Comments by Anonymous Referee #1 and our responses

This is an interesting paper in which the authors discuss the role of photoacclimation and enhanced growth as the underlying mechanism of the DCM in the Mediterranean Sea during the late spring. The study was carried out from 10 May to 11 June during the PEACETIME cruise. The study is exciting; however, I have the following comments/suggestions which will make this manuscript publishable after authors incorporate and modify the paper.

We are grateful to the reviewer for their time and helpful comments.

How can you be sure that the dominance of diatoms at the DCM was resulted from cell sinking from the upper layers due to photoacclimation rather than the new production? I suggest the authors to check any physical mechanism like the role of Rossby wave etc. Analysis of the physical processes in the region is compulsory when you discuss the DCM properties and the underlying mechanism.

We have estimated the diffusive nutrient flux from below the DCM and found that it contributed a small fraction of the nutrient supply required to sustain the deep phytoplankton biomass maximum (see Table 2). In addition, we found no evidence of enhanced phytoplankton growth at the DCM. Hence our conclusion that the DCM was likely to result mainly from cell sinking from above, in a mechanism that has been modelled by Fennel and Boss (2003). The role of photoacclimation is supported not only by the strong decrease in C:Chla ratios at the DCM but also by the fact that our measurements of PP at the DCM imply higher photosynthetic efficiencies than commonly measured at the DCM in oligotrophic regions where small cells dominate (Uitz et al. 2008).

Planetary Rossby waves have been shown to uplift the DCM (Kawamiya and Oschlies 2001) and cause enhanced surface chlorophyll a values (Cippollini et al. 2001) but they are large-scale features that propagate westward over entire ocean basins. Topographic Rossby waves have been observed in the Mediterranean Sea although they are bottom-intensified fluctuations, and therefore not good candidates to explain the occurrence of the DCM. Following the reviewer's advice (see also comment below), we have explored the potential role of other physical mechanisms in originating the DCM. For instance, cells may accumulate in the vertical region of enhanced stability associated with the pycnocline. To explore this possibility, we calculated the depth of maximum Brunt-Väisälä frequency at each long station. We found that the layer of maximum stability lies at a depth of 15-25 m, well above the DCM, which does not support a role for this mechanism during the Peacetime cruise. In the revised version of the manuscript, we will add these data to Table 1.

At stations TYRR and FAST, DCM was deeper than nitracline depth. However, DCM was located above the nitracline depth at ION. From this, I understand that the physical processes operating at ION may be different from the other two stations. Hence, insisting to look into the water column stability in all the three stations during the measurement period.

Throughout the cruise, the depth of the DCM and the depth of the nitracline covaried. For the long stations, these two depths differed, on average, by less than 10 m. Indeed the nitracline was deeper than the DCM (on average, by 9 m) at ION whereas the opposite was true in the other long stations, but the actual difference was subtle and it is not clear that it means a different mechanism for DCM formation, particularly in view of the fact that we do not have information on the seasonal dynamics but rely on snapshot observations conducted during a 1-month cruise. Key properties such as the magnitude of the DCM, and the C:Chla ratio, phytoplankton growth rate and mean PAR at the DCM were the same at all three stations (see Table 1). As mentioned above, we have also looked into the depth of the maximum Brunt-Väisälä frequency, which was the same at ION and FAST (23 m). The fact that the nitracline was deeper than the DCM at ION probably reflects longitudinal differences in the way the DCM and the mixed layer depth are coupled in the Mediterranean Sea, as discussed by Barbieux et al. (2019). These authors concluded, from the analysis of seasonal variability in the DCM using Biogeochemical Argo floats data, that in the Ionian and Levantine basins the deepest winter mixed layer rarely reaches the top of the nutricline and the DCM is persistently well above the nutricline during the stratified season. In the revised version of the manuscript, we will refer to this feature in the first section of the Discussion.

In all the three stations, DCM was just below the 1% PAR depth and below the nitracline depth except the station ION; where nitracline depth was deeper than DCM. Have you noticed any difference in phytoplankton characteristics in the DCM at ION compared to the other two? I feel you can make out the difference from the size of the phytoplankton cell. Please check it and confirm that your hypothesis is true in all the three stations. It is also not clear how the individual role of photoacclimation and biomass contribution was explored? Please mention the way to quantify it?

We have calculated, for the three long stations, the mean values of different variables that help to characterize phytoplankton size structure at the DCM: mean cell biovolume (from the imaging flow cytobot), % contribution of cells >6 µm to total phytoplankton C, and % contribution of cells > 2 µm to total primary production (from size-fractionated PP experiments). This information will be added to Table 1 in the revised manuscript. For all variables, we found no differences between stations, which suggests that, in terms of size structure, the DCM phytoplankton community was comparable among sites. The HPLC pigment data also indicated that in all three stations the contribution of diatoms at the DCM increased markedly in comparison with surface waters, as reflected in the fucoxanthin to total chlorophyll a ratio as well as the fucoxanthin to 19'hex-fuco+19'but-fuco ratio. These data will also be added to the revised version of the manuscript as a new figure in the supplementary information. The HPLC pigment data did suggest some differences among stations in the upper layers. For instance, within the upper mixed layer of ION and TYRR the phytoplankton assemblage was dominated by prymnesiophytes, followed by cyanobacteria, wheres the opposite was the case at FAST.

To estimate the contribution of photoacclimation (increased Chl per unit C biomass) and increased biomass to explain the DCM, we calculated the DCM to surface ratios for chlorophyll a and phytoplankton C concentration, as explained in section 3.2 of the manuscript. For instance, at station TYRR the deep to surface chla concentration ratio was 8.14 (0.57/0.07), while the deep to surface ratio in phytoplankton C was 2.17. This means that 26% (2.17/8.14) of the increased chl a at the DCM resulted from enhanced biomass, while the rest of the increased chl a (74%) resulted from photoacclimation. Repeating the same calculations for the other two stations indicates that, overall, photoacclimation accounted for 66-78% of the increase in chl a concentration from the surface to the DCM.

References (not cited in the ms)

Cippollini et al. (2001) Rossby waves detected in global ocean colour data. Geophys Res Lett 28:323-326

Kawamiya and Oschlies (2001) Formation of a basin-scale surface chlorophyll pattern by Rossby waves Geophys Res Lett 28:4139-4142