Dear Prof. Emilio Marañón,

Thank you for having followed the revision of our manuscript now titled "The bottom mixed layer depth (BMLD) as an indicator of subsurface chlorophyll-a". We received valuable comments from multiple reviewers that clearified and reinforced the aims of the manuscript. Considering the numerous changes that we made on this paper (e.g. exclusion of Chl-a shapes from the first version and definition of BMLD as "bottom mixed layer depth"), we truthfully believe that this final version addresses the ambiguities raised by each of the multiple reviewers and return a knowledgeable manuscript on this topic.

Below we reported the main changes, followed by the responses to specific comments.

Main changes:

- We reinforced the definition of BMLD as "bottom mixed layer depth" following the definition from Pingree and Griffiths (1977) and Sharples et al. (2001). The title, manuscript and abbreviations' table (Table 1) were changed.
- We changed "ecological indicator" with "driver" or "indicator".
- We checked whether the definition of BMLD given in this paper is similar to the bottom boundary layer (BBL) as suggested by reviewer 1. We conclude that the two definitions are different with details in the comment's answer below.
- We improved section 1.3 in order to i) state the use of BMLD (which is different from BBL) in understanding physical and biological processes, ii) reinforce the need of a method identifying for BMLD in shelf complex waters, iii) clarify the aims of the paper (return a method to retrieve MLD and BMLD from highly variable density profiles in shelf waters) and iv) justify the comparison among DLs by comparing the vertical distribution of them and Chl-a maxima (DL=DCM). See below.
- In the discussion and conclusions, we added details of our intentions in comparing the vertical distributions of DL and DCM (e.g. BMLD=DCM) regardless any temporal component. The comparisons were made in absence of any variable controlling for the progression of events affecting the physics and biological dynamics of the water column (e.g. vertical Chl-a shape or water column stability). However, the association between any DL and DCM vary depending on the physical and biological conditions of the water column. Hence, we discussed the potential factors involved in the different associations of DCM with MLDs' and HPDs' indicators, Max N² and BMLD. In general, the MLD is likely to distribute close to DCMs during surface blooms, Max N² might represent a thin layer where phytoplankton gather (13.51% of the profiles) in a less turbulent region, HPD and BMLD showed the highest correlation to DCMs, while BMLD distributed below DCMs in 78.32% of the profiles. Moreover, we specified that the unexplained variance (scatter points along the 1:1 line) in the linear regressions in Figure 4 is most likely related to the different conditions of the water column, such as the vertical distribution of Chl-a (shapes), nutrients profiles, stability of the water column (transition from either stratified to mixed condition or vice versa), tidal phase, grazing factors, phytoplankton dynamics (e.g. cell's light history, species composition and competition). We suggested that further investigations should be carried out including the factors mentioned above.

- We changed and summarized the conclusions according to the comments.

#Reviewer 1

General comment

The Authors made a further effort to include all the suggestions and comments in the manuscript. Some technical aspects have been clarified, while keeping the essential methods and results in the revised manuscript. Discussion has been improved focusing more on the main objectives of the manuscript.

Some minor correction that can further improve the manuscript are listed hereafter:

Consider to do not insert unnecessary acronyms in the abstract, e.g. SCM and MaxN2 are not further used in the abstract itself. On the other hand, MLD (L. 20) is not explained in its first occurrence but then used at the abstract end.

We deleted all the acronymous from the abstract except for BMLD and DCM, which were both defined.

L. 20-21: BMLD and indicators of the halfway pycnocline are compared to MLD indicators but comparison terms are missing in the sentence. I suggest removing "highly predicted" and insert comparison terms (e.g., "more efficiently predicted").

We agree there was not comparison term. We changed "highly predicted" with "better predicted". L. 24: Remove "as a valuable tool".

We changed the whole sentence, and replaced "as a valuable tool" with "as a potential variable to".

L. 86: Here and elsewhere in the manuscript the term "proxy" is used. Since it has been removed from the title, maybe it can be substituted through all the manuscript.

Thank you for pointing this out. We changed "proxy" with "driver" as suggested by the editor.

L. 160: The algorithm and the maximum angle and cluster analysis methods has been introduce above, thus the present sentence can be modified accordingly (e.g., "The algorithm to identify MLD and BLMD was developed in R (available at [...]) and implements i) the maximum angle method [...] and ii) the cluster analysis [...]").

The sentence has been changed as suggested.

L. 351 and elsewhere: check for occurrence of DMC (instead of DCM). Check also the abbreviations and acronyms used in figures (e.g., "CMd" in Fig. 4 should be DCM).

Thank you very much for pointing this out. We changed all the misspelled acronymous into DCM. L. 366: Number of observations is non-dimensional. To be consistent with the measurement units adopted, the standardization should be done with a length quantity (m). Maybe number of observation multiplied by the observation interval (1 m).

We changed the sentence from "The sum of depth-integrated Chl-a mg m⁻³ of all profiles was standardized by the number of observations (mg m⁻³)" to "The sum of depth-integrated Chl-a (mg m⁻²) of all profiles was standardized by the number of sampling intervals (m)".

L. 414: Maybe "effects" is missing between "physics" and "on primary production". We changed the sentence from "the exclusive investigation of the surface physics on primary production" to "the exclusive investigation of the effects of sea surface processes on primary

production".

L. 459: The sentence subject "some of the potential contexts" is placed after the verb "are introduced".

Sentence was corrected as indicated.

#Reviewer 2

Dear Editor.

I have read the new version of manuscript egusphere-2022-140 by Zampollo A. et. al., now entitled: The mixed layer depth below the pycnocline (BMLD) as an ecological indicator of subsurface chlorophyll-a.

I acknowledge improvements from the previous version I reviewed (was first version, sorry I missed an intermediate version), most following the requests of reviewers. Again, I appreciate the author's effort to provide a systematic characterization and a statistical in-depth analysis from a large dataset and to develop methodological tools. Although I have some concerns on the scope and methodology of the ms, I feel the overall outcomes are valuable and the ms deserves publishing after dealing with some issues I consider minor at this stage.

My first general concern involves the definition of BMLD as 'the depth at which the pycnocline ends and deep mixing develops down to the seabed' (sec.1.3). In my view, this definition seems strongly linked to the top limit of what is known as the benthic boundary layer (BBL) (e.g. Lueck et. al. 2019, doi:10.1016/B978-0-12-409548-9.11622-7) but there is neither connection nor even mention to the BBL across the manuscript. The authors should explain if BMLD is the same as the top of the BBL, if not what are the differences while if yes a new definition/name for an already understood concept may be unnecessary.

We used the definition of "bottom mixed layer depth" (BMLD) to indicate the density level separating the pycnocline from the bottom mixed layer, the last being defined by Pingree and Griffiths (1977) as a layer where the temperature change is 0.01 °C, and Sharples et al. (2001) as a layer where the density change is -0.02 kg m⁻³ relative to the closest value to the bed. Furthermore, the BML was used by Palmer et al. (2008), Palmer et al. (2013), Wihsgott et al. (2019), Poulton et al. (2022). On the other hand, the bottom boundary layer (BBL) "refers to a layer flow in the immediate vicinity of the solid sea bottom where the effects of viscosity are significant in determining the characteristics of the flow" (from Zhang (2014), DOI 10.1007/978-94-007-6644-0 134-1). The identification of the BBL would, hence, require data on water velocity, density, and seabed topography to measure the water viscosity near the seabed. Zhang (2014) also writes "in a natural continental shelf", such as the North Sea, "any definition of the BBL structure is not straightforward" and that BBL is "just above the sea bottom" where "there is a homogeneous layer of temperature, salinity and density". Similarly, Trowbridge and Lentz (2018) write that BLL "in the ocean is often stably stratified by temperature and salinity [..], even within 1 m of the seafloor [..], and can also be stably stratified by suspended sediment". They defined the BBL as a boundary layer that "is characterized by turbulent eddies that transport mass, heat and momentum across the streamlines and the density surfaces that are associated with ocean

currents and stratification". Hence, the BBL is measured using a set of Reynolds-averaged equations for mass, which requires data on fluxes of momentum, heat, salt and sediments. For these reasons, we recognize differences between BMLD and BBL, although the two are surely interacting in shelf waters. Therefore, we included a section where we describe the potential interactions between BMLD and BBL in the Discussion (lines XX-XX) and we clarified the definition of BMLD in the introduction (1.3 section).

Moreover, Palmer et al. 2013 investigated the physical oceanography of Jones Bank (Celtic Sea) by measuring several physical variables, including the bottom mixed layer and the bottom boundary layer as two different conditions.

Below we summarized the differences between BMLD and BLL and the changes made in the paper:

- BMLD is defined on density profiles (Sharples et al. 2001), BLL is defined using horizontal and vertical speed components (u,v,w) (Trowbridge and Lentz, 2018)
- BMLD is the base of the pycnocline, and can distribute close to the sea surface by following the pycnocline and bottom mixed layer vertical distribution, BLL distributes close to the seabed, in stably stratified layers even within 1 m of the seafloor (Trowbridge and Lentz, 2018).
- BBL and BMLD can interact with each other in shelf waters since BBL is characterized by "turbulent eddies that transport mass, heat and momentum across the streamlines and the density surfaces that are associated with ocean currents and stratification" (Trowbridge and Lentz, 2018).
- As mentioned in the comment below about the conclusions on BMLD=DMC ("I am a bit unsure [..]", we added in Section 1.3 details and references on the use of BMLD to justify the analyses of this paper (more details below).

I also find a bit misleading the use of "8 density levels DL" to relate to DCM. Some of these only accounts for different methodologies to compute the same thing (MLD, pycnocline depth and BMLD which may be the BBL top).

To avoid misunderstanding, we stated that DLs are "The depths detailing the density structure in the water column are defined here as density levels (DLs)."

We deleted "eight" before "density levels" across the whole paper. In section 1.3, we mentioned that we investigated four different structures of the density profiles "[...] (MLD, halfway pycnocline depth, BMLD, and maximum frequency buoyancy) are analysed using [...]", and we specified in Section 2.6 that "In this study, we investigate the use of the surface mixed layer depth (MLD_{0.01}, MLD_{0.02}, MLD), the maximum squared buoyancy depth (Max N²), halfway pycnocline and bottom mixed layer depths (HPD_{0.01-BMLD}, HPD_{0.02-BMLD}, HPD_{MLD-BMLD}, and BMLD) to derive the vertical distribution of Chl-a".

I am a bit unsure about the conclusions on DCM and density levels. BMLD and mid-pycnocline shows better linkage with DCM than with MLDs so the authors conclude that MLD metrics are weak predictors of DCM (I.406). I guess the reason is the diverse pycnocline shapes and extent that may occur for any given MLD. Moreover, timescales for physics and phytoplanckton dynamics are different, so I would expect spread on chlorophyll profiles characteristics for very similar density profiles and hence I would not expect that any of the DLs proposed should tightly match as predictor of DCM. Aligned with this, I am not sure on the value of evaluating whether DCM=BMLD (or whatever DL, I.261) besides the distribution of MLD-DL also shown in Fig.5. I think that the authors should elaborate on these issues further.

Thank you for bringing this up. We recognize that this paper does not describe the relationship between DCM and DLs under different hydrodynamic conditions and phytoplankton dynamics, and that the time scales of the processes do not necessarily overlap to each other. However, the Chl-a profiles are likely to change in accordance to the density profile (e.g. Carranza et al., 2018) and the paper aimed not only to investigate DCMs, but also the overall vertical distribution of Chla (section 3.2). The association of MLD with phytoplankton has been described in the literature over and regardless the temporal succession of events defining both the physics and phytoplankton aggregations in the water column (see section 1.2). Therefore, the aim of this paper is instead to investigate at which extent the BMLD can inform on the vertical distribution of DCMs in shelf temperate waters during summer, and showing that BMLD is actually returning information on the vertical distribution of Chl-a maxima, independently from the hydrodynamic conditions, pycnocline stability and phytoplankton phenology status.

However, we agree that the temporal component was not well described in the discussion, although it was taken into consideration during the formulation of the research questions. Therefore, we edited the sections 4, 4.1, 4.2 and the conclusions to give a context on the temporal component while discussing the coincidence of DCM at any DL:

<u>Introduction 1.3</u>: Lines 137-140, "Further scrutiny was applied to BMLD to investigate to which extent the BMLD can inform on the vertical distribution of DCMs in temperate shelf waters during summer, regardless of any phytoplankton dynamic (cell's light history regulating photoacclimation) or physical conditions of the water column (e.g. stability)."

<u>Discussion</u>: Lines 474-487, We agree that the small association of MLD with DCM might be related to the many other factors, defined with the phytoplankton dynamic and succession of physical conditions in the water column. Therefore, we specified that:

"It is worth noting that the comparison between any DL and DCM was made independent of the time scales at which physical processes and phytoplankton dynamics develop, which differ from each other and do not necessarily overlap. Therefore, the association of any DL with DCM (e.g. BMLD=DCM) was investigated under different physical (e.g. water column stability) and biological conditions (e.g. cell's light history regulating photoacclimation) which are likely to be responsible for the unexplained variance reported for each linear comparison in Figure 4. As an example, the small association of DCMs with all the investigated surface mixed layers' indicators (MLD_{0.01}, MLD_{0.02} and MLD, Table 3) can relate to temporal aspects of the phytoplankton dynamic and physical data set (e.g. multiple data collection within oligotrophic surface waters in stably stratified conditions after spring blooms) at the time of sampling. Hence, the association between any DL and DCM would vary depending on the progression of events defining the profiles of ChI-a and density. Here, we discussed the location of DCMs in regard to MLD, HPD, BMLD and Max N², considering the potential physical conditions and phytoplankton dynamics at the sampling time (such as water column stability, light history exposure and turbulence) as possible drivers of the resulting associations."

<u>Conclusion</u>: Lines 635-645, "The extent to which subsurface Chl-a maxima distribute in the proximity of any density level was investigated aside from any variable controlling for the progression of

events affecting the physics and biological dynamics of the water column (e.g. vertical Chl-a shape or water column stability) at the sampling time. Hence, the extent of variability retrieved from each comparison (e.g. DCM close to BMLD) is most likely related to the different conditions under which the water columns were investigated, such as the vertical distribution of Chl-a (shapes), nutrients availability, stability of the water column (transition from either stratified to mixed condition or *vice versa*), tidal phase, grazing factors, phytoplankton dynamics (e.g. cell's light history, species composition and competition)."

Moreover, this study can set the basis to develop further questions that would investigate the vertical distribution of Chl-a (shapes), DCM and density features across time when the stratification is set (permanently stratified waters) and ebb/flood cycle increases the distance between MLD and BMLD (enlarge the pycnocline), or when internal waves occur. Hence, we agree that comparing any DL to DCM is as simple as needed in some research field where the physics and phytoplankton dynamics are leaved out, such as deriving the vertical distribution of Chl-a from satellite samplings (Lavigne et al. 2015, 10.5194/bg-12-5021-2015).

Overall, a first description of the vertical distribution of MLD and BMLD can be important to understand whether there are patterns in the vertical distribution of Chl-a under different environmental conditions, e.g. concentrations below MLD in polar regions (Ardyna et al. 2013, 10.5194/bg-10-4383-2013) or below BMLD in coastal waters (< 45 m bathyemtry) (in situ data collected in FoF), close to BMLD in shelf waters (see section 1.1, e.g. Durán-Campos et al., 2019). Since a few methods are described to retrieve BMLD (threshold - Sharples et al. 2001, Wihsgott et al. 2019), the aim of this paper is also to return a useful tool to extract BMLD from high resolution density profiles and potentially state some questions for further investigations (e.g. those mentioned in this reviewer's comment). Therefore, an integrated approach of the threshold method and the maximum angle method (Chu and Fan, 2011) is described and an example on the BMLD's use is given.

Regarding the discussion, climate change and offshore manmade structures are addressed very broadly, highlighting the importance of understanding primary production in a changing environment, but the usefulness of the developed methodology and approach is unclear to me. As mentioned in the last comment made at the end of the document, we clarified how the use of BMLD is useful to investigate potential physical changes related to climate change and man-made structures. We believe that the reviewer referred to the whole method (MLD and BMLD) by writing "developed methodology and approach". However, we want to focus on the reasons behind using BMLD in further studies, as a complementary indicator of the pycnocline position together with MLD and as a driver of subsurface primary production. Moreover, the intention is to shortly summarize the main variations caused by climate change and man-made structure in relation to BMLD/deep mixing processes. We listed below the usefulness of BMLD (and hence the supply of a function/method to retrieve it) in these two contexts:

- Identify the halfway pycnocline depth (HPD), and hence having three indicators of the pycnocline instead of only one, the (surface) mixed layer depth.
- Measures variations in BMLD caused by changes in the deep mixed layer (e.g. changes in the stratification strength due to climate change or increase of the mixing downstream of the turbine foundation)

- Investigate variations in Chl-a abundance, vertical distribution and community composition due to changes in the vertical distribution of BMLD, and its distribution in relation to other factors (e.g. euphotic depth and nutricline).
- Investigating whether grazers, fish or seabirds uses the pycnocline (variations in density throughout the water column) to detect the vertical distribution of food resources, and whether the variation of MLD or BMLD might affect their foraging success.

Specific Comments:

I.18. BMLD should not be explicitly referred in the abstract without providing a definition of the concept, as is done in line 90.

We added the following sentence "(BMLD: depth between the end of the pycnocline and the below mixed layer)". At line 90 was provided the definition: the mixed layer depth below the pycnocline.

1.35. not sure about the relevance of seasonality of tide cycles, but I miss a word on the seasonal heating-cooling cycle.

The seasonal cycle was suggested by a previous reviewer in the second round of revision, and a reference was provided for that (Müller et al. 2014, 10.1007/s10236-013-0679-0). However, the sentence wants to point out that the structure of the stratified water column is highly influenced by the strong daily and biweekly variations in the tidal current (cycles), which ultimately influence the thickness/vertical distribution of the pycnocline's limits and the nutrient-enriched fluxes into the surface waters. For this reason, the seasonal variation of e.g. the lunar component M2 of 10% is not relevant here, and it was deleted from the sentence in this new version. The sentence was also improved to specify that "The vertical distributions of the spring-summer stratification in the water column fluctuate in time and space by the modulation of daily and biweekly strong tidal cycles". Moreover, as indicated by this comment, we included the seasonal heating-cooling cycle in lines 38-41: "The seasonal heating-cooling cycle of the water column regulates the stratification in temperate shelf waters, where the intensified solar radiation in spring-summer increases the difference of temperature and salinity between surface and deep waters and develops a pycnocline dividing surface from deep mixed waters". The seasonal heating-cooling cycle originates the seasonal stratification instead of determining a small temporal scale variation, where BMLD is more relevant. On the other hand, the seasonal heating-cooling cycle is relevant to describe the system where the study is located, and therefore it is added before mentioning the daily/biweekly variation due to tidal cycle.

I.61. It is said that 'the use of MLD is motivated in oceanic sites where the deepest limit of the pycnocline is difficult to draw'. I disagree, the seasonal pycnocline transitions progressively into the permanent thermocline, but it is not difficult to draw a limit, just needed to establish a criteria. This sentence wanted to say that oceanic density profiles can report a smoother rate of change between the pycnocline and the bottom mixed layer (at BMLD). On the other hand, shelf waters are characterized by surface and deep physical processes that make the pycnocline thinner than those in oceanic sites and allow a clearer identification of MLD and BMLD at depths with significant changes in density. Since we wanted to shortly motivate the use of MLD in oceanic sites and introduce the investigation of BMLD in shelf waters, we changed the sentence into "Although the use of MLD is motivated in oceanic sites where the surface processes drive most of the variations in primary production, the biological processes of shelf waters are equally driven by the

physical processes above and below the pycnocline that define the nutrient distribution in a more restricted space. Hence, the identification of the upper and below limits of the pycnocline may improve the understanding of the processes defining the primary production in shelf waters.".

1.79. Interestingly, Kara et.al did not applied a simple threshold but developed an algorithm that involves a previous transformation of the profile (providing a much better result).

We agree with the reviewer, this reference is not entirely correct. We chose Kara et al. 2000 because they listed in table 1 several authors and criteria to measure MLD from the sea surface temperature. We deleted the reference since the threshold method is widely adopted in the scientific community and specific references are only reported to justify the threshold value (as we did in section 2.4 (lines 281-283).

I.480 it is said that 'the role of climate change in increasing stratification is likely to affect the distribution of BMLD and the upward fluxes'. I understand that the main controls of the BBL (hence BMLD) are tidal currents, which will not vary due to climate change. The authors should elaborate further

We addressed this comment together with another comment from the same reviewer "Regarding the discussion, climate change and offshore manmade structures are addressed very broadly [..]". In this section, we mentioned the potential uses of BMLD in future studies. Hence, we listed some of the contexts in which BMLD could be advantageous by referring to:

- Identify the halfway pycnocline depth (HPD), and hence having three indicators of the pycnocline instead of only one, the (surface) mixed layer depth.
- Measures variations in BMLD caused by changes in the deep mixed layer (e.g. changes in the stratification strength due to climate change or increase of the mixing downstream of the turbine foundation)
- Investigate variations in Chl-a abundance, vertical distribution and community composition due to changes in the vertical distribution of BMLD, and its distribution in relation to other factors (e.g. euphotic depth and nutricline).
- Investigating whether grazers, fish or seabirds uses the pycnocline (variations in density throughout the water column) to detect the vertical distribution of food resources, and whether the variation of MLD or BMLD might affect their foraging success.

These points have been better described in section 4.3.