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EARLY PLEISTOCENE NANNOFOSSIL BIOSTRATIGRAPHY IN RHODES ISLAND, AEGEAN SEA

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ΒΙΟΣΤΡΩΜΑΤΟΓΡΑΦΙΑ ΝΑΝΝΟΑΠΟΛΙΘΩΜΑΤΩΝ ΚΑΤΑ ΤΟ ΚΑΤΩΤΕΡΟ ΠΛΕΙΣΤΟΚΑΙΝΟ ΣΤΟ ΝΗΣΙ ΤΗΣ ΡΟΔΟΥ, ΑΙΓΑΙΟ ΠΕΛΑΓΟΣ

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EARLY PLEISTOCENE NANNOFOSSIL BIOSTRATIGRAPHY IN RHODES ISLAND, AEGEAN SEA – *Master Thesis*

ΒΙΟΣΤΡΩΜΑΤΟΓΡΑΦΙΑ ΝΑΝΝΟΑΠΟΛΙΘΩΜΑΤΩΝ ΚΑΤΑ ΤΟ ΚΑΤΩΤΕΡΟ ΠΛΕΙΣΤΟΚΑΙΝΟ ΣΤΟ ΝΗΣΙ ΤΗΣ ΡΟΔΟΥ, ΑΙΓΑΙΟ ΠΕΛΑΓΟΣ – *Μεταπτυχιακή Διπλωματική Εργασία*

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Abstract

Calcareous nannofossils are extremely useful for dating marine sediments as well as geological sections because they provide stable age indicators due to the abundance of nannofossil representatives in even minute amounts of sediment and their increased resistance to dissolution as well as their small life cycles. Three geological sections were studied biostratigraphically: Agathi Beach, Plimiri V and Plimiri VI in terms of calcareous nannoplankton, with the help of indicator species of the Late – Pliocene to Early – Pleistocene interval. In the Agathi Beach section, a greater abundance of the species *Calcidiscus macintyrei* was observed together with the complete absence of *Gephyrocapsa* >=4 μm. *Helicosphaera sellii*, is also abundant (up to 48% of the genus *Helicosphaera*), concluding that this section is older than the top of this species within CNPL 9 (Backman et al. 2012), in the Calabrian stage. Thus, Agathi Beach is assigned to Calcareous Nannofossil Zone (CNPL) 7 (Backman et al., 2012) that covers an interval between 1.93 and 1.71 Ma in the Early Pleistocene (Gelasian-Calabrian boundary) and is correlated with the lower part of Neogene Nannoplankton NN19 (Martini, 1971) biozone and Mediterranean Neogene Nannoplankton MNN19a (Rio et al., 1990). Plimiri V section displays *Discoaster pentaradiatus* up to 70%, together with *Discoaster tamalis* covering up to 48%, *Discoaster brouweri* up to 36% and *Discoaster assymetricus* up to 30%. Based on this assemblage the entire section is assigned to the Neogene Nannoplankton NN16 biozone of Martini (1971), which correlates to the Calcareous Nannofossil Zone (CNPL) 4 (3.81 Ma to 2.76 Ma) of Backman et al. (2012) in the Piacenzian. As far as the Plimiri VI section is considered, the dominant nannofossil is *Calcidiscus macintyrei* with max. more than 60%. The species *Helicospharea sellii* moves in slightly lower percentages, about 18% on average among the species of the genus *Helicospharea*, while *Gephyrocapsa* 3.5-4 μm individuals were measured to represent around 9% of the placoliths. At approximately the same levels as *Gephyrocapsa* 3.5-4 μm, the placolith *Pseudoemiliania lacunosa* is also observed. Finally, the general lack of *Gephyrocapsa* ≥ 4 μm is noteworthy, except of very few specimens in only two of the studied samples. The findings above indicate that the Plimiri VI section is biostratigraphically assigned to the lower part of the Martini biozone NN19, that correlates with biozone MNN19b of Rio et al. (1990) or to the lower part of CNPL8 biozone (Backman et al. 2012), above the base of *Gephyrocapsa* ≥ 4 μm at 1.71 Ma and below the top of the species *Calcidiscus macintyrei* at 1.60 Ma.

Περίληψη

Τα ασβεστολιθικά νανοαπολιθώματα είναι εξαιρετικά χρήσιμα για τη χρονολόγηση θαλάσσιων ιζημάτων καθώς και γεωλογικών τομών, επειδή παρέχουν σταθερούς δείκτες ηλικίας λόγω της αφθονίας των εκπροσώπων νανοαπολιθωμάτων σε ελάχιστες ποσότητες ιζήματος, της αυξημένης αντοχής τους στη διάλυση καθώς και των μικρών κύκλων ζωής τους. Μελετήθηκαν βιοστρωματογραφικά τρεις γεωλογικές τομές: η τομή Agathi Beach, η τομή Plimiri profil V και η τομή Plimiri profil VI ως προς το ασβεστολιθικό ναννοπλαγκτόν, με τη βοήθεια ειδών δεικτών του διαστήματος Ανωτέρου Πλειόκαινου έως Κατώτερου Πλειστόκαινου. Στην τομή Agathi Beach, παρατηρήθηκε μεγαλύτερη αφθονία του είδους *Calcidiscus macintyrei* μαζί με την πλήρη απουσία *Gephyrocapsa* >=4 μm. Το είδος *Helicosphaera sellii*, είναι επίσης άφθονο (έως και 48% του γένους *Helicosphaera)*, καταλήγοντας στο συμπέρασμα ότι αυτή η τομή είναι παλαιότερη από την τελευταία εμφάνιση αυτού του είδους στη CNPL 9 (Backman et al. 2012), στο Καλάβριο. Έτσι, η τομή Agathi Beach αντιστοιχεί στη βιοζώνη CNPL 7 (Backman et al., 2012) που καλύπτει ένα διάστημα μεταξύ 1,93 και 1,71 Ma στο Ανώτερο Πλειστόκαινο (όρια Γελάσιο-Καλάβριο) και συσχετίζεται με το κατώτερο τμήμα των βιοζωνών NN19 (Martini, 1971) και MNN19a (Rio et al. 1990). Στην τομή Plimiri profil V παρατηρήθηκε το είδος *Discoaster pentaradiatus* έως 70%, μαζί με τα *Discoaster tamalis* που καλύπτει έως και 48%, *Discoaster brouweri* έως 36% και *Discoaster assymetricus* έως 30%. Με βάση αυτή τη συνάθροιση, ολόκληρη η τομή αντιστοιχεί στη βιοζώνη NN16 (Martini, 1971) η οποία συσχετίζεται με τη CNPL 4 (3,81 Ma έως 2,76 Ma) των Backman et al. (2012) στο Πλακέντιο. Όσον αφορά την τομή Plimiri VI, το κυρίαρχο είδος είναι το *Calcidiscus macintyrei* με μέγιστα ποσοστά εμφανίσεως που ξεπερνούν το 60%. Το είδος *Helicospharea sellii* ακολουθεί με ελαφρώς χαμηλότερα ποσοστά, περίπου 18% κατά μέσο όρο μεταξύ των ειδών του γένους *Helicospharea*, ενώ τα άτομα *Gephyrocapsa* 3,5-4 μm μετρήθηκαν ότι αντιπροσωπεύουν περίπου το 9% των πλακόλιθων. Στα ίδια περίπου επίπεδα με τη *Gephyrocapsa* 3,5-4 μm, παρατηρείται και το είδος *Pseudoemiliania lacunosa*. Τέλος, αξιοσημείωτη είναι η γενική έλλειψη *Gephyrocapsa* ≥ 4 μm, εκτός από πολύ λίγους αντιπροσώπους σε δύο μόνο από τα δείγματα που μελετήθηκαν. Τα παραπάνω ευρήματα υποδεικνύουν ότι η τομή Plimiri VI αντιστοιχεί βιοστρωματογραφικά με το κατώτερο μέρος της βιοζώνης NN19 (Martini, 1971), που συσχετίζεται με τη βιοζώνη MNN19b των Rio et al. (1990) ή στο κάτω μέρος της βιοζώνης CNPL8 (Backman et al. 2012), πάνω από την πρώτη εμφάνιση του είδους *Gephyrocapsa* ≥ 4 μm στα 1,71 Ma και κάτω από την τελευταία εμφάνιση του είδους *Calcidiscus macintyrei* στα 1,60 Ma.

1. PURPOSE OF THE STUDY

The main purpose of this master thesis is the biostratigraphic study of the calcareous nannofossils of the island of Rhodes during the Late – Pliocene to Early – Pleistocene. The study includes the analysis of 44 samples from three geological sections located in the southeastern part of the island. These three sections are Agathi Beach, Plimiri 5 and Plimiri 6. With the support of previous research, inferences regarding paleoenvironmental conditions can also be inferred from the biostratigraphic examination of these sections, which aims to establish the biozones that signify geological time periods. The determination of biozones is based on the identification of representative index species of coccolithophores of Pliocene and Pleistocene. These are indicator species of different genera, such as the species of the genus *Gephyrocapsa* (<4μm, ≥4μm, >5.5μm) as well as the species *Calcidiscus macintyrei*, *Helicosphaera sellii*, *Pseudoemiliania lacunosa* etc. The specimens that classify to genus *Discoaster* provide a detailed biostratigraphic and chronostratigraphic assignment of the investigated deposits as well.

2. STUDY AREA

2.1. INTRODUCTION

Rhodes, a Dodecanese Island, is located within the warm temperate marine climate zone, at the oceanographic interface between the southern Aegean Sea to the north and the eastern Mediterranean Sea to the south (Betzler et al. 1997). In addition, it is located at the eastern end of the tectonically active Hellenic arc-trench subduction system, which marks the African-Eurasian plate boundary (Nelson et al., 2001). The African Plate was subducted into the Aegean-Anatolian Plate during the Cenozoic period, creating the island of Rhodes. When the Arabian and Eurasian plates collided during the Pliocene, the Aegean Sea began to collapse and the Aegean-Anatolian Plate began to move laterally westward (Facenna et al. 2006, Gautier et al. 1999).

2.2. GEOLOGICAL SETTING

Transgressive Pliocene and Pleistocene strata compensate the northeastern portion of Rhodes, which rests on a distorted and severely degraded, primarily calcareous Mesozoic basement (Lekkas et al. 2001, Mutti et al. 1970). This basement has been faulted, producing a few horsts and grabens that later influenced the kind and distribution of coastal Pliocene-Pleistocene deposits (Hanken et al. 1996). As a result, changes in sedimentary phases are frequent. Three main formations can be distinguished, in northeastern Rhodes, based on the sedimentary phases.

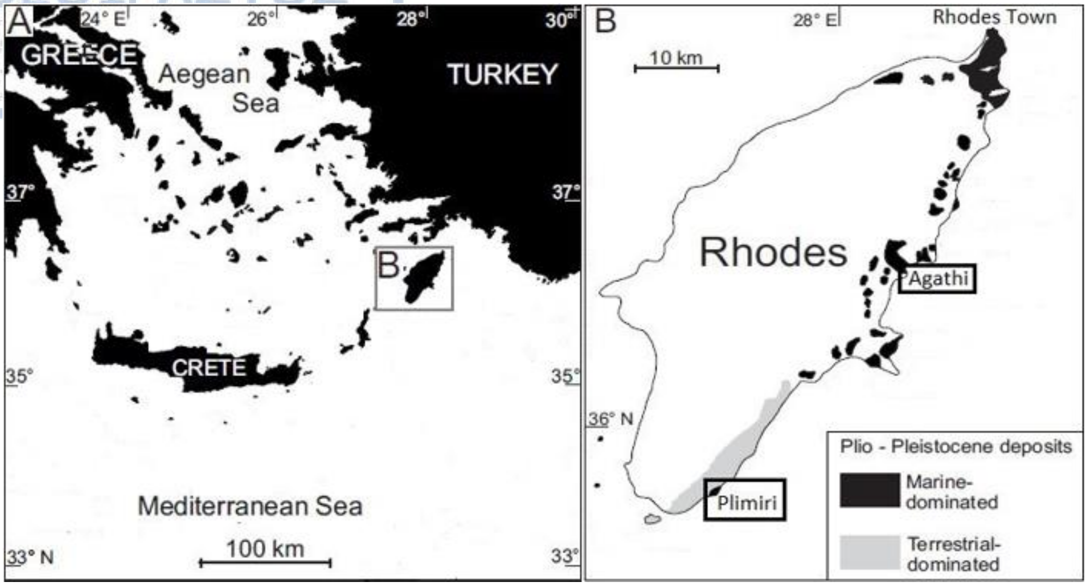


Figure 1. A – Map of southern Aegean Sea, B – Map of the study area, island of Rhodes, demonstrating the sections investigated (modified from Nelson et al. 2001).

Kritika Formation

It is a lithologically variable formation, both vertically and laterally, with silty facies poor in fossils in its southern part. The northern part of the formation consists of coarse- and fine-grained sandstones of continental origin, which include some horizons of polymictic conglomerates (including jasper, limestone and sandstone) with interlayers of silt.

Rhodes Formation

From bottom to top, this formation is made up of six facies groups (Beckman, 1995; Hanken et al., 1996). (a) Haraki limestone. A 3m thick, karstified at the top subtidal red algal and bioclastic bed that is patchily developed. It is most likely Late Pliocene in age. (b) Kolymbia limestone. A fossiliferous bioclastic limestone that ranges in thickness from 4 to 20m and lies unevenly on the bioeroded Mesozoic basement. (Beckman, 1995; Hanken et al., 1996; Spjeldnaes and Moissette, 1997; Steinthorsdottir, 2002) The depositional environment shifts from shoreface at the base to lower offshore in its upper section. Løvlie et al.'s (1989) palaeomagnetic findings support a Late Pliocene age. (c) Lindos Bay clay. Pleistocene sediments with an estimated maximum depth of deposition of 400–600 m, consisting of around 30 m of fossiliferous blue–grey calcareous to silty clays. A 3 Ma age at the base and a 0.7 Ma age at the top of the sequence are inferred from magnetostratigraphic analyses by Løvlie et al. (1989). (d) St. Paul's Bay limestone. Boundstones made of deep-sea coral that cover the perimeter of palaeovalleys that were filled with Lindos Bay clay (Hanken et al., 1996). (e) Ladiko sand. Alternating sands, sandy clays, and silts spanning approximately 30 m. According to Broekman (1972, 1974), the age should not be older than the Late Pliocene. (f) Cape Arkhangelos calcarenite. Grainstones and packstones originated from clinoforms reaching a total thickness of approximately 30 m (Beckman, 1995; Hanken et al., 1996; Hansen, 1999). The highest portion of the Cape Arkhangelos calcarenite has been reduced by erosion. Hanken et al. (1996) estimated that it was deposited during the Pleistocene.

Lindos Acropolis Formation

This formation consists of three groups of lithological phases: irregular blocks of eroded grainstone-packstone with a local matrix of calcarenite called Windmill Bay boulder bed and red-algal biolithite intergrown with laminar stromatolites called Gialos algal biolithite. The last group, called Kleopolu calcirudite, contains planar-laminated, cross-bedded or bioturbated coarse algal grainstones, with packstone beds and clasts that occur throughout, including limestone, calcarenite and algal biolithite.

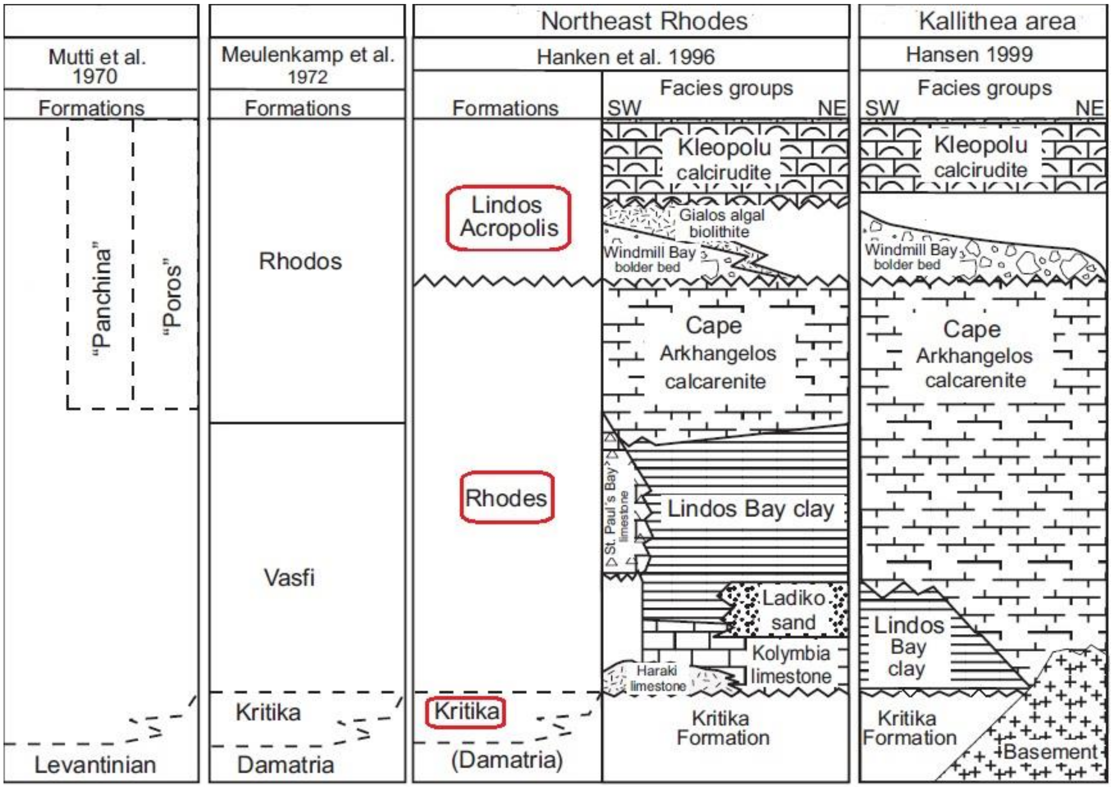


Figure 2. Lithostratigraphy of the Plio-Pleistocene deposits of Northeast Rhodes (modified by Nelson et al. 2001)

The hemipelagic blue-grey calcareous to silty Lindos Bay clay (45 m maximum thickness; Vasfi Formation in Meulenkamp et al., 1972) record the most pronounced tectonic drowning of the eastern part of Rhodes, with a maximum paleo-water depth estimated at 3000 m (Moissette and Spjeldnaes, 1995). According to estimates, the LBF was deposited during the Piacenzian to the Ionian (Lvliee et al., 1989), late Gelasian to early Calabrian (Cornee et al., 2006a, 2006b), Calabrian to early Ionian (Titschack et al., 2013), or late Calabrian (Thomsen et al., 2001). Due to its importance in reconstructing the geodynamic history of the Hellenic sedimentary fore-arc, researchers have been studying the tectonic and sedimentary evolution of Rhodes for a number of decades (Meulenkamp et al., 1972; Pirazzoli et al., 1989; Duermeijer et al., 2000; Ten Veen and Kleinspehn, 2002). Since the Pleistocene, Rhodes has experienced significant vertical movements, anticlockwise rotation, and tectonic activity controlled by major N70º trending sinistral strike-slip faults (e.g., Benda et al., 1977; Flemming and Woodworth, 1988; Hanken et al., 1996; Cornee et al., 2006a; van Hinsbergen et al., 2007; Ten Veen et al. 2009). Additionally, previous research suggests that Rhodes suffered some significant vertical motions between 2.0 and 1.4 Ma and a counterclockwise rotating phase after 1.8 Ma (Duermeijer et al. 2000). Mesozoic to Cenozoic (Jurassic to Paleocene) rocks, primarily carbonates, make up the basement of Rhodes. These rocks are covered with Mio-Pliocene terrestrial clastic deposits (Meulenkamp et al. 1972). Pleistocene marine sediments and Pliocene clastic fluvial and lacustrine sediments were deposited in solitary basins, mainly along Rhodes' eastern coast (Mutti et al. 1970). The island's basement rocks are made up of a diverse range of Mesozoic and Tertiary ophiolites, volcanic, siliciclastic sedimentary, and carbonate rocks that were all folded and faulted during the Alpine orogeny (Mutti et al. 1970). Plio-Pleistocene deposits are widely distributed along the island's eastern coast, stretching from Rhodes town in the north to Cape Prasonision in the south (Meulenkamp et al. 1972; Hanken et al. 1996).

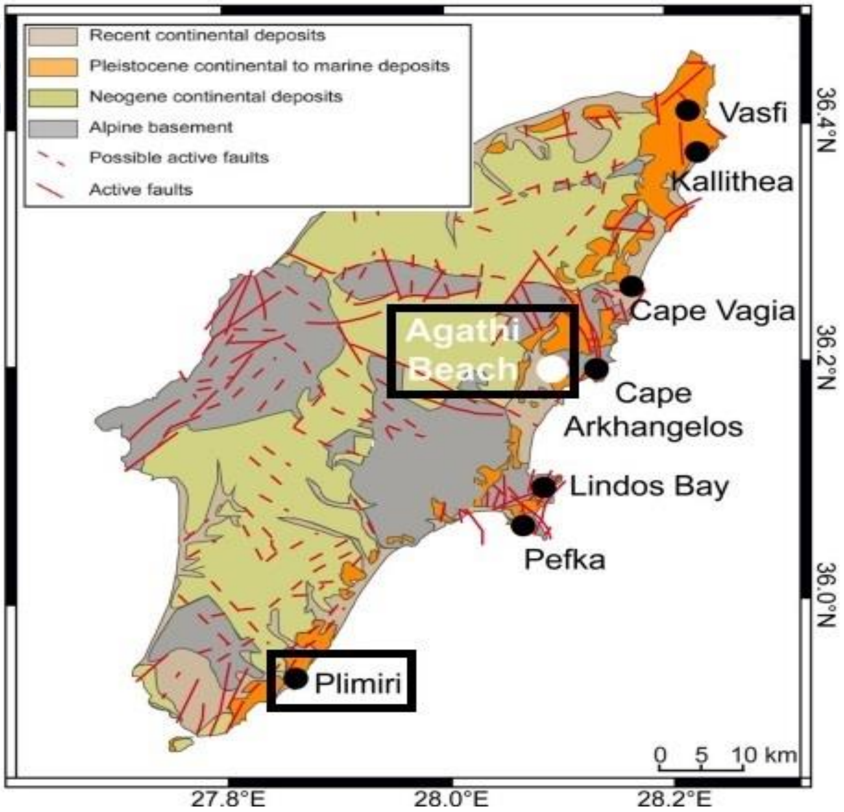


Figure 3. Geological map of Rhodes Island (modified from Eincher et al. 2024) demonstrating the distribution of the geological deposits according to their chronostratigraphic assignment. The sections studied in this work are framed.

Pleistocene deep-sea sedimentary formations that are presently emerging on the island's eastern coast are a result of such vertical motions. According to Mutti et al. (1970) and Lekkas et al. (2001), these deposits lay on a distorted and severely eroded, primarily calcareous Mesozoic basement. A number of steep horsts and grabens were created as a result of the numerous faulting in this basement, which later influenced the kind and location of the Pleistocene deposits (Hanken et al., 1996). Sedimentary facies alterations are frequent inside each graben. The chronostratigraphy of these marine deposits is still poorly understood despite recent advances using a variety of methods, such as bio-, magneto-, and/or radioisotope dating (Thomsen et al., 2001; Cornee et al., 2006a, 2006b). This is because correlations between separated infillings are challenging to establish.

3. CALCAREOUS NANNOFOSSILS

3.1. INTRODUCTION

Coccolithophores are haptophytes with calcified scales on their cell walls at some point throughout their life cycle (coccoliths). They are a key component of oceanic phytoplankton and play an important part in marine carbonate biogeochemistry, and thus in the carbon cycle (Rost and Riebesell 2004). Coccoliths' shape and crystallography are taken into account while classifying them. Among the coccolithophores, there are two structurally distinct types of coccoliths: heterococcoliths and holococcoliths. The heterococcoliths are made up of crystal units of varying shape and size, whereas the holococcoliths are made up of multiple minute (<0.1 μm) crystallites (Young et al. 1999). Coccolithophores primarily reproduce asexually through miotic division, in which the coccoliths are spread among the daughter cells. A complicated life cycle with two or more physically distinct phases has been seen in several haptophytes. The normal coccolithophore life cycle consists of a motile, haploid stage that is typically covered by holococcoliths and a diploid stage that is covered by heterococcoliths (Billard, 1994; Billard and Inuoye, 2004). Studies of culture have shown that the life cycle can change (Parke and Adams, 1960; Cros et al., 2000; Geisen et al., 2002). Calcareous nannofossils are extremely useful as index fossils for dating marine sediments as well as geological sections because they provide consistent age markers. Coccolithophores are common and widespread marine plankton components. Both the calcification of coccoliths and photosynthesis play key roles in the oceanic carbon cycle (Rost and Riebesell 2004). The foundation of the marine food chain is made up of coccolithophores and other phytoplankton. The cell contents of zooplankton-preyed coccolithophores are digested and assimilated, while the calcareous coccoliths are expelled and integrated into fecal pellets. These fecal pellets soon settle to the seafloor, and loose coccoliths and coccospheres frequently mix with other plankton waste to generate marine snow (Honjo 1976). These two mechanisms prevent coccoliths from dissolving in the water column and considerably increase their sinking velocity. Fecal pellets sink 150–570 m per day in the water column, but loose coccoliths sink less than 1 meter per day (Honjo 1976; Steinmetz 1994; Fischer and Karakas 2009). Coccoliths are quickly carried to the sea floor as a result of these two mechanisms and represent one of the major components of marine sediments above the Calcite Compensation Depth (CCD). Despite this, significant dissolution and assemblage alteration do occur, as evidenced by the fact that only roughly 60 coccolithophore types are known from the fossil record out of 280 identified from plankton (Young et al. 2005). Calcareous nannofossils were chosen in this study because research on deposits from different areas of the Mediterranean's Pliocene and Pleistocene periods has demonstrated that this group offers a dependable biostratigraphic framework with a very high stratigraphic resolution (e.g., Raffi & Rio, 1979; Rio et al., 1990; de Keanel et al., 1999).

Calcareous nannofossils are a taxonomically challenging group of microfossils due to the heteromorphic life cycles of the planktonic algae that create them, which allow the organism to excrete various coccoliths, organic scales, or bare cells (e.g., Billard & Inouye 2004). Every species has the potential to create a variety of coverings, each of which, when found in sediments or assemblages, is referred to be a distinct independent species. Moreover, visually similar calcite plates might reflect a cryptospecies rather than the same species (De Vargas et al. 2004). Calcareous nannofossils and coccoliths were originally discovered in 1836 by Christian G. Ehrenberg, who characterized these odd "morpholites" from a Cretaceous chalk outcropping in the Baltic Sea. Following this significant finding, in 1858, Thomas H. Huxley described the coccolithophores as a large number of extremely odd circular structures made up of numerous concentric layers and initially resembling single cells. Three years later, Wallich observed strange spheroidal bodies that he terms "coccospheres" in addition to the coccoliths noted by Huxley. He proposed that the coccoliths originate from the coccospheres (Huxley 1858; Wallich 1861). Henry C. Sorby also released a report, in 1861, outlining the coccoliths he discovered. At that time, coccoliths were recognized as organic in nature, but their significance in the dating of rocks and their part in the biogeochemical cycles were far to be understood, thus coccoliths/nannoliths would have remained just a scientific curiosity for a long time. Following the HMS Challenger Mission (1872–1876), Murray & Blackman (1898) published a significant research on biology and biogeography. Lohmann's work (1902), which first detected the presence of flagella and coined the name "nannoplankton", appeared shortly after. After these strange organisms were thoroughly inspected, researchers soon discovered that they exhibit a wide range of structures and shapes, allowing the creation of taxonomic groups based on similar traits. Yet, the polymorphic life cycle's complexity, changes brought on by the environment, and diagenetic modifications (preservation) may make it challenging to classify them and leave the species notion unclear. The understanding that each taxon has a distinct distribution in space and time was the following step. From the middle of the 20th century, their stratigraphical significance gradually became apparent to the geological world as a result of a seminal publication that demonstrated for the first time their use in biostratigraphy and rock dating (Bramlette & Riedel 1954). These authors did, in fact, give the first stratigraphical distribution of calcareous nannofossil taxa, known as a range chart, which served as a crucial foundation for the advancement of the calcareous nannofossil biostratigraphy.

Dinoflagellates and diatoms are two other planktonic microfossil groupings with a close connection to nannoplankton's geological past. In the Triassic, these three new groups first appeared in the fossil record (Falkowski et al. 2004; De Vargas et al. 2007). The development of biomineralization by nannoplankton and the associated fossil remains marked a crucial turning point in the evolution of the entire marine ecosystem. It also altered pelagic sedimentation and the generation of biogenic carbonate. In the setting of the ocean, their advent in the Triassic marked a turning point, particularly for the marine carbonate system and the global carbon cycle. In the modern ocean, dinoflagellates, diatoms, and calcareous nannoplankton all contribute significantly to the export flux of organic matter to the ocean's sediments. They also produce and export the majority of the ocean's carbonate (Milliman 1993; Falkowski et al. 2004). This group, along with planktonic foraminifera, took action during the Mesozoic to move the main location of carbonate production from shallow water habitats to the open ocean (Brownlee & Taylor 2004; Hay 2004). Around 220 Mya, calcareous nannoplankton began to produce mineralized skeletons and evolved in response to alterations in the geosphere, ocean chemistry, and structure. The Jurassic and Cretaceous experienced increases in biodiversity and rates of evolution, which continued into the Cenozoic. The relatively quick evolutionary changes led to a number of first and last appearance events, which served as the solid basis for high resolution biostratigraphical schemes that were successfully applied over a variety of geological time periods.

3.2. BIOSTRATIGRAPHIC ZONATION

A biostratigraphic unit, also known as a biozone, is a group of strata that are distinguished by their distinctive content, sequential distribution, lack of fossils, or combinations of these factors.

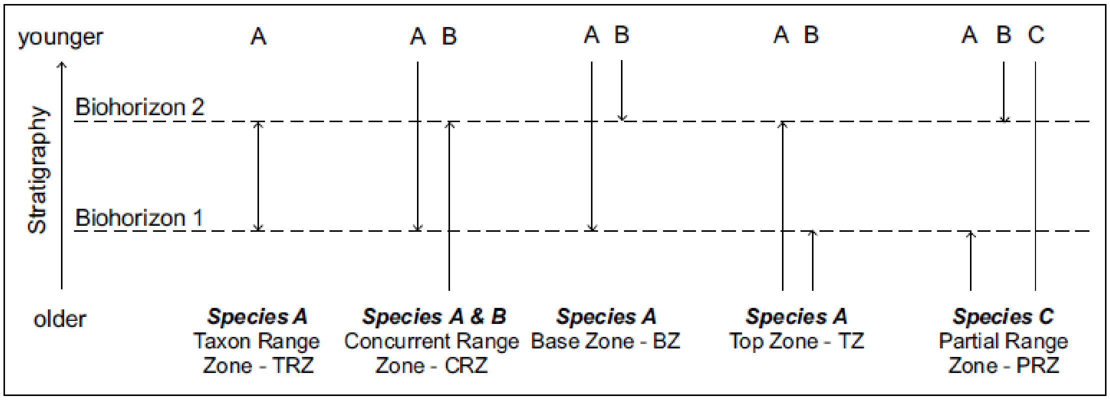


Figure 4. The five logical possibilities for biostratigraphic characterization of biozones (Backman et al., 2012).

A series of calcareous nannofossil biostratigraphic zonations for various areas of the Cenozoic stratigraphic column were established about 40 years ago (Hay et al. 1967; Gartner 1969, 1971). All of these were based on research into marine land sections and/or rotary cored Deep Sea Drilling Project sediments (JOIDES Journal, June 1979). Martini (1971) and Bukry (1973, 1975, 1978) produced biozonations for the entire Cenozoic. Okada and Bukry (1980) codified Bukry's 19 Paleogene and 15 Neogene zones (CP/CN zones), whereas Martini introduced 25 Paleogene and 21 Neogene zones (NP/NN zones). Through the analysis of high-quality deep-sea and on-land sections, numerous authors have improved standard zonation and attempted to increase the biostratigraphic resolution over the past few decades. 20 Paleogene subzones and 24 Neogene subzones were also codified by Okada and Bukry in 1980. According to Bukry (1973a), the material required to adequately examine the stratigraphic and geographic ranges of coccolith species will come from the ongoing recovery of deep-ocean sediment sections at various latitudes. The aim of Bukry's zonation, which merely serves to show the fundamentals of a low-latitude open ocean coccolith zonation, was not to be exhaustive. Instead of providing the best resolution, his objective was to develop a generic framework for relative dating of open ocean sediments. Additional bioevents (biohorizons) have been demonstrated through significant effort, using a variety of taxonomic methodologies and analytical techniques, primarily in the Early and Middle Pleistocene interval. *Gephyrocapsa* is the main genus represented in these more recent biohorizons (Samtleben, 1980; Rio, 1982; Takayama and Sato, 1987; Matsuoka and Okada, 1990). Since the first detailed Pleistocene nannofossil 'standard' zonation was published by Gartner (1977), its usefulness for biostratigraphic classification was tested in many sedimentary successions worldwide (e.g. Raffi and Rio, 1979). The significant biohorizons of the early to Early-Middle Pleistocene interval are the following: the first occurrence (base) of medium-sized *Gephyrocapsa* spp., the last occurrence (top) of *Calcidiscus macintyrei*, the first occurrence (base) of large *Gephyrocapsa* spp., the last occurrence (top) of large Gephyrocapsa spp., the first occurrence (base) of *Reticulofenestra asanoi*, the re-entrance of medium-sized *Gephyrocapsa* spp., the last occurrence (top) of *R. asanoi* (considered by Wei, 1993). The re-entrance of medium-sized *Gephyrocapsa* is equivalent to the first occurrence of *Gephyrocapsa* sp. 3 of Rio (1982). Rio (1974) was the first in an ongoing effort to demonstrate Cenozoic calcareous nannofossils as a tool to generate biostratigraphic data. Backman et al. (2012) synthesized Miocene through Pleistocene data for the establishment of a basic biostratigraphic framework to date marine sediments using calcareous nannofossils.

3.3. BIOSTRATIGRAPHY IN THE MEDITERRENEAN SEA

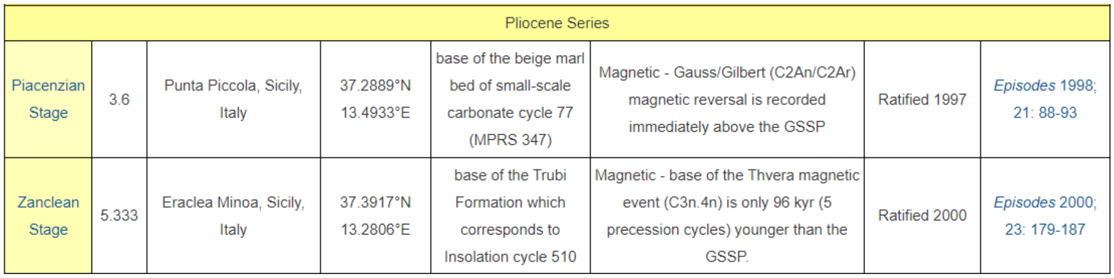
3.3.1. Introduction – historical review

The major characteristics of calcareous nannofossils' immense significance in terms of extensive biostratigraphic correlations are their vast abundance in marine sediments, wide geographic range, and quick evolutionary trends. Calcareous nannofossils have become the main source for biostratigraphic correlations of deep-sea sediments during the DSDP (Deep Sea Drilling Project) due to the abundance of nannofossil representatives in even minute amounts of sediment and their increased resistance to dissolution compared to planktonic foraminifera. The discovery of uninterrupted sequences of deep-sea sediments from the DSDP cores has tremendously benefited efforts to describe and determine the stratigraphic range of a large number of taxa and resulted in the establishment of biozones. The importance of calcareous nannofossils for biostratigraphy was covered in a seminal work written by Milton N. Bramlette and William R. Riedel in 1954. In around

15 years, this field of study had developed to the point where calcareous nannofossil biozonations had started to form for several Cenozoic stages (Hay et al., 1967; Gartner, 1969; Bukry & Bramlette, 1970). In 1968 scientific deep-sea drilling started and the result was the first calcareous nannofossil biozonation based entirely on deep-sea drilling sediments, spanning the whole Cenozoic Era (Burky, 1973). Within twenty years of the development of calcareous nannofossil biostratigraphy as an area of study, several early calcareous nannofossil biozonations were created. The biozonations created by David Bukry (1973, 1975) and Erlend Martini (1971) are still in use today, despite the fact that there is a growing need for revision in light of the new calcareous nannofossil biostratigraphic information that has emerged over the previous four decades. Within the past thirty years, a lot of calcareous nannofossil biostratigraphic information has become accessible. These data mostly represent low- and middle-latitude settings from deep-sea sediments as well as from marine on-land sections in the western Tethys and Mediterranean. Two new calcareous nannofossil biozonations have been resulted, one by Backman et al. (2012) focused on the Neogene and Pleistocene and the other by Agnini et al. (2014) focusing on the Paleogene. The two new biozonations share the critical characteristic that neither of them aims to establish the highest resolution in terms of the number of biozones but instead employs a small number of carefully chosen biohorizons to create an essential and stable biostratigraphic framework for the relative dating of marine sediments in low and middle latitude settings using calcareous nannofossils. 24 biohorizons' ages for the Pliocene–Pleistocene interval are provided by Backman et al. (2012). ODP Sites 926 (latitude 4°N; Ceara Rise, western tropical Atlantic), 925 (latitude 4°N), 607 (middle latitude North Atlantic; latitude 41°N), and 653 (latitude 40°N; Tyrrhenian Sea) were used to calculate 21 of these 24 age estimations.

3.3.2. Pliocene chronostratigraphy and biostratigraphic events

The Pliocene Epoch consists of two stages, the Zanclean and the Piacenzian stage. Outside of the Mediterranean, the beginning of the Pliocene is associated directly (Benson and Hodell, 1994; Benson and Rakic-El Bied, 1996). At the base of the Zanclean Stage in the Eraclea Minoa section, the base of the carbonate bed designating the small-scale stratigraphic cycle 1 (Hilgen, 1987; Hilgen and Langereis, 1988; Langereis and Hilgen, 1991) corresponds to insolation cycle 510 counted from the present, with an astronomically calibrated age of 5.33 Ma (Lourens et al., 1996). In addition to the orbitally forced geochemical and isotopic variations seen in many ODP Sites drilled recently (such as Leg 138: Shackleton et al., 1995; Leg 154: Shackleton and Crowhurst, 1997; Backman and Raffi, 1997), lithostratigraphical evidence for this precise level can also be found in the cyclic sequences of climate-sensitive depositional settings (such as high-productivity basins and abyssal floors). The base of the Thvera magnetic event, which was dated to 5.236 Ma (Lourens et al., 1996) and is only 96 kyrs (5 precession cycles) younger than the postulated GSSP, is a second, legitimate criterion. It also serves as an excellent anchor point for cyclostratigraphic or isotopic calibration of the boundary itself. This is a useful pointer to the boundary in continental, igneous, and non-cyclic marine deposits. The finest marine biostratigraphic tool for the correlation of the border outside the Mediterranean is provided by calcareous nannofossils. Close to the defined Zanclean (and Pliocene) GSSP, there are three significant biological occurrences that are globally recognizable and are included in the standard zonations of Martini (1971) and Okada and Bukry (1980). In order of proximity to the border, the earliest of them is the first appearance of *Ceratolithus acutus*, dated at 5.37 Ma in the equatorial Atlantic (Backman and Raffi, 1997), just 40 kyr before the proposed GSSP. It should be mentioned that *C. acutus* specimens have been found as far south as the very base of the Mediterranean Zanclean (Cita and Gartner, 1973; Castradori, 1998). The second is the disappearance of *Triquetrorhabdulus rugosus*, calibrated at approximately 5.23 Ma in both the equatorial Atlantic Ocean (Backman and Raffi, 1997) and the Mediterranean's lowermost Zanclean layers (Di Stefano et al., 1996; Castradori, 1998). The third is the last occurrence of *Discoaster quinqueramus*. Due to the Salinity Crisis, this event was not recorded in the Mediterranean, but it is dated at 5.537 Ma elsewhere (Backman and Raffi, 1997).



Pliocene Series							
Piacenzian Stage	3.6	Punta Piccola, Sicily, Italy	37.2889°N 13.4933°E	base of the beige marl bed of small-scale carbonate cycle 77 (MPRS 347)	Magnetic - Gauss/Gilbert (C2An/C2Ar) magnetic reversal is recorded immediately above the GSSP	Ratified 1997	*Episodes* 1998; 21: 88-93
Zanclean Stage	5.333	Eraclea Minoa, Sicily, Italy	37.3917°N 13.2806°E	base of the Trubi Formation which corresponds to Insolation cycle 510	Magnetic - base of the Thvera magnetic event (C3n.4n) is only 96 kyr (5 precession cycles) younger than the GSSP.	Ratified 2000	*Episodes* 2000; 23: 179-187

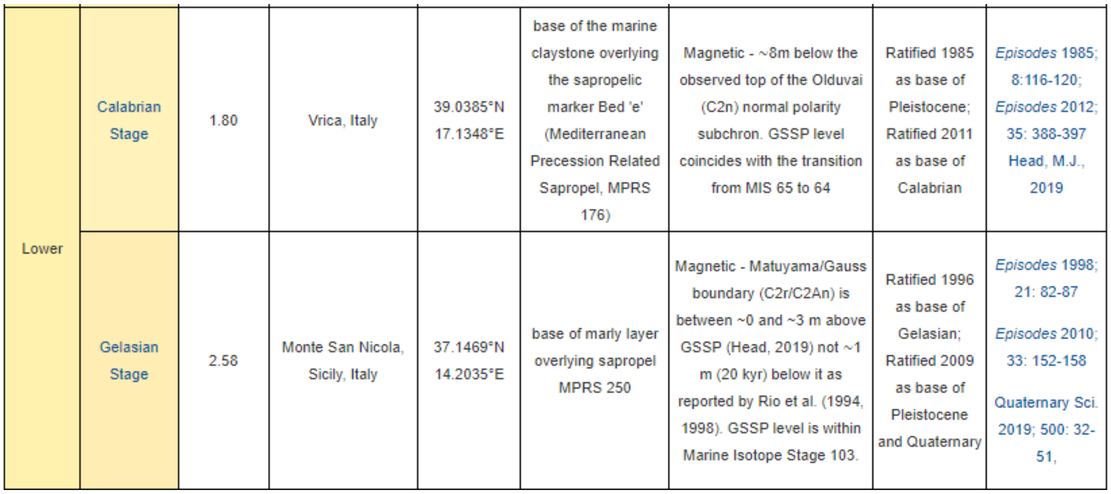
Figure 5. Pliocene Series and the defined stage boundaries, based on the stratigraphic column information from the International Commission on Stratigraphy (www.stratigraphy.org).

The Piacenzian Stage was first described by Mayer-Eymar in 1858, and it has since gained a lot of attention in geologic literature. Pareto (1865) quickly accepted it and made evident that the fossiliferous layers that were outcropping between Castell' Arquato and Lugagnano (Northern Apennine) were representative of the unit. A hiatus is recognized immediately at the base of the type-Piacenzian, as was clearly shown by Rio et al. (1988) and Raffi et al. (1989) in their integrated calcareous plankton biostratigraphic investigation. According to these authors, the Piacenzian base is located between the last known occurrence (LO) of *Reticulofenestra pseudoumbilicus*, which was proven to be 3.89 Ma, and the brief absence of the foraminifera *Globorotalia puncticulata* in the Mediterranean (3.57 Ma). As soon as the focus had to shift away from the Castell'Arquato area for two very clear reasons, it did so to the Punta Piccola area. To begin with, the Italian stratigraphic record is regarded as the type of the Pliocene Series (Langereis & Hilgen, 1991; Hilgen, 1991b; Berggren et al., 1995a; Lourens et al., 1996a). Secondly, the Rossello Composite Section, of which the Punta Piccola is the upper segment, has recently emerged as a reference standard for Pliocene Time Scales. The Caltanissetta Basin is where the Punta Piccola portion is located. From a structural standpoint, the segment is a part of the Gela nappe or Gela thrust system, a significant tectonic structure (Ogniben, 1969; Butler et al., 1995). In the Punta Piccola section calcareous nannofossils were studied by Rio et al. (1984). The section can be related to Zone NN16 of Martini (1971), Zone MNN16a to MNN16b/17 of Rio et al., 1990, and Subzones CN12a and CN12b of Okada & Bukry (1980).

3.3.3. Pleistocene chronostratigraphy and biostratigraphic events

There have typically been three divisions within the Pleistocene Series. At the Second International Congress of the Association pour l'étude du Quaternaire européen, which served as the forerunner to INQUA and its congresses, held in Leningrad in 1932, the names Lower, Middle, and Upper Pleistocene were in use (Woldstedt, 1953). In the former USSR and Russia, a somewhat different scheme developed that, in essence, dates back to the Leningrad Conference and uses the Eopleistocene as a proxy for the Gelasian and Calabrian and the Neopleistocene as a proxy for the Middle and Upper Pleistocene. According to Tesakov et al. (2015) and Head and Gibbard (2015), the Neopleistocene has lower, middle, and upper subdivisions, with the High Neopleistocene being a perfect match for the Upper Pleistocene.

The Lower Pleistocene Subseries comprises the Gelasian Stage followed by the Calabrian Stage (Cita et al., 2012). The GSSP established the base for the Pleistocene and Quaternary Systems at Monte San Nicola, Sicily, Italy (37°8′45.64″N, 14°12′15.22′E), ratifying the Gelasian Stage in August 1996 (Rio et al., 1998). The GSSP is situated at the base of the marly layer immediately above a notable sapropelic bed known as the Nicola bed. The boundary interval, occurring within marly-silty deposits of the Monte Narbone Formation, is interpreted to have been formed within a slope-basin context at a water depth of 500–1000m (Rio et al., 1994). (Rio et al., 1994). At the boundary, the sedimentation rate is 6.1 cm/kyr (Hilgen, 1991). The Gelasian Stage is an important stage in the evolution of the Earth's ocean-climate system because it coincides with the intensification of the Northern Hemisphere glaciation, which controls large obliquity-paced climate cycles (Ruddiman et al., 1986; Lourens et al., 2005). The Nicola bed is approximately 20 cm thick (Rio et al., 1994) and correlates to the Mediterranean Precession-Related Cycle (MPRC) 250 and small-scale cycle 119 at Mount San Nicola (Hilgen, 1991). It is the highest of six sapropels, which occurs when eccentricity is at its peak (Rio et al., 1994). The Punta Piccola section of Sicily, the highest section of the Rosello composite section, likewise has all six sapropelic beds (see figures 3 in Hilgen, 1991; 1 in Lourens et al., 1996a). Obliquity for this period of the Plio-Pleistocene coincides with the global ice volume and, thus, with the marine isotope phases. The astrochronological age of the Nicola bed at its midpoint is 2.588 Ma (Lourens et al., 1996a; Rio et al., 1998, p. 85). Given that the GSSP effectively lies on top of the Nicola bed and the fact that full preservation at this site took place during a period of 7–10 kyr, as indicated by Gibbard and Head (2009), the GSSP's age is around 3.5–5.0 kyr younger than the midpoint age, at 2.58 Ma. That age is currently acknowledged (Cohen et al. 2013, Head and Gibbard 2015a). In the late Cenozoic, the Gauss-Matuyama paleomagnetic boundary is a crucial geochronological indicator. Ohno et al. (2012) discovered that the Gauss-Matuyama border for IODP Site U1314 on the Gardar Drift had a duration of 5 kyr, suggesting an age of 2.590-2.585 Ma. According to Channell and Guyodo (2004) and Lawrence et al. (2013), the northern North Atlantic ODP Site 982 also features the Gauss-Matuyama boundary, which is accepted to have occurred at 2.587 Ma. These two factors led to the precise horizon of the GSSP being chosen (Rio et al., 1994, 1998). First, the Nicola bed is very noticeable in the field, allowing for unambiguous local and regional correlation. The Gauss-Matuyama Chron boundary, which is close to the GSSP, serves as a close approximation of the boundary globally and in both marine and terrestrial deposits.



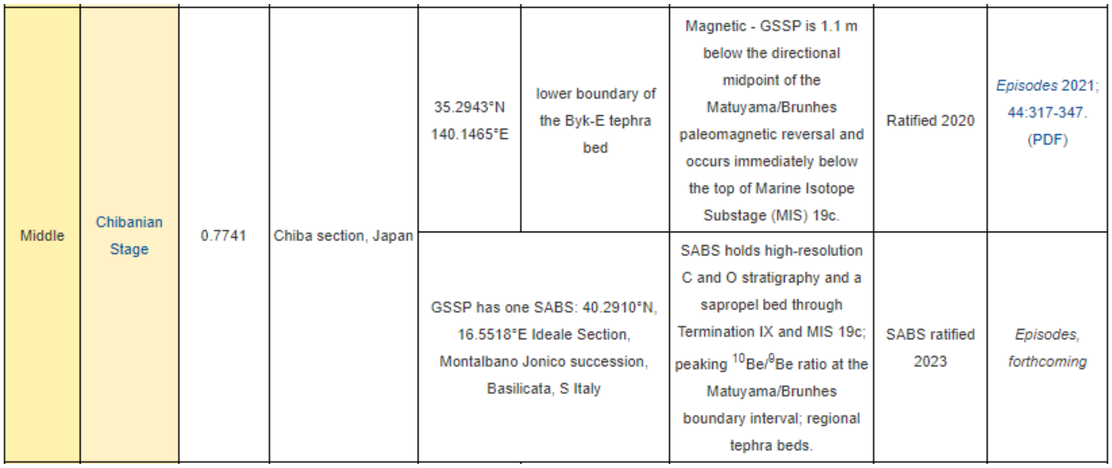
Lower	Calabrian Stage	1.80	Vrica, Italy	39.0385°N 17.1348°E	base of the marine claystone overlying the sapropelic marker Bed 'e' (Mediterranean Precession Related Sapropel, MPRS 176)	Magnetic - ~8m below the observed top of the Olduvai (C2n) normal polarity subchron. GSSP level coincides with the transition from MIS 65 to 64	Ratified 1985 as base of Pleistocene; Ratified 2011 as base of Calabrian	*Episodes* 1985; 8:116-120; *Episodes* 2012; 35: 388-397 Head, M.J., 2019
	Gelasian Stage	2.58	Monte San Nicola, Sicily, Italy	37.1469°N 14.2035°E	base of marly layer overlying sapropel MPRS 250	Magnetic - Matuyama/Gauss boundary (C2r/C2An) is between ~0 and ~3 m above GSSP (Head, 2019) not ~1 m (20 kyr) below it as reported by Rio et al. (1994, 1998). GSSP level is within Marine Isotope Stage 103.	Ratified 1996 as base of Gelasian; Ratified 2009 as base of Pleistocene and Quaternary	*Episodes* 1998; 21: 82-87 *Episodes* 2010; 33: 152-158 Quaternary Sci. 2019; 500: 32-51,

Figure 6. Lower Pleistocene Subseries and the defined stage boundaries, based on the stratigraphic column information from the International Commission on Stratigraphy (www.stratigraphy.org).

The second stage of the Pleistocene, after the Gelasian, is the Calabrian. Its upper limit will be determined by the base of the Middle Pleistocene Subseries stage; the Matuyama/Brunhes polarity chron border is advised as the major guide for this stage because it will make marine/non-marine correlations easier (Head et al., 2008b). The base Calabrian GSSP lies about 4 km south of the town in the Vrica region of the province of Crotone, Calabria (latitude 39°02'18.61" N, longitude 17°08'05.79" E). The GSSP is located close to the base of the marl bed immediately atop sapropel "e", as indicated by a series of silty marls that outcrop in badlands prone to regolith creep and are dark grey or blue grey in color. The marls are abundant in fossils, particularly calcareous nannoplankton and planktonic foraminifers, but they are also abundant in ostracods, benthic foraminifers, and palynomorphs. The Crotone series demonstrates high sedimentation rates, ranging from around 45 cm ka^{-1} near the base to about 29 cm ka^{-1} at about 1.8 Ma (Suc et al., 2010). The main biostratigraphic events of calcareous nannofossils near the GSSP are the LO (Last Occurrence) of *Discoaster brouweri*, recorded in segment A of the section, about 70 m below the boundary stratotype, and the FO (First Occurrence) of a medium-sized *Gephyrocapsa* in segment B, at about 26 m above the GSSP (just above sapropel "h"). The section spreads from zone NN18 (pars) to NN 19 of Martini (1971). Just behind the sapropel "o" upwards, large-sized *Gephyrocapsa* were discovered. The LO of *Calcidiscus macintyrei* occurs in the middle of the sapropel "h" and "n". At the same level, about 10 m above the sapropel "v" in the highest Crotone segment, seem to be the exits of the large-sized *Gephyrocapsa* and *Helicosphaera sellii* (Zijderveld et al., 1991; Lourens et al., 1996b; 1998). With an age of 2.06 Ma in the eastern equatorial Pacific and 1.93 Ma in the western equatorial Atlantic, the LO of *Discoaster brouweri* appears to have the highest potential for worldwide correlation among these events (Lourens et al., 2005a).

The third stage of Pleistocene Series is the Chibanian stage which constitutes the Middle Pleistocene Subseries. The GSSP identifying the coterminous base of the Middle Pleistocene Subseries and the Chibanian Stage at the Chiba section (35°17'39.6" N, 140°08'47.6" E to 35°17'36.9" N, 140°08'47.2" E) of the Chiba composite section, Japan, was authorized by ICS. The Kokumoto Formation of the Kazusa Group is where the Chiba composite portion may be found. It is a continuous marine silty sedimentary record that spans the Lower-Middle Pleistocene and is situated close to the Pacific coast of the central Japanese archipelago and is one of the most extended and chronostratigraphically defined sections ever described (e.g., Kazaoka et al., 2015; Suganuma et al., 2018). According to Kazaoka et al. (2015), the Kazusa Group has a lot of marine and terrestrial micro- and macrofossils that have helped identify the Brunhes Chronozone (Okada and Niitsuma, 1989). Ten Pleistocene nannofossil biohorizons between 2.06 and 0.89 Ma have been identified based on the abundance of well-preserved, calcareous nannofossils that represent at least 15 genera and 27 species throughout the Kazusa Group (Sato et al., 1988; 1999). The highest horizon is located in the middle of the Kasamori Formation, which is thought to be below the LAD (last appearance datum; also known as the highest occurrence datum) of *Pseudoemiliania lacunosa*

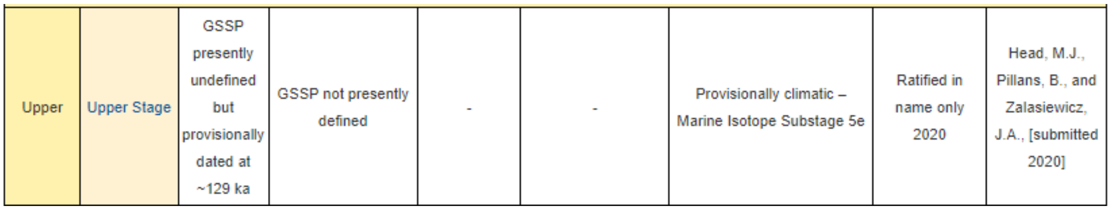
(0.433 ± 0.020 Ma). A lack of trustworthy biohorizons was found in the Chiba composite section (Kameo et al., 2020) due to the brief time period represented. However, fewer occurrences of medium-sized specimens (4-5 μm) and a brief absence of larger specimens (≥5 μm) may correspond to the highest occurrence of the larger form of *Gephyrocapsa* sp. (Matsuoka & Okada, 1990). Alternately, this horizon might correspond with one of *Gephyrocapsa omega*'s two brief disappearances. It was found at the Mediterranean ODP Sites 964, 967, and 977, and the North Atlantic DSDP Site 607, although it seems to be diachronous (Maiorano and Marino, 2004).



Middle	Chibanian Stage	0.7741	Chiba section, Japan	35.2943°N 140.1465°E	lower boundary of the Byk-E tephra bed	Magnetic - GSSP is 1.1 m below the directional midpoint of the Matuyama/Brunhes paleomagnetic reversal and occurs immediately below the top of Marine Isotope Substage (MIS) 19c.	Ratified 2020	*Episodes* 2021; 44:317-347. (PDF)
				GSSP has one SABS: 40.2910°N, 16.5518°E Ideale Section, Montalbano Jonico succession, Basilicata, S Italy		SABS holds high-resolution C and O stratigraphy and a sapropel bed through Termination IX and MIS 19c; peaking ^{10}Be/^{9}Be ratio at the Matuyama/Brunhes boundary interval; regional tephra beds.	SABS ratified 2023	*Episodes, forthcoming*

Figure 7. Middle Pleistocene Subseries and the defined stage boundaries, based on the stratigraphic column information from the International Commission on Stratigraphy (www.stratigraphy.org).

The upper part of Pleistocene Series is known as Upper Stage and its GSSP is presently undefined but provisionally dated at ~129 ka relating to significant warming at the beginning of the Last Interglacial (Head, 2019). There are two potential candidates for the GSSP, the Fronte section, Taranto, Italy (Negri et al., 2015) and the EPICA Dome C Antarctic ice core (Head and Gibbard, 2015a; Head, 2019).



Upper	Upper Stage	GSSP presently undefined but provisionally dated at ~129 ka	GSSP not presently defined	-	-	Provisionally climatic – Marine Isotope Substage 5e	Ratified in name only 2020	Head, M.J., Pillans, B., and Zalasiewicz, J.A., [submitted 2020]

Figure 8. Upper Pleistocene Subseries and the defined stage boundaries, based on the stratigraphic column information from the International Commission on Stratigraphy (www.stratigraphy.org).

4. MATERIALS & METHODS

4.1. INVESTIGATED SECTIONS

The present study describes two geological sections in the island of Rhodes: Agathi Beach section and Plimiri section. Agathi section is located in the northeastern part of the island. Along the road that links Agathi Beach with the main road, the Agathi Beach section outcrops at 36◦10′51.90′′N and 28◦5′21.66′′E. A significant layer of volcanoclastic sediments is visible in the roughly 4.7-m-thick marly succession of the LBF, situated about 2.5 m above the base. A date of 1.89 ± 0.09 Ma was assigned to this layer by Cornee et al. (2006b). Additionally, several fish bones were found within a laminated layer with distinct borders between 1.3 and 1.5 m, and a red layer, or red marker horizon, was formed at about 4 m section height (Eincher et al. 2024). The Agathi Beach location's LBF is covered by recently added terrace.

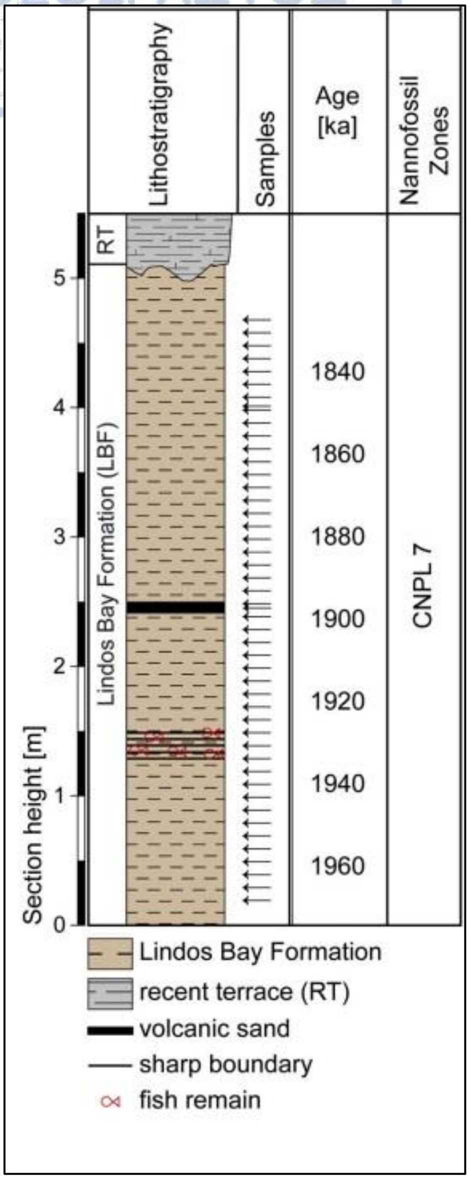


Figure 9. Agathi Beach section lithostratigraphic column (based on *Fig. 2* from Eincher et al. 2024).

Plimiri section is located in the southern coast of Rhodes. Two out of eight Plimiri Section profils are presented in this work: Plimiri profil V and Plimiri profil VI, Unit 17 in stratigraphic columns V and VI (Figs. 10, 11 of Nelson et al. 2001) represents the Lindos Bay Marl. Outwardly, the Plimiri units are most homogeneous in the Lindos Bay Marl, which is mainly composed of blue-grey calcareous mudstone or marl. Based on the existence or lack of interbedding or laminations, five subunits have been identified; however, in general, these structures are uncommon due to bioturbation or the fact that the smectite clay component in the rock expands and contracts with wetting and drying, giving the exposed surfaces a frittered appearance.

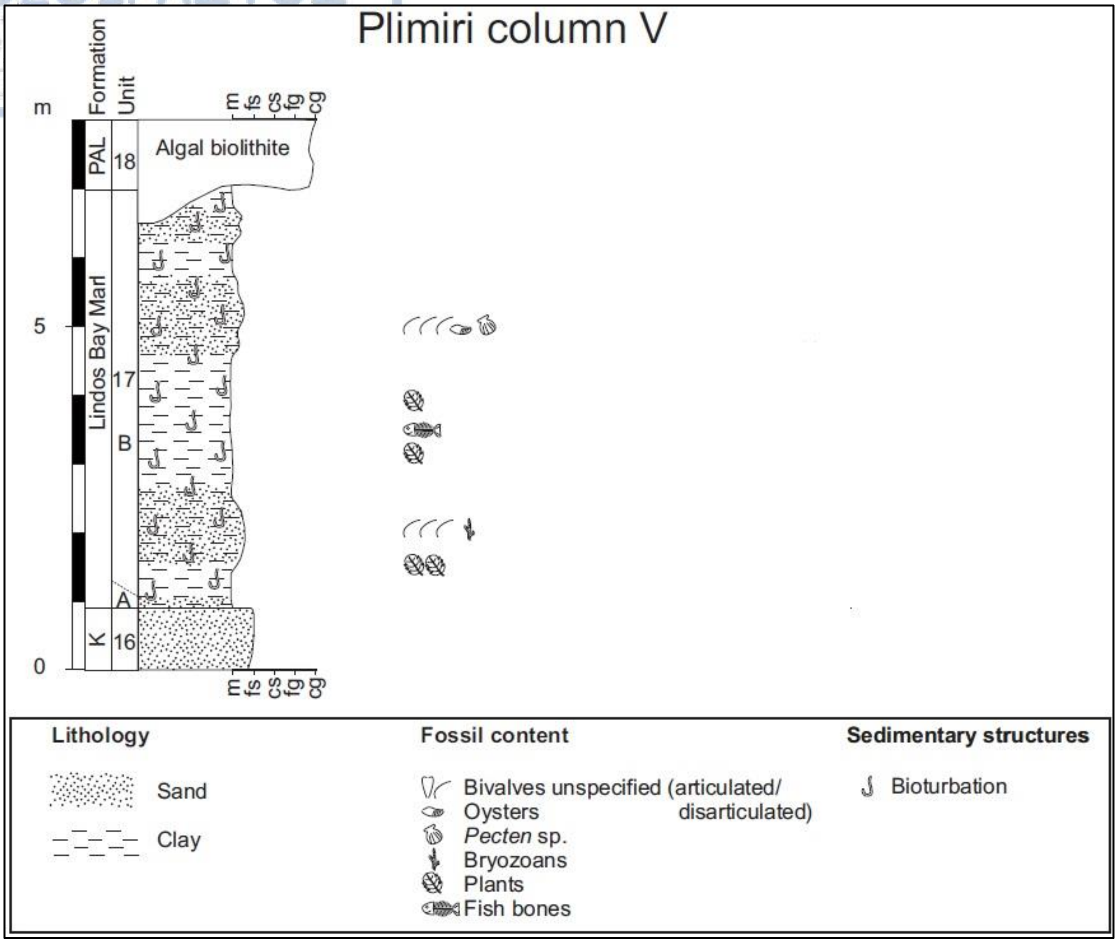


Figure 10. Plimiri Profil V lithostratigraphic column modified from Nelson et al. (2001).

According to Hanken et al. (1996), polished, oiled surfaces of some of the samples examined, show zones that are non-laminated and substantially bioturbated in between laminated and mildly bioturbated zones. Despite their uncommon occurrence in various sections in the northeast of the island, no distinct layers of volcanic ash are visible in the marl at Plimiri (Hanken et al. 1996; pers. observations 2001). The Lindos Bay Marl's lower contact with the Kolymbia Formation is fairly well exposed at Plimiri. Rapid upward fining occurs in the lithological expression, changing from a shelly, muddy sandstone at the summit of the Kolymbia to calcareous sandy mudstone and mudstone at the base of Lindos Bay Marl. It is evident that tectonically driven processes were responsible for the noteworthy deepening observed across this boundary, which is estimated to be a few hundred meters. It has been claimed that Kolymbia-Lindos Bay contacts from northern Rhodes exhibit a similar gradational contact and rapid deepening (Moissette and Spjeldnaes 1995; Hanken et al. 1996).

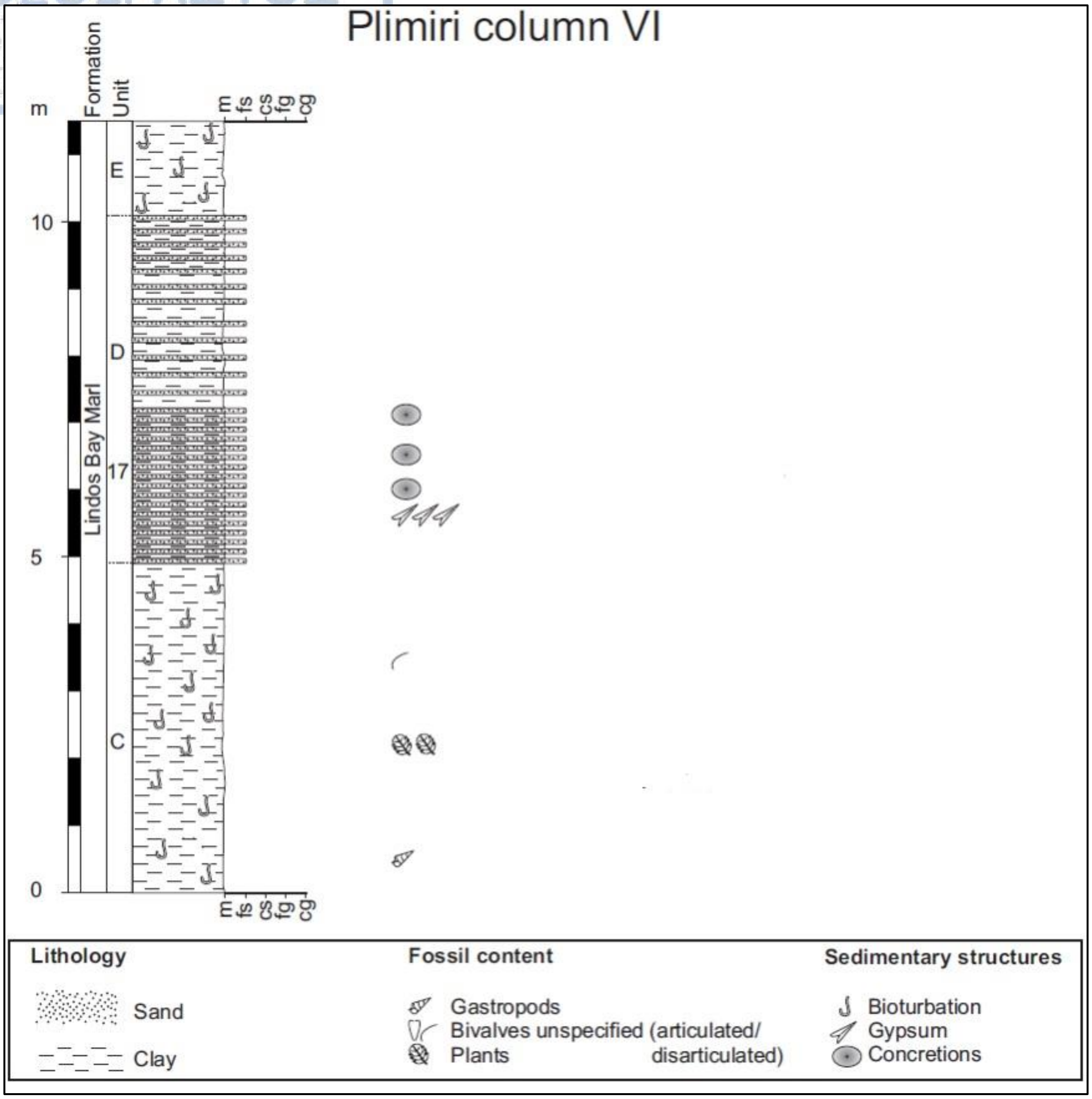


Figure 11. Plimiri Profil VI lithostratigraphic column modified from Nelson et al. (2001).

4.2. METHODS

The quality of the biostratigraphical datums itself has become increasingly important as more calcareous nannofossil biostratigraphical data have been gathered over the past 60 years by various authors utilizing various counting methodology or abundance evaluations. For any given species, semi-quantitative and relative abundance counts (Backman & Shackleton 1983; Rio et al. 1990, Triantaphyllou 1996) can depict stratigraphical ranges very precisely. This enables the proper identification and positioning of a biohorizon, which can increase the reproducibility of datums and provide higher degree of correlativity over broad, regional to superregional areas.

The philosophy of Hay (1967) was used to determine a species' abundance. According to Hay, species with more than one individual per field of view are considered abundant, those with one individual every 1–10 fields of view are very common, those with one individual every 10 fields of view are common, and those with only a few individuals every 100 fields of view are considered rare. Based on the philosophy of Rio et al (1990b) for the quantification of associated Plio-Pleistocene calcareous nannofossils, the definition of biostratigraphic events and the estimation of the frequencies of distinctive index species were determined. A species' frequency in fossil assemblages is influenced by a number of parameters, including the species' productivity, how other microfossils alter the species' content in the sediment, how the sediments affect the terrestrial environment, and secondary sedimentary processes like dissolution, recrystallization, and fragmentation. Concentrations of calcareous nannofossils change with time due to environmental changes that cause migrations (upper and lower occurrences) and fluctuations in the presence and frequency of different species. Such flora changes when distinct and correlated can constitute biostratigraphic events. There are many factors that affect the biostratigraphic reliability of an event, which can be divided into two categories: those related to physical causes and those related to methodological causes (Olafsson, 1991). In the first category, the reliability is a function of the change in the productivity of the species, the preservation of the event as well as the possibility of its correlation over long geographical distances. Methods that define an event and that affect its biostratigraphic reliability are limited to sampling distances, sample processing, and quantitative/qualitative approaches to the frequency of indicator species. The counts of the characteristic index species against the total concentrations, the counts of the index species against a predetermined number of taxonomically related forms, and finally the number of individuals of the indicator species in a predetermined area of the prepared sample can all be used to express the occurrence frequencies of species. The first is also the most objective method, according to Rio et al. (1990b), which allows the confidence level for the detection of a species to be determined based on its frequency in the original population and sample size. It is claimed that this method is independent of changes in concentrations due to terrestrial effects and is applied with relative ease in most of the samples (Dennison & Hays, 1967). The second method offers benefits such independence from the nannofossil's preservation status, implementation outcomes, and paleoenvironmental parameters. The presence of enough members of the reference group in the total concentrations is a crucial component of the execution of this method. The third method involves measuring a group of individuals in a predetermined number of fields of view, each one corresponds to a certain region of the preparation. While it is argued to correspond to absolute frequencies (number of individuals/weight unit), this method has the potential to provide data with astounding accuracy. It is impacted by the density of the material used in the sample preparation, the effects of dissolution, and terrigenous effects on sediments (Rio et al. 1990b, Triantaphyllou, 1996).

In the present study a total of 44 samples from 3 Plio-Pleistocene geological sections in the island of Rhodes were studied. The samples were prepared using the smearing technique. A small amount of sediment from a clean surface of the sample is smeared on a slide using a drop of distilled water and a toothpick. When the slide is dry, the cover slip is affixed with the help of a mounting medium. The smearing technique, which does not involve breaking the material, enables the preparations to adequately represent the burial societies of the calcareous nannofossils contained in the studied samples (Schmidt, 1978). The qualitative and quantitative analysis of the calcareous nannofossil content of the samples was done under a polarizing microscope, under 1000x magnification. Also, a manual counter was used to count the nannofossils as well as the website Nannotax 3 (https://www.mikrotax.org/Nannotax3/) for the identification of the nannofossil species. The quantitative methods of biostratigraphic analysis refer to: 1) measurements of the characteristic indicator species in relation to the total concentrations of nannofossils, 2) measurements of the indicator species in relation to a fixed number of taxonomically related forms and 3) semi-quantitative estimates of the presence of indicator species in relation to a specific number of fields of view of the preparation. The second approach was easily applied to regulate the frequency of occurrence of the species *Helicosphaera sellii*, *Calcidiscus macintyrei*, *Gephyrocapsa* spp. (< 4μm,>= 4μm), as well as representative species of discoasterids, in taxonomic groups well represented in the analyzed material. More specifically, for the presence of *Gephyrocapsa* 3.5–4 μm there have been counted the *Gephyrocapsa* specimens in 100 placoliths, for *Calcidiscus macintyrei* the species specimens in 100 coccoliths of genus *Calcidiscus* and for *Helicosphaera sellii*, the species specimens in 100 helicoliths of genus *Helicosphaera*. The preparation of samples followed the standard smear slide techniques for calcareous nannofossil analysis (Perch- Nielsen, 1985; Bown and Young, 1998). All samples were routinely examined at 1250×, using a LEICA DMLSP light microscope (LM).

5. RESULTS

5.1. AGATHI BEACH SECTION

For Agathi Beach, 9 samples were examined for calcareous nannofossils in the 4.5 m thick section at Agathi beach in eastern Rhodes. Graphs A, B, C and D show the outcomes from the quantitative analysis of a few different indicator species. These graphs present the index species of Plio-Pleistocene sediments *Calcidiscus macintyrei, Helicosphaera sellii* and *Pseudoemiliania lacunosa*. There is no graph for *Gephyrocapsa* ≥ 4μm. because there were not observed any, instead

Gephyrocapsa 3.5-4 μm have appeared in significant numbers. A remarkable result is that *Calcidiscus macintyrei* appears a maximum of 72% at the lower part of the section (sample 42550) but as we move upwards it shows a gradually decreasing trend. The same trend is observed to all other index species with *Helicosphaera sellii* showing a maximum of 48% and both *Gephyrocapsa* 3.5-4 μm and *Pseudoemiliania lacunosa* ~25% in the same sample.

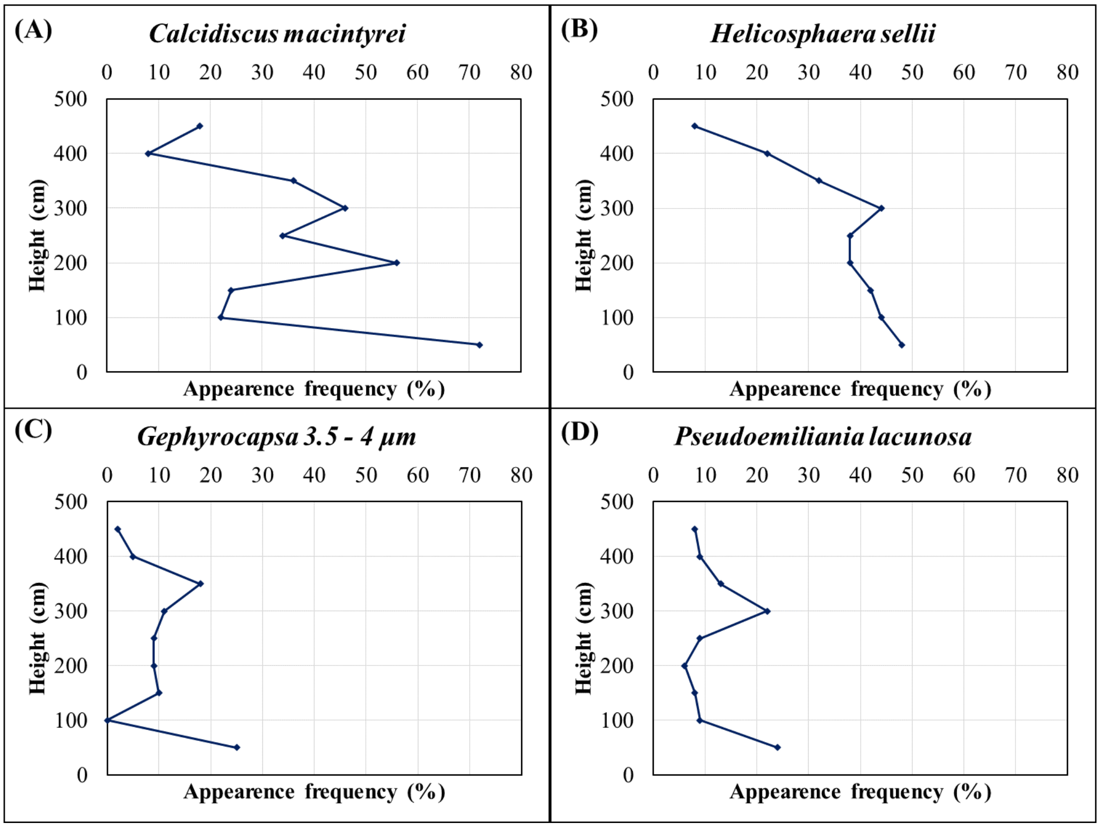


Figure 12. Graphs demonstrating the nannofossils of Agathi Beach section. (A) *Calcidiscus macintyrei* (B) *Helicosphaera sellii* (C) *Gephyrocapsa* 3.5-4 μm (D) *Pseudoemiliania lacunosa.*

5.2. PLIMIRI PROFIL V

For Plimiri Profil V, 13 samples have been observed from a 6,5 m thick part of the main section. The graphs below demonstrate the % appearance frequency of six main *Discoaster* index species: *Discoaster tamalis, Discoaster assymetricus, Discoaster pentaradiatus, Discoaster brouweri, Discoaster variabilis* and *Discoaster surculus.. Discoaster tamalis* and *Discoaster assymetricus* show a decreasing trend as we move upwards in the section. In contrast, all other discoasterids observed show and increasing trend with *D. pentaradiatus* to be the most abundant. It reached a maximum of 70% at the upper part of the section (5m, sample 42529).

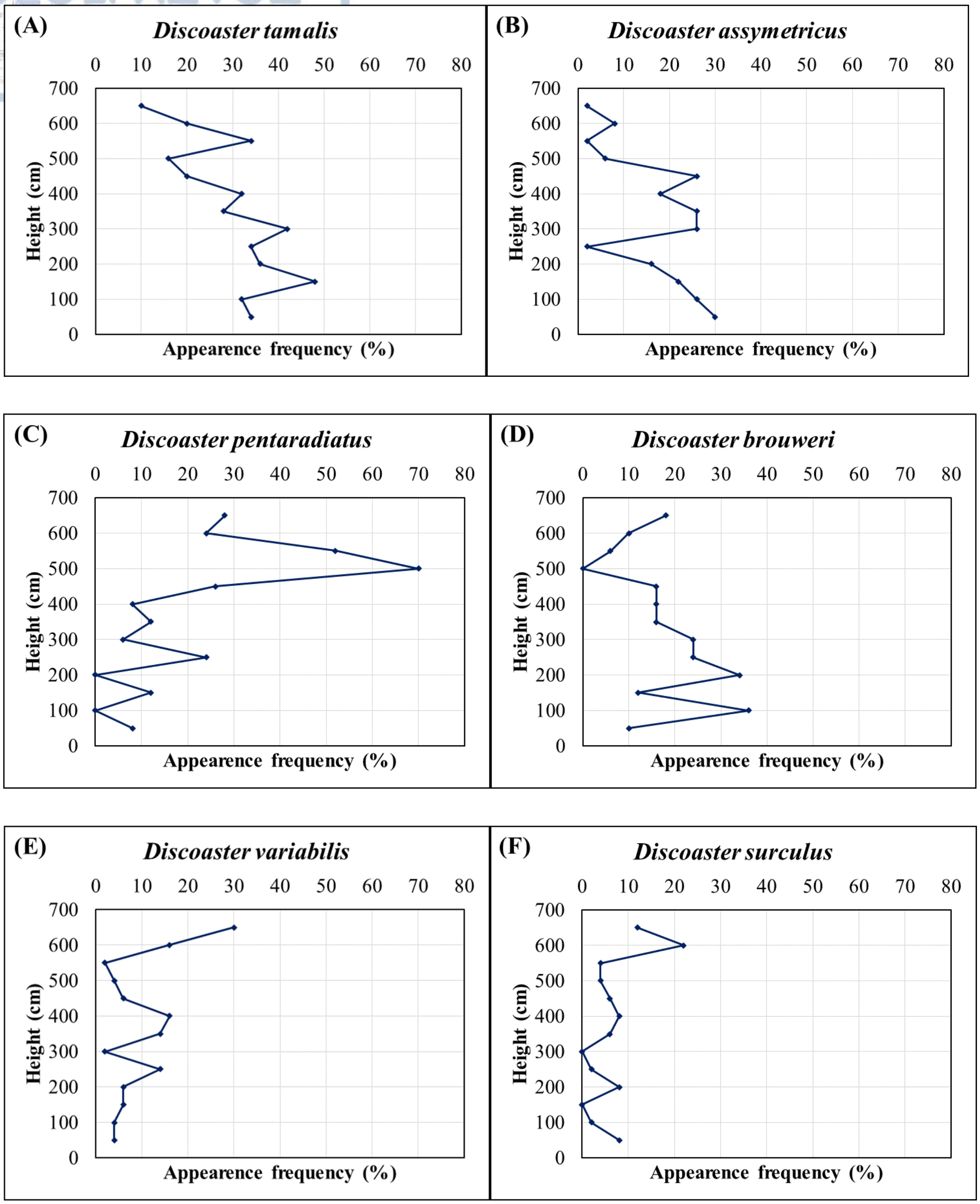


Figure 13. Graphs demonstrating the nannofossils of Plimiri V section. (A) *Discoaster tamalis* (B) *Discoaster assymetricus* (C) *Discoaster pentaradiatus* (D) *Discoaster brouweri* (E) *Discoaster variabilis* (F) *Discoaster surculus.*

5.3. PLIMIRI PROFIL VI

From this 11 m thick section, also a part of the main South Plimiri section, 22 samples were observed for the same calcareous nannofossil index-species as the Agathi Beach section. *C. macintyrei* seems to be the most dominant, once again, and appears a maximum percentage of 64% at the bottom of the section. The peculiarity of this section, as can be seen in the graphs below, is that nannofossils are observed up to 4.5 m, while from that point on there is a complete lack of them, apparently due to the lithological features of the sequence that imply shallow water depositional setting, see the sandstone beds in the lithological column of the section (Fig. 10).

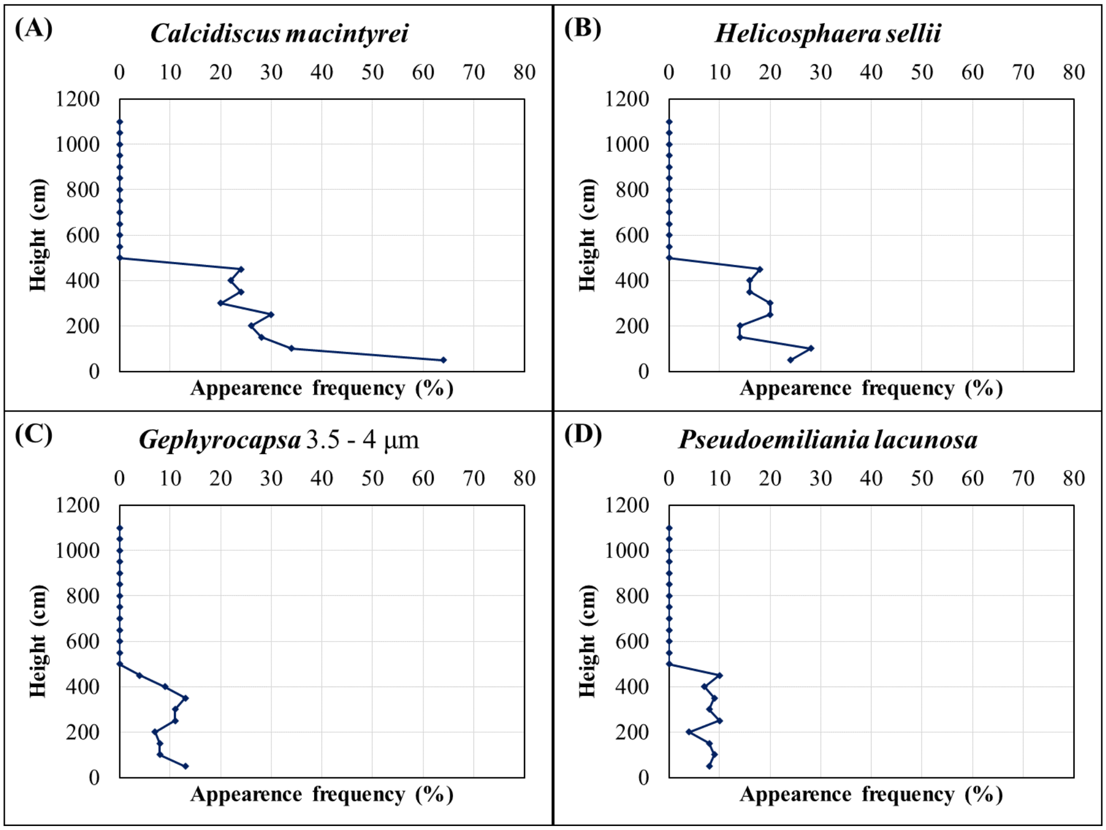


Figure 14. Graphs demonstrating the nannofossils of Plimiri VI section. (A) *Calcidiscus macintyrei* (B) *Helicosphaera sellii* (C) *Gephyrocapsa* 3.5-4 μm (D) *Pseudoemiliania lacunosa.*

6. DISCUSSION

By observing the results, we can distinguish that the Agathi Beach section is featured by the complete absence of *Gephyrocapsa* >=4 μm which means that the sediments of this section date below the upper part of biozone NN19 (Martini et al. 1971) and/or CNPL 8 (Backman et al. 2012) in the late Gelasian/early Calabrian boundary. This is also confirmed by the high abundance of the species *Calcidiscus macintyrei* (up to 72 individuals of *C. macintyrei* per 100 individuals of the genus *Calcidiscus*). In addition, several individuals of the species *Pseudoemiliania lacunosa* were found, which limits the range, in geological time, from which the sediments originate, between Martini biozone NN14+NN15 (CNPL3, Backman et al. 2012) and NN19/NN20 boundary (top CNPL10, Backman et al. 2012). Finally, the fact that the species *Helicosphaera sellii*, which appears for the first time during the Zanclean stage (4.5 Ma), is abundant (up to 48% of the genus *Helicosphaera*) leads us to conclude that this section is older than the top of this species within CNPL 9 (Backman et al. 2012), in the Calabrian stage. *Gephyrocapsa* ≥4 μm, however, were absent in any sample and, thus, Agathi Beach is assigned to Calcareous Nannofossil Zone (CNPL) 7 (Backman et al., 2012) that covers an interval between 1.93 and 1.71 Ma in the Early Pleistocene (Gelasian-Calabrian boundary; Fig. 2) and is correlated with the lower part of Neogene Nannoplankton NN19 (Martini, 1971) biozone and Mediterranean Neogene Nannoplankton MNN19a (Rio et al., 1990).

Looking at the graphs of Plimiri V section, it is observed that the species *Discoaster pentaradiatus* shows the highest rates of occurrence, up to 70%, among the 50 individuals of the genus *Discoaster* measured in each of the thirteen samples. The appearances of the remaining species are also noteworthy, with *Discoaster tamalis* covering up to 48%, *Discoaster brouweri* up to 36% and *Discoaster assymetricus* up to 30% of the individuals measured. The regular presence of *Discoaster assymetricus* suggests that the Plimiri Profil V is chronologically placed above the NN14+NN15/CNPL3 biozone in the late Zanclean. During the microscopic observation of the samples, some remarkable occurrences of index species were noted which in some cases characterize different biozones, both Pliocene and Pleistocene. In the upper part of the section, in the first 50cm (sample 42505), a large number of placoliths (small *Gephyrocapsa*) were observed, together with the relative lack of species of the genus *Discoaster*. Continuing, in the 100cm sample (sample 42484), the species *Calcidiscus leptoporus*, *Helicosphaera sellii*, *Ceratolithus cristatus* as well as various species of placoliths were observed. Also noteworthy was the appearance of some transported Oligocene-Miocene species such as: *Reticulofenestra bisecta*, *Cyclicargolithus floridanus* and *Sphenolithus belemnos*. The 150cm sample (sample 42491) is characterized by the occurrence of the species *D. pentaradiatus* and *D. tamalis* with the latter showing great abundance. Conversely, in the 200 cm sample (sample 42494), discoasterids (e.g. *D. surculus*, *D. pentaradiatus*) rarely occur, while *H. sellii*, *C. macintyrei* and some placoliths were observed in greater abundance, with the small-sized *Gephyrocapsa* being more common. In the 250, 300 and 350cm samples (sample 42499, 42509 and 42514 respectively), discoasterids are mostly abundant with the exception of *D. pentaradiatus* which is rare. As for 400cm (sample 42519), this is a more productive sample in which small *Gephyrocapsa*, few individuals of the genus *Braarudosphaera* as well as siliceous spicules were observed among others. The relative absence of discoasterids in this particular sample was characteristic. The sample taken at 450cm (42524) of the section is characterized by the presence of the species *H. sellii*, *C. macintyrei*, *D. tamalis*. The next one (500cm, sample 42529) is characterized by the presence of slabstones, by a great abundance of species of the genus *Discoaster*, while a reworked species, *Reticulofenestra hillae*, was also identified. Continuing, at 550cm (sample 42534), discoasterids are abundant with *D. pentaradiatus* and *D. tamalis* more dominant in contrast to the 600cm sample (42539) in which discoasterids are absent and placoliths are abundant. Finally, at 650cm (sample 42544), species of various genera were observed (e.g. placoliths, *C. macintyrei*, *H. sellii, D. tamalis*, *D. pentaradiatus*, *D. brouweri*, *C. cristatus*) therefore this is a productive sample.

In conclusion, the Plimiri V nannofossil sediment samples are characterized by the presence of common *Discoaster tamalis* (up to 48%) and *Discoaster pentaradiatus* (up to 70%), as well as the presence of *Discoaster asymmetricus* (up to 30%; Fig. 2). Based on this assemblage the entire section is assigned to the Neogene Nannoplankton NN16 biozone of Martini (1971), which correlates to the Calcareous Nannofossil Zone (CNPL) 4 (3.81 Ma to 2.76 Ma) of Backman et al. (2012) in the Piacenzian.

As far as the Plimiri VI section is considered, the dominant nannofossil is *Calcidiscus macintyrei* with max. more than 60%. On average, this species appeared with a percentage of 28% in the 4.5 m of the section. The species *Helicospharea sellii* moves in slightly lower percentages, about 18% on average among the species of the genus *Helicospharea*, while *Gephyrocapsa* 3.5-4 μm individuals were measured to represent around 9% of the placoliths. At approximately the same levels as *Gephyrocapsa* 3.5-4 μm the placolith *Pseudoemiliania lacunosa* also appears (about 8 individuals per 100 placoliths). Finally, the general lack of *Gephyrocapsa* ≥ 4 μm is noteworthy, except of very few specimens (max 3 specimens at sample 42600). The findings above indicate that the samples studied originate from the Early Pleistocene in the Calabrian stage. More specifically, they date to the lower part of the Martini biozone NN19, that correlates with biozone MNN19b of Rio et al. (1990) or to the lower part of CNPL8 biozone (Backman et al. 2012), above the base of *Gephyrocapsa* ≥ 4 μm at 1.71 Ma and below the top of the species *Calcidiscus macintyrei* at 1.60 Ma.

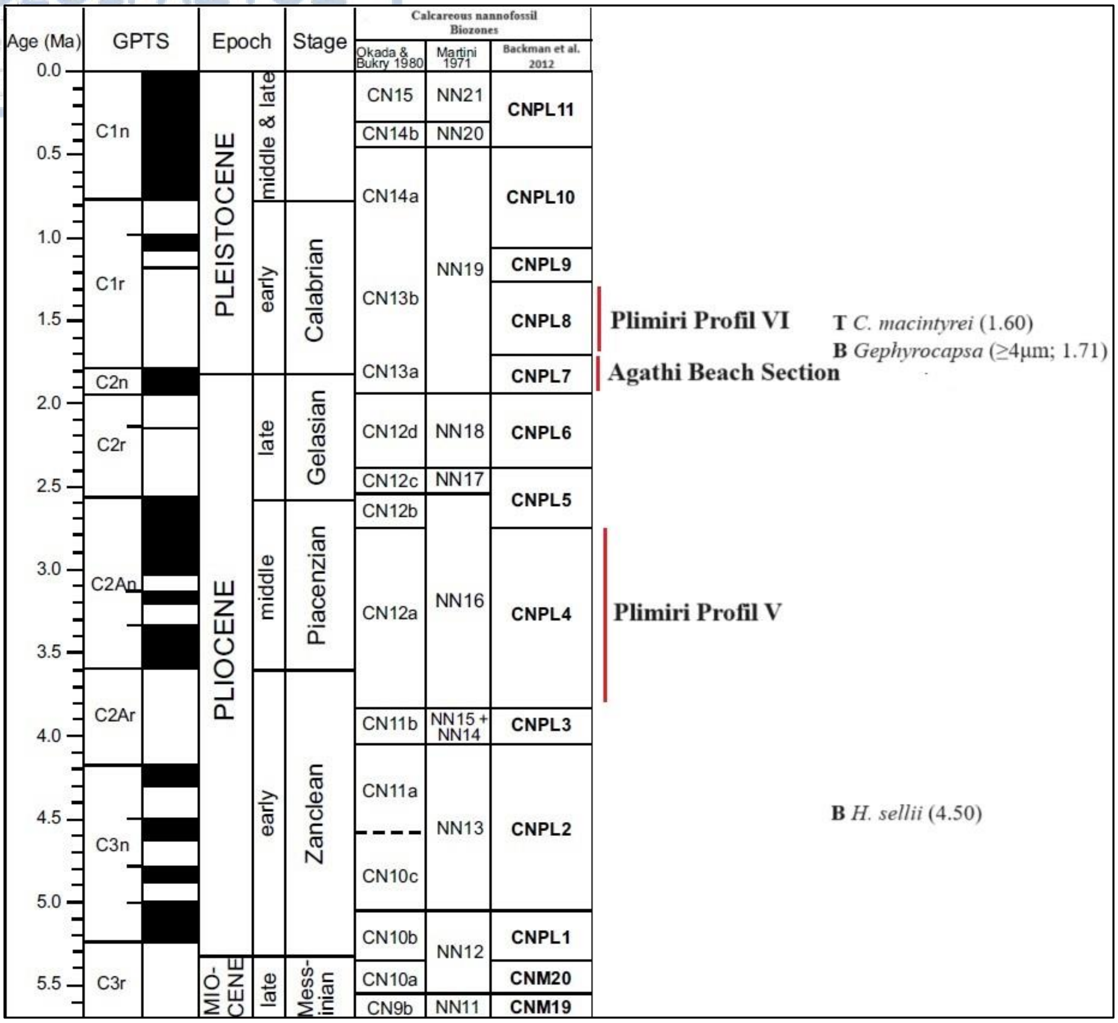


Figure 15. Summary figure depicting sections mapped to the biozones with which they were associated. Modified from Backman et al. (2012).

The comparison with other studies is considered appropriate for the study of biozones as similar biozones are also observed in different regions of Greece. For example, Triantaphyllou (1996) studied biozones on three Ionian islands, Corfu, Zakynthos and Kefalonia. In particular, biozone NN19, where it was identified in the present work in the sections Plimiri VI and Agathi Beach, was also identified in Corfu (Alonaki section), in Zakynthos (sections "Seliniako Topio", Gerakas 1, Cape Gerakas, Cape Kalogeras, Citadelle, Cape Kryoneri), as well as in Kefallinia (sections Ammes, Lofos, Minies, Mousata, Mihalitsata). The results of Triantaphyllou (1996) are similar to the results of this study in terms of nannofossil concentrations in the sections associated with biozone NN19, with *C. macintyrei* being one of the dominant species (e.g. 29% in Alonaki section). The same applies to *H. sellii*. Also, some of the sections studied by Triantaphyllou (1996) do not spread over the entire range of NN19 of Martini (1971), but are limited to MNN19a (Raffi and Rio,

1979) which can be chronologically placed close to 1.89 Ma. These are the sections Alonaki, "Seliniako Topio", Gerakas 1, Citadelle, Ammes and Lofos. Regarding biozone NN16, where it was located in the Plimiri V section, Triantaphyllou (1996) established the presence of this biozone in Corfu (sections Contres and Agios Georgios) and Zakynthos (section Cape Alikanas). The dominance of *D. pentaradiatus* in these sections (e.g. 58% in section Agios Georgios) is remarkable as it is in Plimiri V (70%, sample 42529).

7. CONCLUSIONS

With the intention of conducting a biostratigraphic examination of Plio-Pleistocene sediments based on calcareous nannofossils, three sections of the island of Rhodes were studied for this master's thesis. Calcareous nannofossils are extremely useful as index fossils for dating sediments because they provide consistent age markers due to their small life cycles, increased resistance to dessolution as well as other biological features. The study included the analysis of 44 samples from Agathi Beach section and two profils (out of eight from Nelson et al. 2001) of Plimiri section, Plimiri profil V and Plimiri profil VI. At both Agathi Beach and Plimiri VI the most dominant fossil is that of *C. macintyrei* and the general lack of *Gephyrocapsa* ≥ 4 μm is noteworthy. At Plimiri V the observation of discoasterids was noticable, therefore the index species studied were species of genus *Discoaster* with *D. pentaradiatus* to be the most dominant. The biochronological determination of biozone (Martini, 1971) NN16 has been concluded for Plimiri V section. Agathi beach section is assigned to biozone NN19a, and NN19b has been defined for the Plimiri VI section. Comparison with the results of Triantaphyllou (1996) has shown similar patterns in terms of dominant index species in sections characterized by the same biozones.

8. SYSTEMATIC CLASSIFICATION OF THE MAIN REPRESENTATIVES OF CALCAREOUS NANNOFOSSILS

Genus ***Calcidiscus*** Kamptner, 1950

Circular coccoliths characterized by two shields that defining their margin, with the distal shield being larger than the proximal one. These forms have a center hole and a circle of elements forming their central area.

Calcidiscus macintyrei (Bukry and Bramlette, 1969) Loeblich and Tappan, 1978

Large circular placoliths made up of two shields connected by a tubular structure. Each shield is made out of a cycle that has approximately forty crystal elements in it. On the distal surface of the distal shield, the sutures in between the elements are radially straight, but on the other surfaces, they are slightly bent. Only the proximal, smaller shield appears bright in polarized light. The systematic classification of *C. macintyrei* was previously ambiguous. While some researchers recognized it as a variety of the species *Calcidiscus leptoporus* (Mcintyre et al. 1967, Bartolini 1970, Ellis et al. 1972), Bukry & Bramlette (1969) defined it as a distinct species due to its larger size (~10μm) and higher element number. When examining the species under an optical microscope, the overall size of the placolith can be a useful tool for species separation (Backman & Shackleton 1983). Although similar morphotypes do exist in the Miocene, the term *C. macintyrei* is typically limited to the large specimens seen in the Pliocene and early Pleistocene. According to Raffi et al. (2006), the species' last occurrence (in mid part of MIS-58 Isotope stage, 1.7Ma in Calabrian stage) has served as a sub-zonal marker. Individuals larger than 10 μm were classified as *C. macintyrei* in this study.

Genus ***Discoaster*** Tan Sin Hok, 1927

The nannoliths that comprise the genus Discoaster are composed of a limited quantity of fine rays that decrease as one advances t towards younger ages. All species in this genus, with the exception of *D.pPentaradiatus*, are not bright in polarized light unless they are significantly recrystallized.

Discoaster assymetricus Gartner, 1969

These nannoliths have five long, thin rays that, in some cases, bend strongly and end in sharp ends. The species' primary feature is the uneven angles that form between the rays. They feature a tiny central area from which short dorsal projections extending in the direction of the rays often originate from an evolved node. According to Gartner (1969), this species is not symmetrical and has only five rays, unlike *D. brouweri*, from which it may have originated. Asymmetric five-rayed individuals are produced by the majority of six-rayed species, and *D. assymetricus* is distributed across *D. brouweri*'s range. On the other hand, mid-Pliocene sediments contain them in far higher quantities and with greater consistency. It appears to be a limited stratigraphical acme intraspecific variety. Both the beginning and the end of the acme can be used in biostratigraphy (Bukry 1973a, Backman 1986).

Discoaster brouweri (Tan Sin Hok 1927) Bramlette and Riedel, 1954

Six long, thin rays that form umbrella-shaped structures when their edges tilt moderately to strongly in the direction of the more proximal surface. There might or might not be a knob on the outer face of the central area, which is tiny in comparison to the length of the rays. Conversely, there could not be any unique characteristics in the proximal of the core region. In contrast to other discoasterids, *D. brouweri* does not have bifurcation at the end of each ray.

Discoaster pentaradiatus Tan Sin Hok, 1927

Five long, thin, progressively thinner rays that terminate in a distinctive branching and lean slightly in the direction of the individual's more secure side are what define this species. The angle formed by the two parts of the branch varies. With low central nodes on both its proximal and distal surfaces, the core region is relatively short to the length of the rays. *D. pentaradiatus* is characterized by optical discontinuity, wherein each ray functions as a distinct crystal, and presents birefringence in polarized light. Because of the rays' curvature, the complete species appears in polarized light with varying intensities of luminosity in its various portions, and the suture lines appear as dark dividing surfaces between the various parts.

Discoaster surculus Martini & Bramlette, 1963

These kinds of nannoliths typically consist of six rays with a modest central area compared to the length of the rays. A star-shaped, rounded node is seen on the proximal and distal side of the center region, and the ray ends are distinguished by a triple apophysis (trifurcation). In other words, a third projection that is at a different level and pointed in the direction of the proximal side is seen between the branched ends of the rays. This species' triple branching distinguishes it from *D. variabilis*.

Discoaster tamalis Kamptner, 1967

Discoasterid having a very small core area and four thin to moderately thick rays intersecting vertically. The rays have a small convexity and progressively get smaller in diameter as they approach the ends, where there is no sign of branching. This species is widespread in the lower part of Upper Pliocene and is regarded as a member of the *D. brouweri* group (Roth 1973). Unlike *D. asymmetricus*, this form is very rare outside the mid Pliocene. It seems to have x first and last occurrences at, or very near to, the levels of the beginning and end of the *D. asymmetricus* acme.

Within this range its abundance is closely related to that of *D. asymmetricus* (Bergen 1984, Backman 1986, Chapman & Chepstow-Lusty 1997).

Discoaster variabilis Martini & Bramlette, 1963

The majority of *D. variabilis* consists of six long rays, each of which has a diameter that slightly reduces towards the tips, where a strong bifurcation or some thickening rather than evident branching is seen. Typically, the rays curve in the direction of the nannolith's closest side. The branches feature tangential protrusions and are somewhat lengthy. The area between the branches of the branches sometimes has a calcitic membrane. Typically, the rays curve in the direction of the nannolith's proximal side. The branches feature tangential protrusions and are somewhat lengthy. There may occasionally be a calcitic membrane in the space between a branch's branches. Dorsal projections run down the middle of the rays from a low, round to stellate central knob that is visible on the proximal surface of the central region. All six-rayed *Discoasters* with more or less noticeable branches at the extremities of the rays, varying in size, are classified in this species.

Genus ***Gephyrocapsa*** Kamptner, 1943

The group of gephyrocapsids according to most authors comprises placoliths characterized by the existence of a bridge that crosses the central area (Gartner 1977b, Raffi et al. 1993). Particular issues are noted with regard to identification at the species level because, given the small size of the individuals, the characteristics used for species identification (number of crystal particles, number of shield particle circles, thickness, structure, diagonal bridge orientation) can only be ascertained with the assistance of an electronic microscope. Raffi et al. (1993) contend that Rio's (1982) observation—that the size of gephyrocapsids progressively increased during the Early Pleistocene—represents a real evolutionary trend that happened synchronously in all major oceans and marginal seas.

small *Gephyrocapsa* spp.

The diminutive members of the *Gephyrocaps*a genus exhibit a wide range in diameter length, with diameters ranging from 2 to 3.5 μm. Individuals sized between 3.5 and 4 μm are regarded as transitional forms for *G. oceanica* s.1. The diagonal bridge exhibits significant variation in size, direction, and brightness intensity in polarized light. Some small *Gephyrocaps*a spp. have been found in many cases to have an especially bright bridge.

***Gephyrocapsa oceanica* s.1. / Normal-sized gephyrocapsids**

The elliptical placoliths that are associated with the gephyrocapsids categorized as *G. oceanica* s.1 have two shields that are convex on the distal side and separated by a short distance. The aforementioned individuals exhibit a central open area and a noticeable diagonal bridge that makes an angle of less than 45 degrees with the placolith's small axis. They are larger than 4μm and smaller than 5.5μm.

Genus ***Helicosphaera*** Kamptner, 1984 emend. Theodoridis, 1984

This genus contains the helicoliths, or coccoliths having a helical edge. They are divided into three portions that comprise the coccolith's central area and margin. The flange and the blanket on the coccolith's distal side, as well as the central area of the proximal plate and the cover, when necessary, comprise the margin. The central area may be covered, be empty, or separated by a continuous or discontinuous bar. In polarized light, helicoliths shine brightly. According to Theodoridis (1984), the bar in the center of the helicolith exhibits distinct optical behavior from the remainder of the structure and is bright in response to the sample's rotation in polarized light. Of the species of the genus, only *Helicosphaera sellii* is employed in the Plio-Pleistocene biostratigraphy.

Helicosphaera sellii (Bukry and Bramlette, 1969) Jafar and Martini, 1975

These small to medium-sized, symmetrically elliptical helicoliths are distinguished by their short, tiny flange. A nearly horizontal or slightly inclined bar that forms two large central holes and is visually continuous with the rest of the coccolith is present on the proximal plate. The blunket covers most of the distal side of the helicolith, delivering brightness to polarized light over the whole surface of the individual (Theodoridis 1984, Aubry 1988).

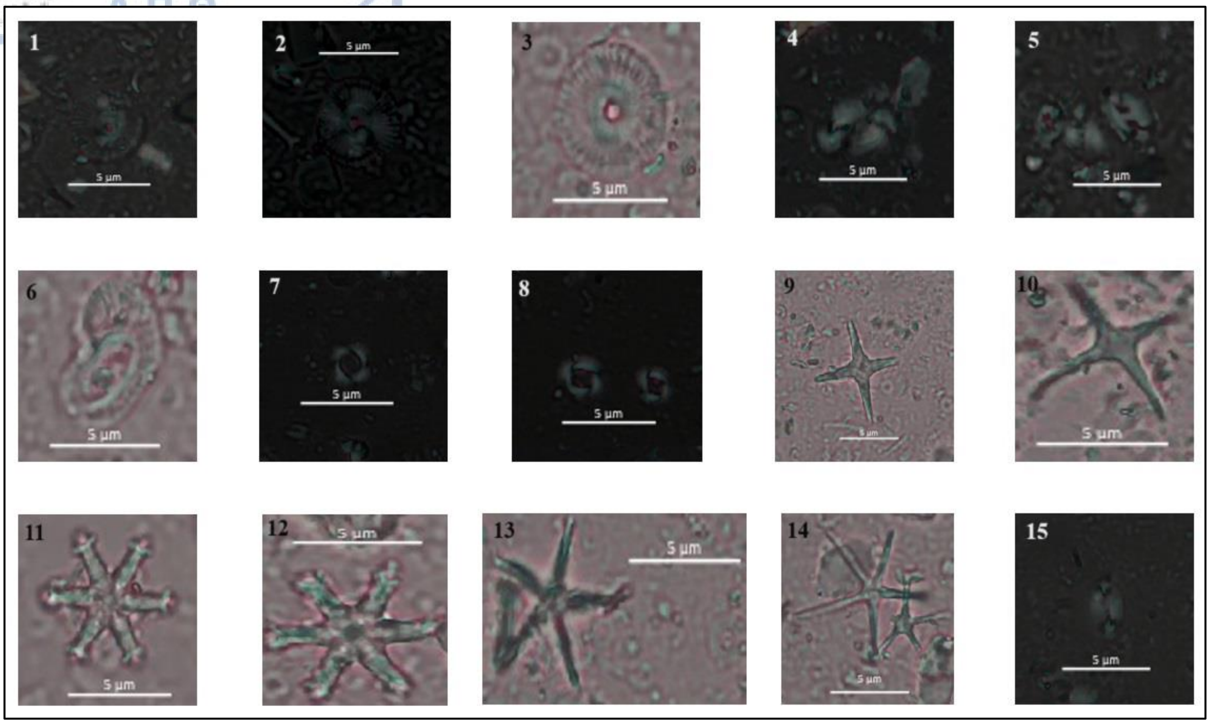
Genus ***Pseudoemiliania*** Gartner, 1969

Elliptical to circular placoliths, with few to numerous slits in their shields. The central hole can be open or covered by a fine mesh.

Pseudoemiliania lacunosa (Kamptner, 1963) Gartner, 1969

Placoliths that are subcircular to elliptical and have two shields with distal shield with a few to several slits. The coccoliths have a size range of 2.5μm to almost 6μm at their maximum (Driever, 1988).

TABLE



1, 2, 3: *Calcidiscus macintyrei* (Plimiri profil V, sample 42544)

5, 15*: Helicosphaera sellii* (Plimiri profil V, sample 42544)

4,6: *Helicosphaera carteri* (Plimiri profil V, sample 42544)

7,8*: Pseudoemiliania lacunosa* (Plimiri profil VI, sample 42600)

9, 10*: Discoaster tamalis* (Plimiri profil V, sample 42544)

11, 12*: Discoaster surculus* (Plimiri profil VI, sample 42600)

13, 14: *Discoaster pentaradiatus* (Plimiri profil V, sample 42544)

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