

ECOfast - an integrative ecological evaluation index for an ecosystem-based assessment of shallow rocky reefs

Abstract

The degradation of marine ecosystems is a growing concern worldwide, emphasizing the need for efficient tools to assess their ecological status. Herein, a novel ecosystem-based ecological evaluation index of shallow rocky reefs is introduced and tested in the Aegean and Ionian Seas (NE Mediterranean). The index focuses on a specific set of pre-selected species, including habitat-forming, key, commercially important, and non-indigenous species, across a wide range of trophic levels (1.00 – 4.53). Data acquisition is conducted through rapid non-destructive SCUBA diving surveys to assess all macroscopic food web components (macroalgae, invertebrates, and fish). Two versions of the index, ECOfast and ECOfast-NIS, were developed, each applying a different approach to account for the impact of non-indigenous species. In our case study, the correlations between the two versions of the index and sea surface temperature, protection status, occurrence of carnivorous fish, and non-indigenous herbivores were assessed through generalized additive models (GAMs). The assessment assigned 93% (ECOfast) or 96% (ECOfast-NIS) of the sites to a *moderate to bad* ecological status, indicating an alarming situation in the shallow rocky reefs of the NE Mediterranean. Sites evaluated as *poor* or *bad* were characterized by extensive coverage of ephemeral macroalgae, absence or minimal presence of large indigenous carnivorous fish, and complete absence of one to three out of five invertebrate functional trophic groups. The community composition of macroalgae, herbivorous species, and carnivorous fishes differed between the 5 m and 15 m depth zones. Surface temperature and carnivorous fish occurrence were the most important tested predictors of the ecological status of shallow rocky reefs. The best GAMs showed that the ECOfast score declined with sea surface temperature and increased with the occurrence of carnivorous fish; ECOfast-NIS declined with sea surface temperature and the occurrence of non-indigenous fish and increased with the occurrence of carnivorous fish. The non-destructive and integrative nature of this approach, its speed of data acquisition and analysis, and its capacity to account for highly mobile predatory fish and non-indigenous species render the ECOfast index a novel, robust, and valuable tool for assessing the ecological status of shallow rocky reefs.

Keywords: non-destructive methods, food webs, visual surveys, ecosystem structure, ecological status, Marine Strategy Framework Directive

1. Introduction

Understanding the structure and function of shallow rocky reefs under the influence of natural and anthropic stressors is essential for evaluating their ecological status (ES) (Bevilacqua et al., 2021). Marine food webs describe trophic interactions of species

assemblages, depicting ecosystems' biotic structure and function (Valiela, 2015). Within a complex rocky reef food web, the trophic structure can be top-down controlled by predation (Prato et al., 2013), bottom-up controlled by resource limitation (Paine, 1974, Estes, 1996), or determined by the combined effect of both (Menge, 2000). Coastal food webs are affected by persistent cumulative pressures from human activities in terrestrial and marine environments (Hall, 2002; Kytinou et al., 2020), which can impact communities and food web dynamics (Sala, 2004), alter the ecosystem's structure and function (Thibaut et al., 2017), and cause sudden shifts between alternate ecosystem states (Jungblut et al., 2018). Such dramatic, abrupt, and persistent changes in the ecosystems are defined as regime shifts (Beisner et al., 2003; Scheffer and Carpenter, 2003), with often catastrophic consequences and major losses of biodiversity and ecosystem services (Rocha et al., 2015; Conversi et al., 2015; Möllmann et al., 2015).

Characterized as one of the most heavily impacted marine regions globally (Halpern et al., 2008; Lejeune et al., 2010; Micheli et al., 2013), the Mediterranean Sea is strongly affected by the cumulative effects of overfishing (Sala et al., 2012), climate change (Rilov, 2016), pollution (Tornero and Hanke, 2016), non-indigenous invasive species (Katsanevakis et al., 2014; Tsirintanis et al., 2022), coastal development (Meinesz et al., 1991), and destructive fishing practices (Guidetti, 2011). Large-scale regime shifts of reef ecosystems from a state of diverse algal forests dominated by perennial, canopy-forming species to an alternate, less structurally complex state of ephemeral turf-forming species or barren grounds have been extensively reported in the Mediterranean Sea (Sala et al., 2012; Thibaut et al., 2017; Bevilacqua et al., 2021). Mediterranean canopy-forming macroalgae belong primarily to the perennial genera of *Cystoseira sensu lato (s.l.)* (Sales et al., 2012; Bevilacqua et al., 2021) and *Sargassum* (Thibaut et al., 2005, 2015, 2017), which form complex forest-like structures, and act as a refuge for numerous other macroalgae, invertebrates (Sales et al., 2012; Piazzini et al., 2018), and fishes (Cheminée et al., 2013). The regime shift of Mediterranean reefs to turf-dominated or barren grounds is accompanied by a dramatic loss of associated biodiversity, ecosystem functions, and services (Cheminée et al., 2013; Thibaut et al., 2017; Bevilacqua et al., 2021).

Historically, marine resources of the Mediterranean Sea have been heavily exploited, leading to fishing down the Mediterranean food webs (Briand, 2000) and the simplification of food web structure (Lotze et al., 2011). The decrease of key predatory fishes in shallow Mediterranean rocky reefs is known to promote the overpopulation of native sea urchins, which in turn overgraze macroalgal beds (Sala et al., 1998), leading to highly degraded rocky reefs and simplified coastal food webs (Pinnegar, 2000; Bevilacqua et al., 2021). On top of the already complicated set of interactions and cascade effects governing the structure and function of shallow rocky reef food webs, the introduction of invasive non-indigenous species (NIS) has been a critical driver of change. The structure of eastern Mediterranean coastal food webs has been strongly affected by invasive NIS (Sala et al., 2011), which have already dominated the rocky reefs of the Levantine basin (Rilov et al., 2018). Overpopulation of the invasive herbivorous fish *Siganus luridus* and *Siganus rivulatus* threatens *Cystoseira s.l.* beds and the communities they support (Vergés et al., 2014; Gianni et al., 2017; Rilov et al.,

2018). As an umbrella of multiple stressors, climate change massively impacts Mediterranean Sea ecosystems (Cramer et al., 2018). The entire Mediterranean basin is facing high rates of sea surface warming (Pisano et al., 2020), with the highest values estimated in the eastern Mediterranean ecoregions (Skliris et al., 2012). Persistent marine heatwaves have been associated with mass mortality events, severely decreasing invertebrate and macroalgal assemblages (Garrabou et al., 2019, 2022). Ocean warming and acidification can affect *Cystoseira s.l.* assemblages (Celis-Plá et al., 2017; Falace et al., 2021), multiple invertebrates (Pandori and Sorte, 2019), fish species and rocky reefs' structure and function (Rilov et al., 2019).

Integrated ecosystem-based approaches are essential when assessing the ES of rocky reefs under the impact of multiple stressors, as they account for multiple food web compartments and their interactions and can detect regime shifts often affecting many trophic levels (TLs) (Libralato et al., 2014; Rocha et al., 2015; Thibaut et al., 2017). In contrast to earlier approaches (e.g., Water Framework Directive 2000/60/EC, Marine Strategy Framework Directive 2008/56/EC) focusing on single taxonomic groups (e.g., macroalgae – CARLIT: Ballesteros et al., 2007; EEI-c: Orfanidis et al., 2011), most recent European policies have highlighted the urgent need for the development of integrated indices (Bevilacqua et al., 2021). Only recently, an ecosystem-based index was developed in the NW Mediterranean Sea for the ecological assessment of shallow rocky reefs (reef-EBQI: Thibaut et al., 2017).

This study introduces ECOfast, a novel ecosystem-based ecological index, and evaluates its application on rocky reefs in the NE Mediterranean, specifically the Aegean and Ionian Seas. The proposed approach is based on assessing multiple macroscopic components of rocky reef food webs (i.e., macroalgae, invertebrates, and fish), based on pre-selected key species and predefined macroalgal morphofunctional groups, using non-destructive visual survey methods. The ECOfast index is characterized by its ease of field application, fast field and analysis procedures, and capacity to account for highly mobile predatory fishes and NIS, commonly overlooked by other methodological approaches.

2. Methods

2.1. ECOfast index

2.1.1 General description

The ECOfast is a rapid ES assessment tool designed for shallow (0-20m), well-lit rocky reefs. The index focuses on a specific set of pre-selected species, including habitat-forming, key, commercially important, and non-indigenous species, across a broad range of TLs (1.00 – 4.53). The field protocol is applied through scientific diving and exclusively uses non-destructive visual survey methods. A key feature of this approach is that no fixed transects are required. Instead, each site is assessed by investigating an extensive area through six 10-min intervals of active search (hereafter referred to as transects) at a steady swimming speed. Two divers scan the same six transects; the first diver swims ahead, recording mobile fish species, while the second follows a few

meters behind, recording macroalgae and actively searching for targeted invertebrates and cryptic fish. The two observers meet at the end of each transect for safety check and to set the time and starting point of the next 10-min transect. Three replicate transects are conducted at the 15 m and three at the 5 m depth zone. This approach allows for recording of highly mobile fish species, such as high TL carnivores (Table 1), detected at greater distances, which are typically ignored in standard strip transects used for fish visual surveys (Prato et al., 2017). However, to reduce potential bias due to reduced visibility, it is recommended to conduct fish transects when visibility is at least 10 m. Additionally, the approach of freely moving and surveying extensive areas enables the detection and recording of scarcely distributed invertebrates, which is often the case for many large-sized high TL carnivores (Table 2).

Field data are readily available for the index estimation straight after fieldwork. The index estimation consists of three distinct sub-indices: ECOfast-m for macroalgae, ECOfast-f for fish, and ECOfast-i for invertebrates, the calculation of which follows a simple scoring system. Eventually, the ECOfast index classifies shallow rocky reefs into five ES categories (i.e., *bad*, *poor*, *moderate*, *good*, *high*). Furthermore, there are two proposed versions of the ECOfast index with distinct ecological assumptions. (1) In the basic version of the ECOfast index, non-indigenous species (NIS) are assessed based on their functional role in the rocky reef food web. Any potential negative impact of NIS on the local food web is considered indirectly through the absence or reduced presence of other species that may be affected. (2) The second version, namely ECOfast-NIS, assumes that certain NIS have significant negative impacts on the local food web and are thus given negative values *a priori*. The main methodological aspects of ECOfast index are summarized in Figure 1.

2.1.2 Macroalgae sub-index

The ECOfast-m sub-index is based on the condition that high ES is characterized by a structurally complex macroalgal community that provides many different microhabitats and supports a high level of species diversity. The percentage cover of each macroalgal group (based on Thibaut et al., 2017) and NIS (Table S1, Figure 1) is visually estimated *in situ*, and scores are given for the entire length of each transect. ECOfast-m classifies the status of macroalgal assemblages into five grades of ecosystem status (0-4) based on the percentage cover of four different macroalgal morphofunctional groups (i.e., arborescent perennial, shrubby, turf, encrusting; Thibaut et al., 2017) (Table S1). Transects with an extensive coverage ($\geq 50\%$) of arborescent perennial macroalgae are assigned to the highest ES (i.e., a value of 4). In contrast, turf- or encrusting-dominated transects are linked to the lowest ES (i.e., a value of 0). The value attributed to each transect is hierarchically determined, prioritizing the percentage cover value of the highest macroalgal morphofunctional group in the hierarchy: arborescent perennial > shrubby > turf/encrusting. For example, 10% coverage of arborescent perennial macroalgae and 90% of shrubby macroalgae in a given transect will result in a value of 3 (Table S1). Each transect is evaluated individually; the overall ECOfast-m score per site is defined as the average value of the scores of the six surveyed transects (see supplementary excel file, sheet: ECOfast-m).

The ECOfast-NIS-m sub-index penalizes non-indigenous macroalgal species. The highest the percentage cover of NIS within a transect, the higher the penalties (Table S1, Figure 1). Negative values due to NIS are subtracted from each transect's score before estimating the final ECOfast-NIS-m score. In both versions of the ECOfast-m index, the morphofunctional role of non-indigenous macroalgae is considered. For example, *Styopodium schimperi* (Kützing) Verlaque and Boudouresque, 1991 and *Asparagopsis* spp. are classified as shrubby, whereas *Caulerpa cylindracea* Sonder, 1845 as turf.

2.1.3 Fish sub-index

The ECOfast-f sub-index is based on the condition that high ES corresponds to a complex food web with large individuals, key species, and high species richness of selected fishes across all functional trophic groups (FTGs). The visual survey protocol follows the main principles of the Fish Assemblage Survey Technique (FAST: Seytre and Francour, 2008, 2009; Ben Lamine et al., 2018), with additions to the species list to adapt it to the rocky reefs of the NE Mediterranean. The presence/absence of two size classes (small and large) of 30 selected fish species is investigated in each transect. The ECOfast fish species list (Table 1) includes species across all TLs (2.00 – 4.53), key species (i.e., critically contributing to the maintenance of the food web's structure and function, like high TL carnivores), species targeted by professional and recreational fishing (nets, angling, spearfishing) based on expert judgment and Froese and Pauly (2022), and certain invasive NIS (following Tsirintanis et al., 2022).

Fish are assigned into two-size classes, *large*: i.e., longer than two-thirds of the maximum species size recorded in the eastern Mediterranean (Froese and Pauly, 2021), and *small*: i.e., shorter than two-thirds of the maximum species size (Table 1). If only *small* individuals of a species are present in a transect, the species contributes to the index with one scoring unit, whereas when *large* individuals are present, it contributes with two units. Certain species are given higher values when they meet one of the following criteria: they are (1) carnivores of TL higher than 3.70 (Froese and Pauly, 2022; Karachle and Stergiou, 2017; Savva et al., 2020), (2) species of the IUCN categories Near Threatened, Vulnerable, Endangered, Critically Endangered (IUCN, 2022), and (3) species with high vulnerability to fishing (i.e., $\geq 50\%$ *sensu* Froese and Pauly, 2022). When at least one of these criteria is valid, the species is attributed a higher value: i.e., two units if only *small* individuals are present and three units if *large* individuals are present (Table 1, Figure 1). In the ECOfast-NIS-f sub-index, non-indigenous species are assigned the value of -1 if only *small* individuals are present and -2 if *large* individuals are present (Table 1, Figure 1). The final score of fish for each transect is the sum of values of all detected species. The ECOfast-f score per site is the average value of the scores of all six transects (supplementary excel file, sheet: ECOfast-f).

Moreover, fish species are classified into four FTGs according to their feeding habits (similar to Karachle and Stergiou, 2017; Froese and Pauly, 2022): H: herbivores or omnivores with a preference for phytobenthos, $2.00 \leq TL < 2.90$; O: omnivores, $2.90 \leq TL < 3.70$; C_L: low TL carnivores, $3.70 \leq TL < 4.00$; C_H, high TL carnivores, $TL \geq 4.00$

(Table 1). If any of the four FTGs is entirely missing from a site, a penalty of 15% of the site's score is subtracted from the ECOfast-f score (supplementary excel file, sheet: ECOfast-f). Penalizing the ECOfast-f score is based on the assumption that the better the ES, the more complex food webs are, with a good representation of all FTGs. Two high TL carnivore species (i.e., *Euthynnus alletteratus* and *Sarda sarda*), which are primarily pelagic with a loose association to reefs, only contributed to avoiding the FTG-related penalty and not to the ECOfast-f score (supplementary excel file, sheet: ECOfast-f).

2.1.4 Invertebrate sub-index

The ECOfast-i sub-index is based on the condition that high ES corresponds to a complex food web with key species and high species richness of selected invertebrates across all FTGs. The presence/absence of 33 pre-defined invertebrates (hereafter ECOfast invertebrates) (Table 2) is recorded along the same six transects. Detection of cryptic invertebrates is strengthened by using underwater torches and actively searching within the selected species' microhabitats. The selected taxa include representative species from a wide range of TLs (2.00 – 4.27), key species (i.e., critically contributing to the maintenance of the food web's structure and function, like high TL carnivores), species targeted by professional and recreational fisheries (Palomares and Pauly, 2022; expert judgment) and certain non-indigenous species. Given the sedentary lifestyle of most macroinvertebrates and their inability to move over large distances once environmental conditions become unfavourable, species with a high vulnerability to climate change and local stressors (Jones and Cheung, 2018; expert judgment) are also considered.

Certain invertebrates (Table 2) contribute with higher values in the ECOfast-i index (twice the value of the other taxa) if at least one of the following three criteria is met: they are (1) species of a TL higher than 2.90 (Palomares and Pauly, 2022; DORIS, 2021; species-specific literature as in Table 2), (2) species of the IUCN categories Near Threatened, Vulnerable, Endangered, Critically Endangered (IUCN, 2022), and (3) species of high vulnerability to fishing ($\geq 50\%$ *sensu* Palomares and Pauly, 2022). In the ECOfast-NIS-i sub-index, the NIS are given a value of -1 (Table 2, Figure 1). The ECOfast score for each transect results from the sum of all values assigned to the detected taxa. The ECOfast-i score per site is defined as the average score of the six transects (supplementary excel file, sheet: ECOfast-i).

Invertebrate taxa are further classified into five FTGs according to their feeding habits (Palomares and Pauly, 2022; DORIS, 2021; species-specific literature as in Table 2): H: herbivores or omnivores with a preference for phytobenthos, $2.00 \leq TL < 2.90$; D: detritivores; F: filter-feeders and suspension-feeders; C_L: low TL carnivores, $2.90 \leq TL < 3.70$; C_H: high TL carnivores, $TL \geq 3.70$ (Table 2). If an invertebrate FTG is entirely missing from a site, a penalty equal to 5% of the site's score is subtracted from the ECOfast-i score (supplementary excel file, sheet: ECOfast-i).

2.1.5 Integrating ECOfast sub-indices

To combine all three sub-indices in an integrated index, these are standardized to a common zero-to-one scale corresponding to the following five ES classes: *bad* [0, 0.2], *poor* (0.2, 0.4], *moderate* (0.4, 0.6], *good* (0.6, 0.8], *high* (0.8, 1], in line with the Marine Strategy Framework Directive 2008/56/EC. To standardize the sub-indices, the boundaries among the five ES classes (*bad*, *poor*, *moderate*, *good*, *high*) for each sub-index are determined by five experts on the ecology of NE Mediterranean rocky reef ecosystems, based on hypothetical community compositions (supplementary excel file, sheet: ECOfast). For example, to determine the boundary between *good* and *high* ES for fish communities, experts answer the question ‘What should be the minimum set of fish records in the six transects of the ECOfast-f index so that the site qualifies for *high* ES?’. Each expert assesses the boundaries between consecutive ES classes independently, and the average among all experts is estimated (Figure S1). Linear models are fitted to the pairs of average boundary values of each ES class to standardize sub-index scores (Figure 2, supplementary excel file, sheet: ECOfast). The final scores of ECOfast and ECOfast-NIS are estimated as the average value of the standardized three scores of the respective macroalgae, fish and invertebrates’ sub-indices (Figure 1, supplementary excel file, sheet: ECOfast).

2.2. Case study of ECOfast implementation

2.2.1. Fieldwork

The *ECOfast* visual survey protocol was applied to assess 28 shallow rocky reef sites in the Aegean and Ionian Seas (Greece, NE Mediterranean) (Figure 3) during the warm season (June to October) of 2019-2021 (Table S3). During this time of the year, macroalgal stands have grown enough to allow identification and representative coverage estimation, and certain fish species show their maximum association with rocky reefs. The study area included shallow rocky reefs (0-20 m) of Greece. Sampling sites were selected based on the availability of rocky substrates in areas of varying fishing pressure and logistic constraints. Two sites were located inside the partially enforced (since 2003) Marine Protected Area (MPA) of the “National Marine Park of Alonissos Northern Sporades” (N. Aegean), three inside the recently (2019) enforced MPA of Gyaros Island (S. Aegean), and the rest in non-protected areas (Figure 3).

2.2.2. Depth zones

Wilcoxon signed-rank tests were conducted to investigate significant differences in the occurrence of key species and species groups between 15 m and 5 m. The selected species and groups play an essential role in the structure and function of shallow rocky reefs. They include perennial and shrubby macroalgae, turf macroalgae, the herbivorous sea urchins *Arbacia lixula* and *Paracentrotus lividus*, the herbivorous fish species *Sarpa salpa*, Siganidae (i.e., *S. luridus* and *S. rivulatus*), and a species group of typical, carnivorous, rocky reef fish species, exerting top-down control in Mediterranean, shallow rocky reef food webs (i.e., *Dentex dentex*, *Seriola dumerili*, *Epinephelus costae*, *Scorpaena scrofa*, *Pterois miles*, *Muraena helena*, *Labrus viridis*, *Epinephelus marginatus*). An occurrence index was estimated for each of the above

species / species groups based on their presence/absence within each of the three replicate transects at each depth. *Large* fishes contributed to their occurrence indices with two units, while *small* ones with one unit. The rest species / species groups contributed to their occurrence indices with one unit.

2.2.3. ECOfast explanatory variables

Limited available information from the study area shows that overfishing (Sala et al., 2012, Sini et al., 2019), climate change (Garrabou 2022), invasive species (Tsirintanis et al. 2022), and pollution (Tsiamis et al., 2013) can affect the structure and function of shallow rocky reefs. Generalized additive models (GAMs) were applied to investigate the variance of ECOfast and ECOfast-NIS scores in response to four explanatory variables and their combined effects. The selected variables were: (1) the protection status of each site (i.e., located within or outside an MPA), (2) average sea surface temperature (T) per site for the years 2019-2021, extracted from Copernicus Marine daily gap-free (L4) satellite maps at ultra-high (0.01°) spatial resolution over the Mediterranean Sea (DOI product - <https://doi.org/10.48670/moi-00172>, Nardelli et al., 2013), (3) *Siganidae occurrence index*, estimated as the number of transects among the six replicates of each site where *S. luridus* and *S. rivulatus* were present (*large* fish contributing with two units when present, while small ones with one), (4) *carnivores occurrence index*, estimated as before, for high TL carnivorous fishes (Table 1).

For the selection of the best possible model, the information theory approach (Burnham and Anderson, 2002) was applied: two sets of 16 candidate models each for the ECOfast index (Table S6) and ECOfast-NIS version (Table S7) were constructed respectively, with candidate models including all possible combinations of the four explanatory variables affecting ECOfast and ECOfast-NIS scores. Model selection was based on the Akaike's information criterion (AIC: Akaike, 1973). The AIC differences, $\Delta_i = AIC_i - AIC_{min}$, were computed over all candidate models. The 'Akaike weight' w_i of each model was calculated to quantify the plausibility of each model, where $w_i = \exp(-0.5\Delta_i) / \sum_j \exp(-0.5\Delta_j)$. The 'Akaike weight' is considered as the weight of evidence in favour of model i being the actual best model of the available set of models (Akaike, 1983; Katsanevakis et al., 2010). The sum of Akaike weights of all models where each variable occurs (w_{i+}) reflects the relative importance of variable j ($j = 1$ to 4); the larger the sum of Akaike weights the more important that variable is, relative to the other variables (Burnham and Anderson, 2002).

Furthermore, ECOfast and ECOfast-NIS scores were compared in sites located within and outside the MPAs, with Wilcoxon rank-sum tests. R v.4.2.1 (R Core Team, 2022) was used for model fitting (mgvc package), and the Wilcoxon rank-sum and signed-rank tests.

3. Results

3.1. Macroalgae sub-index

According to the standardized ECOfast-m sub-index (range: 0.00 – 0.89), four sites were classified in *bad*, 12 in *poor*, ten in *moderate*, one in *good*, and one in *high* ES

(Figure 4B, Figure 5C, Table S2). The standardized ECOfast-NIS-m sub-index (range: 0.00 – 0.65) resulted in seven sites of *bad*, 14 *poor*, six *moderate*, and one *good* ES (Figure 4F, Figure 5D, Table S2). The variability among experts on the number of sites belonging to each ES, based on their set boundaries, was low for both sub-indices, particularly ECOfast-m (Figure S1 B, F).

The arborescent perennial morphofunctional group had high coverage ($\geq 50\%$) in only 7% of the sites, intermediate coverage (5-50%) in 32%, and complete absence or minimal coverage ($< 5\%$) in 61% of the sites. Non-indigenous macroalgae were present in 75% of the sites, had low coverage (0-5%) in 39%, intermediate coverage [5-20%) in 25%, high coverage [20-50%) in 7%, and very high coverage ($\geq 50\%$) in 4% of the sites. The most prolific non-indigenous macroalgae were *S. schimperi* (coverage exceeding 50% in some cases), *Asparagopsis taxiformis* (Delile) Trevisan de Saint-Léon, 1845 (coverage up to 50%), *C. cylindracea* (coverage less than 5%) and the cryptogenic *Ganonema farinosum* (J.V.Lamouroux) K.-C.Fan and Y.-C.Wang, 1974 (coverage up to 20%).

3.2. Fish sub-index

The standardized ECOfast-f sub-index (range: 0.41-0.68) classified 23 sites in *moderate* and five sites in *good* ES (Figure 4C, Figure 5E, Table S2). The standardized ECOfast-NIS-f sub-index (range: 0.09 – 0.65) evaluated three sites in *bad*, eight in *poor*, 16 in *moderate*, and one in *good* ES (Figure 4G, Figure 5F, Table S2). The variability among experts on the number of sites belonging to each ES, based on their set boundaries, was moderate for both sub-indices (Figure S1 C, G).

All four fish FTGs were present in all sites. All sites classified as *good* had at least one *large* indigenous high TL carnivore. Sites evaluated as *bad* by the ECOfast-NIS-f sub-index had *large* herbivorous Siganidae present in all surveyed transects. All targeted high TL fishes were absent from a large number of sites, [e.g., *Phycis phycis* (96%), *S. dumerili* (82%), *Sphyraena sphyraena* (82%), *S. scrofa* (71%), *D. dentex* (64%), *M. helena* (64%), *E. marginatus* (46%)], as well as certain low TL carnivores [i.e., *Sciaena umbra* (96%), *L. viridis* (68%)]. Instead, other low TL carnivores, such as *Serranus cabrilla* and *Serranus scriba*, were common. Site AL1, located in Zone A of the National Marine Park of Alonissos, ranked first among all surveyed sites in ECOfast-f and ECOfast-NIS-f and hosted at least one (maximum three) high TL carnivore in each transect. The omnivorous fishes *Diplodus sargus* (100% presence), *Diplodus vulgaris* (96%) and *Oblada melanura* (96%) were widespread in the study area, although not present in all replicate transects or often only present in *small* sizes. The omnivores *Symphodus tinca* (100% presence) and *Mullus surmuletus* (75%) were also common. Both indigenous herbivores, i.e., *S. salpa* and *Sparisoma cretense*, were common, with the former being present in all sites and the latter in all but one site. Herbivorous, non-indigenous Siganidae were present in 75% of all sites, primarily absent from northern latitudes. Between the two species of Siganidae, *S. luridus* (present in 75% of sites) was more common in the study area and was recorded in *large* sizes at all transects and sites of the SE Aegean Sea. The non-indigenous high TL carnivores *P. miles* and *Fistularia commersonii* were present in 43% and 4% of all sites, respectively (for all species, see

Table S4). Usually, sites with a species presence, hosted at least one *large* individual of this species. But *large*, indigenous high TL carnivores were scarce in the study area and most stations (64%) hosted none or only one *large* individual in only one transect.

3.3. Invertebrate sub-index

The standardized ECOfast-i sub-index (range: 0.13-0.74) classified six sites in *bad*, seven in *poor*, 12 in *moderate*, and three in *good* ES (Figure 4D, Figure 5G, Table S2). The standardized ECOfast-NIS-i sub-index (range: 0.07 – 0.74) evaluated eight sites in *bad*, six in *poor*, 11 in *moderate*, and three in *good* ES (Figure 4H, Figure 5H, Table S2). The variability among experts on the number of sites belonging to each ES, based on their set boundaries, was moderate for both sub-indices (Figure S1 D, H).

All sites that the sub-index classified in *good* ES were characterized by numerous ECOfast invertebrates ($n \geq 15$) of most FTGs (≥ 4). Instead, all sites assigned to a *bad* ES sheltered fewer ECOfast invertebrates ($n \leq 9$) of limited FTGs (≤ 4). Of the 28 sites, 93% lacked at least one of the five invertebrate FTGs. High TL carnivores were missing in 75% of the sites, detritivores in 43%, low TL carnivores in 25%, whereas herbivores/omnivores with a preference for phytobenthos and filter-/suspension-feeders were present in all sites. Furthermore, 29% of all sites lacked both detritivores and high TL carnivores, and 21% completely lacked either low or high TL carnivorous invertebrates. Three FTGs (i.e., detritivores, high and low TL carnivores) were simultaneously absent in 11% of all sites. At the species level, certain invertebrates were absent from most sites, i.e. all high TL carnivores, such as *Octopus vulgaris* (absence in 93% of all sites), *Sepia officinalis* (96%), *Charonia variegata* (93%), *Scyllarides latus* (96%), *Palinurus elephas* (96%), most low TL carnivores such as *Marthasterias glacialis* (82%), *Maja squinado* (96%), *Ophidiaster ophidianus* (68%), both corals *Cladocora caespitosa* (96%) and *Balanophyllia (Balanophyllia) europaea* (89%), three bivalves *Pinna nobilis* (100%), *Ostrea edulis* (100%) and *Arca noae* (96%), and the exploited tunicates *Microcosmus* spp. (68%) (for all species, see Table S5). The herbivorous alien sea urchin *Diadema setosum* was common in nine S. Aegean sites, while the herbivorous, non-indigenous decapod *Percnon gibbesi* was limited to two S. Aegean sites.

3.4. ECOfast and ECOfast-NIS versions

According to the standardized ECOfast index (score range: 0.28-0.69), half of the sites were classified as *poor*, 12 as *moderate*, and two as *good*, while no sites were assigned a *bad* or *high* ES (Figure 4A, Figure 5A). According to the standardized ECOfast-NIS version of the index (score range: 0.08-0.63), five sites were classified as *bad*, 13 as *poor*, nine as *moderate*, one as *good*, with no sites assigned a *high* ES (Figure 4E, Figure 5B, Table S2). The variability among experts on the number of sites belonging to each ES, based on their set boundaries, was generally low for both versions of the index, with all differences referring to consecutive categories (Figure S1 A, E).

Sites classified as *good* by the ECOfast and ECOfast-NIS presented extensive coverage of perennial macroalgae, *large* indigenous carnivorous fishes, multiple ECOfast invertebrates ($n \geq 12$), and most FTGs thereof (≥ 4). On the other hand, sites evaluated as *poor* or *bad* presented extensive coverage of turf/encrusting macroalgae and/or low coverage by shrubby macroalgae, usually absence or minimal presence of *large* sizes of indigenous high TL carnivorous fishes, and absence of up to three invertebrate FTGs. Site AL1, located in zone A of the National Marine Park of Alonissos and SK - Skyros (N. Aegean), had the highest scores among all surveyed sites and were classified in *good* ES by ECOfast index (Table S2, Figure 5A). The lowest scores of both versions of the index were assigned to sites of the SE Aegean Sea, specifically the islands of Karpathos, Kalymnos, and Crete (Table S2).

Non-indigenous species that primarily affected the ECOfast and ECOfast-NIS scores were the macroalgae *S. schimperi* and *A. taxiformis*, which displayed extended coverage in specific sites, and the herbivorous fishes *S. luridus* and *S. rivulatus* with repetitive presence of *large* individuals in many sites. In all cases of different ES classifications between ECOfast and ECOfast-NIS and among respective sub-indices, the ECOfast-NIS version assessed the lowest ES. The ECOfast and ECOfast-NIS versions of the index assigned different ES in 36% of all sites. The different ES classifications between the ECOfast and ECOfast-NIS sub-indices were 32% of sites for ECOfast-m, 54% for ECOfast-f, and 11% for ECOfast-i (Table S2, Figure 5).

3.5. Depth zones

In 75% of all sites, macroalgal morphofunctional groups differed between the surveyed depths of 5 and 15 m. In 61% of all sites, 5 m had lower perennial and shrubby macroalgae coverage, in 25% of the sites both depth zones had similar macroalgal groups, while in 14%, 5 m had higher perennial and shrubby macroalgae coverage compared to 15 m (Figure 6). Regarding non-indigenous macroalgae, 46% of all sites had a similar percentage cover at both 5 m and 15 m, 39% had a lower percentage cover at 5 m, and 14% had a higher percentage cover at 5 m. Many carnivorous fishes typical of rocky reefs had lower occurrence at 5 m (i.e., *D. dentex*, *S. dumerili*, *E. costae*, *S. scrofa*, *P. miles*, *M. helena*, *L. viridis*, *E. marginatus*). Hence, in sites with species presence, *large* carnivores were very often absent from 5 m [e.g., *D. dentex* and *S. dumerili* (100% absence), *E. costae* (90%), *E. marginatus* (87%), *S. scrofa* (88%), *P. miles* (67%), *M. helena* (70%)], and were only present at the 15 m depth zone. Absence percentages of all ECOfast fishes and invertebrates at both depths, are listed in the supplementary file (Tables S4, S5). Wilcoxon signed-rank tests statistically confirmed that the occurrence index at 5 m was lower for carnivorous fish typical of rocky reefs ($p = 0.00$) and *large* carnivorous fish typical of rocky reefs ($p = 0.00$), as well as for perennial and shrubby macroalgae ($p = 0.00$) (Figure 6, Figure S2). In contrast, occurrence was higher at 5 m for turf macroalgae ($p = 0.01$), the herbivorous sea urchins *A. lixula* ($p = 0.00$) and *P. lividus* ($p = 0.00$), and for the herbivorous fishes *S. salpa*, *S. rivulatus*, and *S. luridus* ($p = 0.00$) (Figure 6, Figure S2).

3.6. MPAs

Average ECOfast and ECOfast-NIS scores were numerically higher in MPAs than in non-protected sites (Figure S3). However, no statistically significant differences were found between the average ECOfast scores of sites in MPAs (0.50 ± 0.13) and in non-protected sites (0.41 ± 0.09) ($p = 0.12$), or between the average ECOfast-NIS scores in MPAs (0.45 ± 0.12) and in non-protected sites (0.33 ± 0.13) ($p = 0.11$).

3.7. GAM results

For ECOfast, among the 16 GAMs tested, models m_{10} (with T and *carnivores occurrence index* as predictor variables) and m_4 (with T , *carnivores occurrence index*, and *Siganidae occurrence index* as predictor variables) gave the lowest AIC score (Table S6). Since m_4 had one variable (*Siganidae*) that was non-significant ($p = 0.23$), the model m_{10} was selected as the best model (Table S6). Furthermore, based on the set of candidate models, carnivores' occurrence ($w_{i+} = 0.99$) and surface temperature ($w_{i+} = 0.98$) were highly supported predictor variables of ECOfast score, whereas *Siganidae* occurrence and protection status had less support from the data (Table S8), further supporting the selection of m_{10} . According to this model, the ECOfast score declines with temperature and generally increases with carnivores' occurrence (Figure S4).

Among the 16 ECOfast-NIS GAMs tested, m_4 gave the lowest AIC score and was selected as the best model (Table S7). According to this model, ECOfast-NIS declines with temperature and siganids' occurrence, and generally increases with carnivores' occurrence (Figure S5). Based on the set of candidate models, the surface temperature was the most important predictor variable of ECOfast-NIS score ($w_{i+} = 0.99$), followed by carnivores' occurrence ($w_{i+} = 0.83$) and *Siganidae* occurrence ($w_{i+} = 0.62$) (Table S8).

4. Discussion

4.1 Application of ECOfast

The ECOfast index can be easily applied with a single two-observer SCUBA dive, and results are available upon completion of fieldwork (Table 3) without further need for laboratory analysis. Both divers require a basic knowledge of the assemblages under study since the protocol demands the *in situ* identification of macroalgal morphofunctional groups, together with certain fish and invertebrate species. The index is based on a semi-quantitative description of shallow rocky reef food webs (other examples in Paine, 1988; Sala, 2004) and assess their status through basic functional or morphofunctional trophic groups alongside a predefined list of selected key species. Such a representation prevents discrepancies due to the spatio-temporal differences in species density, which can dramatically vary independently of ecosystem health (Sala et al., 2004). Nevertheless, the ECOfast-f and ECOfast-i sub-indices assume that the more abundant a species is at any given site, the more transects of this site it will be

detected at, until a point where there is saturation of the occurrence metric, and all transects are populated by at least one individual of the specific species. From that moment on, higher population densities of the same species have no effect to the sub-index. There can be an overweighting bias in that approach, as the discrimination between sites with medium to high target species densities is not possible. However, having configured the weighting of the sub-indices in a way to promote high species richness rather than just higher trophic group biomasses, the aforementioned bias is somewhat controlled in the final assessment of the ecological state.

Moreover, the use of non-fixed survey transects, a distinctive characteristic of the ecosystem-based ECOfast methodology (Table 3), allows for better detection of high TL mobile carnivores, targeted by small-scale or recreational fisheries (Albouy et al., 2010; Prato et al., 2016). Such species are often difficult to detect within a typical, fixed strip transect of a certain width (e.g., 4-m width transects in reef-EBQI), and their low detectability has been highlighted in previous fish assessments (Seytre and Francour, 2008, 2009; Ben Lamine et al., 2018; Thanopoulou et al., 2018).

The ECOfast field protocol covers two depth zones, 5 m and 15 m, to jointly assess the status of rocky reef communities, while accounting for depth-related differences in community structure and human pressures (e.g., Sini et al., 2019). Differences between the two depth zones were indeed detected for macroalgal morphofunctional groups, certain herbivorous and carnivorous fishes, and herbivorous invertebrates, thus affecting the ES assessment. Indices based exclusively on vegetation data from limited shallow depths (0-1 m), such as EEI-c (Orfanidis et al., 2011) and CARLIT (Ballesteros et al., 2007), can readily detect coastal pollution, but cannot adequately account for other human pressures, such as overfishing or climate change, or the way these affect the deeper parts of coastal rocky reefs (Table 3) (Bevilacqua et al., 2021; Savin et al., 2023). For example, the 0-1 m depth zone can often be covered with dense *Cystoseira s.l.* stands, while deeper zones are overgrazed and degraded (Sala et al., 2012; Salomidi et al., 2016; Savin et al., 2023).

One of the main advantages of ecosystem-based approaches is the ability to disaggregate complex indices to check their individual components (Thibaut et al., 2017). The ECOfast index allows the independent examination of three sub-indices to detect the most vulnerable community features at each location, thus enabling the identification of adequate region-specific conservation targets and the development of relevant management measures. Moreover, unlike the ecosystem-based index reef-EBQI developed in the western Mediterranean (Thibaut et al., 2017), the ECOfast index, developed in the oligotrophic and warmer eastern basin, further considers NIS (Table 3), an increasingly important component of the region's coastal ecosystems (Rilov et al., 2018).

4.2 Ecological status assessment

The ECOfast-m sub-index classified most sites in *poor* to *bad* ES, while the ECOfast-NIS-m assessments were even stricter. These results corroborate previous findings (e.g., Sala et al., 2011; Thibaut et al., 2015; Bevilacqua et al., 2020; Savin et al., 2023),

reporting the extensive degradation of shallow Mediterranean reef ecosystems. Within our dataset, this was particularly common at sites of the SE Aegean Sea where the dominance of low homogenous turf or encrusting forms at the expense of 3D structured heterogeneous perennial species, results in simplified reef structure and food webs and loss of biodiversity and ecosystem services.

The ECOfast-f and ECOfast-NIS-f sub-indices classified most sites in *moderate* ES, primarily due to the increased absence of all high TL ECOfast carnivores. Low density and biomass of carnivorous fishes, mainly attributed to overfishing, have been recorded in several areas of the Mediterranean (Sala et al., 2012; Sini et al., 2019). The depletion of high-level carnivores recorded herein can affect their top-down predatory role and trigger cascading effects, which alter the structure and function of the entire shallow rocky reef food web (Sala et al., 2012; Prato et al., 2013). The recorded decline of once-common large Mediterranean reef fishes is consistent with the findings of other studies in the Mediterranean (Lotze et al., 2011; Sbragaglia et al., 2021; Damalas et al., 2022). Nevertheless, the presence of all four fish FTGs at all sites and a few *large* high-level carnivores at specific sites indicates a potential for recovery upon effective protection.

The ECOfast-i and ECOfast-NIS-i sub-indices classified many sites in *poor* to *bad* ES due to the absence of certain FTGs (mainly high TL carnivores and detritivores) and all high TL carnivorous invertebrates. Despite the crucial role of megabenthic invertebrates on ecosystem structure and functioning (Jones, 1973), their contribution to marine food webs and status assessments is globally scarcely investigated (Anderson et al., 2011; Eddy et al., 2017). For example, even though there are available estimations for all ECOfast fish species regarding their TL, fishing vulnerability (Froese and Pauly, 2022), and IUCN Red List status (IUCN, 2022), the respective information is not available for the majority of ECOfast invertebrates (Palomares and Pauly, 2022; IUCN, 2022). At a Mediterranean scale, it is only recently that megabenthic invertebrates started being considered in shallow rocky reef status assessments (Thibaut et al., 2017). Like carnivorous fish, the extensive exploitation of invertebrates (Anderson et al., 2011) can severely alter the rocky reef trophic structure, with subsequent strong ecosystem effects (Eddy et al., 2017).

Overall, the ECOfast and ECOfast-NIS versions of the index classified most sites in *poor* to *bad* ES, depicting the low ES of rocky reefs and their hosted food webs. The ES outcome often differed among the three ECOfast sub-indices applied within the same site for each index (Table S2), highlighting the importance of integrated approaches. The observed dominance of turf or encrusting macroalgae with a low presence of high TL carnivorous fishes and invertebrates reflects a substantial divergence from reference conditions (Sala et al., 2012), and a shift of the whole rocky reef food web to more simplified states. This results in the overall alteration of the rocky ecosystem's functions, including a lack of available habitat for certain species due to decreased 3D complexity in macroalgal assemblages (Cheminée et al., 2013). These findings complement the results of previous studies regarding the decline in the ES of shallow rocky reefs across the entire basin (Bevilacqua et al., 2020, 2021).

In the Greek Seas, integrated assessments of shallow rocky reefs through ecosystem-based biotic indices are limited in space and quantity. Available data classify most sites

in *poor* ES (Bevilacqua et al., 2020; Salomidi et al., 2021). Application of the reef-EBQI index in multiple 5 m sites within Gyaros MPA, including the three sites surveyed herein, revealed *poor* ES of rocky reefs (Salomidi et al., 2021). Similarly, ECOfast-NIS classified two sites of Gyaros MPA in *poor* and one in *moderate* ES, while ECOfast assessed one *poor* and two moderate sites. Signs of degradation have also been previously reported in the few available assessments of different functional compartments in the shallow rocky reefs of the Aegean Sea. They refer to a shift from complex algal forests to homogenized turf or barren substrates, low presence of high TL carnivores, and a high abundance of herbivorous non-indigenous species (Sala et al., 2012; Giakoumi et al., 2012; Bianchi et al., 2014; Salomidi et al., 2016, 2021; Sini et al., 2019; Bevilacqua et al., 2020). Most sites of the Greek Ionian Sea were assigned to moderate ES, but there are no previously published data from this area to compare the ECOfast results.

4.3 Pressures

Mediterranean rocky reefs are known to be severely transformed because of climate change and biological invasions, with multiple collapses of cold-affinity, habitat-forming and keystone species (Bevilacqua et al., 2021; Albano et al., 2021; Tsirintanis et al., 2022), while marine heatwaves are threatening the rocky reef communities through mass mortality events (Garrabou et al., 2022). Herein both ECOfast and ECOfast-NIS significantly declined with temperature, and *poor* or *bad* ES was recorded particularly in rocky reefs of the SE Aegean Sea, where the effects of climate change and biological invasions are more pronounced (Katsanevakis et al., 2020).

Fishing pressure is critical in shaping the structure of Mediterranean coastal food webs (Sala et al., 2004). The presence of carnivorous fish was particularly low at 5 m, possibly due to the greater intensity of spearfishing activities, a critical threat for specific keystone carnivorous fishes, such as groupers (Mavruk et al., 2018; Sbragaglia et al., 2021). Higher abundance and biomass of high TL carnivores at 15 m than 5 m have been highlighted in previous studies in the Aegean Sea (Sini et al., 2019). However, no significant ES difference was evident between MPAs and non-protected sites, while this predictor variable was the least supported from our data in GAMs (Table S8). This is likely due to the low sample size of protected areas and inadequate protection in the sampled MPAs. In the National Marine Park of Alonissos Northern Sporades, enforcement is low; nevertheless, in the no-take zone, *good* ES was found. In Gyaros MPA, a no-take marine reserve was only recently established and enforced; thus, there was inadequate time for recovery. Fish were in a slightly higher status, and the ECOfast-f sub-index classified the fish assemblages of G1 and G3 in *good* ES. Additionally, the ECOfast-f sub-index was higher for sites of Gyaros island compared to the adjacent non-protected site of Andros Island (AN). On the same page, a two-year study after the Gyaros MPA establishment reported an increase of carnivorous fishes, a high occurrence of certain carnivorous invertebrates, and overall positive signs of recovery (Salomidi et al., 2021; Damalas et al., 2022). Despite such promising exceptions, the lack of long-term well-enforced MPAs in the Greek Aegean and Ionian

Seas is an obstacle to understanding the effect of protection (or lack thereof) on the structure of rocky reef food webs.

4.4 Concluding remarks

An ecosystem-based index, based on key food web compartments of the eastern Mediterranean Sea is developed and applied for the first time, confirming the alarming low ES of shallow rocky reefs. The ECOfast index and field protocol are designed to improve the detection and proper consideration of key species and functional groups of the coastal food web, to integratively assess the ecological status of rocky reefs. Two versions of the index (i.e., ECOfast and ECOfast-NIS) account for the different impacts that non-indigenous species can have on shallow rocky reef food webs. One of the two versions can be selected, depending on the focus of relevant studies and the potential negative impact of widespread non-indigenous species in each study area. Separate assessments of three sub-indices, allow for the identification of the most vulnerable ecosystem compartments and setting the appropriate conservation targets. One-time application of ECOfast index can detect shifts between ecosystem states, while application in the same site through consecutive years can monitor the evolution of an ecosystem shift.

The further adaptation of the ECOfast index to fit other reef ecosystems of the world, through careful selection of region-specific keystone and non-indigenous species, and their corresponding maximum size characteristics, would enable its effective application across a broader geographic scale. However, a prerequisite for any future adaptation of the index is that the most important local ecosystem components are predefined along with the main anthropogenic drivers. The ECOfast index can be a valuable monitoring tool for the implementation of the EU Marine Strategy Framework Directive (MSFD, 2008/56/EC) since existing assessments of multiple ecosystem components and their key ecosystem functions are spatially discrete and limited to selected sites in the eastern Mediterranean. Furthermore, it can be used to monitor the ecological status of rocky reefs along the lines of the Habitats Directive (92/43/EC) or upon establishment of Marine Protected Areas to detect how and when fish, invertebrate and macroalgal communities respond.

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Attributions for icons of Figure 6: Fish icons created by Freepik - Flaticon for Carnivorous fishes icon, Integration and Application Network (ian.umces.edu/media-library) with Joanna Woerner, Tracey Saxby and Jane Hawkey attributors for Shrubby, Perennial and Herbivorous sea urchins icons respectively.

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Table 1: Fish species, selected characters, and their contribution to the score of the ECOfast-f sub-index. 2/3ML: 2/3 of the maximum length of the species in the eastern Mediterranean (according to Froese and Pauly, 2021); TL: trophic level (according to Froese and Pauly, 2022; Karachle and Stergiou, 2017; Savva et al., 2020); IUCN: IUCN Red List categories (according to IUCN, 2022), DD: Data Deficient, LC: Least Concern, NT: Near Threatened VU: Vulnerable, EN: Endangered; vulnerability: percentage of vulnerability to fishing (according to Froese and Pauly, 2022); Value S: species contribution to ECOfast-f score for “small” individuals; Value L: species contribution to ECOfast-f score for *large* individuals, additional negative values correspond to the ECOfast-NIS-f scoring system; FTG: functional trophic group, H: herbivores or omnivores with a preference for phytobenthos $2.00 \leq TL < 2.90$, O: omnivores $2.90 \leq TL < 3.70$, C_L: low TL carnivores $3.70 \leq TL < 4.00$, C_H: high TL carnivores $TL \geq 4.00$. Marked in bold are species that are given a higher score, and the respective criterion/a that led to this classification (i.e., $TL > 3.70$; IUCN categories: NT, VU, EN; vulnerability to fishing $\geq 50\%$).

Family	Species	2/3 ML (cm)	TL	IUCN	vulnerability	Value S	Value L	FTG
Siganidae	<i>Siganus luridus</i>	19	2.00	LC	39%	1/-1	2/-2	H
	<i>Siganus rivulatus</i>	16	2.00	LC	36%	1/-1	2/-2	H
Scaridae	<i>Sparisoma cretense</i>	23	2.86	LC	36%	1	2	H
	<i>Sarpa salpa</i>	28	2.00	LC	41%	1	2	H
Sparidae	<i>Diplodus puntazzo</i>	17	3.22	LC	34%	1	2	O
	<i>Spondylisoma cantharus</i>	26	3.34	LC	37%	1	2	O
	<i>Oblada melanura</i>	20	3.38	LC	43%	1	2	O
	<i>Diplodus sargus</i>	22	3.38	LC	63%	2	3	O
	<i>Diplodus vulgaris</i>	20	3.52	LC	32%	1	2	O
	<i>Dentex dentex</i>	52	4.53	VU	66%	2	3	C _H
Labridae	<i>Symphodus tinca</i>	18	3.25	LC	40%	1	2	O
	<i>Labrus merula</i>	20	3.55	LC	44%	1	2	O
	<i>Labrus viridis</i>	22	3.93	VU	34%	2	3	C _L
Mullidae	<i>Mullus surmuletus</i>	22	3.45	LC	42%	1	2	O
Sciaenidae	<i>Sciaena umbra</i>	20	3.75	NT	64%	2	3	C _L
	<i>Serranus cabrilla</i>	17	3.76	LC	55%	2	3	C _L
Serranidae	<i>Serranus scriba</i>	16	3.82	LC	44%	2	3	C _L
	<i>Epinephelus costae</i>	37	3.87	DD	66%	2	3	C _L
	<i>Epinephelus marginatus</i>	40	4.43	EN	64%	2	3	C _H
	<i>Mycteroperca rubra</i>	52	4.13	LC	67%	2	3	C _H
Scorpaenidae	<i>Pterois miles</i>	21	4.00	LC	33%	2/-1	3/-2	C _H
	<i>Scorpaena scrofa</i>	26	4.25	LC	68%	2	3	C _H
Sphyraenidae	<i>Sphyraena sphyraena</i>	67	4.04	LC	49%	2	3	C _H
Muraenidae	<i>Muraena helena</i>	81	4.18	LC	79%	2	3	C _H
Phycidae	<i>Phycis phycis</i>	35	4.25	LC	45%	2	3	C _H
Fistulariidae	<i>Fistularia commersonii</i>	77	4.26	LC	42%	2/-1	3/-2	C _H
Congridae	<i>Conger conger</i>	60	4.31	LC	86%	2	3	C _H
Carangidae	<i>Seriola dumerili</i>	85	4.50	LC	60%	2	3	C _H
Scombridae (only for FTG)	<i>Euthynnus alletteratus</i>	67	4.49	LC	41%	-	-	C _H
	<i>Sarda sarda</i>	60	4.50	LC	33%	-	-	C _H

Table 2: Invertebrates, selected characters, and their contribution to the score of the ECOfast-i sub-index. TL: trophic level (according to Palomares and Pauly, 2022; DORIS, 2021 and species specific literature on feeding habits); IUCN: IUCN Red List categories (according to IUCN, 2022), NE: Not Evaluated, DD: Data Deficient, LC: Least Concern, VU: Vulnerable, EN: Endangered, CR: Critically Endangered; vulnerability: percentage of vulnerability to fishing (according to Palomares and Pauly, 2022); value: proposed value for each taxon, negative values correspond to the ECOfast-NIS-i scoring system; FTG: functional trophic group, H: herbivores or omnivores with a preference for phytobenthos $2.00 \leq TL < 2.90$, D: detritivores, F: filter-feeders and suspension-feeders, C_L: low TL carnivores $2.90 \leq TL < 3.70$, C_H: high TL carnivores $TL \geq 3.70$. Marked in bold are species that are given a higher score, and the respective criterion/a that led to this classification (i.e., $TL > 2.90$; IUCN categories: VU, EN, CR; vulnerability to fishing $\geq 50\%$).

Class	Species	TL	Literature on feeding habits	IUCN	vulnerability	value	FTG
Echinoidea	<i>Sphaerechinus granularis</i>	2.00	Palomares and Pauly, 2022; Vafidis et al., 2020	NE	NE	1	H
	<i>Diadema setosum</i>	2.00	Palomares and Pauly, 2022	NE	NE	1/-1	H
	<i>Paracentrotus lividus</i>	2.40	Palomares and Pauly, 2022	NE	NE	1	H
	<i>Centrostephanus longispinus</i>	2.50	Palomares and Pauly, 2022; Pawson and Miller, 1983; Reguieg et al., 2020	NE	NE	1	H
Malacostraca	<i>Arbacia lixula</i>	2.70	Wangensteen et al., 2011	NE	NE	1	H
	<i>Percnon gibbesi</i>	2.60	Marić et al., 2016; Félix-Hackradt et al., 2018	NE	NE	1/-1	H
Demospongiae	<i>Aplysina aerophoba</i>	2.10	Moutopoulos et al., 2013; Tsagarakis et al., 2010, 2020	NE	NE	1	F
	Black massive sponges	2.10	Moutopoulos et al., 2013; Tsagarakis et al., 2010, 2020; Palomares and Pauly, 2022	NE	NE, 10-20%	1	F
	<i>Halocynthia papillosa</i>	2.10	Moutopoulos et al., 2013; Tsagarakis et al., 2010, 2020; Ribes et al., 1998; Palomares and Pauly, 2022	NE	NE	1	F
Asciacea	<i>Microcosmus</i> spp.	2.10	Moutopoulos et al., 2013; Tsagarakis et al., 2010, 2020	NE	NE, 0-10%	1	F
	Gymnolaemata	Bryozoa erect	2.10	Deehr et al., 2014; Moutopoulos et al., 2013; Tsagarakis et al., 2010, 2020	NE	NE	1
Bivalvia	<i>Arca noae</i>	2.20	Deehr et al., 2014	NE	NE	1	F
	<i>Spondylus gaederopus</i>	2.20	Deehr et al., 2014	NE	NE	1	F
	<i>Ostrea edulis</i>	2.20	Deehr et al., 2014	NE	NE	1	F
	<i>Pinna nobilis</i>	2.20	Deehr et al., 2014	CR	NE	2	F
Holothuroidea	<i>Holothuria</i> spp.	2.20	Deehr et al., 2014	DD/LC	NE, 10-20%	1	D
	<i>Sabella pavonina</i>	2.20	Deehr et al., 2014; Banaru et al., 2013; Tsagarakis et al., 2010, 2020; Palomares and Pauly, 2022	NE	NE	1	F
Polychaeta	<i>Sabella spallanzanii</i>	2.20	Deehr et al., 2014; Banaru et al., 2013; Tsagarakis et al., 2010, 2020; Palomares and Pauly, 2022	NE	48%	1	F
	<i>Hermodice carunculata</i>	3.00	Didierlaurent and Desvignes, 2021	NE	NE	2	C _L
Anthozoa	<i>Balanophyllia (Balanophyllia) europaea</i>	3.00	Lafourcade et al., 2021; Sherwood et al., 2008; Goffredo et al., 2008	LC	NE	2	F

	<i>Cladocora caespitosa</i>	3.00	Allard et al., 2021; Sherwood et al., 2008	EN	NE	2	F
	<i>Echinaster (Echinaster) sepositus</i>	2.50	Lamare and Bertoncello, 2021; Villamor and Becerro, 2010	NE	NE	1	D
Asteroidea	<i>Hacelia attenuata</i>	3.00	Le Granché and Foveau, 2021	NE	NE	2	C _L
	<i>Ophidiaster ophidianus</i>	3.00	Ziemski et al., 2020a; Di Trapani et al., 2020	NE	NE	2	C _L
	<i>Coscinasterias tenuispina</i>	3.10	Ziemski et al., 2020b	NE	NE	2	C _L
	<i>Marthasterias glacialis</i>	3.30	Palomares and Pauly, 2022	NE	NE	2	C _L
Malacostraca	<i>Maja squinado</i>	2.94	Palomares and Pauly, 2022; Welden et al., 2018	NE	12%	2	C _L
	<i>Palinurus elephas</i>	3.34	Palomares and Pauly, 2022; Maran et al., 2021	VU	40%	2	C _L
	<i>Homarus gammarus</i>	3.70	Palomares and Pauly, 2022; Sohler et al., 2020	LC	46%	2	C _H
	<i>Scyllarides latus</i>	3.86	Palomares and Pauly, 2022	DD	35%	2	C _H
Gastropoda	<i>Charonia variegata</i>	4.10	Palomares and Pauly, 2022; Hall et al., 2017	NE	NE	2	C _H
Cephalopoda	<i>Octopus vulgaris</i>	3.74	Palomares and Pauly, 2022; Aussel et al., 2021; Ambrose and Nelson, 1983	LC	78%	2	C _H
	<i>Sepia officinalis</i>	4.27	Palomares and Pauly, 2022	LC	30%	2	C _H

Table 3: Differences of mostly used non-destructive indices for the ecological status assessment of Mediterranean shallow rocky reefs.

Index	Ecosystem-based approach	High detectability of mobile key species	Fast available outcome	Application in >1m depth	Inclusion of NIS	Developed in East Med.	Developed in West Med.
EEI-c	-	-	-	-	+	+	-
Carlit	-	-	+	-	-	-	+
reef-EBQI	+	-	-	+	-	-	+
ECOfast	+	+	+	+	+	+	-

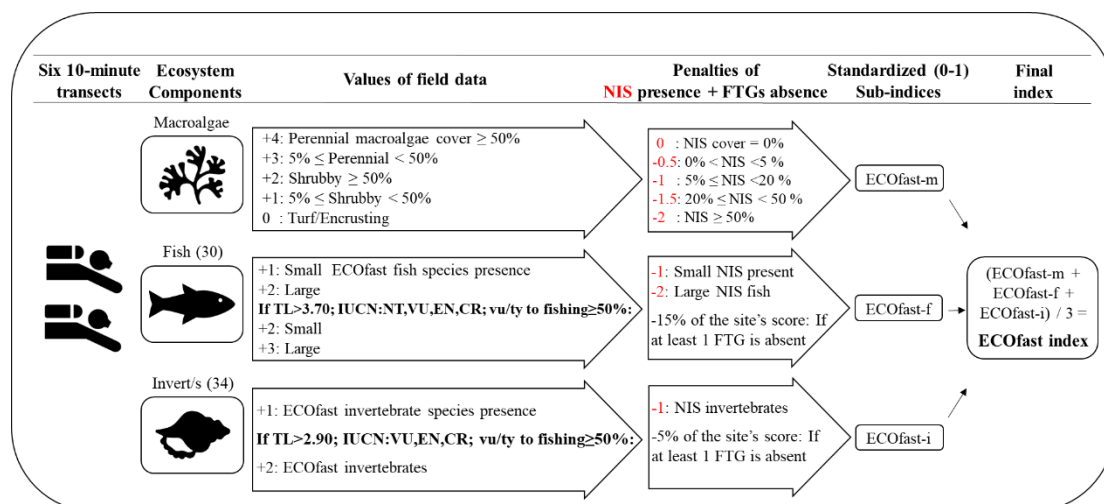


Figure 1: Flowchart summarizing the main methodological aspects of ECOfast index. NIS: Non-indigenous species; FTGs: functional trophic groups; TL: trophic level; IUCN: IUCN Red List categories, NT: Near Threatened, VU: Vulnerable, EN: Endangered, CR: Critically Endangered; vu/ty to fishing: percentage of vulnerability to fishing. Red colour corresponds to the values of the ECOfast NIS version of the index. For a detailed description see also 2.1 Methods. All equations for estimation of ECOfast index are available in the supplementary excel file.

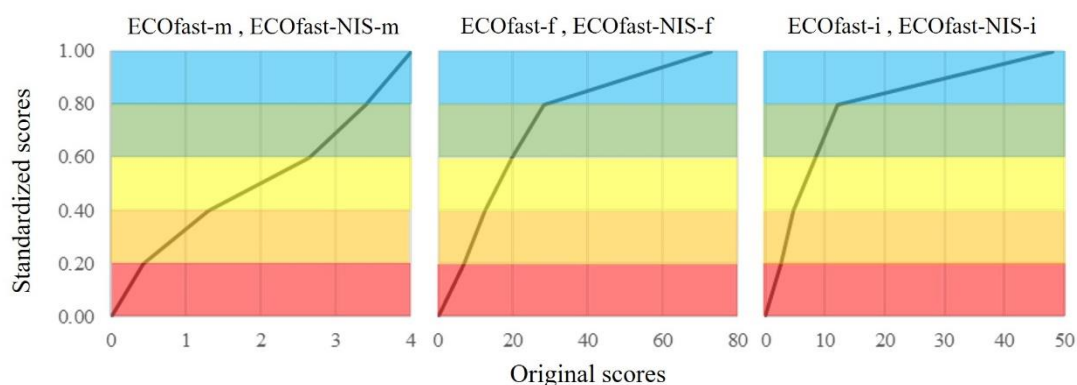


Figure 2: Standardized scores (0.00-1.00) of each sub-index in relation to the original scores, for the ECOfast and ECOfast-NIS versions. Different colours represent *bad* (red), *poor* (orange), *moderate* (yellow), *good* (green), and *high* (blue) ecological status of each sub-index. The boundaries of the original scores and the functions for the score standardization are provided in the supplementary excel (sheet: ECOfast).

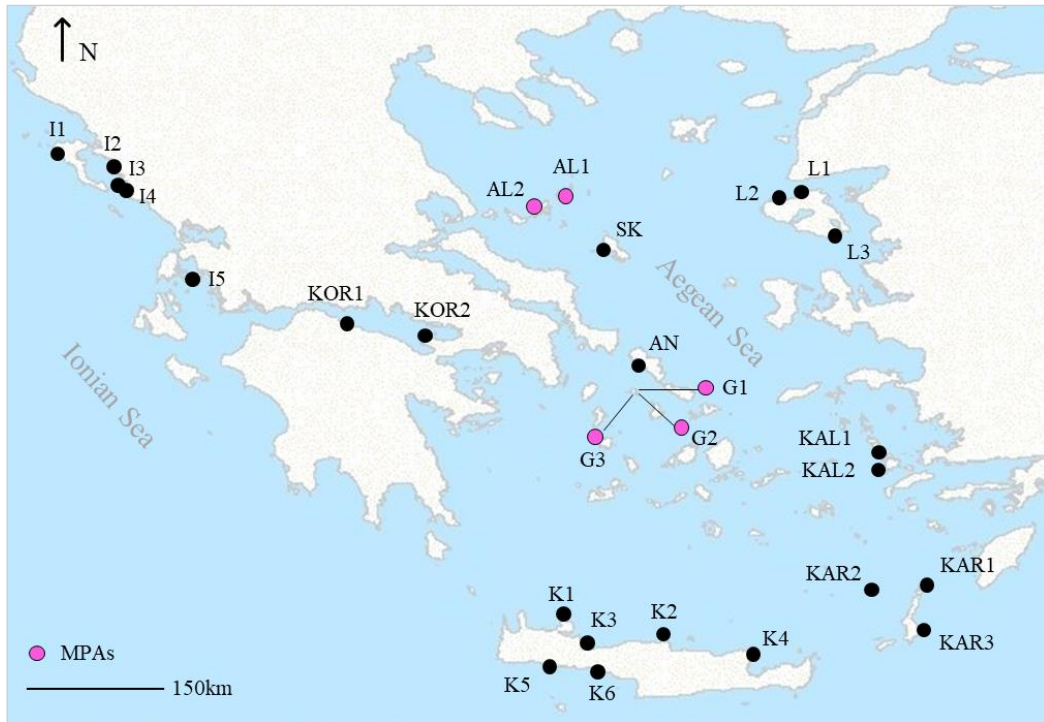


Figure 3: Map of Greece (NE Mediterranean Sea) depicting the 28 rocky reef sites. Pink colour corresponds to sites found within marine protected areas.

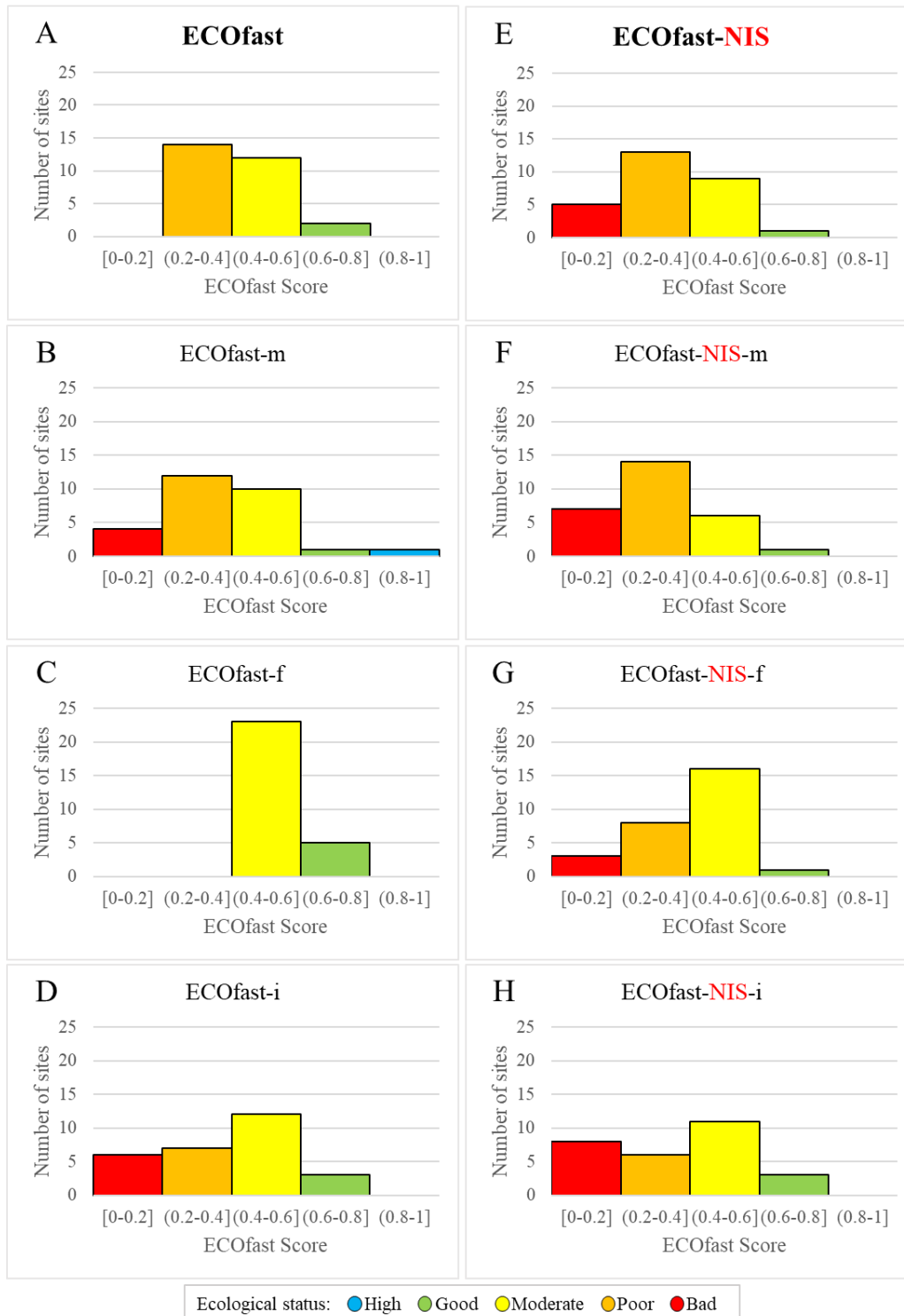


Figure 4: Number of sites per ecological status evaluated by the A) ECOfast index and its B) ECOfast-m, C) ECOfast-f and D) ECOfast-i sub-indices, E) ECOfast-NIS version of the index and its respective three sub-indices (F, G, H).

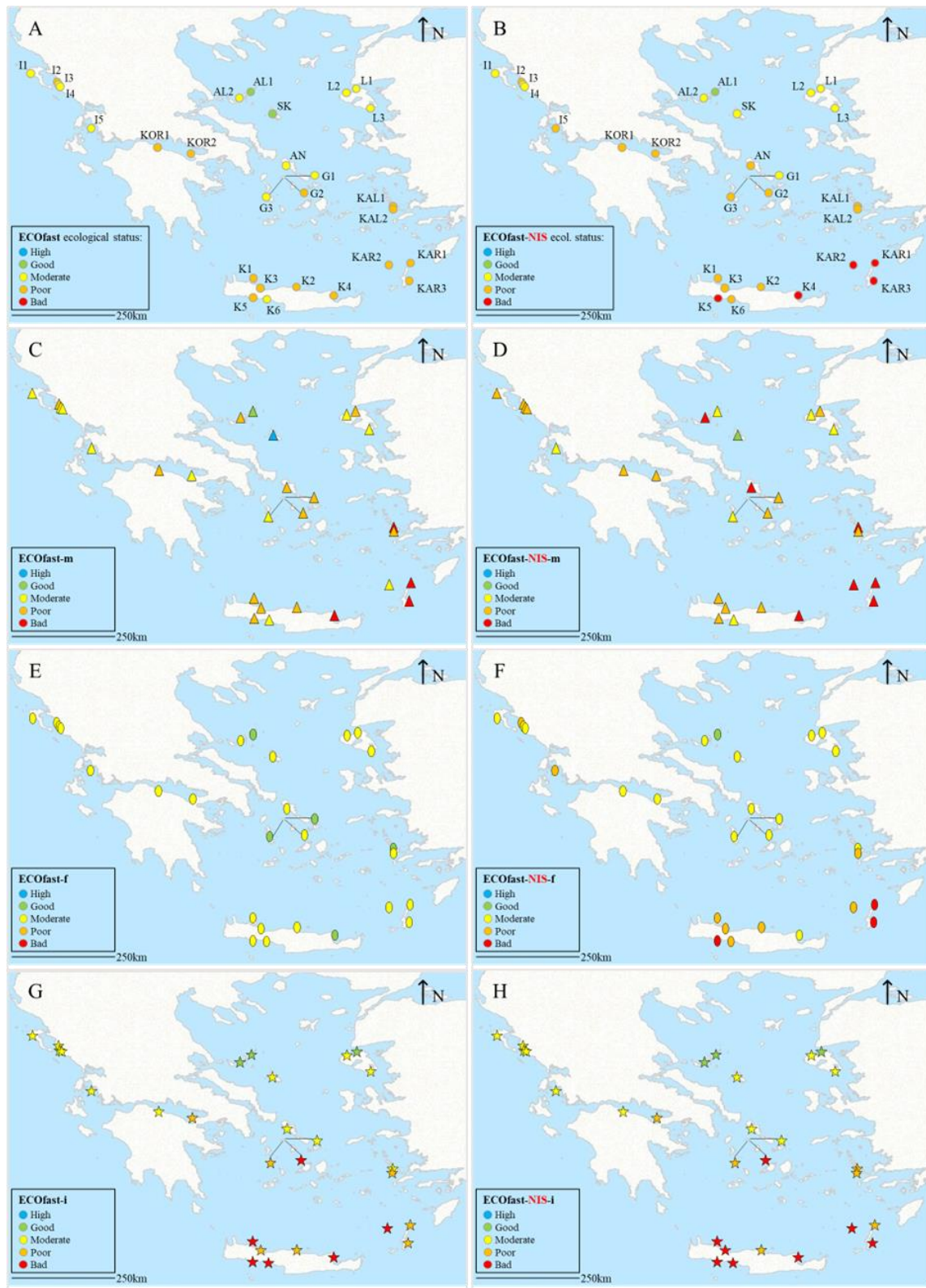


Figure 5: Ecological status of 28 rocky reef sites according to the A) ECOfast index B) ECOfast-NIS version of the index C) ECOfast-m sub-index D) ECOfast-NIS-m sub-index E) ECOfast-f sub-index F) ECOfast-NIS-f sub-index G) ECOfast-i sub-index H) ECOfast-NIS-i sub-index.

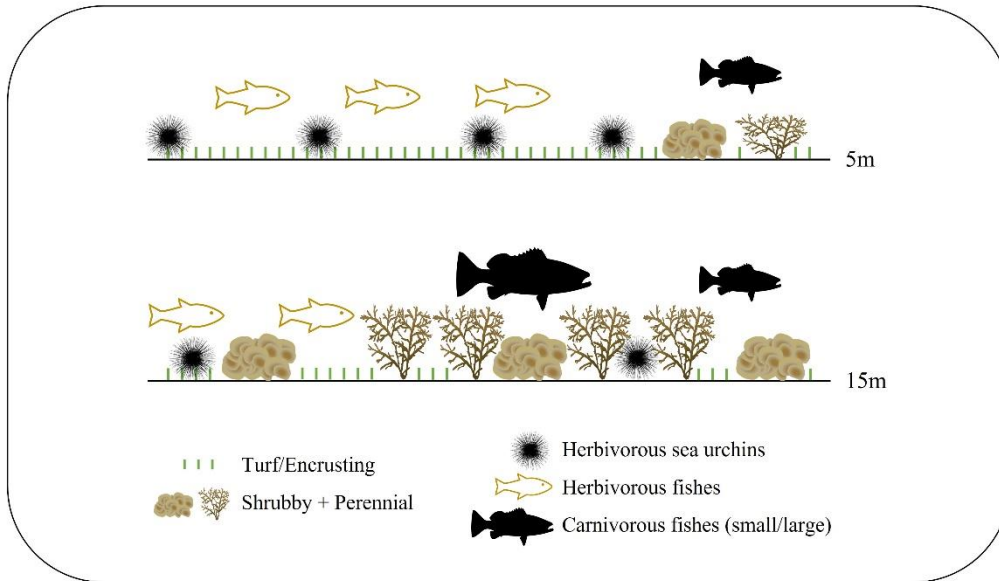


Figure 6: Simplified depiction of the main statistical differences identified by the ECOfast protocol, between the 5m and 15m depth zones. In 5m: (1) perennial and shrubby macroalgae show lower coverage, (2) herbivores (i.e., the sea urchins *P. lividus* and *A. lixula* and the fishes *S. salpa*, *S. rivulatus* and *S. luridus*) are more often present, (3) lower presence of carnivorous fishes typical of rocky reefs (i.e., *D. dentex*, *S. dumerili*, *E. costae*, *S. scrofa*, *P. miles*, *M. helena*, *L. viridis*, *E. marginatus*), (4) very low presence (up to 0%) of large carnivorous fishes typical of rocky reefs (for more details see results, Figure S2, Tables S4, S5).