Respond to RC1

General information: The line numbers (given for changes in text) refer to lines in the manuscript **with** Track Changes accepted!

RC1: 'Comment on egusphere-2025-5385', Anonymous Referee #1, 20 Dec 2025

This manuscript presents a comprehensive field study on the biomolecular composition of the sea surface microlayer (SML) in the Central Baltic Sea, with a particular focus on the role of cyanobacteria and phytoplankton size structure. The dataset is rich and combines multiple complementary approaches, including microscopy, flow cytometry, detailed biomolecular analyses, and surfactant measurements. The study has clear potential to advance our understanding of how phytoplankton community structure imprints on SML chemistry and potentially affects air–sea exchange processes.

We thank the referee for this positive and constructive assessment of our manuscript. We appreciate the recognition of the comprehensive dataset, the combination of complementary methodological approaches, and the potential relevance of our results for understanding how phytoplankton community structure influences SML chemistry and air–sea exchange processes. We believe that the referee's comments have helped to substantially improve the manuscript.

However, several issues need to be addressed before the manuscript can be considered for publication. Most importantly, the classification scheme based on HPA/LPA and phytoplankton size classes is conceptually confusing and, in its current form, hampers the interpretation of results and discussion. The conflation of size range and abundance category, as well as the use of cross-definitions (e.g. HPA>20 µm corresponding to LPA<20 µm), should be clarified or simplified, ideally by adopting more explicit ecological descriptors.

Thank you for highlighting this important point. We agree that the current classification scheme was conceptually confusing, particularly due to the conflation of size classes and abundance categories and the use of cross-definitions (e.g., HPA>20 µm corresponding to LPA<20 µm). Both reviewers found the abbreviations difficult to understand, which caused confusion throughout the manuscript. We have revised the abbreviations to enable clearer classification and have improved the structure of the manuscript.

We introduce phytoplankton abundance conditions and how they have been classified (Line 297-310). We decided to use abbreviations for microphytoplankton (MP) and pico – and nanophytoplankton (P/NP). We revised and restructured Section 3.1 and explained classification of the phytoplankton abundance conditions in more detail:

New abbreviations:

- **High MP /Low MP**
- MP: Microphytoplankton (> 20µm); refers to the net samples.
- **High P/NP /Low P/NP**
- P/NP: pico- and nanophytoplankton (≤ 20µm); refers to the flow cytometry samples.

Changes in text:

Line 306-315: […] Accordingly, stations above the respective medians were classified as a condition characterized by high, while those below were classified as a condition characterized by low abundances. Subsequently, stations with high P/NP abundance were addressed as 'High P/NP' conditions, while stations with low P/NP abundance were addressed as 'Low P/NP' conditions (Fig. 2b). The same categories were implemented to classify conditions based on MP abundances, either referring to high ('High MP') or low ('Low MP') abundances (Fig. 2c). In summary, the following four conditions were introduced and will be used throughout the text: 'High P/NP' vs. 'Low P/NP' and 'High MP' vs. 'Low MP'. It is important to highlight that in 10 out of 13 common stations, the abundances of P/NP corresponded inversely to those of the MP, i.e. stations characterized by elevated abundances of P/NP

(High P/NP) generally showed reduced abundances of MP (Low MP), and vice versa. At two stations, no net samples were derived and thus no MP condition could be assigned.

Accordingly, HPA>20µm and LPA>20µm have been replaced with High MP and Low MP, and HPA<20µm and LPA<20µm have been replaced with High P/NP and Low P/NP throughout the manuscript (text and figures). We believe that these changes have substantially improved the clarity and interpretability of the results and discussion.

When presenting results that describe differences between the abundances of a size fraction, e.g., High P/NP vs. Low P/NP (e.g. Fig. 5), we focused exclusively on these comparisons and avoided reiterating inverse relationships for the corresponding smaller size fraction. This removed the need for cross-definitions and further improved clarity.

The manuscript contains numerous punctuation and formatting inconsistencies throughout the text, which should be carefully checked and corrected by the authors.

We thank the reviewer for carefully reading the manuscript and for noticing the punctuation and formatting inconsistencies! We appreciate this detailed attention and have carefully corrected these issues throughout the manuscript.

Overall, this is a potentially impactful contribution, but substantial revisions are required to improve conceptual clarity.

1. Specific scientific comment

Line100: The manuscript discusses surfactants, amino acids, and carbohydrates in the SML in relation to cyanobacteria. However, it remains unclear whether these compounds are produced directly by cyanobacteria or whether they primarily result from bacterial processing and degradation of cyanobacterial biomass and exudates. The relative roles of cyanobacteria versus heterotrophic bacteria in shaping the SML biomolecular composition require clearer clarification.

Based on the data presented, we cannot differentiate whether the detected biomolecular compounds originate directly from cyanobacterial production or from bacterial processing and degradation of cyanobacterial biomass and exudates. Our dataset does not allow us to disentangle these pathways mechanistically.

In the revised manuscript, we clarified this limitation and adjusted the respective interpretations. We expanded the Introduction and Discussion to more clearly acknowledge the potential contributions of both cyanobacteria and heterotrophic bacteria in shaping the biomolecular composition of the SML and to emphasized that our results reflect associations rather than direct source attribution. The Introduction was also restructured in response to comments from Reviewer 1 (Line 98-118)

Changes in introduction:

Line 98-105: Laboratory and field studies suggest that cyanobacteria produce EPS and glycolipids with pronounced surface activity, facilitating microlayer film formation and altering interfacial properties (De Philippis and Vincenzini, 1998), while experimental evidence highlights further the rapid turnover of phytoplankton bloom-derived organic matter by heterotrophic bacteria. During a filamentous cyanobacterial bloom in the Gulf of Finland (Baltic Sea), heterotrophic bacteria were shown to utilize approximately 25–55% of dissolved organic carbon (DOC) released from the plankton community, with more than half of the accumulated DOC degraded within seven days (Hoikkala et al., 2016). This suggests that a substantial fraction of cyanobacteria-derived compounds entering the SML is short-lived unless stabilized through transformation or incorporation into particulate or gel-like structures.

Line 118: […] cyanobacteria can represent major producers [..]

Changes in discussion:

Line 724-733: […] On the other hand, heterotrophic bacteria release surfactants to enhance substrate availability and uptake (Satpute et al., 2010). Higher surfactant enrichment has been shown to occur in alignment with advanced organic matter degradation (Wurl et al., 2011; Barthelmeß and Engel, 2022). GABA characterized the organic matter pool during High P/NP and has been associated with, first, advanced bacterial processing (Davis et al., 2009) and, second, the accumulation of surfactants at the air-sea interface (Engel et al., 2018b).

Direct attribution of individual biomolecules to cyanobacterial production versus heterotrophic processing remains limited, because most available studies rely on correlations with community composition or gene expression rather than direct compound-specific source tracing (Berg et al., 2018; Barthelmeß and Engel, 2022). Thus, the SML surfactant pool, explicitly during High P/NP conditions, should be understood as the outcome of an interplay between cyanobacterial inputs and subsequent microbial reworking, rather than as a signal attributable to one group alone. […]

Line103-105: The SML was sampled using a glass plate or Garrett Screen, whereas the ULW was sampled using nets and/or discrete water samples at ~1 m depth. Given the use of different sampling methods, it is unclear to what extent the observed molecular differences between the SML and ULW may be influenced by methodological biases, particularly with respect to particulate versus dissolved fractions. In addition, Table A1 suggests that ULW samples were collected using a Niskin bottle, which raises some ambiguity as to whether both net- and bottle-based approaches were used for ULW sampling. The authors should clarify the sampling strategy and discuss potential methodological effects on the SML–ULW comparison.

We thank the reviewer for this thoroughly comment! The underlying water (ULW) samples in this study were collected exclusively using a manually deployed water sampler (more appropriate definition than Niskin bottle) at approximately 1 m depth for surfactant measurements. No net-based sampling was applied for surfactant studies, and the text was revised to remove any ambiguity and to ensure consistency with Table A1.

Changes in text:

Line 126-127: […] Garrett Screen (Garrett, 1965). Underlying water (ULW) samples, used as reference for the SML, were collected at approximately 1 m depth using a manually deployed water sampler. […]

Line 143-144: […] Seawater was sampled from the SML using a glass plate or a Garrett Screen and from the ULW by a manually operated 2L water sampler (Hydrobios, according to Ruttner).

Line 155: […] water sampler (2 L) and […]

We recognize that there may have been some confusion regarding which parameters were obtained from the different sampling methods. We have therefore revised the manuscript accordingly and now provide a list of the parameters associated with each sampling method directly following their description.

Line 156-158: […] The following parameters were derived from seawater sampled with the glass plate or Garrett Screen (SML) and the manually deployed water sampler (ULW): P/NP abundance, total organic carbon (TOC), total combined carbohydrates (TCCHO), total amino acids (TAA), and surfactants (SA). […]

Line 180-182: […] Sample sizes were n = 13 for each depth with the following parameters derived: MP biomass and abundance, particulate organic carbon >20 µm (POC >20 µm), particulate amino acids >20 µm (PAA >20 µm), and particulate combined carbohydrates >20 µm (PCCHO >20 µm). […]

Line 762: […] with a manually deployed water sampler. […]

In table A 1, “Nikin bottle” was replaced by “Manual water sampler”.

The use of a Garrett Screen and, where conditions permitted, a glass plate, for sampling the sea surface microlayer (SML), together with deployment of a water sampler for the underlying waters, represents a well-established and widely accepted methodological framework in SML research, as documented in the SCOR Working Group 141 guide (SCOR, 2014). These methods are considered standard for SML–ULW comparisons. Whenever meteorological conditions allowed, SML sampling was conducted directly from a small boat to ensure optimal sampling conditions, otherwise the Garrett Screen was used from the bow of the mother ship. Although different sampling approaches may preferentially capture particulate versus dissolved fractions, the applied methods follow community standards, and potential methodological effects are unlikely to fully explain the observed molecular differences between the SML and ULW. We addressed and discussed potential molecular differences arising from methodological or meteorological influences in the Supplement (Fig. S8). We also adjusted Fig. S8 and to also show methodological differences in the total amino acid data.

Changes in text:

Supplementary material

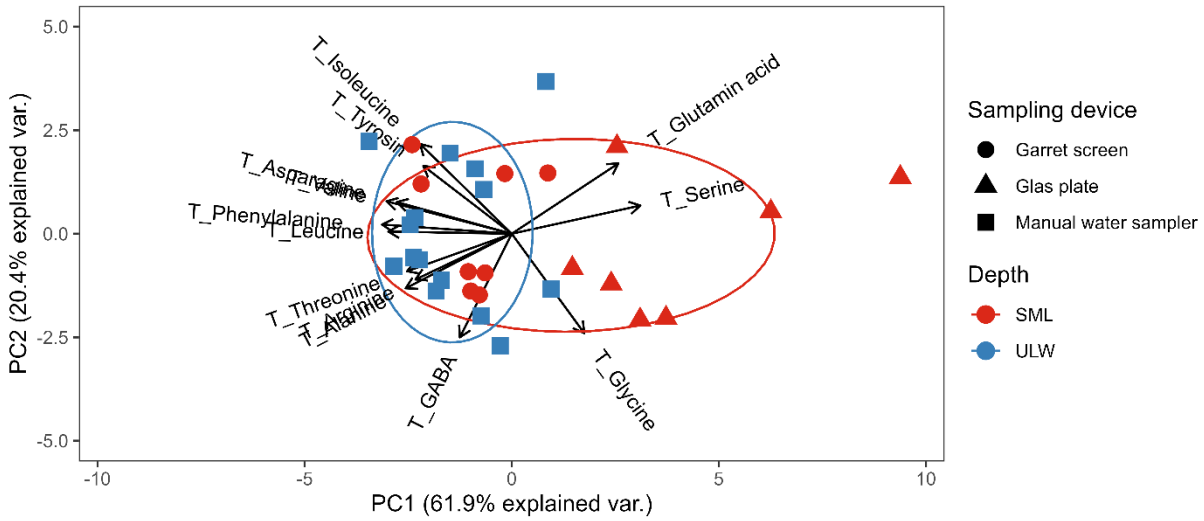

Figure S 8: PCA biplots of total amino acids (TAA). Scores are colored by SML (red) and ULW (blue) and sampling devices are indicated with different shapes: Garrett screen in circles, glass plate in triangles and manual water sampler in squares. Arrows show variable loadings; axes are scaled to unit variance and annotated with percent variance explained (PC1 61.9%, PC2 20.4%).

Line 60-92: […] Comparison of Glass Plate and Garrett-Screen SML Sampling

Seawater was sampled from the sea surface microlayer (SML) using a glass plate or a Garrett Screen and from the underlying water (ULW) by a manually operated 2L water sampler (Hydrobios, according to Ruttner). Glass plate sampling of the SML and samples from corresponding ULW was conducted from a working boat. When bad weather conditions prevented the deployment of the working boat, SML and ULW samples were instead collected from the bow of the vessel. In these cases, the SML was sampled using a Garrett screen. The amino acid composition obtained from the different sampling methods is presented in Fig. S8. While the compositions derived from Garrett screen and ULW samples largely overlap, those obtained using the glass plate show a separation from the other.

Meteorological conditions exert a strong control on the physical structure, composition, and persistence of the SML. Wind speed is a primary driver, influencing both SML thickness and organic matter enrichment. While earlier studies suggested disruption above ~5 m s^{-1}, subsequent work has shown that surfactant enrichment can persist at wind speeds of up to 10 m s^{-1} (Wurl et al., 2011). Nevertheless, increasing wind enhances mixing with underlying water, reducing SML thickness and the enrichment of particulate organic matter (Liu and Dickhut, 1998), and alters biogenic particle structure, with gel-like particles accumulating under calm conditions but breaking down or being removed at higher wind speeds (Sun et al., 2017). Meteorological conditions also affect sampling outcomes. Under high wind conditions, methods such as glass plates and Garrett screens may collect thicker layers and include more subsurface water, potentially diluting the SML signal (Cunliffe et al., 2009; Wong et al., 2021).

Precipitation further modifies SML properties by introducing freshwater signals and transient stratification, particularly under low wind conditions, while higher winds promote rapid mixing (Gassen et al., 2024).

Sampling approaches such as glass plates and Garrett screens collect layers of substantially different thickness (~30–60 µm vs. 200–500 µm) (Kuznetsova et al., 2004; Engel, and Galgani, 2016), which can lead to dilution of surface-enriched compounds, e.g. amino acids and carbohydrates, with underlying water in thicker samples (Dreshchinskii and Engel, 2017). Glass plates preferentially collect hydrophobic amino acids and surface-active materials, while screens better recover phytoplankton organisms and particulate matter (Momzikoff et al., 2004). Overall, while qualitative compositional patterns between SML and ULW are generally robust, absolute differences e.g. for dissolved fractions should be interpreted with caution due to potential method-dependent dilution effects. This difference may partly reflect the meteorological conditions under which the respective sampling methods were applied, as Garrett screen sampling was primarily conducted during bad weather conditions (lots of wind and waves, sometimes rain), while glass plate sampling was only possible to perform under calmer conditions from the working boat.

Overall, the SML is a highly dynamic system, and both its properties and their measurement are strongly influenced by short-term meteorological forcing. As glass plate and Garrett screen sampling were not conducted in parallel in our study, it is not possible to disentangle methodological effects from environmental variability in the present dataset. However, all methods applied here follow established community standards. Therefore, methodological differences alone are unlikely to fully account for the observed molecular contrasts between SML and ULW samples.

In Section 2.2, net samples containing the >20 µm particulate fraction were diluted with filtered seawater at ratios ranging from 2:1 to 1:2 prior to subsampling. It is unclear whether this dilution procedure may have altered the structure of colloids or extracellular polymeric substances (EPS), potentially affecting aggregation state and surface activity measurements. The authors should briefly discuss whether dilution could influence EPS integrity and surfactant analyses.

- General comment on Section 2.2: This section has been restructured to first describe SML and ULW sampling, followed by net sampling. This order now aligns with the Results section, which first presents biomolecules from the SML and ULW and subsequently the particulate fraction derived from net samples. Also, the station numbering has been revised. The previous station numbers referred to ship-based event logs. However, since no event logs were recorded in DSHIP for the working boat sampling, a new station scheme was introduced for this manuscript. These stations encompass both SML sampling and the collection of the particulate fraction using nets.

We are grateful for this detailed comment. Surfactants were only taken from the SML and ULW samples, which were not diluted with filtered seawater. Only the net samples were diluted with filtered seawater due to very high biomass. We clarified in the method section, which collection method has been used to measure which parameter.

Changes in text:

Line 156-158: […] The following parameters were derived from seawater sampled with the glass plate or Garrett Screen (SML) and the manually deployed water sampler (ULW): P/NP abundance, total organic carbon (TOC), total combined carbohydrates (TCCHO), total amino acids (TAA), and surfactants (SA). […]

Line 180-182: […] Sample sizes were n = 13 for each depth with the following parameters derived: MP biomass and abundance, particulate organic carbon >20 µm (POC >20 µm), particulate amino acids >20 µm (PAA >20 µm), and particulate combined carbohydrates >20 µm (PCCHO >20 µm). […]

Furthermore, we do not present EPS data in this manuscript.

In the microphytoplankton microscopy analysis, community composition and biomass estimates were restricted to the eight most dominant species. While these taxa likely dominate in abundance, it remains unclear whether less abundant species with potentially large cell volumes may still contribute disproportionately to biomass, EPS production, or surface-active compounds.

Thanks for this comment! Since we sampled during the main summer cyanobacterial bloom- time, we expected to find the phytoplankton community would be dominated by cyanobacteria in abundance and biomass. This expectation was based on several studies from previous years (HELCOM, 2025; Ploug, 2008; Zettler et al., 2024), which we also referenced. During the sampling period, cyanobacteria accounted for the majority (about 96%) of total microphytoplankton in terms of both abundance and volume, i.e. biomass. Nevertheless, we agree that rarer species may still play a role in shaping SML biogeochemistry. This potential limitation was explicitly acknowledged, and a corresponding clarification was added to the manuscript.

Changes in text:

Line 74-81: […] In most studies focusing on phytoplankton bloom dynamics, fresh organic matter production is attributed to phytoplankton communities that dominate in terms of biomass and abundance (Mannino & Harvey, 2002; Ploug 2008; Berg et al., 2018). Bibi et al. (2025) explicitly focused on the SML and attributed the release of fresh organic matter and surfactants to blooming phytoplankton taxa, dominating in abundance. Despite the recognized influence of phytoplankton production on organic matter composition in the SML (Wurl et al., 2011; Galgani and Engel, 2013), little is known about how specific phytoplankton species alter the molecular and biogeochemical composition of the SML under natural conditions. Accordingly, we address in our study how dominating, bloom-forming taxa influence the composition of the SML, while acknowledging that less abundant species potentially further contribute.

In Section 2.7, the manuscript states that triplicate 18 mL SML and ULW samples were prepared, but only one of the three replicate samples was analyzed. It is unclear why only a single replicate was measured and what the purpose of the remaining replicates was.

We thank the reviewer for spotting this inconsistency. We took triplicates for surfactant samples, of which two replicates were stored at 4 °C and one replicate at -20°C. Our intention was to measure the fresh surfactant samples directly after the cruise. However, due to logistical challenges this could not be realized. We decided to only include the surfactant replicate which was stored at -20°C and in alignment with the storage protocol of the amino acid and carbohydrate samples. We clarified this in the method section: We did not mention that replicates were taken and stored at difference temperatures. We will revise accordingly and write: e.g. One sample of 18 mL SML and ULW was taken and stored at -20°C.

Changes in text:

Line 258: […], an 18 mL aliquot of SML and ULW was […]

In Section 3.1, the classification of phytoplankton abundance is confusing. The low-abundance category of phytoplankton >20 µm is labeled as “LPA<20 µm”, which is identical to the low-abundance category of phytoplankton <20 µm. This notation conflates size class and abundance level and may mislead readers to interpret the low-abundance >20 µm group as belonging to a different size fraction. Low-abundance categories should retain the same size designation (i.e. >20 µm) and differ only in abundance. In addition, Aphanizomenon sp. is listed twice in the species list. Finally, the notation “300 x 10^3 cells mL^{-1}” does not follow standard scientific formatting and should be written as “300 × 10^3”.

We thank the reviewer for this detailed comment. We agree that the classification was confusing and that errors such writing <20µm when >20µm was intended further contributed to this confusion. We therefore revised the terminology of the categorization (see above, revision of Section 3.1) and consider that these changes improve readability and reduce ambiguity.

Aphanizomenon sp. is now listed only once in the revised manuscript (Line 305).

Thank you for pointing out the formatting issue. The manuscript was revised accordingly, and the notation was corrected to follow the appropriate scientific format.

Changes in text: e.g. Line 296, 298, 304,. and in any further lines where this had to be corrected.

In the second half of this section, the logic of the HPA/LPA classification becomes increasingly difficult to follow and begins to affect the interpretation of the results. Statements such as “under HPA>20 µm (which corresponds mostly with LPA<20 µm conditions)” and “LPA>20 µm conditions (which corresponds to HPA<20 µm conditions)” introduce cross-definitions that obscure which ecological states are actually being compared.

We thank the reviewer for this comment and agree that cross-definition was confusing and could lead to misinterpretation. We revised this aspect in the manuscript. When comparing specific size fractions (e.g., High MP and Low MP), we focused strictly on the comparison between these two groups without implying direct equivalence. We believe that a clearer explanation of the relationship we observe between the pico-, nano-, and microphytoplankton fractions improves clarity. We therefore described in more detail that, as shown in Table A2, an inverse pattern was observed at 10 out of 15 stations: high abundance of microphytoplankton is associated with low abundance of pico- and nanophytoplankton, and vice versa. However, this pattern was not observed at three stations (03, 08, and 12), and at two stations no microphytoplankton was sampled, preventing classification into a condition (High MP or Low MP). We explicitly stated this in the manuscript but avoided statements such as “under High MP (which corresponds mostly with Low P/NP conditions)”. Instead, we referred only to the differences between the two groups as shown in the respective figures.

Changes in text:

Line 312-314: […] It is important to highlight that in 10 out of 13 common stations, the abundances of P/NP corresponded inversely to those of the MP, i.e. stations characterized by elevated abundances of P/NP (High P/NP) generally showed reduced abundances of MP (Low MP), and vice versa. […]

In Section 3.2, there is an inconsistency between the statistical significance and the descriptive language used to interpret the results. For TAA, terms such as “tendency,” “slightly higher,” and “more pronounced” are repeatedly used, while the corresponding statistical tests are either not significant (e.g. $p = 0.076$) or not explicitly reported.

We agree with the reviewer that these terms were not suitable given the lack of statistical significance and that they are often overused. We made appropriate changes in the manuscript and revised and restructured Section 3.2. As part of this, we have also revised Fig. 5 and present parameters derived from SML and ULW only under P/NP conditions (Line 374-375).

Changes in text to prevent terms such as “tendency”, etc:

Line 389-390: […] however, this difference was not statistically significant (Fig. 5b). Similarly, under High P/NP conditions, TAA concentrations did not differ significantly, […]

Line 394: […] Surfactant concentrations were comparable between SML (0.52 ± 0.06 mg L^{-1} TX-100 equiv.) and ULW […]

- Within the Results section, the text and reported values have been revised and carefully checked to ensure consistency with the Materials and Methods. Further, Fig. 8 has been revised to correctly assign the unassigned data point (na) to High P/NP conditions.

In Section 4.2, the manuscript repeatedly emphasizes that enrichment factors (EFs) are close to 1, while at the same time using relatively strong wording such as “significantly higher,” “substantially higher,” or “highest average EF” to describe differences. For example, the mean TAA EF of 1.2 ± 0.4 largely overlaps with the previously reported range of 0.8–1.2, raising the question of whether this truly represents enhanced enrichment in a statistical or process-based sense. It may be more appropriate to

describe these values as being at the upper end of the historical range or as consistent with, but slightly elevated relative to, previous studies, rather than implying a clearly enhanced enrichment.

We state that enrichment of TAA and TCCHO are in line with previous studies and that TAA concentrations were higher compared to the western Baltic Sea.

Line 596-597: […] The enrichments of TAA and TCCHO in the SML of the Central Baltic Sea found in this study are thus in line with previous reports […]

Line 598-600: […] At the time series station Boknis Eck, only one SML sample showed TAA concentration >3.0 µM during two consecutive years (Dreshchinskii and Engel, 2017). Hence, TAA concentrations observed during this study (e.g. with a mean of 3.72 ± 1.35 µM in the SML during High MP condition) were thus substantially higher than concentrations previously recorded in the Western Baltic Sea.

4.3 Section, In the discussion of POC composition, the authors state that cellular biomass accounts for only ~13.5% of POC>20 µm, with the remainder attributed to heterotrophic plankton, detritus, and extracellular material. This interpretation is conceptually sound and well supported by the literature. However, the subsequent statement that "POC>20 µm was related to cyanobacterial cellular biomass >20 µm (HPA>20 µm: 1.6 ± 1.7; LPA>20 µm: 0.7 ± 0.7%)" is unclear. It is not evident whether these percentages refer to correlation strength, explained variance, or relative contribution to POC. The authors should clarify the meaning of these values and how they were derived. In addition, "Synecococcus sp." appears to be a typographical error and should be corrected to "Synechococcus sp.".

We thank the reviewer for this this detailed comment. We corrected this part in the manuscript, as the percentage values refer to POC contributing to TOC and were incorrect in the manuscript. We have revised the manuscript accordingly.

Changes in text:

Line 645-646: […] this study, POC >20 µm concentration contributed in average 1.2 ±1.4 % to TOC and varied with conditions (High MP: 1.6 ±1.7 % of TOC; Low MP: 0.7 ±0.7 % of TOC). Our results confirm the […]

We corrected the typographical error and change "*Synecococcus* sp." to "*Synechococcus* sp." In line 671.

- General information for the Discussion: The Discussion has been revised. Parts of the Discussion have been moved to the Introduction and vice versa, and additional statements have been included to improve overall consistency throughout the manuscript. Since TAA concentrations and enrichment factors in this study were compared to previous studies, the same approach was consistently applied to TCCHO.

1. Other comments and technical corrections

Line19: Please define the abbreviation "HPA" at its first occurrence.

Thank you for this. Since we have decided to change the abbreviations, we revised the manuscript accordingly.

Changes in text:

Line 19-20: […] high abundance of pico- and nanophytoplankton (P/NP; *Synechococcus*-dominated), […]

Line36: Please add a space after the citation "(Engel et al., 2017)".

A space was added after the citation “(Engel et al., 2017)”. – Line 38

Line53: The phrase “A large-scale oceanic in the Atlantic demonstrated” appears incomplete. A noun such as “study”, “survey”, or “investigation” seems to be missing.

We thank the reviewer for this comment. The sentence will be revised by adding the missing noun “study” to complete the phrase. – Line 53

Line 121: In “>20µm”, a space is missing before “µm”. Please check and correct similar formatting issues throughout the manuscript.

Thank you for pointing out the formatting issue. A space will be added before “µm” in “>20 µm” (Line 124), and all similar issues were corrected throughout the manuscript.

Line 123: “Garret” should be corrected to “Garrett”.

Thank you for catching this typo. “Garret” will be corrected to “Garrett”. – Line 144

Line 152: The units “knt” and “knots” are used interchangeably. Please standardize the unit notation.

The unit notation was standardized and used consistently throughout the manuscript. We chose to use “knots”. – Line 167

Line 153: In “~14min”, please add a space before “min”.

Thank you for noting this. A space was added before “min”. - Line 168

Line 146: “within ≤2 h” is recommended to be revised to “within 2 h”.

The phrasing was revised to “within 2 h”. – Line 151

Line 153: “Bottle” should be “bottle”.

We decided to use the term “manually deployed water sampler” instead of “manually Niksin bottle.

Changes in text:

Line 160: […] with the manually deployed water sampler.

Line 185: Please provide a reference for the chlorophyll *a* measurement method.

Thank you for this comment. The reference for the chlorophyll a measurement method was added. - Line 217

Line 215: “sample” should be corrected to “samples”.

“sample” was corrected to “samples”. – Line 241

Line 223-224: Thirteen amino acids are stated, but only twelve are listed. In addition, please revise the punctuation in the references according to journal style.

We thank the reviewer for this careful observation. The list of amino acids was corrected to include all thirteen compounds (GABA was missing in the list), and the reference punctuation was revised according to journal style.

Changes in text:

Line 251-252: […] phenylalanine, leucine, and and γ-aminobutyric acid (GABA).

Line 225: In "(Dittmar et al., 2009.; Engel & Händel, 2011)", there is an extra punctuation mark after "2009".

Thank you for pointing out the extra punctuation. This was corrected. – Line 254

Line 231: "Sample were measured" should be corrected to "Samples were measured".

Thank you for noting this grammatical error. The sentence will be corrected to "Samples were measured". – Line 259

Line 236: "the samples ionic strength was standardized" should be revised to "the sample's ionic strength was standardized".

The sentence will be revised to "the sample's ionic strength was standardized". – Line 264

Line 327, In the sentence "Cylindrotheca closterium and Chaetoceros sp. occurred at HPA>20 µm only with < 0.2 µg C L^{-1})", there is an extra closing parenthesis.

Thank you for identifying this issue. The extra closing parenthesis was removed. – Line 364

Line 407: The term "later amino acids" is incorrect and should be replaced with "latter amino acids".

Thank you for pointing this out. "later amino acids" was replaced with "latter amino acids". – Line 453

Line 419: "glucosamin" is missing the final "e" and should be corrected to "glucosamine".

Thank you for noting this typo. "glucosamin" was corrected to "glucosamine". – Line 466

Line 435: The phrase "excluded … due to their low and abundance" is incomplete and should read "low abundance".

Thank you for highlighting this. The phrase was revised to "low abundance". – Line 484

Line 698: "Moring" should be corrected to "Morning".

Thank you for catching this typo. "Moring" was corrected to "Morning" – Line 762

Respond to RC2

RC2: 'ReviewerComment-2025-5385', Anonymous Referee #2, 22 Jan 2026

General information: The line numbers (given for changes in text) refer to lines in the manuscript **with** Track Changes accepted!

1) General assessment

The manuscript presents a detailed investigation of biochemical characteristics of the sea surface microlayer in the central Baltic Sea, with a particular focus on potential links to cyanobacterial blooms. The study addresses an important and understudied interface in marine biogeochemistry, and the authors do a good job of placing their results within the context of existing literature. However, aspects of the manuscript would benefit from improved clarity and clearer framing. Additionally, some conclusions appear to be stated more strongly than is supported by the presented data, and certain interpretations would benefit from additional clarification. In several sections, especially in the Discussion, more explicit references to figures, supplemental material, and the specific datasets being discussed would substantially improve readability and help guide the reader through the results. These issues appear readily addressable through focused revisions of the text and its organization.

We are particularly grateful for the detailed feedback regarding clarity, framing, and interpretation. The reviewer's comments have helped us to critically reassess several sections of the manuscript. In response, we carefully revised the text to improve clarity and structure, especially in the Discussion and we have modified several statements where conclusions had previously been phrased too strongly relative to the supporting data.

Overall, we believe that the reviewer's constructive comments have significantly improved the clarity and readability of the manuscript, and we are grateful for the opportunity to revise it accordingly.

- General comment on Section 2.2: This section has been restructured to first describe SML and ULW sampling, followed by net sampling. This order now aligns with the Results section, which first presents biomolecules from the SML and ULW and subsequently the particulate fraction derived from net samples. Also, the station numbering has been revised. The previous station numbers referred to ship-based event logs.
- General comment on the Results Section: The text has been restructured and reported values have been revised and carefully checked to ensure consistency with the Materials and Methods. Further, Fig. 8 has been revised to correctly assign the unassigned data point (na) to High P/NP conditions.
- General information for the Discussion: Parts of the Discussion have been moved to the Introduction and vice versa, and additional statements have been included to improve overall consistency throughout the manuscript. Since TAA concentrations and enrichment factors in this study were compared to previous studies, the same approach was consistently applied to TCCHO.
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2) Comments on the Abstract

The abstract generally reads well. However, unlike other acronyms, POC is not defined and should be introduced explicitly.

We defined acronyms at it first appearance.

Changes in text:

Line 25: […] mirroring particulate organic carbon >20 µm (POC >20 µm) […]

Regarding the sentence in the abstract: "While SML enrichment patterns and carbohydrate concentrations were comparable to those previously reported for the Western Baltic Sea, concentrations of total amino acids (TAA) and surfactants were substantially higher, suggesting enhanced production by cyanobacteria." The sentence reads well stylistically, but the concluding phrase "suggesting enhanced production by cyanobacteria" appears too early in the abstract. At this point, the abstract has

not yet established the mechanistic/observational links between surfactants and cyanobacterial presence. The following sentences provide information that would support this inference, but as currently written this conclusion seems insufficiently justified at this stage. Consider either softening this statement or restructuring the abstract so that the supporting context precedes this statement.

We thank the reviewer for this comment and agree that this statement appears too early in the abstract. We revised this sentence to avoid a direct causal relationship between the observed elevated concentrations and cyanobacterial production

Changes in text:

Line 18: higher in this study and under cyanobacteria-dominated phytoplankton conditions. […]

3) Comments on the Body of the Manuscript (substantive)

General comment: When trends or results from this study are discussed in the Discussion section, it would be very helpful to consistently reference the corresponding figures or supplemental material. The discussion frequently refers to data generated here, but it is not always clear where the reader can quickly locate those results. Aside from this, the manuscript does an excellent job of engaging with and referencing related literature.

We systematically referred to figures or supplementary material to ensure that statements discussing the results are easier to follow and that the reader can directly access the relevant figures and clearly identify the data being referenced.

Lines 84–86: “Biochemical studies in the Baltic Sea have shown that the SML is enriched in amino compounds and carbohydrates relative to the underlying water (Van Pinxteren et al., 2012). In mesocosm experiments, proteinaceous gels (CSP) abundance was five times higher than that of carbohydrate-rich gels in the SML (Galgani and Engel, 2013).”

- The first sentence compares enrichment between the SML and the underlying water, whereas the second sentence compares proteinaceous versus carbohydrate-rich material within the SML. Since the first sentence already establishes that both compound classes are enriched in the SML, it would be helpful if the second sentence either (i) explicitly stated how CSP abundance compared between the SML and underlying water (if available), or (ii) provided clearer justification for why the relative abundance of proteins versus carbohydrates is important in this context. For example, how does this comparison advance understanding of phytoplankton bloom dynamics or surfactant production?

Thanks for this comment. We explained the relevance of the higher abundance of proteinaceous gels compared to carbohydrate-rich gels in the SML in this context. A statement from the Discussion has also been moved to this section, as it is more appropriate in this context.

Changes in text:

Line 58-64: […] A recent meta-analysis revealed that pronounced enrichment of organic matter in the SML is rather uncommon, while nitrogen-containing biomolecules, including amino acids, preferentially accumulate in the SML (Silva et al., 2025). During a diatom culture experiment it has been shown that both, carbohydrate-rich gels (TEP) and proteinaceous gels (CSP) were enriched in the SML, with the latter showing five times higher abundances in the SML (Galgani and Engel, 2013). This distinct enrichment of proteinaceous material, which is typically more amphiphilic and surface-active (Cosović & Vojvodić, 1998; Laß et al., 2013; Laß and Friedrichs, 2011, Barthelmeß and Engel, 2022), could contribute disproportionately to physiochemical properties of the SML. […]

- More broadly, the paragraph spanning lines 84–98 contains interesting and relevant material, but the narrative does not clearly guide the reader toward a central takeaway. The key point appears to be articulated later in line 100 (“cyanobacteria represent major producers of

extracellular organic matter"). If this is indeed the main message, consider introducing this idea earlier in the paragraph to better frame the supporting evidence.

Thanks for pointing this out. We agree that the guidance provided through this paragraph is lacking. Lines 100-101 were intended to summarize lines 84-98, however, we agree that this came too late. We revised the paragraph (Line 95-119 in the revised version) to better guide the reader and clarify the relevance of the information presented, thereby leading more effectively into our central message.

Changes in text:

Line 95-98: Their filamentous morphology is sustained by specific filament-forming proteins (Springstein et al., 2020), and protein-rich EPS, including CSP, have been observed in *Nodularia spumigena* cultures (Endres et al., 2013; Zhi et al., 2023), suggesting a potentially significant contribution to the extracellular amino acid fraction, which constitutes to the broader extracellular organic matter pool. […]

Lines 97 – 98: "Seasonal records further indicate that surfactant concentrations peak during summer months following the spring bloom and reflecting intensified solar radiation (Laß et al., 2013)."

- A slightly weak statement to have as the final sentence of a paragraph. Also, is not entirely clear what the implication of intensified solar radiation is here. For example, is this meant to link to climate warming, enhanced biological activity, or altered near-surface physical processes as mentioned in the previous sentence ("The release of such compounds can result in visible surface slicks, which locally warm the upper sub-millimeter ocean skin and modify nearsurface physical processes")? Clarifying this connection and providing a stronger closing sentence would improve the paragraph's coherence.

This section has been restructured, including the addition of a discussion on heterotrophic bacteria as suggested by Reviewer 1. The paragraph now ends with a stronger statement followed by a concise summary.

Changes in text:

Line 98- 118: […] Laboratory and field studies suggest that cyanobacteria produce EPS and glycolipids with pronounced surface activity, facilitating microlayer film formation and altering interfacial properties (De Philippis and Vincenzini, 1998), while experimental evidence highlights further the rapid turnover of phytoplankton bloom-derived organic matter by heterotrophic bacteria. During a filamentous cyanobacterial bloom in the Gulf of Finland (Baltic Sea), heterotrophic bacteria were shown to utilize approximately 25–55% of dissolved organic carbon (DOC) released from the plankton community, with more than half of the accumulated DOC degraded within seven days (Hoikkala et al., 2016). This suggests that a substantial fraction of cyanobacteria-derived compounds entering the SML is short-lived unless stabilized through transformation or incorporation into particulate or gel-like structures.

On the one hand, the release of such compounds can result in visible surface slicks, which locally modify near-surface physical processes (Wurl et al., 2018, Mustaffa et al., 2020). Under low-wind conditions in the Baltic Sea, the SML rapidly reorganizes into slick-like, polymer- and particle-rich states, consistent with enhanced surface activity during bloom periods (Stolle et al., 2010). During such periods, particulate organic carbon can contribute up to 55% of the surfactant pool (Gašparović and Ćosović, 2003). Seasonal records further indicate that surfactant concentrations in the Western Baltic Sea peak during summer months following the spring bloom (Laß et al., 2013).On the other hand, biochemical studies in the Baltic Sea have shown that the SML is enriched in carbohydrates and amino compounds relative to the underlying water (Van Pinxteren et al., 2012). Time-series data from the coastal Boknis Eck station in the Western Baltic Sea further demonstrated recurring enrichment of carbohydrates, amino acids, TEP and CSP in the SML during spring (Dreshchinskii and Engel, 2017). Phytoplankton-derived biopolymers, including amino acid- and carbohydrate-like components, are thus potential key contributors to surface activity in this region, and are characterized by short turnover times and diel variability typical of fresh phytoplankton exudates (Van Pinxteren et al., 2012; Barthelmeß and Engel, 2022).

Taken together, cyanobacteria can represent major producers […]

Lines 266–268: “Accordingly, phytoplankton >20µm abundance was grouped into high (HPA>20µm) and low (LPA<20µm) categories (Fig. 2b), and phytoplankton <20µm abundance into high (HPA<20µm) and low (LPA<20µm) categories (Fig. 2c). A summary of this classification is provided in Table A2.”

- There may be an inconsistency here: should the first low category be LPA>20µm rather than LPA<20µm?

Thanks for pointing that out. Yes, it should say >20 µm (in the revised version Low MP).

Both reviewers found the abbreviations difficult to understand, which caused confusion throughout the manuscript. We have revised the abbreviations to enable clearer classification and improve the structure of the manuscript.

We introduce phytoplankton abundance conditions and how they have been classified (Line 297-310). We decided to use abbreviations for microphytoplankton (MP) and pico – and nanophytoplankton (P/NP). We revised Section 3.1 and explained classification of the phytoplankton abundance conditions in more detail:

New abbreviations:

- **High MP /Low MP**
- MP: Microphytoplankton (> 20µm); refers to the net samples.
- **High P/NP /Low P/NP**

P/NP: pico- and nanophytoplankton (≤ 20µm); refers to the flow cytometry samples.

Accordingly, HPA>20µm and LPA>20µm have been replaced with High MP and Low MP, and HPA<20µm and LPA<20µm have been replaced with High P/NP and Low P/NP throughout the manuscript (text and figures). We believe that these changes have substantially improved the clarity and interpretability of the results and discussion.

- More generally, I found the rationale for distinguishing between HPA and LPA categories difficult to follow. If stations with ULW concentrations above the median are classified as “high” and those below as “low,” it seems that each station would belong to only one category (either HPA or LPA). The purpose of this classification, and how it captures variability across stations, would benefit from clearer explanation.

We agree with the reviewer that this classification is rather confusing and that a clearer explanation will improve the manuscript. We therefore described in more detail that, as shown in Table A2, an inverse pattern was observed at 10 out of 15 stations: high abundance of microphytoplankton is associated with low abundance of pico- and nanophytoplankton, and vice versa. However, this pattern was not observed at three stations (03, 08, and 12), and at two stations no microphytoplankton were sampled, preventing classification into a condition (High MP or Low MP). We explicitly stated this in the manuscript but avoided statements such as “under High MP (which corresponds mostly with Low P/NP conditions)”. Instead, we referred only to the differences between the two groups as shown in the respective figures.

Changes in text:

Line 312-314: […] It is important to highlight that in 10 out of 13 common stations, the abundances of P/NP corresponded inversely to those of the MP, i.e. stations characterized by elevated abundances of P/NP (High P/NP) generally showed reduced abundances of MP (Low MP), and vice versa. […]

When presenting results that describe differences between the abundances of a size fraction, e.g., High P/NP vs. Low P/NP (e.g. Fig. 5), we focused exclusively on these comparisons and avoided reiterating inverse relationships for the corresponding smaller size fraction. This removed the need for cross-definitions and further improved clarity. As part of this, we have also revised Fig. 5 and present parameters derived from SML and ULW only under P/NP conditions (Line 374-375).

Line 345: “Despite the increase, EFs were close to 1 across stations (1.1 ± 0.1).”

- This sentence refers to enrichment in TOC levels. It would be helpful to try and explain why TOC concentrations differ significantly while enrichment factors do not.

We explained why the EF of TOC were close to 1 despite differences in concentrations between depth. This resulted from overall elevated TOC concentrations, which reduced the relative enrichment, while absolute differences between the SML and ULW still remain statistically significant.

Changes in text:

Line 384-387: […] Despite the significant difference, TOC EFs were close to 1 across stations (1.1 ± 0.1), which resulted from the overall elevated and unusually high TOC concentrations encountered in the Baltic Sea. Consequently, overall high TOC concentrations reduced the relative SML enrichment, while absolute differences between the SML and ULW remain statistically significant. […]

Lines 354–357: "During HPA<20µm conditions, surfactant concentrations (Fig. 5c) were elevated in the SML (0.56 ± 0.09 mg L^{-1} TX-100 equiv.) relative to the ULW (0.52 ± 0.14 mg L^{-1} TX-100 equiv.), corresponding to an EF of 1.1 ± 0.2 (median = 1.1). In contrast, surfactant EFs were lower under LPA<20µm conditions (1.1 ± 0.2; median = 1.0)."

- This comparison is somewhat confusing, as the mean EF values are identical and the median EFs are very similar. The concentrations are also not statistically different between the two groups, making the use of terms like "elevated" and "lower" a bit of an overstatement.

We formulated these statements more cautiously and point out that, although the surfactant concentration tends to be higher in the SML than in the ULW, this difference was not significant and EF values during High P/NP and Low P/NP were similar.

Changes in text:

Line 394-398: […] Surfactant concentrations were comparable between SML (0.52 ± 0.06 mg L^{-1} TX-100 equiv.) and ULW (0.50 ± 0.05 mg L^{-1} TX-100 equiv.) under Low P/NP conditions, with an EF of 1.1 ± 0.2 (median = 1.0), as shown in Fig. 5c. Under High P/NP conditions, surfactant concentrations were elevated in the SML (0.56 ± 0.08 mg L^{-1} TX-100 equiv.) compared to the ULW (0.52 ± 0.12 mg L^{-1} TX-100 equiv.), however, the difference was not significant. This corresponded to an EF of 1.1 ± 0.2 (median = 1.1), which is similar to the EFs observed during Low P/NP conditions. […]

Line 505: "Our data showed that Chl a concentration increased over time"

- This statement seems potentially inaccurate. As I understand it, concentrations were measured at different stations, with each station sampled only once during the cruise (with two stations sampled daily). If I am understanding the sample collection methodology correctly, then these concentrations of ~1.63 µg L^{-1} and ~2.47 µg L^{-1} would represent different sampling locations, correct? Because the current statement reads as though Chl-a levels at a set location/ station were increasing over time during the duration of the cruise.
- If I am misinterpreting the sample collection methodology, please add some clarifying information on how many times the stations were visited and at what time.

this seems inaccurate since you were measuring concentrations in different stations, correct? Each station was only measured once, with two stations measured every day? If not, then where is the temporal data shown, since this statement does not contain that information. And if it is temporal data then during what time period did the concentrations measure 1.63 ug/L and when did they measure ~2.47 ug/L?

The referee is right. The described change refers to the difference in Chl a concentration between station 01 and station 15 (Fig. 2a) and does not represent a temporal Chl a increase at a specific station. We changed this accordingly.

Changes in text:

Lines 552-553: […] increased over the course of the cruise from 1.63 ± 0.015 µg L-1 at the first station to 2.47 ± 0.1 µg L-1 at the last station (Fig. 2a). […]

Also, the station numbering has been revised. The previous station numbers referred to ship-based event logs. However, since no event logs were recorded in DSHIP for the working boat sampling, a new station scheme was introduced for this manuscript. These stations encompass both SML sampling and the collection of the particulate fraction using nets.

Lines 538–539: “TAA concentrations in the SML peaked in autumn with a maximum EF of 2.4 (Dreshchinskii and Engel, 2017), which is comparable to the highest EF reported in this study”

- This highest EF appears to be shown in the supplemental material (likely Fig. S7 based on line 362). Please clarify where and when this highest EF was observed in the present study and provide the corresponding value.

We agree with the reviewer, referred to the supplement material and provided the highest TAA EF.

Changes in text:

Line 592-593: […], which is comparable to the highest TAA EF of 2.3 (Fig. S7; station 06) reported in this study. […]

Lines 547–548: “The enrichment of surfactants in the SML differed in dependence of phytoplankton abundance, as the EF during HPA<20µm (1.1 ± 0.2; median = 1.1) exceeded the EF during LPA<20µm (1.1 ± 0.2; median = 1.0).”

- This statement is awkward given how similar the EF values are. It is unclear whether these differences are statistically or mechanistically meaningful, and this should be addressed explicitly.

We agree with both reviewers on this point and adjusted our statement accordingly.

Changes in text:

Line 601-603: The enrichment of surfactants in the SML was similar across phytoplankton abundance conditions, with comparable EF values observed for High P/NP (1.1 ± 0.2; median = 1.1) and Low P/NP (1.1 ± 0.2; median = 1.0). Although the median EF was slightly higher during High P/NP conditions, the overall differences were small and not statistically significant. […]

Lines 629–630: “Data separated only into the defined categories of HPA and LPA<20µm along PC2, which explained considerably less variance of the TAA data”

- It is not clear to me what new information this sentence is intended to convey. Additionally, while the amino acids associated with PC1 are listed (line 625), a similar description is not provided for PC2. Clarifying this would improve interpretability.

We understand that it is not clear what this sentence intends to convey. We therefore provided a more detailed explanation to clarify what was meant.

We wanted to convey that the PCA shows a more pronounced separation of TAA between SML and ULW (Fig. S8) than a separation between the categories High P/NP and Low P/NP (Figure 8c). We revised the manuscript accordingly and added a clear explanation.

Changes in text:

Line 685-689: […] Consistently, the PCA reveals a more pronounced separation of TAA composition between SML and ULW samples (Fig. S8; PC1) than between the High P/NP and Low P/NP condition (Fig. 8b; PC2). This pattern suggests that depth-related differences exert a stronger influence on TAA variability than the distinction between P/NP-associated conditions. However, filamentous and colonial cyanobacteria dominated in the ULW during High MP conditions (Fig. 4), while […]

4) Other Minor / Editorial Comments that were noticed

Line 53: “A large-scale oceanic in the Atlantic demonstrated…” This sentence appears to be missing a word (e.g., “study” or similar), which disrupts readability.

We thank the reviewer for this comment. The sentence was revised by adding the missing noun “study” to complete the phrase. – Line 53

Lines 352–354: “When samples were pooled across categories (i.e., not distinguished by HPA/LPA), TAA concentrations differed significantly between depths….resulting in an average EF of 1.2 ± 0.4 (median = 1.1).”

It is unclear whether this statement refers to >20µm or <20µm conditions. Based on the preceding sentence, it appears to refer to <20µm, but clarification would be helpful.

Thanks for this comment. This statement was removed during the revision of Section 3.2.

Line 406: GABA could be redefined here or in the Figure 7 caption/legend, as it was last mentioned much earlier in the introduction.

Line 452: […] contributions of γ-aminobutyric acid (GABA) […]

Line 470: Instead of stating that correlations “appeared,” a more definitive term such as “were observed” may be more appropriate.

We replaced “appeared” with “were observed”. – Line 495

Line 573: “summerly” does not appear to be a standard usage word.

Thanks for pointing this out. We changed the sentence appropriately.

Changes in text:

Line 629-630: […] The observed summer TOC variability can likely be attributed to phytoplankton production. […]

Line 590: “Central Baltic Sea and in spite the absence of a bloom.” This sentence appears to have an extra “and” which is causing some confusing grammar.

Indeed, it sounds confusing. We changed that so that the meaning of the sentence is clear.

Change in text:

Line 646: […] Central Baltic Sea, even in the absence of a bloom.

Figures:

Figure 5 caption: Regarding the statement “surfactants (c) and total combined carbohydrates (TCCHO) (d) were differentiated for high and low phytoplankton <20µm abundance (HPA<20µm and LPA<20µm).”

On initial reading this caption was confusing because the lowercase “s” made it appear to continue the previous sentence. This also led to confusion because the x-axis (at least on the left side of the figure) refers to >20µm categories. Consider leading with a clearer framing, for example: “High and low phytoplankton <20µm abundance groups (HPA<20µm and LPA<20µm) were compared for differences in surfactant concentrations (c) and total combined carbohydrates (TCCHO) (d).”

We thank the reviewer for this constructive comment. The “s” in “Surfactants” should indeed be capitalized, as it begins a new sentence. In addition, we decided to revise Fig. 5 and present the given parameters with respect to depth and P/NP conditions. As part of this revision the caption of Fig. 5 changed.

Please note here again that we have also revised Fig. 5 and present parameters derived from SML and ULW only under P/NP conditions (Line 374-375).

Changes in text:

Line 378-381: Concentration differences during low pico- and nanophytoplankton abundance (Low P/NP) and high pico- and nanophytoplankton abundance (High P/NP) conditions in the sea surface

microlayer (SML) and underlying water (ULW) for (a) total organic carbon (TOC), (b) total amino acids (TAA), (c) surfactant (SA) concentrations, and (d) total combined carbohydrates (TCCHO) are presented. Enrichment factors (EFs) for each parameter are shown for the respective condition.