47						
KW2 P1 & P3	Group 1	Group 3				
Metalinhomoeus	16.33	0.67	14.57	0.68	23.7	23.7
Crenopharynx	18	15	9.85	1.73	16.03	39.73
Phanoderma	7.67	1.33	7	0.97	11.38	51.11
AD = 46.02						
KW2 P2 & P3	Group 2	Group 3				
Crenopharynx	7.33	15	15.3	1.6	33.24	33.24
Metacomesoma	1	4	5.68	1.65	12.34	45.57
Enoplus	6	8.67	3.72	1.1	8.07	53.65

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
AD = 54.28						
KC1 P1 & P2	Group 1	Group 2				
Enoplus	2	18.33	11.04	1.27	20.34	20.34
Metalinhomoeus	13.67	6.33	8.05	1.21	14.82	35.16
Phanoderma	2.67	11.67	6.77	1.49	12.47	47.63
AD= 66.10						
KC1 P1 & P3	Group 1	Group 3				
Euchromadora	6	50.67	25.79	5.12	39.01	39.01
Enoplus	2	32.33	17.35	5.33	26.25	65.26
Metalinhomoeus	13.67	0	7.57	2.66	11.44	76.71
AD = 62.81						
KC1 P2 & P3	Group 2	Group 3				
Euchromadora	1	50.67	27.48	4.34	43.75	43.75
Enoplus	18.33	32.33	10.24	1.21	16.31	60.06
Phanoderma	11.67	1.67	4.82	1.46	7.68	67.74
AD = 58.52						
KC2 P1 & P2	Group 1	Group 2				
Enoplus	8	0	7.86	2.08	13.44	13.44
Chromaspirina	10.33	4.33	7.05	1.02	12.05	25.49
Cyatholaimus	9	2.33	6.87	1.26	11.75	37.24
AD = 62.36						

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
KC2 P1 & P3	Group 1	Group 3				
Chromaspirina	10.33	0.33	12.61	1.74	20.22	20.22
Cyatholaimus	9	0.33	10.25	1.8	16.43	36.66
Enoplus	8	0.67	8.79	1.96	14.09	50.75
AD = 58.76						
KC2 P2 & P3	Group 2	Group 3				
Crenopharynx	8	7	8.5	1.44	14.46	14.46
Phanoderma	8.33	4.67	5.79	1.29	9.86	24.31
Euchromadora	4	0.67	5.75	1.06	9.79	34.1
AD = 49.71						
KE1 P1 & P2	Group 1	Group 2				
Euchromadora	31.33	53	14.32	1.48	28.8	28.8
Enoplus	15.33	17	9.05	0.97	18.2	47
Metalinhomoeus	17	1	8.48	0.72	17.07	64.07
AD = 67.65						
KE1 P1 & P3	Group 1	Group 3				
Euchromadora	31.33	17.33	12.24	1.64	18.09	18.09
Metalinhomoeus	17	5	9.89	0.95	14.61	32.71
Phanoderma	3	15.67	8.08	0.83	11.95	44.66
AD = 60.98						
KE1 P2 & P3	Group 2	Group 3				

Table A1. 1 Result of one way SIMPER analysis of nematode abundance data in patches level

Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Euchromadora	53	17.33	19.44	3.71	31.88	31.88
Enoplus	17	0.33	8.38	0.92	13.74	45.62
Phanoderma	3	15.67	7.99	0.86	13.1	58.72
AD = 57.97						
KE2 P1 & P2	Group 1	Group 2				
Metalinhomoeus	16.67	0	9.77	1.13	16.85	16.85
Eurystomina	19.67	14.67	5.03	1.17	8.67	25.52
Phanoderma segmentum	5	8.67	4.17	1.29	7.19	32.71
AD = 60.98						
KE2 P1 & P3	Group 1	Group 3				
Metalinhomoeus	16.67	12.33	9.11	1.17	14.94	14.94
Euchromadora	2.33	14.67	6.85	1.07	11.23	26.17
Eurystomina	19.67	9	6.35	1.23	10.41	36.58
AD = 60.08						
KE2 P2 & P3	Group 2	Group 3				
Euchromadora	4	14.67	7.99	1.16	13.31	13.31
Metalinhomoeus	0	12.33	7.29	0.75	12.13	25.44
Eurystomina	14.67	9	4.36	1.38	7.26	32.7

Appendix 2

Table A2. 1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison listing the main three discriminating species, the average abundance in each shore (Av. Abund), average of dissimilarity (Av. Diss), standard deviation of dissimilarity (Diss/SD), contribution (Contrib%), accumulation (Cum%) and average dissimilarity (AD); group 1: control, group 2: RRR, group 3: BBB, group 4: GGG, group 5: GRB, group 6: BRR, group 7: GRR, group 8: RGG, group 9: RBB, group 10: BGG, group 11: GBB, group 12: sediment, R: red algae, *Corallina officinalis*, G: green algae, *Ulva intestinalis*, and B: brown algae, *Fucus vesiculosus*.

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Groups 1 & 2						
AD = 33.15	Group 1	Group 2				
Adoncholaimus	2.27	2.3	2.68	1.65	8.1	8.1
Metachromadora	1.69	0.33	1.88	2.14	5.66	13.76
Oncholaimus	3.65	3.53	1.84	1.79	5.55	19.31
Groups 1 & 3						
AD = 55.70	Group 1	Group 3				
Euchromadora	5.49	2	6.84	3.64	12.28	12.28
Oncholaimus	3.65	1.28	4.76	1.49	8.55	20.82
Crenopharynx	2.98	1.08	3.65	1.78	6.55	27.38
Groups 2 & 3						
AD = 51.70	Group 2	Group 3				
Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Euchromadora	6.02	2	7.77	6.86	15.03	15.03
Oncholaimus	3.53	1.28	4.39	3.48	8.49	23.52

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Crenopharynx	2.94	1.08	3.58	1.83	6.93	30.45
Groups 1 & 4						
AD= 33.20	Group 1	Group 4				
Adoncholaimus	2.27	1.61	2.64	1.43	7.95	7.95
Oncholaimus	3.65	2.99	2.33	1.74	7.02	14.97
Metachromadora	1.69	0.47	1.93	1.47	5.8	20.77
Groups 2 & 4						
AD = 30.39	Group 2	Group 4				
Euchromadora	6.02	4.56	2.28	2.25	7.49	7.49
Axonolaimus	1.22	0	1.82	1.27	5.97	13.47
Adoncholaimus	2.3	1.61	1.66	1.77	5.45	18.92
Groups 3 & 4						
AD = 44.13	Group 3	Group 4				
Euchromadora	2	4.56	5.59	10.83	12.66	12.66
Crenopharynx	1.08	2.67	3.95	1.53	8.94	21.6
Oncholaimus	1.28	2.99	3.82	2.21	8.65	30.25
Groups 1 & 5						
AD= 44.41	Group 1	Group 5				
Euchromadora	5.49	3.63	3.25	1.61	7.32	7.32
Adoncholaimus	2.27	1.61	3.22	1.59	7.25	14.57
Metachromadora	1.69	0	2.86	4.28	6.45	21.02

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Groups 2 & 5						
AD= 40.65	Group 2	Group 5				
Euchromadora	6.02	3.63	4.1	2.54	10.08	10.08
Enoplus	1.79	3.04	2.16	1.51	5.31	15.39
Crenopharynx	2.94	1.8	2.07	1.49	5.09	20.48
Groups 3 & 5						
AD= 43.70	Group 3	Group 5				
Euchromadora	2	3.63	4.09	2.22	9.36	9.36
Viscosia	0.94	2.39	3.69	1.31	8.45	17.81
Oncholaimus	1.28	2.64	3.5	4.47	8	25.82
Groups 4 & 5						
AD= 33.10	Group 4	Group 5				
Monoposthia	1.28	0	2.41	7.1	7.28	7.28
Crenopharynx	2.67	1.8	2.35	1.2	7.11	14.39
Cyatholaimus	1.88	1.67	2.28	1.33	6.9	21.29
Groups 1 & 6						
AD = 44.02	Group 1	Group 6				
Neochromadora	0	2.98	3.84	2.84	8.72	8.72
Theristus	0.67	3.16	3.25	4.06	7.39	16.11
Chromaspirina	1.35	3.4	2.71	1.62	6.15	22.26

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Groups 2 & 6						
AD= 37.45	Group 2	Group 6				
Chromaspirina	1.05	3.4	3.01	2.99	8.04	8.04
Euchromadora	6.02	3.74	2.94	2.38	7.86	15.9
Neochromadora	0.94	2.98	2.62	1.56	6.99	22.9
Groups 3 & 6						
AD= 53.44	Group 3	Group 6				
Chromaspirina	0.33	3.4	5.33	5.12	9.98	9.98
Neochromadora	0	2.98	5.14	2.92	9.61	19.59
Chromadora	0	2.1	3.68	3.52	6.88	26.47
Groups 3 & 6						
AD = 53.44	Group 3	Group 6				
Chromaspirina	0.33	3.4	5.33	5.12	9.98	9.98
Neochromadora	0	2.98	5.14	2.92	9.61	19.59
Chromadora	0	2.1	3.68	3.52	6.88	26.47
Groups 4 & 6						
AD = 38.65	Group 4	Group 6				
Chromaspirina	0.8	3.4	3.71	3.09	9.6	9.6
Neochromadora	0.67	2.98	3.19	2.06	8.26	17.86
Chromadora	0	2.1	2.98	3.52	7.7	25.56
Groups 5 & 6						

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
AD= 43.71	Group 5	Group 6				
Chromaspirina	0	3.4	5.27	8.23	12.06	12.06
Neochromadora	0	2.98	4.57	2.9	10.45	22.52
Theristus	0.91	3.16	3.5	2.79	8	30.52
Groups 1 & 7						
AD = 50.74	Group 1	Group 7				
Spirinia	0	2.87	5.22	0.67	10.28	10.28
Euchromadora	5.49	3.25	4.32	0.95	8.52	18.8
Odontophora	1	2.12	3.3	1.08	6.51	25.31
Groups 2 & 7						
AD = 47.24	Group 2	Group 7				
Spirinia	0	2.87	5.18	0.67	10.97	10.97
Euchromadora	6.02	3.25	4.72	0.98	9.99	20.96
Odontophora	0.58	2.12	3.19	1.05	6.76	27.72
Groups 3 & 7						
AD= 58.81	Group 3	Group 7				
Spirinia	0	2.87	8.1	0.66	13.78	13.78
Euchromadora	2	3.25	5.72	3.22	9.72	23.49
Odontophora	0.33	2.12	5.05	1.02	8.59	32.08
Groups 4 & 7						

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
AD = 47.07	Group 4	Group 7				
Spirinia	0.33	2.87	6	0.73	12.74	12.74
Euchromadora	4.56	3.25	4.02	1	8.54	21.28
Odontophora	0.33	2.12	3.78	1.02	8.03	29.31
Groups 5 & 7						
AD = 52.36	Group 5	Group 7				
Spirinia	0	2.87	6.73	0.67	12.85	12.85
Euchromadora	3.63	3.25	4.44	1.26	8.49	21.34
Odontophora	0.33	2.12	4.25	1.04	8.12	29.46
Groups 6 & 7						
AD= 48.61	Group 6	Group 7				
Spirinia	0.33	2.87	4.82	0.73	9.91	9.91
Chromaspirina	3.4	0.94	3.61	2.26	7.42	17.34
Neochromadora	2.98	0.58	3.47	1.66	7.14	24.48
Groups 1 & 8						
AD = 39.91	Group 1	Group 8				
Odontophora	1	2.72	3.32	1.25	8.32	8.32
Adoncholaimus	2.27	3.53	3.2	1.28	8.02	16.34
Oncholaimus	3.65	2.57	2.37	1.52	5.95	22.29
Groups 2 & 8						
AD = 34.90	Group 2	Group 8				

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Odontophora	0.58	2.72	3.46	1.47	9.91	9.91
Adoncholaimus	2.3	3.53	2.67	1.69	7.65	17.57
Enoplolaimus	1	1.63	1.81	1.39	5.18	22.74
Groups 3 & 8						
AD = 48.47	Group 3	Group 8				
Euchromadora	2	4.83	5.62	3.36	11.59	11.59
Odontophora	0.33	2.72	4.91	1.45	10.12	21.71
Adoncholaimus	1.99	3.53	3.66	1.59	7.56	29.27
Groups 4 & 8						
AD = 34.68	Group 4	Group 8				
Odontophora	0.33	2.72	3.93	1.39	11.33	11.33
Adoncholaimus	1.61	3.53	3.09	1.41	8.9	20.23
Enoplolaimus	0.8	1.63	2.09	1.7	6.02	26.25
Groups 5 & 8						
AD= 43.13	Group 5	Group 8				
Odontophora	0.33	2.72	4.32	1.45	10.01	10.01
Adoncholaimus	1.61	3.53	3.73	1.71	8.64	18.65
Chromaspirina	0	1.49	2.56	3.84	5.93	24.59
Groups 6 & 8						
AD = 40.03	Group 6	Group 8				

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Neochromadora	2.98	0.33	3.41	2.33	8.51	8.51
Odontophora	1.33	2.72	2.93	2.43	7.31	15.82
Chromaspirina	3.4	1.49	2.51	3.73	6.26	22.09
Groups 7 & 8						
AD = 46.92	Group 7	Group 8				
Spirinia	2.87	0.47	5.42	0.74	11.56	11.56
Euchromadora	3.25	4.83	3.81	0.94	8.12	19.68
Adoncholaimus	1.82	3.53	3.7	1.38	7.88	27.56
Groups 1 & 9						
AD= 44.25	Group 1	Group 9				
Theristus	0.67	2.94	3.25	2.15	7.35	7.35
Oncholaimus	3.65	1.73	3.02	1.46	6.83	14.18
Adoncholaimus	2.27	1.82	2.48	1.48	5.61	19.79
Groups 2 & 9						
AD = 34.04	Group 2	Group 9				
Euchromadora	6.02	4.15	2.68	2.11	7.87	7.87
Oncholaimus	3.53	1.73	2.56	2.19	7.52	15.39
Rhabditis	0.33	1.39	1.76	1.02	5.17	20.56
Groups 3 & 9						
AD = 48.33	Group 3	Group 9				
Euchromadora	2	4.15	4.19	3.01	8.66	8.66

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Crenopharynx	1.08	2.51	3.18	1.5	6.57	15.23
Theristus	1.33	2.94	3.13	1.6	6.47	21.7
Groups 4 & 9						
AD= 35.46	Group 4	Group 9				
Theristus	1.28	2.94	2.57	1.87	7.24	7.24
Rhabditis	0	1.39	2.1	1	5.93	13.16
Oncholaimus	2.99	1.73	2.05	1.54	5.77	18.93
Groups 5 & 9						
AD = 42.00	Group 5	Group 9				
Theristus	0.91	2.94	3.52	1.72	8.38	8.38
Chromaspirina	0	1.91	3.25	2.33	7.74	16.12
Neochromadora	0	1.38	2.37	4.71	5.65	21.76
Groups 6 & 9						
AD= 30.40	Group 6	Group 9				
Neochromadora	2.98	1.38	2.07	1.42	6.81	6.81
Chromaspirina	3.4	1.91	1.99	1.56	6.53	13.34
Rhabditis	0.33	1.39	1.64	1.02	5.39	18.73
Groups 7 & 9						
AD = 49.23	Group 7	Group 9				
Spirinia	2.87	0	5.28	0.67	10.73	10.73

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Euchromadora	3.25	4.15	3.66	1.15	7.44	18.17
Theristus	1.82	2.94	2.85	1.22	5.79	23.96
Groups 8 & 9						
AD= 35.80	Group 8	Group 9				
Odontophora	2.72	1.61	3.08	3.36	8.61	8.61
Adoncholaimus	3.53	1.82	2.89	1.86	8.07	16.68
Theristus	1.58	2.94	1.97	1.35	5.49	22.17
Groups 8 & 9						
AD = 35.80	Group 8	Group 9				
Odontophora	2.72	1.61	3.08	3.36	8.61	8.61
Adoncholaimus	3.53	1.82	2.89	1.86	8.07	16.68
Theristus	1.58	2.94	1.97	1.35	5.49	22.17
Groups 1 & 10						
AD = 38.68	Group 1	Group 10				
Neochromadora	0	1.99	2.85	5.82	7.36	7.36
Adoncholaimus	2.27	0.67	2.85	1.09	7.36	14.72
Theristus	0.67	2.51	2.67	2.95	6.9	21.61
Groups 2 & 10						
AD = 31.61	Group 2	Group 10				
Adoncholaimus	2.3	0.67	2.63	1.67	8.31	8.31
Chromadora	0.47	1.94	2.09	1.42	6.6	14.91

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Chromaspirina	1.05	2.43	1.91	1.67	6.05	20.96
Groups 3 & 10						
AD = 53.28	Group 3	Group 10				
Euchromadora	2	4.75	5.41	10.63	10.15	10.15
Viscosia	0.94	3.54	5.09	3.2	9.55	19.7
Chromaspirina	0.33	2.43	4.12	3.42	7.74	27.44
Groups 4 & 10						
AD = 34.96	Group 4	Group 10				
Chromadora	0	1.94	2.99	2.69	8.55	8.55
Chromaspirina	0.8	2.43	2.58	2.03	7.39	15.94
Viscosia	2.08	3.54	2.25	3.44	6.43	22.37
Groups 5 & 10						
AD = 41.43	Group 5	Group 10				
Chromaspirina	0	2.43	4.2	5.85	10.15	10.15
Neochromadora	0	1.99	3.46	5.54	8.36	18.51
Chromadora	0	1.94	3.31	2.77	7.99	26.49
Groups 6 & 10						
AD = 28.48	Group 6	Group 10				
Cyatholaimus	0.8	2.3	1.92	1.65	6.74	6.74
Adoncholaimus	1.88	0.67	1.8	1.54	6.31	13.05

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Euchromadora	3.74	4.75	1.53	1.77	5.36	18.4
Groups 7 & 10						
AD = 46.94	Group 7	Group 10				
Spirinia	2.87	0.67	5.53	0.78	11.79	11.79
Euchromadora	3.25	4.75	3.76	0.99	8.02	19.81
Odontophora	2.12	0.67	3.25	1.09	6.92	26.73
Groups 7 & 10						
AD = 46.94	Group 7	Group 10				
Spirinia	2.87	0.67	5.53	0.78	11.79	11.79
Euchromadora	3.25	4.75	3.76	0.99	8.02	19.81
Odontophora	2.12	0.67	3.25	1.09	6.92	26.73
Groups 8 & 10						
AD= 38.80	Group 8	Group 10				
Adoncholaimus	3.53	0.67	4.25	1.84	10.94	10.94
Odontophora	2.72	0.67	3.5	1.59	9.02	19.97
Neochromadora	0.33	1.99	2.36	3.04	6.09	26.05
Groups 8 & 10						
AD = 38.80	Group 8	Group 10				
Adoncholaimus	3.53	0.67	4.25	1.84	10.94	10.94
Odontophora	2.72	0.67	3.5	1.59	9.02	19.97
Neochromadora	0.33	1.99	2.36	3.04	6.09	26.05

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Groups 8 & 10						
AD = 38.80	Group 8	Group 10				
Adoncholaimus	3.53	0.67	4.25	1.84	10.94	10.94
Odontophora	2.72	0.67	3.5	1.59	9.02	19.97
Neochromadora	0.33	1.99	2.36	3.04	6.09	26.05
Groups 9 & 10						
AD = 30.75	Group 9	Group 10				
Oncholaimus	1.73	3.23	2.25	1.69	7.31	7.31
Adoncholaimus	1.82	0.67	2.02	1.48	6.56	13.87
Rhabditis	1.39	0.33	1.79	1.03	5.81	19.67
Groups 9 & 10						
AD = 30.75	Group 9	Group 10				
Oncholaimus	1.73	3.23	2.25	1.69	7.31	7.31
Adoncholaimus	1.82	0.67	2.02	1.48	6.56	13.87
Rhabditis	1.39	0.33	1.79	1.03	5.81	19.67
Groups 1 & 11						
AD = 47.14	Group 1	Group 11				
Spirinia	0	2.82	3.93	1.28	8.34	8.34
Euchromadora	5.49	3.49	2.84	2.11	6.03	14.37
Adoncholaimus	2.27	3.07	2.77	1.59	5.88	20.25

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Groups 2 & 11						
AD = 40.12	Group 2	Group 11				
Spirinia	0	2.82	3.91	1.28	9.75	9.75
Euchromadora	6.02	3.49	3.54	3.94	8.83	18.59
Odontophora	0.58	2.14	2.7	1.44	6.74	25.32
Groups 3 & 11						
AD = 48.75	Group 3	Group 11				
Spirinia	0	2.82	5.41	1.29	11.1	11.1
Odontophora	0.33	2.14	3.9	1.47	8.01	19.1
Viscosia	0.94	2.7	3.41	1.34	7	26.1
Groups 3 & 11						
AD = 48.75	Group 3	Group 11				
Spirinia	0	2.82	5.41	1.29	11.1	11.1
Odontophora	0.33	2.14	3.9	1.47	8.01	19.1
Viscosia	0.94	2.7	3.41	1.34	7	26.1
Groups 4 & 11						
AD = 39.92	Group 4	Group 11				
Spirinia	0.33	2.82	4.13	1.4	10.34	10.34
Odontophora	0.33	2.14	3.12	1.4	7.81	18.15
Linhomoeus	0	1.63	2.48	1.2	6.21	24.36
Groups 5 & 11						

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
AD= 44.24	Group 5	Group 11				
Spirinia	0	2.82	4.75	1.29	10.74	10.74
Odontophora	0.33	2.14	3.43	1.47	7.74	18.49
Linhomoeus	0	1.63	2.74	1.21	6.19	24.68
Groups 5 & 11						
AD = 44.24	Group 5	Group 11				
Spirinia	0	2.82	4.75	1.29	10.74	10.74
Odontophora	0.33	2.14	3.43	1.47	7.74	18.49
Linhomoeus	0	1.63	2.74	1.21	6.19	24.68
Groups 5 & 11						
AD = 44.24	Group 5	Group 11				
Spirinia	0	2.82	4.75	1.29	10.74	10.74
Odontophora	0.33	2.14	3.43	1.47	7.74	18.49
Linhomoeus	0	1.63	2.74	1.21	6.19	24.68
Groups 5 & 11						
AD = 44.24	Group 5	Group 11				
Spirinia	0	2.82	4.75	1.29	10.74	10.74
Odontophora	0.33	2.14	3.43	1.47	7.74	18.49
Linhomoeus	0	1.63	2.74	1.21	6.19	24.68
Groups 6 & 11						

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
AD = 39.70	Group 6	Group 11				
Spirinia	0.33	2.82	3.49	1.4	8.79	8.79
Neochromadora	2.98	0.33	3.39	2.27	8.55	17.34
Chromaspirina	3.4	1.38	2.62	4.11	6.61	23.94
Groups 6 & 11						
AD = 39.70	Group 6	Group 11				
Spirinia	0.33	2.82	3.49	1.4	8.79	8.79
Neochromadora	2.98	0.33	3.39	2.27	8.55	17.34
Chromaspirina	3.4	1.38	2.62	4.11	6.61	23.94
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD= 49.42	Group 7	Group 11				

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD = 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 7 & 11						
AD= 49.42	Group 7	Group 11				
Spirinia	2.87	2.82	6.14	1.3	12.43	12.43
Euchromadora	3.25	3.49	3.44	1.37	6.96	19.39
Odontophora	2.12	2.14	3.05	1.27	6.18	25.58
Groups 8 & 11						
AD = 35.28	Group 8	Group 11				
Spirinia	0.47	2.82	3.75	1.38	10.64	10.64

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Odontophora	2.72	2.14	2.86	1.21	8.12	18.75
Linhomoeus	0	1.63	2.29	1.2	6.48	25.23
Groups 9 & 11						
AD = 35.90	Group 9	Group 11				
Spirinia	0	2.82	3.97	1.28	11.06	11.06
Odontophora	1.61	2.14	2.26	3.8	6.3	17.37
Linhomoeus	0.82	1.63	2.13	1.25	5.94	23.31
Groups 10 & 11						
AD = 40.44	Group 10	Group 11				
Spirinia	0.67	2.82	3.68	1.51	9.11	9.11
Adoncholaimus	0.67	3.07	3.44	1.86	8.51	17.62
Odontophora	0.67	2.14	2.73	1.63	6.75	24.37
Groups 10 & 11						
AD = 40.44	Group 10	Group 11				
Spirinia	0.67	2.82	3.68	1.51	9.11	9.11
Adoncholaimus	0.67	3.07	3.44	1.86	8.51	17.62
Odontophora	0.67	2.14	2.73	1.63	6.75	24.37
Groups 1 & 12						
AD = 73.50	Group 1	Group 12				
Euchromadora	5.49	1.22	9.28	2.54	12.63	12.63

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Oncholaimus	3.65	0.33	7.15	1.86	9.73	22.36
Crenopharynx	2.98	0.33	5.5	4.27	7.49	29.85
Groups 2 & 12						
AD = 68.44	Group 2	Group 12				
Euchromadora	6.02	1.22	10.27	3.12	15.01	15.01
Oncholaimus	3.53	0.33	6.72	3.6	9.82	24.83
Crenopharynx	2.94	0.33	5.42	4.63	7.91	32.74
Groups 3 & 12						
AD = 59.02	Group 3	Group 12				
Enoplus	1.99	0.67	5.15	1.68	8.72	8.72
Adoncholaimus	1.99	1.8	4.79	1.46	8.11	16.83
Cyatholaimus	1.14	0.47	3.8	1.03	6.44	23.27
Groups 4 & 12						
AD = 67.10	Group 4	Group 12				
Euchromadora	4.56	1.22	8.3	2.6	12.37	12.37
Oncholaimus	2.99	0.33	6.47	2.66	9.65	22.02
Crenopharynx	2.67	0.33	5.65	1.92	8.42	30.44
Groups 5 & 12						
AD = 65.01	Group 5	Group 12				
Euchromadora	3.63	1.22	7.15	1.79	11	11

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Enoplus	3.04	0.67	7.03	2.46	10.81	21.81
Oncholaimus	2.64	0.33	6.59	3.67	10.13	31.94
Groups 6 & 12						
AD = 68.69	Group 6	Group 12				
Chromaspirina	3.4	0	6.4	5.67	9.32	9.32
Neochromadora	2.98	0	5.53	2.78	8.06	17.38
Euchromadora	3.74	1.22	4.89	1.74	7.12	24.5
Groups 7 & 12						
AD = 73.70	Group 7	Group 12				
Spirinia	2.87	0.33	9.43	0.7	12.8	12.8
Euchromadora	3.25	1.22	6.63	1.6	8.99	21.79
Oncholaimus	2.52	0.33	5.43	2.13	7.37	29.17
Groups 8 & 12						
AD = 65.78	Group 8	Group 12				
Euchromadora	4.83	1.22	7.98	2.31	12.14	12.14
Adoncholaimus	3.53	1.8	4.99	1.51	7.59	19.72
Odontophora	2.72	1.05	4.92	1.71	7.48	27.21
Groups 9 & 12						
AD= 64.36	Group 9	Group 12				
Euchromadora	4.15	1.22	6.41	2.14	9.96	9.96
Enoplus	2.88	0.67	4.82	2.91	7.49	17.46

Table A2.1 Result of one way SIMPER analysis of nematode abundance data in each mash bag treatment comparison

Genus	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Crenopharynx	2.51	0.33	4.6	2.06	7.15	24.6
Groups 10 & 12						
AD = 72.74	Group 10	Group 12				
Euchromadora	4.75	1.22	7.77	2.73	10.68	10.68
Oncholaimus	3.23	0.33	6.28	2.74	8.63	19.31
Viscosia	3.54	0.67	6.23	3.29	8.56	27.87
Groups 11 & 12						
AD = 62.27	Group 11	Group 12				
Spirinia	2.82	0.33	5.64	1.39	9.07	9.07
Enoplus	3.19	0.67	5.4	1.69	8.67	17.73
Euchromadora	3.49	1.22	4.92	1.92	7.9	25.63

List of References

Alkemade R, Van Rijswijk P (1993) Path analyses of the influence of substrate composition on nematode numbers and on decomposition of stranded seaweed at an Antarctic coast. Netherlands Journal of Sea Research 31:63-70

Alkemade R, Wielemaker A, De Jong S, Sandee A (1992a) *Diplolaimella dievengatensis* in stimulating the mineralization of *Spartina anglica* detritus. Marine Ecology Progress Series 90:149-155.1992

Alkemade R, Wielemaker A, Hemminga M (1993) Correlation between nematode abundance and decomposition rate of *Spartina anglica* leaves. Marine Ecology Progress Series 99:293-300

Alkemade R, Wielemaker A, Hemminga MA (1992b) Stimulation of decomposition of Spartina anglica leaves by the bacterivorous marine nematode *Diplolaimelloides bruciei* (Monhysteridae). Journal of Experimental Marine Biology and Ecology 159:267-278

Allan JD (1975) Components of diversity. Oecologia 18:359-367

Andrews JH (1977) Observations on the pathology of seaweeds in the Pacific Northwest. Canadian Journal of Botany 55:1019-1027

Armenteros M, Ruiz-Abierno A, Fernández-Garcés R, Pérez-García JA, Díaz-Asencio L, Vincx M, Decraemer W (2009) Biodiversity patterns of free-living marine nematodes in a tropical bay: Cienfuegos, Caribbean Sea. Estuarine, Coastal and Shelf Science 85:179-189

Arrontes J (1999) On the evolution of interactions between marine mesoherbivores and algae. Botanica Marina 42:137-155

Ash C, Crompton D, Keymer A (1984) Nature's unfair food tax. New scientist

Barreiro F, Gómez M, López J, Lastra M, la Huz R (2012) Coupling between macroalgal inputs and nutrients outcrop in exposed sandy beaches. Hydrobiologia 700:73-84

Bauer JE, Cai W-J, Raymond PA, Bianchi TS, Hopkinson CS, Regnier PA (2013) The changing carbon cycle of the coastal ocean. Nature 504:61-70

Bell SS, Coen LD (1982) Investigations on epibenthic meiofauna. II influence of microhabitat and macroalgae on abundance of small invertebrates on *Diopatra cuprea* (Bosc)(Polychaeta: Onuphidae) tube-caps in Virginia. Journal of Experimental Marine Biology and Ecology 61:175-188

Bhadury P, Austen MC, Bilton DT, Lambshead PJD, Rogers AD, Smerdon GR (2006) Development and evaluation of a DNA-barcoding approach for the rapid identification of nematodes. Marine Ecology Progress Series 320:1-9

Blome D, Schleier U, van Bernem K-H (1999) Analysis of the small-scale spatial patterns of free-living marine nematodes from tidal flats in the East Frisian Wadden Sea. Marine Biology 133:717-726

Bongers T, Alkemade R, Yeates G (1991) Interpretation of disturbance-induced maturity decrease in marine nematode assemblages by means of the Maturity Index. Marine Ecology Progress Series 76:135-142

Bongers T, Ferris H (1999) Nematode community structure as a bioindicator in environmental monitoring. Trends in Ecology & Evolution 14:224-228

Boström C, O'Brien K, Roos C, Ekebom J (2006) Environmental variables explaining structural and functional diversity of seagrass macrofauna in an archipelago landscape. Journal of Experimental Marine Biology and Ecology 335:52-73

Boucher G (1990) Pattern of nematode species diversity in temperate and tropical subtidal sediments. Marine Ecology 11:133-146

Boucher G (1997) Structure and biodiversity of nematode assemblages in the SW lagoon of New Caledonia. Coral Reefs 16:177-186
Bracken ME, Gonzalez-Dorantes CA, Stachowicz JJ (2007) Whole-community mutualism: associated invertebrates facilitate a dominant habitat-forming seaweed. Ecology 88:2211-2219
Brauko KM, de Souza FM, Muniz P, de Camargo MG, da Cunha Lana P (2015) Spatial variability of three benthic indices for marine quality assessment in a subtropical estuary of Southern Brazil. Marine Pollution Bulletin 91:454-460
Bremner J, Rogers S, Frid C (2003) Assessing functional diversity in marine benthic ecosystems: a comparison of approaches. Marine Ecology Progress Series 254:11-25
Britton-Simmons KH (2004) Direct and indirect effects of the introduced alga Sargassum muticum on benthic, subtidal communities of Washington State, USA. Marine Ecology Progress Series 277:61-78
Brown AC, McLachlan A (2010) The ecology of sandy shores. Academic Press
Buchsbaum R, Valiela I, Swain T, Dzierzeski M, Allen S (1991) Available and refractory nitrogen in detritus of coastal vascular plants and macroalgae. Marine ecology progress series 72:131-143
Burgess R (2001) An improved protocol for separating meiofauna from sediments using colloidal silica sols. Marine Ecology Progress Series 214:161-165
Buschbaum C, Chapman AS, Saier B (2006) How an introduced seaweed can affect epibiota diversity in different coastal systems. Marine Biology 148:743-754
Byrnes JE, Gamfeldt L, Isbell F, Lefcheck JS, Griffin JN, Hector A, Cardinale BJ, Hooper DU, Dee LE, Emmett Duffy J (2014) Investigating the relationship between biodiversity and ecosystem multifunctionality: challenges and solutions. Methods in Ecology and Evolution 5:111-124
Cacabelos E, Olabarria C, Incera M, Troncoso JS (2010a) Do grazers prefer invasive seaweeds? Journal of Experimental Marine Biology and Ecology 393:182-187
Cacabelos E, Olabarria C, Incera M, Troncoso JS (2010b) Effects of habitat structure and tidal height on epifaunal assemblages associated with macroalgae. Estuarine, Coastal and Shelf Science 89:43-52
Cadotte MW, Fukami T (2005) Dispersal, spatial scale, and species diversity in a hierarchically structured experimental landscape. Ecology Letters 8:548-557
Cai YW, Cheng XY, Xu RM, Duan DH, Kirkendall LR (2008) Genetic diversity and biogeography of red turpentine beetle Dendroctonus valens in its native and invasive regions. Insect Science 15:291-301
Caley MJ, Schluter D (1997) The relationship between local and regional diversity. Ecology 78:70-80
Cameron EK, Bayne EM, Clapperton MJ (2007) Human-facilitated invasion of exotic earthworms into northern boreal forests. Ecoscience 14:482-490
Cardinale BJ, Duffy JE, Gonzalez A, Hooper DU, Perrings C, Venail P, Narwani A, Mace GM, Tilman D, Wardle DA (2012) Biodiversity loss and its impact on humanity. Nature 486:59-67
Chan M-S (1997) The global burden of intestinal nematode infections—fifty years on. Parasitology today 13:438-443
Chapin III FS, Matson PA, Vitousek P (2011) Principles of terrestrial ecosystem ecology. Springer Science & Business Media
Chapman M, Tolhurst T, Murphy R, Underwood A (2010) Complex and inconsistent patterns of variation in benthos, micro-algae and sediment over multiple spatial scales. Marine Ecology Progress Series 398:33-47
Chown S (1996) Kelp degradation byParactora trichosterna (Thomson)(Diptera: Helcomyzidae) at sub-Antarctic South Georgia. Polar Biology 16:171-178

Clarke A (1992) Is there a latitudinal diversity cline in the sea? Trends in Ecology & Evolution 7:286-287
Clarke K, Gorley R (2006) PRIMER v6: user manual/tutorial (Plymouth routines in multivariate ecological research). Plymouth: Primer-E Ltd
Clarke K, PRIMER GR (2006) V6: user manual/tutorial. Primer-E Ltd Plymouth-2006
Clarke KR (1993) Non-parametric multivariate analyses of changes in community structure. Australian journal of ecology 18:117-143
Coleman FC, Williams SL (2002) Overexploiting marine ecosystem engineers: potential consequences for biodiversity. Trends in Ecology & Evolution 17:40-44
Coleman RA, Underwood AJ, Benedetti-Cecchi L, Åberg P, Arenas F, Arrontes J, Castro J, Hartnoll RG, Jenkins SR, Paula J (2006) A continental scale evaluation of the role of limpet grazing on rocky shores. Oecologia 147:556-564
Colombini I, Aloia A, Fallaci M, Pezzoli G, Chelazzi L (2000) Temporal and spatial use of stranded wrack by the macrofauna of a tropical sandy beach. Marine Biology 136:531-541
Cornell HV (1985) Local and regional richness of cynipine gall wasps on California oaks. Ecology 66:1247-1260
Cornell HV (1993) Unsaturated patterns in species assemblages: the role of regional processes in setting local species richness. Species diversity in ecological communities University of Chicago Press, Chicago, Illinois, USA:243-252
Coull BC (1999) Role of meiofauna in estuarine soft-bottom habitats*. Australian Journal of Ecology 24:327-343
Coupland GT, Duarte CM, Walker DI (2007) High metabolic rates in beach cast communities. Ecosystems 10:1341-1350
Coupland GT, McDonald JI (2008) Extraordinarily high earthworm abundance in deposits of marine macrodetritus along two semi-arid beaches. Marine Ecology Progress Series 361:181-189
Creer S, Fonseca V, Porazinska D, GIBLIN-DAVIS R, Sung W, Power D, Packer M, Carvalho G, Blaxter M, Lambshead P (2010) Ultrasequencing of the meiofaunal biosphere: practice, pitfalls and promises. Molecular Ecology 19:4-20
Crisp D, Southward AJ (1958) The distribution of intertidal organisms along the coasts of the English Channel. Journal of the Marine Biological Association of the United Kingdom 37:157-203
Critchley AT, Farnham W, Morrell S (1983) A chronology of new European sites of attachment for the invasive brown alga, Sargassum muticum, 1973–1981. Journal of the Marine Biological Association of the United Kingdom 63:799-811
Crowe T, Thompson R, Bray S, Hawkins S (2000) Impacts of anthropogenic stress on rocky intertidal communities. Journal of Aquatic Ecosystem Stress and Recovery 7:273-297
Curvelo RR, Corbisier TN (2000) The meiofauna asssociated whith *Sargassum cymosum* at Lázaro Beach, Ubatuba, São Paulo. Revista Brasileira de Oceanografia 48:119-130
Da Rocha C, Venekey V, Bezerra T, Souza J (2006) Phytal marine nematode assemblages and their relation with the macrophytes structural complexity in a Brazilian tropical rocky beach. Hydrobiologia 553:219-230
De Laender F, Rohr JR, Ashauer R, Baird DJ, Berger U, Eisenhauer N, Grimm V, Hommen U, Maltby L, Meliàn CJ (2016) Reintroducing Environmental Change Drivers in Biodiversity–Ecosystem Functioning Research. Trends in Ecology & Evolution 31:905-915

De Ley P, Blaxter M (2002) Systematic position and phylogeny. The biology of nematodes. CRC Press

De Mesel I, Derycke S, Moens T, Van der Gucht K, Vincx M, Swings J (2004) Top-down impact of bacterivorous nematodes on the bacterial community structure: a microcosm study. Environmental Microbiology 6:733-744

De Mesel I, Derycke S, Swings J, Vincx M, Moens T (2003) Influence of bacterivorous nematodes on the decomposition of cordgrass. Journal of Experimental Marine Biology and Ecology 296:227-242

De Mesel I, Derycke S, Swings J, Vincx M, Moens T (2006) Role of nematodes in decomposition processes: Does within-trophic group diversity matter? Marine Ecology Progress Series 321:157-166

De Oliveira DA, Derycke S, Da Rocha CM, Barbosa DF, Decraemer W, Dos Santos GA (2016) Spatiotemporal variation and sediment retention effects on nematode communities associated with *Halimeda opuntia* (Linnaeus) Lamouroux (1816) and *Sargassum polyceratium* Montagne (1837) seaweeds in a tropical phytal ecosystem. Marine Biology 163:1-13

De Schryver P, Vadstein O (2014) Ecological theory as a foundation to control pathogenic invasion in aquaculture. The ISME journal 8:2360-2368

Delibes M, Clavero M, Prenda J, del Carmen Blázquez M, Ferreras P (2004) Potential impact of an exotic mammal on rocky intertidal communities of northwestern Spain. Biological Invasions 6:213-219

Deprez T (2007) NeMys. Worldwide web electronic publication.

Derycke S, Backeljau T, Moens T (2013) Dispersal and gene flow in free-living marine nematodes. Frontiers in zoology 10:1

Derycke S, Van Vynckt R, Vanoverbeke J, Vincx M, Moens T (2007) Colonization patterns of Nematoda on decomposing algae in the estuarine environment: Community assembly and genetic structure of the dominant species *Pellioditis marina*. Limnology and oceanography 52:992-1001

Dray S, Pélissier R, Couteron P, Fortin M-J, Legendre P, Peres-Neto P, Bellier E, Bivand R, Blanchet FG, De Cáceres M (2012) Community ecology in the age of multivariate multiscale spatial analysis. Ecological Monographs 82:257-275

Dugan JE, Hubbard DM, McCrary MD, Pierson MO (2003) The response of macrofauna communities and shorebirds to macrophyte wrack subsidies on exposed sandy beaches of southern California. Estuarine, Coastal and Shelf Science 58:25-40

Edgar GJ, Klumpp DW (2003) Consistencies over regional scales in assemblages of mobile epifauna associated with natural and artificial plants of different shape. Aquatic Botany 75:275-291

Ekschmitt K, Bakonyi G, Bongers M, Bongers T, Boström S, Dogan H, Harrison A, Nagy P, O'Donnell AG, Papatheodorou EM (2001) Nematode community structure as indicator of soil functioning in European grassland soils. European Journal of Soil Biology 37:263-268

Engelen AH, Primo AL, Cruz T, Santos R (2013) Faunal differences between the invasive brown macroalga *Sargassum muticum* and competing native macroalgae. Biological invasions 15:171-183

Farnham W, Fletcher R, IRVINE LM (1973) Attached Sargassum found in Britain. Nature 243:232-233

Findlay S, Tenore KR (1982) Effect of a Free-Living Marine Nematode(*Diplolaimella chitwoodi*) on Detrital Carbon Mineralization. Marine Ecology Progress Series 8:161-166

Fire A, Xu S, Montgomery MK, Kostas SA, Driver SE, Mello CC (1998) Potent and specific genetic interference by double-stranded RNA in Caenorhabditis elegans. nature 391:806-811

Fisher RA, Corbet AS, Williams CB (1943) The relation between the number of species and the number of individuals in a random sample of an animal population. The Journal of Animal Ecology:42-58
Fleishman E, Fay JP, Murphy DD (2000) Upsides and downsides: contrasting topographic gradients in species richness and associated scenarios for climate change. Journal of Biogeography 27:1209-1219
Frame K, Hunt G, Roy K (2007) Intertidal meiofaunal biodiversity with respect to different algal habitats: a test using phytal ostracodes from Southern California. Hydrobiologia 586:331-342
Franklin JF (1993) Preserving biodiversity: species, ecosystems, or landscapes? Ecological applications 3:202-205
Freckman DW (1988) Bacterivorous nematodes and organic-matter decomposition. Agriculture, Ecosystems & Environment 24:195-217
Frey MA (2010) The relative importance of geography and ecology in species diversification: evidence from a tropical marine intertidal snail (*Nerita*). Journal of Biogeography 37:1515-1528
Fridley J, Stachowicz J, Naeem S, Sax D, Seabloom E, Smith M, Stohlgren T, Tilman D, Holle BV (2007) The invasion paradox: reconciling pattern and process in species invasions. Ecology 88:3-17
Gamfeldt L, Lefcheck JS, Byrnes JE, Cardinale BJ, Duffy JE, Griffin JN (2015) Marine biodiversity and ecosystem functioning: what's known and what's next? Oikos 124:252-265
Garcia-Robledo E, Corzo A, de Lomas JG, Van Bergeijk S (2008) Biogeochemical effects of macroalgal decomposition on intertidal microbenthos: a microcosm experiment. Marine Ecology Progress Series 356:139-151
Gartner TB, Cardon ZG (2004) Decomposition dynamics in mixed-species leaf litter. Oikos 104:230-246
Gee J, Warwick R (1994a) Metazoan community structure in relation to the fractal dimensions of marine macroalgae. Marine Ecology Progress Series 103:141-150
Gee JM, Warwick RM (1994b) Body-size distribution in a marine metazoan community and the fractal dimensions of macroalgae. Journal of Experimental Marine Biology and Ecology 178:247-259
Gering JC, Crist TO (2002) The alpha–beta–regional relationship: providing new insights into local–regional patterns of species richness and scale dependence of diversity components. Ecology Letters 5:433-444
Gerlach SA (1978) Food-chain relationships in subtidal silty sand marine sediments and the role of meiofauna in stimulating bacterial productivity. Oecologia 33:55-69
Gestoso I, Olabarria C, Troncoso JS (2010) Variability of epifaunal assemblages associated with native and invasive macroalgae. Marine and Freshwater Research 61:724-731
Gibbons M (1988) The impact of wave exposure on the meiofauna of *Gelidium pristoides* (Turner) Kuetzing (Gelidiales: Rhodophyta). Estuarine, Coastal and Shelf Science 27:581-593
Gibbons M, Griffiths C (1986) A comparison of macrofaunal and meiofaunal distribution and standing stock across a rocky shore, with an estimate of their productivities. Marine Biology 93:181-188
Giere O (2008) Meiobenthology: the microscopic motile fauna of aquatic sediments. Springer Science & Business Media
Giere O (2009) Meiobenthology: the microscopic motile fauna of aquatic sediments. Springer
Gobin JF, Warwick RM (2006) Geographical variation in species diversity: a comparison of marine polychaetes and nematodes. Journal of Experimental Marine Biology and Ecology 330:234-244

Godbold JA, Bulling MT, Solan M (2011) Habitat structure mediates biodiversity effects on ecosystem properties. Proceedings of the Royal Society of London B: Biological Sciences 278:2510-2518

Godbold JA, Solan M, Killham K (2009) Consumer and resource diversity effects on marine macroalgal decomposition. Oikos 118:77-86

Gonzalez A, Cardinale BJ, Allington GR, Byrnes J, Arthur Endsley K, Brown DG, Hooper DU, Isbell F, O'Connor MI, Loreau M (2016) Estimating local biodiversity change: a critique of papers claiming no net loss of local diversity. Ecology 97:1949-1960

Gray JS (1997) Marine biodiversity: patterns, threats and conservation needs. Biodiversity & Conservation 6:153-175

Gray JS (2000) The measurement of marine species diversity, with an application to the benthic fauna of the Norwegian continental shelf. Journal of Experimental Marine Biology and Ecology 250:23-49

Griffin JN, Jenkins SR, Gamfeldt L, Jones D, Hawkins SJ, Thompson RC (2009) Spatial heterogeneity increases the importance of species richness for an ecosystem process. Oikos 118:1335-1342

Griffiths C, Stenton-Dozey J (1981) The fauna and rate of degradation of stranded kelp. Estuarine, Coastal and Shelf Science 12:645-653

Griffiths C, Stenton-Dozey J, Koop K (1983) Kelp wrack and the flow of energy through a sandy beach ecosystem. Sandy beaches as ecosystems. Springer

Griffiths D (1999) On investigating local–regional species richness relationships. Journal of Animal Ecology 68:1051-1055

Guégan J-F, Lek S, Oberdorff T (1998) Energy availability and habitat heterogeneity predict global riverine fish diversity. Nature 391:382-384

Gurevitch J, Padilla DK (2004) Are invasive species a major cause of extinctions? Trends in Ecology & Evolution 19:470-474

Hakenkamp CC, Morin A (2000) The importance of meiofauna to lotic ecosystem functioning. Freshwater biology 44:165-175

Hallegraeff G, Bolch C (1992) Transport of diatom and dinoflagellate resting spores in ships' ballast water: implications for plankton biogeography and aquaculture. Journal of plankton research 14:1067-1084

Halpern BS, Walbridge S, Selkoe KA, Kappel CV, Micheli F, D'agrosa C, Bruno JF, Casey KS, Ebert C, Fox HE (2008) A global map of human impact on marine ecosystems. Science 319:948-952

Hardison AK, Canuel EA, Anderson IC, Veuger B (2010) Fate of macroalgae in benthic systems: carbon and nitrogen cycling within the microbial community. Marine Ecology Progress Series 414:41-55

Harries DB, Harrow S, Wilson JR, Mair JM, Donnan DW (2007) The establishment of the invasive alga *Sargassum muticum* on the west coast of Scotland: a preliminary assessment of community effects. Journal of the Marine Biological Association of the UK 87:1057-1067

Hättenschwiler S, Tiunov AV, Scheu S (2005) Biodiversity and litter decomposition in terrestrial ecosystems. Annu Rev Ecol Evol Syst 36:191-218

Hawkins SJ, Jones HD (1992) Rocky shores, Vol 1. Sea Challengers

Hawkins SJ, Moore PJ, Burrows MT, Poloczanska E, Mieszkowska N, Herbert R, Jenkins SR, Thompson RC, Genner MJ, Southward AJ (2008) Complex interactions in a rapidly changing world: responses of rocky shore communities to recent climate change. Climate research 37:123-133

Heip C, Vincx M, Vranken G (1985) The ecology of marine nematodes. Aberdeen University Press

Hejda M, Pyšek P, Jarošík V (2009) Impact of invasive plants on the species richness, diversity and composition of invaded communities. Journal of Ecology 97:393-403

Herbert RJ, Southward A, Clarke RT, Sheader M, Hawkins S (2009) Persistent border: an analysis of the geographic boundary of an intertidal species. Marine Ecology Progress Series 379:135-150
Hewitt JE, Thrush SF, Halliday J, Duffy C (2005) The importance of small-scale habitat structure for maintaining beta diversity. Ecology 86:1619-1626
Hillebrand H (2004) Strength, slope and variability of marine latitudinal gradients. Marine Ecology Progress Series 273:251-267
Holeck KT, Mills EL, MacIsaac HJ, Dochoda MR, Colautti RI, Ricciardi A (2004) Bridging troubled waters: biological invasions, transoceanic shipping, and the Laurentian Great Lakes. BioScience 54:919-929
Holmquist JG (1997) Disturbance and gap formation in a marine benthic mosaic: influence of shifting macroalgal patches on seagrass structure and mobile invertebrates. Marine Ecology Progress Series 158:121-130
Holovachov O, Schmidt-Rhaesa A (2014) Handbook of Zoology. Gastrotricha, Cycloneuralia, Gnathifera. Volume 2: Nematoda. de Gruyter
Hooper DU, Adair EC, Cardinale BJ, Byrnes JE, Hungate BA, Matulich KL, Gonzalez A, Duffy JE, Gamfeldt L, O'Connor MI (2012) A global synthesis reveals biodiversity loss as a major driver of ecosystem change. Nature 486:105-108
Hopper B, Meyers S (1967) Population studies on benthic nematodes within a subtropical seagrass community. Marine Biology 1:85-96
Hopper BE (1967) Foliicolous marine nematodes on turtle grass, Thalassia testudinum Konig, in Biscayne Bay, Florida. Bulletin of Marine Science 17:471-517
Horner-Devine MC, Carney KM, Bohannan BJ (2004) An ecological perspective on bacterial biodiversity. Proceedings of the Royal Society of London B: Biological Sciences 271:113-122
Huff TM, Jarett JK (2007) Sand addition alters the invertebrate community of intertidal coralline turf. Marine Ecology Progress Series 345:75-82
Huh OK (1982) Spring season flow of the Tsushima Current and its separation from the Kuroshio: Satellite evidence. Journal of Geophysical Research: Oceans 87:9687-9693
Huston MA (1999) Local processes and regional patterns: appropriate scales for understanding variation in the diversity of plants and animals. Oikos:393-401
Ieno EN, Solan M, Batty P, Pierce GJ (2006) How biodiversity affects ecosystem functioning: roles of infaunal species richness, identity and density in the marine benthos. Marine Ecology Progress Series 311:263-271
Ince R, Hyndes GA, Lavery PS, Vanderklift MA (2007) Marine macrophytes directly enhance abundances of sandy beach fauna through provision of food and habitat. Estuarine, Coastal and Shelf Science 74:77-86
Inglis G (1989) The colonisation and degradation of stranded Macrocystis pyrifera (L.) C. Ag. by the macrofauna of a New Zealand sandy beach. Journal of Experimental Marine Biology and Ecology 125:203-217
Inglis WG, Coles JW (1961) The species of Rhabditis (Nematoda) found in rotting seaweed on British beaches. Bulletin of the British Museum (Natural History) D Zoology 7:320-333
Isbell F, Calcagno V, Hector A, Connolly J, Harpole WS, Reich PB, Scherer-Lorenzen M, Schmid B, Tilman D, van Ruijven J (2011) High plant diversity is needed to maintain ecosystem services. Nature 477:199-202
Jaramillo E, Huz RDL, Duarte C, Contreras H (2006) Algal wrack deposits and macroinfaunal arthropods on sandy beaches of the Chilean coast. Revista chilena de historia natural 79:337-351
Jedrzejczak M (2002) Spatio-temporal decay'hot spots' of stranded wrack in a Baltic sandy coastal system. Part I. Comparative study of the pattern: 1 type of wrack vs 3 beach sites. Oceanologia 44:491-512

Jenkins S, Åberg P, Cervin G, Coleman R, Delany J, Della Santina P, Hawkins S, LaCroix E, Myers A, Lindegarth M (2000) Spatial and temporal variation in settlement and recruitment of the intertidal barnacle *Semibalanus balanoides* (L.)(Crustacea: Cirripedia) over a European scale. Journal of Experimental Marine Biology and Ecology 243:209-225

Jenkins S, Coleman R, Della Santina P, Hawkins S, Burrows M, Hartnoll R (2005) Regional scale differences in the determinism of grazing effects in the rocky intertidal. Marine Ecology Progress Series 287:77-86

Jensen P (1987) Feeding ecology of free-living aquatic nematodes. Marine Ecology Progress Series 35:187-196

Jeong E (2011) Applying an underwater photography technique to nearshore benthic mapping: A case study in a rocky shore environment. Journal of Coastal Research:1764-1768

Joint I, Gee J, Warwick R (1982) Determination of fine-scale vertical distribution of microbes and meiofauna in an intertidal sediment. Marine Biology 72:157-164

Kelaher B, Chapman M, Underwood A (2001) Spatial patterns of diverse macrofaunal assemblages in coralline turf and their associations with environmental variables. Journal of the Marine Biological Association of the UK 81:917-930

Kelaher BP, Castilla JC, Seed R (2004) Intercontinental test of generality for spatial patterns among diverse molluscan assemblages in coralline algal turf. Marine Ecology Progress Series 271:221-231

Kendall MA, Aschan M (1993) Latitudinal gradients in the structure of macrobenthic communities: a comparison of Arctic, temperate and tropical sites. Journal of Experimental Marine Biology and Ecology 172:157-169

Kim H-g, Rho HS, Oh C-W (2013) Seasonal and spatial variations in nematode assemblages affected by thermal influence of nuclear power plant in Korea (East Sea, Pacific Ocean). Marine Biology Research 9:725-738

Koh C-H, Kim Y, Kang S-G (1993) Size distribution, growth and production of Sargassum thunbergii in an intertidal zone of Padori, west coast of Korea. Hydrobiologia 260:207-214

Koop K, Lucas M (1983) Carbon flow and nutrient regeneration from the decomposition of macrophyte debris in a sandy beach microcosm. Sandy beaches as ecosystems. Springer

Lande R (1996) Statistics and partitioning of species diversity, and similarity among multiple communities. Oikos:5-13

Lastra M, Page HM, Dugan JE, Hubbard DM, Rodil IF (2008) Processing of allochthonous macrophyte subsidies by sandy beach consumers: estimates of feeding rates and impacts on food resources. Marine Biology 154:163-174

Leduc D, Rowden AA, Bowden DA, Nodder SD, Probert PK, Pilditch CA, Duineveld GC, Witbaard R (2012) Nematode beta diversity on the continental slope of New Zealand: spatial patterns and environmental drivers. Marine Ecology Progress Series 454:37-52

Levin PS, Coyer JA, Petrik R, Good TP (2002) Community-wide effects of nonindigenous species on temperate rocky reefs. Ecology 83:3182-3193

Levin SA (1992) The problem of pattern and scale in ecology: the Robert H. MacArthur award lecture. Ecology 73:1943-1967

Levinton J, Kelaher B (2004) Opposing organizing forces of deposit-feeding marine communities. Journal of Experimental Marine Biology and Ecology 300:65-82

Lewis J (1964) The ecology of rocky shores. Universities Press, London:323

Loreau M (2000) Are communities saturated? On the relationship between α, β and γ diversity. Ecology letters 3:73-76

Loreau M, Mouquet N (1999) Immigration and the maintenance of local species diversity. The American Naturalist 154:427-440
Loreau M, Mouquet N, Gonzalez A (2003) Biodiversity as spatial insurance in heterogeneous landscapes. Proceedings of the National Academy of Sciences 100:12765-12770
Lorenzen S (1981) Entwurf eines phylogenetischen Systems der freilebenden Nematoden. Kommissionverlag Franz Leuwer
Mackie AS, Oliver PG, Darbyshire T, Mortimer K (2005) Shallow marine benthic invertebrates of the Seychelles Plateau: high diversity in a tropical oligotrophic environment. Philosophical Transactions of the Royal Society of London A: Mathematical, Physical and Engineering Sciences 363:203-228
Mayr E (1942) Systematics and the origin of species, from the viewpoint of a zoologist. Harvard University Press
McClain CR, Barry JP (2010) Habitat heterogeneity, disturbance, and productivity work in concert to regulate biodiversity in deep submarine canyons. Ecology 91:964-976
McDonald PS, Bingham BL (2010) Comparing macroalgal food and habitat choice in sympatric, tube-building amphipods, *Ampithoe lacertosa* and *Peramphithoe humeralis*. Marine Biology 157:1513-1524
McEwen GF, Johnson MW, Folsom TR (1954) A statistical analysis of the performance of the Folsom plankton sample splitter, based upon test observations. Archiv für Meteorologie, Geophysik und Bioklimatologie, Serie A 7:502-527
McLachlan A (1985) The biomass of macro-and interstitial fauna on clean and wrack-covered beaches in Western Australia. Estuarine, Coastal and Shelf Science 21:587-599
Meiners SJ, Pickett ST, Cadenasso ML (2001) Effects of plant invasions on the species richness of abandoned agricultural land. Ecography 24:633-644
Mews M, Zimmer M, Jelinski DE (2006) Species-specific decomposition rates of beach-cast wrack in Barkley Sound, British Columbia, Canada. Marine Ecology Progress Series 328:155-160
Michaud E, Desrosiers G, Aller RC, Mermillod-Blondin F, Sundby B, Stora G (2009) Spatial interactions in the Macoma balthica community control biogeochemical fluxes at the sediment-water interface and microbial abundances. Journal of Marine Research 67:43-70
Moens T, dos Santos GAP, Thompson F, Swings J, Fonsêca-Genevois V, Vincx M, De Mesel I (2005) Do nematode mucus secretions affect bacterial growth? Aquatic Microbial Ecology 40:77-83
Mouillot D, Mason WN, Dumay O, Wilson JB (2005) Functional regularity: a neglected aspect of functional diversity. Oecologia 142:353-359
Mukai H (1971) The phytal animals on the thalli of *Sargassum serratifolium* in the *Sargassum* region, with reference to their seasonal fluctuations. Marine Biology 8:170-182
Muniz P, Hutton M, Kandratavicius N, Lanfranconi A, Brugnoli E, Venturini N, Giménez L (2012) Performance of biotic indices in naturally stressed estuarine environments on the Southwestern Atlantic coast (Uruguay): a multiple scale approach. Ecological Indicators 19:89-97
Murphy R, Tolhurst T, Chapman M, Underwood A (2009) Seasonal distribution of chlorophyll on mudflats in New South Wales, Australia measured by field spectrometry and PAM fluorometry. Estuarine, Coastal and Shelf Science 84:108-118
Naeem S, Bunker DE, Hector A, Loreau M, Perrings C (2009) Biodiversity, ecosystem functioning and human wellbeing. Oxford university press

Nehring S, Hesse K-J (2008) Invasive alien plants in marine protected areas: the *Spartina anglica* affair in the European Wadden Sea. Biological Invasions 10:937-950

Nevo E (1995) Asian, African and European biota meet at'Evolution Canyon'Israel: local tests of global biodiversity and genetic diversity patterns. Proceedings of the Royal Society of London B: Biological Sciences 262:149-155

Norkko J, Bonsdorff E, Norkko A (2000) Drifting algal mats as an alternative habitat for benthic invertebrates:: Species specific responses to a transient resource. Journal of Experimental Marine Biology and Ecology 248:79-104

Norling K, Rosenberg R, Hulth S, Grémare A, Bonsdorff E (2007) Importance of functional biodiversity and species-specific traits of benthic fauna for ecosystem functions in marine sediment. Marine Ecology Progress Series 332:11-23

North WJ (1973) Regulating marine transplantation. Science 179:1181-1181

Norton TA, Benson MR (1983) Ecological interactions between the brown seaweed *Sargassum muticum* and its associated fauna. Marine Biology 75:169-177

Nozais C, Perissinotto R, Tita G (2005) Seasonal dynamics of meiofauna in a South African temporarily open/closed estuary (Mdloti Estuary, Indian Ocean). Estuarine, Coastal and Shelf Science 62:325-338

O'Connor NE, Crowe TP (2005) Biodiversity loss and ecosystem functioning: distinguishing between number and identity of species. Ecology 86:1783-1796

O'Hara TD, Poore GC (2000) Patterns of distribution for southern Australian marine echinoderms and decapods. Journal of Biogeography 27:1321-1335

Olabarria C, Lastra M, Garrido J (2007) Succession of macrofauna on macroalgal wrack of an exposed sandy beach: effects of patch size and site. Marine Environmental Research 63:19-40

Organization IH (1953) Limits of Oceans and Seas. International Hydrographic Organization

Orr M, Zimmer M, Jelinski DE, Mews M (2005) Wrack deposition on different beach types: spatial and temporal variation in the pattern of subsidy. Ecology 86:1496-1507

Park C, Hwang E, Sohn C (2003) A stable seeding method for Porphyra pseudolinearis Ueda (Rhodophyta): developing a new species for cultivation of Porphyra in Korea. Aquaculture Research 34:895-898

Parker JD, Duffy JE, Orth RJ (2001) Plant species diversity and composition: experimental effects on marine epifaunal assemblages. Marine Ecology Progress Series 224:55-67

Pejchar L, Mooney HA (2009) Invasive species, ecosystem services and human well-being. Trends in ecology & evolution 24:497-504

Pennings SC, Carefoot TH, Zimmer M, Danko JP, Ziegler A (2000) Feeding preferences of supralittoral isopods and amphipods. Canadian Journal of Zoology 78:1918-1929

Petchey OL, Gaston KJ (2002) Functional diversity (FD), species richness and community composition. Ecology Letters 5:402-411

Petchey OL, Gaston KJ (2006) Functional diversity: back to basics and looking forward. Ecology letters 9:741-758

Phillips N (1995) Biogeography of Sargassum (Phaeophyta) in the Pacific basin. Taxonomy of economic seaweeds 5:107-145

Pianka ER (1966) Latitudinal gradients in species diversity: a review of concepts. American Naturalist:33-46

Pingree R, Griffiths D (1980) Currents driven by a steady uniform wind stress on the shelf seas around the British-Isles. Oceanologica Acta 3:227-236

Piot A, Nozais C, Archambault P (2014) Meiofauna affect the macrobenthic biodiversity–ecosystem functioning relationship. Oikos 123:203-213

Platt H, Warwick R (1980) The significance of free-living nematodes to the littoral ecosystem.
Platt H, Warwick R, Price J, Irvine D, Farnham W (1980) The significance of free-living nematodes to the littoral ecosystem. Academic Press.
Platt HM, Warwick RM (1983) Freeliving marine nematodes. Part 1: British enoplids. Pictorial key to world genera and notes for the identification of British species. Cambridge University Press, for the Linnean Society of London and the Estuarine and Brackish-water Sciences Association
Plouguerné E, Le Lann K, Connan S, Jechoux G, Deslandes E, Stiger-Pouvreau V (2006) Spatial and seasonal variation in density, reproductive status, length and phenolic content of the invasive brown macroalga *Sargassum muticum* (Yendo) Fensholt along the coast of Western Brittany (France). Aquatic Botany 85:337-344
Polis GA, Anderson WB, Holt RD (1997) Toward an integration of landscape and food web ecology: the dynamics of spatially subsidized food webs. Annual review of ecology and systematics 28:289-316
Poore GC (1993) Marine species richness. Nature 361:597-598
Posey MH (1988) Community changes associated with the spread of an introduced seagrass, Zostera japonica. Ecology 69:974-983
Prado P, Thibaut T (2008) Differences between epiphytic assemblages on introduced *Caulerpa taxifolia* and coexisting eelgrass (*Zostera capricorni*) in Botany Bay (NSW, Australia). Scientia Marina 72:645-654
Puig P, Sardà F, Palanques A, Latasa M, Scharek R (2008) Climate influence on deep sea populations. PloS one 3:e1431
R CK (1993) Non-parametric multivariate analyses of changes in community structure. Australian Journal of Ecology 18:117-143
Race MS (1982) Competitive displacement and predation between introduced and native mud snails. Oecologia 54:337-347
Raffaelli DDG, Hawkins SJ (1996) Intertidal ecology. Springer
Ranjitham NS, Thirumaran G, Anantharaman P, Nightingale VDR, Balasubramanian R (2008) Associated Fauna of Seaweeds and Seagrasses in Vellar Estuary.
Rapoport E (1994) Remarks on marine and continental biogeography: an areographical viewpoint. Philosophical Transactions of the Royal Society of London B: Biological Sciences 343:71-78
Rex MA, Stuart CT, Hessler RR, Allen JA, Sanders HL, Wilson GD (1993) Global-scale latitudinal patterns of species diversity in the deep-sea benthos. Nature 365:636-639
Ricklefs RE (1987) Community diversity: relative roles of local and regional processes. Science(Washington) 235:167-171
Rodil IF, Olabarria C, Lastra M, López J (2008) Differential effects of native and invasive algal wrack on macrofaunal assemblages inhabiting exposed sandy beaches. Journal of Experimental Marine Biology and Ecology 358:1-13
Romeyn K, Bouwman L (1983) Food selection and consumption by estuarine nematodes. Aquatic Ecology 17:103-109
Roy K, Jablonski D, Valentine JW (2000) Dissecting latitudinal diversity gradients: functional groups and clades of marine bivalves. Proceedings of the Royal Society of London B: Biological Sciences 267:293-299
Rueness J (1989) *Sargassum muticum* and other introduced Japanese macroalgae: biological pollution of European coasts. Marine Pollution Bulletin 20:173-176
Rysgaard S, Christensen PB, Sørensen MV, Funch P, Berg P (2000) Marine meiofauna, carbon and nitrogen mineralization in sandy and soft sediments of Disko Bay, West Greenland. Aquatic Microbial Ecology 21:59-71

Salvaterra T, Green DS, Crowe TP, O'Gorman EJ (2013) Impacts of the invasive alga *Sargassum muticum* on ecosystem functioning and food web structure. Biological invasions 15:2563-2576

Schimel J, Balser TC, Wallenstein M (2007) Microbial stress-response physiology and its implications for ecosystem function. Ecology 88:1386-1394

Schmid-Araya J, Hildrew A, Robertson A, Schmid P, Winterbottom J (2002) The importance of meiofauna in food webs: evidence from an acid stream. Ecology 83:1271-1285

Schmidt AL, Scheibling RE (2006) A comparison of epifauna and epiphytes on native kelps (*Laminaria* species) and an invasive alga (*Codium fragile* ssp. *tomentosoides*) in Nova Scotia, Canada. Botanica marina 49:315-330

Schratzberger M, Bolam S, Whomersley P, Warr K (2006) Differential response of nematode colonist communities to the intertidal placement of dredged material. Journal of Experimental Marine Biology and Ecology 334:244-255

Schratzberger M, Warr K, Rogers S (2007) Functional diversity of nematode communities in the southwestern North Sea. Marine Environmental Research 63:368-389

Schreider M, Glasby T, Underwood A (2003) Effects of height on the shore and complexity of habitat on abundances of amphipods on rocky shores in New South Wales, Australia. Journal of Experimental Marine Biology and Ecology 293:57-71

Shirayama Y, Kaku T, Higgins RP (1993) Double-sided microscopic observation of meiofauna using an HS-slide. Benthos Research 1993:41-44

Smith B, Foreman R (1984) An assessment of seaweed decomposition within a southern Strait of Georgia seaweed community. Marine Biology 84:197-205

Smith JR, Vogt SC, Creedon F, Lucas BJ, Eernisse DJ (2014) The non-native turf-forming alga *Caulacanthus ustulatus* displaces space-occupants but increases diversity. Biological Invasions 16:2195-2208

Soininen J, Lennon JJ, Hillebrand H (2007) A multivariate analysis of beta diversity across organisms and environments. Ecology 88:2830-2838

Solan M, Aspden RJ, Paterson DM (2012) Marine biodiversity and ecosystem functioning: Frameworks, methodologies, and integration. Oxford University Press

Son MH, Hong SY (1998) Reproduction of Littorina brevicula in Korean waters. Marine Ecology Progress Series 172:215-223

Southward A (1991) Forty years of changes in species composition and population density of barnacles on a rocky shore near Plymouth. Journal of the Marine Biological Association of the United Kingdom 71:495-513

Southward A, Hawkins S, Burrows M (1995) Seventy years' observations of changes in distribution and abundance of zooplankton and intertidal organisms in the western English Channel in relation to rising sea temperature. Journal of thermal Biology 20:127-155

Srivastava DS (1999) Using local–regional richness plots to test for species saturation: pitfalls and potentials. Journal of Animal Ecology 68:1-16

Stachowicz JJ, Fried H, Osman RW, Whitlatch RB (2002) Biodiversity, invasion resistance, and marine ecosystem function: reconciling pattern and process. Ecology 83:2575-2590

Stachowicz JJ, Whitlatch RB (2005) Multiple mutualists provide complementary benefits to their seaweed host. Ecology 86:2418-2427

Stæhr PA, Pedersen MF, Thomsen MS, Wernberg T, Krause-Jensen D (2000) Invasion of *Sargassum muticum* in Limfjorden (Denmark) and its possible impact on the indigenous macroalgal community. Marine Ecology Progress Series 207:79-88

Stenton-Dozey J, Griffiths C (1983) The fauna associated with kelp stranded on a sandy beach. Sandy beaches as ecosystems. Springer

Tataranni M, Lardicci C (2010) Performance of some biotic indices in the real variable world: a case study at different spatial scales in North-Western Mediterranean Sea. Environmental pollution 158:26-34
Taylor RB, Cole RG (1994) Mobile epifauna on subtidal brown sea-weeds in northeastern New Zealand. Marine Ecology Progress Series 115:271-271
Thébault E, Huber V, Loreau M (2007) Cascading extinctions and ecosystem functioning: contrasting effects of diversity depending on food web structure. Oikos 116:163-173
Thistle D, Lambshead P, Sherman K (1995) Nematode tail-shape groups respond to environmental differences in the deep sea. Vie et Milieu 45:107-116
Thistle D, Sherman KM (1985) The nematode fauna of a deep-sea site exposed to strong near-bottom currents. Deep Sea Research Part A Oceanographic Research Papers 32:1077-1088
Thompson R, Wilson B, Tobin M, Hill A, Hawkins S (1996) Biologically generated habitat provision and diversity of rocky shore organisms at a hierarchy of spatial scales. Journal of Experimental Marine Biology and Ecology 202:73-84
Thorson G (1957) Bottom communities (sublittoral or shallow shelf). Geological Society of America Memoirs 67:461-534
Tietjen JH, Lee JJ (1973) Life history and feeding habits of the marine nematode, *Chromadora macrolaimoides* Steiner. Oecologia 12:303-314
Tittensor DP, Mora C, Jetz W, Lotze HK, Ricard D, Berghe EV, Worm B (2010) Global patterns and predictors of marine biodiversity across taxa. Nature 466:1098-1101
Tuomisto H (2010a) A diversity of beta diversities: straightening up a concept gone awry. Part 1. Defining beta diversity as a function of alpha and gamma diversity. Ecography 33:2-22
Tuomisto H (2010b) A diversity of beta diversities: straightening up a concept gone awry. Part 2. Quantifying beta diversity and related phenomena. Ecography 33:23-45
Tuomisto H, Ruokolainen K, Kalliola R, Ari L (1995) Dissecting amazonian biodiversity.
Underwood AJ (1997) Experiments in ecology: their logical design and interpretation using analysis of variance. Cambridge University Press
Underwood AJ, Chapman MG (2013) Design and analysis in benthic surveys in environmental sampling. Methods for the Study of Marine Benthos, Fourth Edition:1-45
Urban-Malinga B, Burska D (2009) The colonization of macroalgal wrack by the meiofauna in the Arctic intertidal. Estuarine, Coastal and Shelf Science 85:666-670
Urban-Malinga B, Gheskiere T, Degraer S, Derycke S, Opalinski KW, Moens T (2008) Gradients in biodiversity and macroalgal wrack decomposition rate across a macrotidal, ultradissipative sandy beach. Marine Biology 155:79-90
Valiela I (2013) Marine ecological processes. Springer Science & Business Media
Valiela I, Teal JM, Allen SD, Van Etten R, Goehringer D, Volkmann S (1985) Decomposition in salt marsh ecosystems: the phases and major factors affecting disappearance of above-ground organic matter. Journal of Experimental Marine Biology and Ecology 89:29-54
Vasas V, Jordán F (2006) Topological keystone species in ecological interaction networks: considering link quality and non-trophic effects. Ecological Modelling 196:365-378
Vázquez-Luis M, Sanchez-Jerez P, Bayle-Sempere J (2008) Changes in amphipod (Crustacea) assemblages associated with shallow-water algal habitats invaded by *Caulerpa racemosa var. cylindracea* in the western Mediterranean Sea. Marine Environmental Research 65:416-426

Veiga P, Rubal M, Sousa-Pinto I (2014) Structural complexity of macroalgae influences epifaunal assemblages associated with native and invasive species. Marine environmental research 101:115-123
Veiga P, Sousa-Pinto I, Rubal M (2016) Meiofaunal assemblages associated with native and non-indigenous macroalgae. Continental Shelf Research 123:1-8
Vetter E, Dayton P (1999) Organic enrichment by macrophyte detritus, and abundance patterns of megafaunal populations in submarine canyons. Marine Ecology Progress Series 186:137-148
Viejo RM (1999) Mobile epifauna inhabiting the invasive *Sargassum muticum* and two local seaweeds in northern Spain. Aquatic botany 64:131-149
Walker B, Kinzig A, Langridge J (1999) Original articles: plant attribute diversity, resilience, and ecosystem function: the nature and significance of dominant and minor species. Ecosystems 2:95-113
Walker RH, Brodie J, Russell S, Irvine LM, Orfanidis S (2009) Biodiversity of coralline algae in the north eastern Atlantic including *Corallina caespitosa* sp. Nov.(corallinoideae, rhodophyta) 1. Journal of phycology 45:287-297
Warwick R (1977) The structure and seasonal fluctuations of phytal marine nematode associations on the Isles of Scilly. Marine Biology of Benthic Organisms Keegan Pergamon Press, London:577-585
Warwick R (1987) Comparative study of the structure of some tropical and temperate marine soft-bottom macrobenthic communities. Marine Biology 95:641-649
Warwick R, Platt H, Somerfield P (1998) Free living marine nematodes. Part III. British Monhysterida. Synopses of the British fauna no 53. Field Studies Council, Shrewsbury
Wernberg T, Thomsen MS, Staehr PA, Pedersen MF (2004) Epibiota communities of the introduced and indigenous macroalgal relatives *Sargassum muticum* and *Halidrys siliquosa* in Limfjorden (Denmark). Helgoland Marine Research 58:154-161
Whittaker RH (1960) Vegetation of the Siskiyou mountains, Oregon and California. Ecological monographs 30:279-338
Whittaker RH (1967) Gradient analysis of vegetation. Biological reviews 42:207-264
Whittaker RH (1970) Communities and ecosystems. London, Macmillan Company, Collier-Macmillan Limited
Whittaker RJ, Willis KJ, Field R (2001) Scale and species richness: towards a general, hierarchical theory of species diversity. Journal of Biogeography 28:453-470
Wiens JA (1989) Spatial scaling in ecology. Functional ecology 3:385-397
Wieser W (1952) Investigations on the microfauna inhabiting seaweeds on rocky coasts (Untersuchungen über die algenbewohnende Mikrofauna mariner Hartböden). IV. Studies on the vertical distribution of the fauna inhabiting seaweeds below the Plymouth Laboratory. Journal of the Marine Biological Association of the United Kingdom 31:145-174
Wieser W (1953) Die Beziehung zwischen Mundhöhlengestalt, Ernährungsweise und Vorkommen bei freilebenden marinen Nematoden: eine ökologisch-morphologische Studie. Universitätsbibliothek Johann Christian Senckenberg:439-484
Wieser W (1959) The effect of grain size on the distribution of small invertebrates inhabiting the beaches of Puget Sound. Limnol Oceanogr 4:181-194
Wikström SA, Kautsky L (2004) Invasion of a habitat-forming seaweed: effects on associated biota. Biological Invasions 6:141-150
Wikström SA, Steinarsdóttir MB, Kautsky L, Pavia H (2006) Increased chemical resistance explains low herbivore colonization of introduced seaweed. Oecologia 148:593-601
Wilson EO, MacArthur RH (1967) The theory of island biogeography. Princeton, NJ

Witman JD, Etter RJ, Smith F (2004) The relationship between regional and local species diversity in marine benthic communities: a global perspective. Proceedings of the National Academy of Sciences of the United States of America 101:15664-15669

Worm B, Sommer U (2000) Rapid direct and indirect effects of a single nutrient pulse in a seaweed-epiphyte-grazer system. Marine Ecology Progress Series 202:283-288

Wright JP, Jones CG, Flecker AS (2002) An ecosystem engineer, the beaver, increases species richness at the landscape scale. Oecologia 132:96-101

Yoo J-S (2003) Biodiversity and community structure of marine benthic organisms in the rocky shore of Dongbaekseom, Busan. Algae 18:225-232

Zinger L, Amaral-Zettler LA, Fuhrman JA, Horner-Devine MC, Huse SM, Welch DBM, Martiny JB, Sogin M, Boetius A, Ramette A (2011) Global patterns of bacterial beta-diversity in seafloor and seawater ecosystems. PLoS One 6:e24570