

First of all, I really appreciate the effort that the authors went to in responding to my overlong review. On the whole, I'm happy with the changes that they've made to the manuscript. However, I do still have some concerns with a few of their responses. I've listed the main, overarching ones immediately below, but then give more specific comments to particular responses afterwards.

We want to thank the reviewer for the positive feedback and highly appreciate the additional comments and suggestions for clarifications. We addressed all comments as outlined below and believe that the changes improved the manuscript considerably.

Some of the comments pointed to weaknesses of the model itself that we corrected (e.g. How labile and refractory DON are separated, how bacteria biomass is incorporated, Chl degradation after Si limitation). Due to the changes we repeated the model tuning and got slightly different parameters, and fits. Overall, the quality of the model fits stays very similar and the discussion stays overall the same, but the model is now clearer, more realistic and applicable on a wider scale. Some weaknesses of the models that we needed to discuss disappeared (lack of Chl degradation, low sensibility of excretion and remineralization parameters). Because of these changes we updated the parameter, sensitivity analyses and collinearity test tables and updated all plots showing model fits. We also updated all error estimates in the text, and removed weaknesses of the model, that were previously pointed out in the text, but which are now not apparent anymore.

The missing equations and state variables for labile and refractory DON pointed to an assumption in our model that excreted labile DON is quickly converted to refractory DON. We realized that this quick conversion is unrealistic and changed it to 2 separate DON pools (See equations below), where labile DON does not aggregate to refractory DON during the time of the experiment. This change led to a higher sensitivity of the DON degradation parameters and a worse fit of the EXT<sub>-exr</sub> model. This points to a higher importance of autochthonous DOM degradation compared to allochthonous degradation, which is more realistic. The finding of our previous version was the opposite, which we previously discussed as a potentially problematic simplification in our study.

The higher sensitivity of the excretion and remineralization parameters also lead to a higher importance of EPS aggregation that was previously pointed out by reviewer 3. Thus, we re-evaluated the model extensions of the supplement and can now state that EPS degradation is indeed a potentially important process as suspected by reviewer 3. When considering EPS aggregation we now get a better fit than possible without it. The extension with increased excretion in a biofilm shows the same result as before (Full compensation/collinearity by/with PSSi). We removed the 3<sup>rd</sup> extension of increased NH<sub>4</sub> regeneration due to the problems of the NH<sub>4</sub> data pointed out by this review. There may be more NH<sub>4</sub> regeneration in the biofilm, but at the same time more immobilization. So the trend can go both ways and a model assuming increased remineralization is unrealistic.

The lack of Chlorophyll degradation of the EXT model points simply to a missing formulation in one of the model equations.  $dChl/dt = 0$  (under Si limitation), which should be  $dChl/dt = 0 - RChl * Chl$  (RChl, being Chlorophyll degradation), which is modelled in all other Chl synthesis related equations of the model. After the correction, the Chl degradation of the EXT model is now well represented. In the previous manuscript, this absence of degradation was pointed out as a weakness of the model. Hence, the correction strengthened the model and we could remove the statement that Chl degradation is not well modelled.

The confusion about the units of the bacterial carrying capacity pointed us to a more realistic approach of giving bacteria biomass (and carrying capacity) in Carbon units, which makes their abundances directly comparable with algae POC and biomass and the model easier to understand and replicate. We also added the logistic growth curve fit to the bacteria biomass in the supplement.

The starting conditions (the state variables) were indeed not clear. We now clarified that the experiments start at day 1 (excluding day 0 where we assume artifacts directly after the transfer of cultures). For NO<sub>x</sub> and Si we do not have measurements for day 1, but only for day 0 and 2. We now clarified that we use the mean data between day 0 and day 2 as starting condition. After adding the information to the methods, with the experimental data in the plots of the manuscript and the raw data being publically available on Dataverse, the process is now transparent, clear and repeatable.

Some of the explanations for the experimental data still do not make sense (e.g. occurrence of a

PO4 spike). Explanations should be consistent between the axenic and non-axenic cultures, and the authors should not be afraid to note where a feature in the data appears inexplicable (i.e. is more likely an experimental / measurement error).

Concerning the high experimental NH<sub>4</sub> values, we explained in-depth, why we suggest that the issue lies with immobilized ammonium that is released via filtration (and thereby part of the measured NH<sub>4</sub>), while being unavailable for the diatoms (immobilized in EPS). This is indeed an experimental issue as we also mention in the revised manuscript.

Concerning the phosphate peak we agree that the sudden increase of 100% can not solely be explained by reduced Diatom uptake of PO<sub>4</sub> while bacterial PO<sub>4</sub> remineralisation increases. We also do not have an explanation for the sudden drop at the last day. Since PO<sub>4</sub> is not part of the model, we do not consider these unexplained variations to be a problem for the model and manuscript. However, we now acknowledge these patterns as mostly inexplicable in the manuscript as suggested by the reviewer (See below). We agree that it is better to not data/measurement issues that we cannot fully explain, than only discussing potential problems that may explain part of these inexplicable PO<sub>4</sub> changes.

We also clarified the outliers in the figures more clearly by excluding them from the polygons and simply marking them as asterisks. Some of the concerns are related to single outlier values (e.g. sudden Chl drop at day 8, large variance of phosphate at day 14). The figures are now much clearer and less misleading (outliers are apparent without the need to read the legend).

The model equations are still not complete as far as I can see. There are also some cosmetic issues around the use of (unexplained) constants in the equations – it would be better to assign (fixed) conversion parameters.

We are very grateful for the effort of RW2 to check our model equations in detail. Some equations were indeed not complete and/or unclear.

We now added the missing DON equations separating labile and refractory DON and the conversion of DOC (which we measured) to DON (which we used in the model). The constants are now parameterized before the equations (Redfield ratio, Molar mass of nitrogen).

We also corrected the equation about logistic growth of bacteria and now use Carbon units instead of cells, which makes it more quantitative and comparable to other model parameters. For Chl degradation after Si limitation, we added the term  $dChl/dt = - RChl * Chl$ , which now allows us to model the degradation in the stationary phase.

The explanation of the tuning method is much appreciated. However, as it stands, it's very dense text that's been included. It might be better to add a supplementary section that breaks this up into bullet-pointed steps – that would be ugly for the main body, but would probably be very helpful as a supplement interested readers could consult.

We thank the reviewer for the suggestion and added such a supplementary section with bullet point in the Supplement.

While the authors have rephrased their hypotheses, they still make no use of them, nor even refer to them, elsewhere in the manuscript. Plus, and especially because of the way they're used, these read more like conclusions of the work than hypotheses that the paper actively tests.

We do return to the hypothesis but we realized that we did it in a very intertwined way with no clear references back to the specific hypothesis. We agree that it should be more clear and suggest additional references to the hypotheses in the discussion. We now added clear references back to the hypotheses where it was useful. (See details below)

Overall, I think that the manuscript is now close to being acceptable once these remaining details are addressed. I would advise accept after minor revisions.

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Ln. 161, "The strong spike of phosphate after day 8 corresponds with the end of the exponential phase for algal growth and a spike of ammonium. At the same time bacteria abundances start increasing considerably. Thus, we explain the phosphate peak by increased bacterial regeneration (source of phosphate) and decreased algal uptake (sink of phosphate) at the same day." - \*Qualitatively\* I can see this line of argument, but it doesn't really stack up \*quantitatively\*. First, the timing's not quite right, with ambient PO<sub>4</sub> jumping ~50% on day 8, when phytoplankton are at their peak and bacteria have only just got moving. - It would be more convincing to me if there was an attempt to budget the elements to make this shift between the medium, the phytoplankton and the bacteria clearer; it remains difficult for me to shake the impression that something has gone wrong in the experiment; and this should be done for both experiments and models; among other things, if it can't be done for whatever reason, it does tend to suggest that, contrary to the authors "hypothesis 3", experiments do not provide a good testbed for ecosystem functionality

Concerning the phosphate peak we agree that the sudden increase of 100% can quantitatively not solely be explained by reduced Diatom uptake of PO<sub>4</sub> while bacterial PO<sub>4</sub> remineralisation increases. We also do not have an explanation for the sudden drop at the last day. We can not budget the entire P pool within bacterial P, algae P and PO<sub>4</sub> since a large part is within the DOM pool. Mineralization processes may however be another explanation. Since PO<sub>4</sub> is not part of the model, we do not consider these unexplained variations to be a problem for the manuscript or model. We now acknowledge these patterns as mostly inexplicable in the manuscript.

The mismatch of NH<sub>4</sub> and PO<sub>4</sub> remineralisation may be caused by N being more limiting (more N retained by bacteria than released as NH<sub>4</sub>) and PO<sub>4</sub> not (less PO<sub>4</sub> retained and more excreted via remineralization), but our data do not allow to answer this hypothesis conclusively.

Changes: L472f in the revised MS: "After NO<sub>3</sub> depletion at day 15, also PO<sub>4</sub> concentrations drop, indicating a coupling of NO<sub>3</sub>:P metabolism, but not of NH<sub>4</sub>:P metabolism. Thus, the sudden drop may also indicate dynamics of bottle experiments, not accounted for, showing potential limitations of these experiments."

Changes: L467f in the revised MS: "With the start of the stationary phase, NH<sub>4</sub> and PO<sub>4</sub> concentrations doubled, presumably due to decreased assimilation by the silicate starved diatoms and increased regeneration by bacteria, supplied with increasing labile DOM (doubled remineralisation rate in EXT) excreted by the stressed algae. However, NH<sub>4</sub> concentrations double in 4 days, while PO<sub>4</sub> concentrations double in only 1 day, indicating some unexplained internal dynamics, potentially via different bacterial uptake and release of N and P."

Changes: L511f in the revised MS: "Overall, our cultivation experiment is powerful to represent some major spring bloom dynamics, but has its limitations, thereby confirming our third hypothesis only to some extent."

Ln. 183, "Excretion of organic phosphate by diatoms is also common in cultures with surplus orthophosphate (Admiraal and Werner, 1983), which can be another explanation of the phosphate peak after silicate becomes limiting." - but it only happens in the case with bacteria, so this doesn't make sense; if anything the case without bacteria should be more stressed because there's no nutrient regeneration

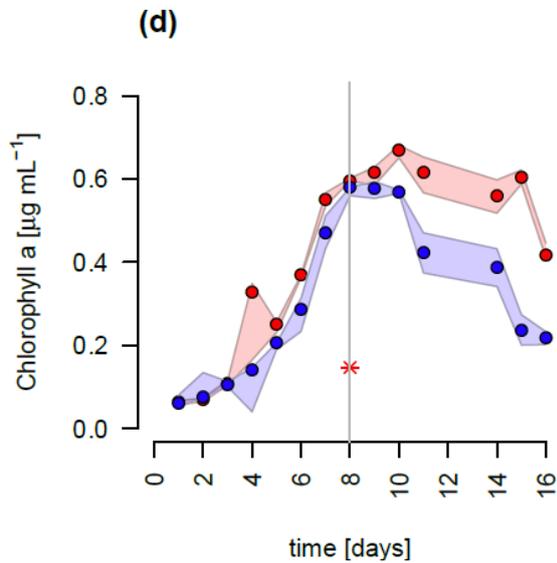
We do see a spike in both treatments. However, we agree that the spike in the axenic culture is much less pronounced and removed the argument for streamlining the manuscript and to avoid confusion.

Ln. 193, "Figure 4, 5, B1) "...Chlorophyll a concentration in experimental cultures with a potential outlier at day 8, presumably due to photodegradation, causing a negative spike." - "photodegradation"? it looks like a straight error to me as it bounces right back; if you are going with photodegradation, expand on this

We assume indeed that the outlier is a measurement error. We think that this specific sample might have been exposed to light during the extraction, which degrades Chlorophyll and leads to lower values. So, yes, it is a straight error. We avoided simply removing outlier values of measured data from plots, simply because they do not fit to our (or the models) expectations, which we consider

data manipulation. However, we acknowledge that this data point appears misleading. In fact, it is not clear that it is based on an outlier without reading the entire legend. We show all outliers now as an asterisk in the plot, while excluding it from the area of the polygon.

Changed legend: "Figure 4, 5, B1) "...Chlorophyll a concentration in experimental cultures (the asterisk indicates a presumed measurement error)."



Ln. 202, "We added the missing equation and double-checked for any other incomplete model descriptions." - the equations for DOM are still inadequate; background DOM is listed as a state variable but its equation is omitted

We did not list DOM as state variable, but assume the reviewer refers to the DON state variable. We are indeed lacking the conversion from DOC (measured variable) to DON, which we estimated via the Redfield ratio:  $DON = DOC / 16$ .

We did following changes in the appendix tables:

- We added the state variable **DOC** in table A1
- We defined the redfield ratio (**RR**) C:N = 106/16 in as parameter in table A3
- We added following equation to table A7:  **$DONr = DOC / RR$**

We also realized that we did not differ between DONr and DONl in the equations in table A7 and corrected the equations in the following way (Replacing the number 14 with the parameter molar mass of N ( $M_N$ ) as suggested below). The previous model also had an assumption of labile excreted DON becoming refractory shortly after it is released to the medium, which we realized is not realistic without including a 3<sup>rd</sup> DON pool. Thus, we adjusted the equations in order to keep all excreted DON labile throughout the experiment. The model changes required a new model fitting, plotting, and tuning. The results are nearly the same.

Changes in table A7:

8)	Ammonium uptake and production	$IF \left( \frac{C}{N} < 10 \right)$
	<i>(Threshold after Tezuka 1989, and Gilpin 2004)</i>	$\frac{dNH_4}{dt} = \frac{-\left(\frac{V_{NH_4}^C}{Q}\right)N + Bact\ DONl\ rem + Bact\ DONr\ rem_d}{M_N 10^3}$
		$ELSE\ IF\ (NH_4 > nh_4_{thresj})$

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$$\frac{dNH_4}{dt} = \frac{-\left(\frac{V_{NH_4}^C}{Q}\right)N - \frac{dBact}{dt}RR}{M_N 10^3}$$

ELSE

$$\frac{dNH_4}{dt} = \frac{-\left(\frac{V_{NH_4}^C}{Q}\right)N}{M_N 10^3}$$

- 9) DON uptake and production

$$IF \left(\frac{C}{N} < 10\right)$$

$$\frac{dDONl}{dt} = \frac{-Bact DONl rem + xf N - \frac{dBact}{dt}RR}{M_N 10^3}$$

$$\frac{dDONr}{dt} = \frac{-Bact DONr rem_d}{M_N 10^3}$$

ELSE

$$\frac{dDONl}{dt} = \frac{xf N}{M_N 10^3}$$

$$\frac{dDONr}{dt} = 0$$


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Ln. 206, "Equation 7a) Silicate uptake" - working through the units in this equation, using the given units for state variables and parameters, I find that there's a discrepancy of 3 orders of magnitude; I think one of the sets of units must be listed incorrectly

Vmax = (mol Si / d) / (mg C)  
 Sid = umol / L  
 Smin = umol / L  
 Ksi = umol / L  
 C = mg C / m3

dSid / dt = (Vmax \* Sid \* ((Sid - Smin) / (Ksi \* Smin)) \* C  
 = ((mol Si / d) / (mg C)) \* umol / L \* (umol / L) / (umol / L \* umol / L) \* mg C / m3  
 = ((mol Si / d) / (mg C)) \* ((umol / L)^2) / ((umol / L)^2) \* (mg C / m3)  
 = ((mol Si / d) / (mg C)) \* (mg C / m3)  
 = mol Si / d / m3  
 = mmol Si / L / d  
 ≠ Sid ≠ umol / L

We are thankful for the reviewer found this error and corrected the unit for Vmax to: (umol Si/ d) / (mg C). as mentioned below we also changed the units of C,N, and Chl to mg L-1

dSid / dt = (Vmax \* Sid \* ((Sid - Smin) / (Ksi \* Smin)) \* C  
 = ((umol Si / d) / (mg C)) \* umol / L \* (umol / L) / (umol / L \* umol / L) \* mg C / L  
 = ((umol Si / d) / (mg C)) \* ((umol / L)^2) / ((umol / L)^2) \* (mg C / L)  
 = ((umol Si / d) / (mg C)) \* (mg C / L)  
 = umol Si / d / L  
 = umol Si / L / d  
 = Sid / dt = umol / L / d

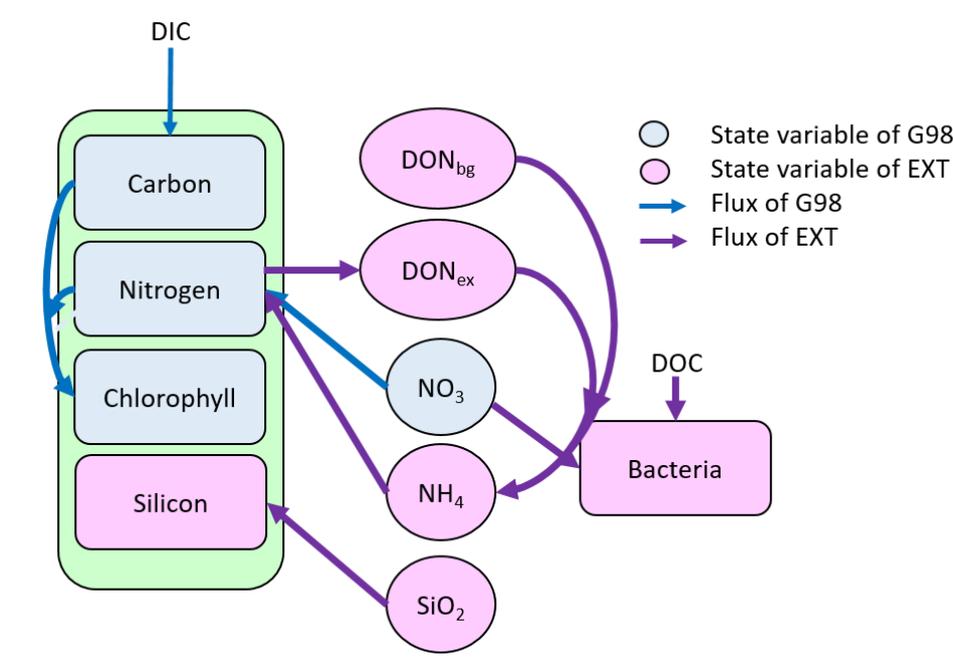
Ln. 207, "Equation 7b)" - the 14 comes from nowhere here; you should make it clear that it's a molar mass

We agree and added the term M<sub>Si</sub> as parameter in table A3 and replaced 14 in eq 7b with M<sub>Si</sub>

Ln. 235, "Figure 1"

- what is meant by control here?; these lines in particular make this diagram difficult to follow; when I suggested a diagram it was to (a) make clear what the state variables are, and (b) indicate the relationships between them so that the simple food web in the experiment vessels was; this diagram is overcomplicated, while also seeming to omit detail (e.g. bacterial metabolism)  
- in this diagram the bacteria - which are meant to be heterotrophic - consume only nitrate; that doesn't make sense biogeochemically

We agree that the diagram is too complex with the lines indicating the controls and suggest removing the controls and simplifying the diagram by simply showing the state variables and how they are connected. Regarding bacteria we add an unmodelled DOC pool to make the diagram biogeochemically more meaningful.



Ln. 258, "2.3 Modelling fitting"

- while thorough, this is very difficult to parse. I would suggest writing it out clearly as a bullet pointed step-by-step process (you've actually begun this in your response elsewhere), and adding this description to the supplementary material. Make clear which models, which parameters, which target datasets and whether tuning was manual or automatic  
- On a separate note, I don't think initial conditions used are covered; and some of the plots suggest this is not done as one might expect; e.g. Figure 6 seems to show the model starting at values quite different from the experiments

We agree that the "Modelling fitting" chapter became quite detailed after considering all suggestions from the first review round and added a step-by-step process as bullet points to the Supplement. Since the bullet points do not include the justifications for the different approaches, we do not think that they can replace Ch. 2.3, but we agree that it helps to understand the approach more clearly.

We added following chapter to the Supplement:

### Model tuning protocol (short version)

#### 1. Fitting of the G98 model to the BAC- experiment

##### 1.1. Model programming (R)

##### 1.2. Model solving (ode function, Runge-Kutta method, deSolve package)

### 1.3. G98 parameter fitting on BAC- experiment

#### 1.3.1. visual tuning (visual comparison with plots)

#### 1.3.2. RMSE tuning (manual minimizing the RMSE Eq C2)

#### 1.3.3. Automated tuning (FME package)

##### 1.3.3.1. Normalize data (POC, PON & Chl x 10, DIN / 10)

##### 1.3.3.2. Sensitivity analysis (SensFun function)

##### 1.3.3.3. Parameter identifiability/ collinearity (Collin function)

##### 1.3.3.4. Parameter selection (identifiable, in doubt higher sensitivity)

##### 1.3.3.5. Automated tuning ( $R_C$ , $K_{NO_3}$ , $n$ , and $\alpha_{Chl}$ , modfit function)

###### 1.3.3.5.1. Global optimum (Pseudorandom search algorithm)

###### 1.3.3.5.2. Local optimum (Nelder Mead algorithm)

#### 1.3.4. Compare automated parameters with initial fit (visual & RMSE)

### 2. Fitting of the EXT model to the BAC+ experiment

#### 2.1.1. Model programming (R)

#### 2.1.2. Model solving (See 1.2)

#### 2.1.3. EXT parameter fitting to BAC+ experiment (parameters from 1 unchanged)

##### 2.1.3.1. Tuning of Bacterial growth parameters on bacteria ( $\mu_{bact}$ , $bact_{max}$ )

##### 2.1.3.2. Tuning of Silicate parameters on silicate ( $K_{Si}$ , $V_{max}$ )

##### 2.1.3.3. Tuning of remaining EXT parameters (See 1.3)

Concerning the separate note, the model starts at day 1 in order to avoid artifacts caused by the stress of transfer. For POC,Chl,PON and  $NH_4$  the values were measured at day 1 and this was straightforward.  $NO_3$  values were measured at day 0 and day 2, but not directly at day 1. Hence, we used the mean between the values of day 0 and 2 as start  $NO_3$  values for the model (Separate calculations for BAC- and BAC+ -> different starting conditions). Fig 6 still shows the values at day 0 of the measured data, since we argue that it helps to evaluate the overall fit more thoroughly. We added the information (including why we start the model at day 1) to chapter 2.3.

Change: L270f in the revised MS: "The model fitting started at day 1 in order to avoid artifacts during acclimation of the cultures after transfer to a new medium. Si and  $NO_3$  were not measured at day 1 and the mean of day 0 and day 2 was used."

Ln. 381, "2. Diatoms continue photosynthesis under silicate limitation at a reduced rate if DIN is available"

- what does this mean for cell division?; if not here, you should certainly comment on this somewhere

We added the information more specifically in the discussion:

Change: Line 526f in the revised MS: "Si is only needed for frustule formation and cell division, mostly during a specific time in the cell cycle (G2 and M phase,...)..."

Ln. 389, "Ammonium is most likely immobilized in the biofilm via adsorption to the EPS and accumulation in pockets unavailable to diatoms"

- this needs a bit of expansion - the model has less  $NH_4$  than the observations, and if it was being immobilised in the experiments within the biofilm (something that's absent in the model), one might instead expect lower concentrations, no?

We suggest that the immobilized ammonium is not available for diatoms, but that it will be part of the measurements after release of the ammonium during filtration and pH changes. Thus, we measure ammonium which is not available for diatoms, while the model only considers bioavailable ammonium. In the model, ammonium is taken up preferably and quickly after its release keeping the ammonium levels low, while we suggest that this is not possible for immobilized ammonium. Consequently, the model will assume more ammonium uptake and less  $NO_3$  uptake.

- also, it's not clear which of this text has made it into the revised manuscript

We added this information of the revised manuscript submitted already after the first round of reviews in the following sections. We also added a few more details as suggested in this review (marked in yellow below):

Ch 2.3: "The model was only fitted to total DIN, due to the potential uncertainties related to ammonium immobilization in the biofilm, which is released during filtration and part of the measured data."..." The main effect of the biofilm that we could not model with the available data appears to be ammonium immobilization in the biofilm, either due to adsorption, accumulation in pockets, or conversion to ammonia due to the locally reduced pH caused by increased bacterial respiration."

Ch 3.1: "While not all ammonium measured is also available for algae growth, discussion of the dynamics (decrease in the start, increase with the onset of the stationary phase), especially if also shown in the EXT model, are still useful to understand multinutrient dynamics (e.g. regeneration)"

Ch 3.2: "This indicates that the problem lies with the ammonium data, which include immobilized ammonium in the biofilm, unavailable for diatoms growth, while the model assumes that all ammonium is available."

Ln. 520, "In previous cultivation experiments, no efforts for obtaining axenic cultures were mentioned, which hints to bacteria contaminated cultures."

- I take the point; from my own experience of the tedious nature of maintaining axenic cultures, it's not unreasonable to assume that a study where this isn't mentioned is probably not working with axenic cultures

We thank the reviewer for the acknowledgement and personal experience as support for our suggestion.

Ln. 601, "Microscopy showed bacteria attached to the diatoms (mostly in the stationary phase), but mostly free-living."

- as the audience of this paper may be unclear on this point, it would be good to add a sentence noting your microscopy results so that readers understand you are largely referring to free-living bacteria

We added the information to the text: Line 120 in the revised MS: "...inoculation with mostly free-living bacteria cultures, isolated beforehand from the non-axenic culture."

Ln. 621, "As mentioned above, we changed the hypotheses in the following way:"

- you still don't return to them!; normally, hypotheses are stated ahead of work then reassessed at the end of the work; these are almost conclusions that are appearing in the introduction!

We do return to the hypothesis but we realized that we did it in a very intertwined way with no clear references back to the specific hypothesis. We agree that it should be more clear and suggest following additions in the discussion, which mainly add a clear reference back to the hypotheses where it was useful.

In the previous version we summarized how each hypothesis was confirmed in the study as introduction to the Discussion: "The experimental incubations represented typical spring bloom dynamics for coastal Arctic systems, including an initial exponential growth phase terminated by N and Si limitation and the potential for an extended growth period via regenerated production."

We changed this introduction in the following way to make it more clear that it refers back to the hypotheses: "The experimental incubations showed that in the presence of bacteria both the growth period and gross carbon fixation can be extended (Hypothesis I). The diatoms were able to continue photosynthesis under silicate limitation at a reduced rate as long as silicate was present (Hypothesis II). Overall, the incubations represented typical spring bloom dynamics for coastal Arctic systems, including an initial exponential growth phase terminated by N and Si limitation (Hypothesis III) and the potential for an extended growth period via regenerated production."

For making the red line and relevance of the hypothesis even clearer we also reference back to the hypothesis in the more detailed discussion.

L. 459-462: "As suggested by our second hypothesis, photosynthesis was reduced by approx. 70% after silicate became limiting, which is comparable to earlier experimental studies (Tezuka, 1989). However, as suggested by our first hypothesis, the secondary bloom was extended in time by bacterial regeneration of ammonium, allowing regenerated production to contribute about 69% of the total production (f-ratio=0.31) ..."

L469f: "The presence of bacteria and thus regenerated production allowed diatom growth to continue 8 days after silicate became limiting (Figs. 2, 3 & 4), nearly doubling the growth period similar to observations in the field (e.g. Legendre and Rassoulzadegan, 1995; Johnson et al., 2007), which supports our Hypotheses I and III."

L481f: "While we do not expect the f-ratio in our bottle experiment to be directly comparable to open ocean system, which does include a variety of algal taxa beyond *C. socialis*, a comparison can aid to identify limitations in our experiment and model. The f-ratio also allows a discussion of how representative the cultivation study is for typical spring bloom dynamics (Hypothesis III)."

L500f: "Hence, ecosystem scale models will need to consider these dynamics regarding bacterial abundances, microbial networks and particle export in addition to bacterial remineralization in order to model realistic ammonium regeneration in the euphotic zone. Overall, our cultivation experiment is powerful to represent some major spring bloom dynamics, but has its limitations and thereby confirming our third hypothesis partly."

Ln. 754, "Thus, we implemented a labile (DONl) and refractory (DONr) DON pool with different remineralization rates (rem, remd)."

- as noted the equations for the DONr pool are not included

We added the equations as mentioned above.

Ln. 760, "We do not suggest a complete stop of remineralisation ..."

- OK - this sounds more reasonable

Thanks for the confirmation.

Ln. 766, "DOM C/N mass ratio"

- why use "mass ratio" when "molar ratio" is more common?

We changed the definition to molar ratio including line 775.

Ln. 775, "We refer to DOM as substrate for bacteria and clarified it: DOM C/N ratios...."

- Please be clear if C:N is mass or molar throughout if you're going to chop and change

We changed the manuscript in a way that all ratios relate to molar ratios, unless given otherwise (gC:gN). gC:gN ratios are mostly needed for the modelling and kept for comparability of parameters with earlier fits of models relating to the G98 construct. In the methods we still mention "molar C/N ratios" (e.g. molar C/N ratio <10) while we do not add this information, when we are writing about ratios in general. A "higher" molar C/N ratio has the same implications as a "higher" mass C/N ratio. What matters and what needs to be clear are ratios with values given. We proof-read the text and clarified it where needed.

Ln. 884, "Collinearity is a measure for the parameter identifiability ..."

- this is good, but has it made it into the text?

Yes it is part of ch 2.3: "Prior to the automated fitting, parameters were tested for local sensitivity (SensFun) and collinearity, or parameter identifiability (collin; e.g. Wu et al., 290 2014)."... The sensFun output is further used as input for the collinearity, or parameter identifiability analyses. Parameters were considered collinear and not identifiable in combination with a collinearity index higher than 20 (Brun et al., 2001). In this case, only the more sensitive parameter was used for 295 further tuning."

Ln. 975, "due to the potential uncertainties related to immobilized ammonium"

- as noted above the immobilisation is curious given that the measurements include this extra NH<sub>4</sub>; is the NH<sub>4</sub> only effectively immobilised for the phytoplankton such that when the vessel is stirred, it becomes apparent?

Yes, as mentioned above, we suggest that the ammonium is only immobilized for the algae, but released during filtration (vacuum disintegrating EPS structure, change in temperature and pH). As mentioned above this information is now given more clearly in the text as well.

Ln. 1129, "The R code is now available at github"  
- if you want to create a permanent record of the model with a DOI, consider using the Zenodo link from Github; this will preserve a version of the exact version used here  
- if you do this, make sure to add the DOI to your final manuscript revision

We thank the reviewer for the valuable suggestion and archived the code permanently on zenodo. The doi number is given under data availability in the manuscript.

Ln. 1154, "We argue that the large range is plausible since it is i) ..."  
- on point (i), outliers are usually excluded because they are suspected of being false (e.g. measurement error); on point (ii), you're talking about the size of the range of PO<sub>4</sub> on day 14, when there's literally zero range of bacterial variability at day 14 - this doesn't make sense

We now excluded outliers from the polygons and marked them as an asterisk to avoid confusion. (See comment on Chl above).

Regarding point 2 we overall agree and did not mention it in the manuscript. Instead we acknowledge some inexplicable variation pointing to limitations of the experiment as suggested earlier by the reviewer.

Ln. 1165, "As mentioned in the results, the bacteria growing towards the end are still in so low ..."  
- that's OK - it's tough keeping things axenic

We thank the reviewer for the confirmation

Ln. 1192, "we added the output of a prolonged model run in the supplement"  
- thanks!

We are thankful for the helpful suggestion.

Ln. 1280, "We shortened the table slightly and explained all columns in detail in the corrected version"  
- the mean, min and max columns are still not properly explained; for instance, why are there negative values?

Since the information about the sensitivity ranges is not needed for the manuscript we removed the last columns (min, mean max). Negative or positive values indicate the direction in which the output variable changes after parameter perturbations (decrease or increase).

Ln. 1313, "We changed the form of eq 14 to the same format as in eq 15"  
- OK; but the use of (unexplained) numbers in these equations rather than parameters remains unhelpful

As mentioned above we replaced the numbers with parameters that we explain now in table A4 (e.g. RR (Redfield C:N ratio) = 16; MN (Molar mass of nitrogen) = 14)

Ln. 1345, "Table A8"  
- this table is still confusing; do the rows need identifying labels?

The rows indicate which set of two parameters were tested for collinearity. For consistency with other tables in this paper, we replaced 0 with X (for absence) and 1 with V (for presence). We also added

a vertical line before the last column to make clear that the first columns are part of a matrix and the last column is the result (Collinearity index).

- also, what's with the strange number more than 1 million?

A collinearity of more than 1 million means that these two parameters are highly collinear and clearly unidentifiable. The parameters are I (light) and alphaChl (C assimilation per Chl and light), which shows already in their units that a change in light can be almost fully compensated by a change in alphaChl. With the threshold of 20 given by Brun et al. we argue that the table header is sufficient for interpreting this high value as unidentifiable parameter combination.

Changed table:

Parameter combinations										collinearity
$\zeta$	$R^C$	$\theta_{max}^N$	$Q_{min}$	$Q_{max}$	$\alpha^{Chl}$	I	n	$K_{no3}$	$P_{ref}^C$	
V	X	V	X	X	X	X	X	X	X	27
V	X	X	X	V	X	X	X	X	X	98
V	X	X	X	X	V	X	X	X	X	31
V	X	X	X	X	X	V	X	X	X	31
V	X	X	X	X	X	X	V	X	X	93
X	V	X	X	X	X	X	X	X	V	25
X	X	V	X	V	X	X	X	X	X	34
X	X	V	X	X	V	X	X	X	X	101
X	X	V	X	X	X	V	X	X	X	101
X	X	V	X	X	X	X	V	X	X	38
X	X	V	X	X	X	X	X	X	V	32
X	X	X	X	V	V	X	X	X	X	40
X	X	X	X	V	X	V	X	X	X	40
X	X	X	X	V	X	X	V	X	X	146
X	X	X	X	X	V	V	X	X	X	455473
X	X	X	X	X	V	X	V	X	X	46
X	X	X	X	X	V	X	X	X	V	28
X	X	X	X	X	X	V	V	X	X	46
X	X	X	X	X	X	V	X	X	V	28

Figure 2

- I'd not really paid attention before, but the experiments have a large span of initial nutrient concentrations - shouldn't this be properly controlled?; or am I misunderstanding the plots?

We agree that the variation of the nutrient concentrations are quite high, but that are the data we measured. However, since our model starts at day 1 and not day 0 (Due to expected stress responses in the 1<sup>st</sup> day), we argue that these variations do not have strong implications for the discussion of the manuscript.

- (and thinking back the point made previously) not only does PO<sub>4</sub> increase inexplicably between days 6-10 (+100%), it also drops off a cliff between days 14-15 (-80%)

See comments above were we acknowledge some inexplicable internal dynamics in the manuscript.

- your plots consistently run out of x-axis; the experiments are at least 15 days long, but your axis stops at 14 days

We extended the x axis to 16 days

- Panel d runs out of y-axis (as well as x-axis)

We also extended the y-axes were needed

Other changes based on the comments above include that outlier are excluded from the polygons and marked with an asterisk

Figure 5

- nothing about bacterial biomass and how it fares when excretion is included / omitted

Modelling bacteria biomass is not the main objective of the paper. As described earlier we modelled bacteria biomass production via a logistic growth curve. DON does not directly affect the fitted growth curve. As described in our previous response, we do not think that an added complexity of bacterial growth modelling is justified by the data or aim of the paper. Thus, we do not think that adding the effect of DON excretion on bacterial growth would add value to the manuscript, but add complexity with rather poor support in the model. See also our response to the first review:

“The main improvement of the model is to include a remineralisation rate controlled by: i) bacteria biomass, ii) substrate (DOM) C:N ratios, and iii) substrate origin (autochthonous, allochthonous). Other models typically have a fixed remineralisation rate either only dependent on the DOM/POM, or not controlled by any environmental variable. Thus, we still see our extension as a considerable improvement and consider a simple logistic growth estimate sufficient.

We could of course model bacteria growth via Michaelis-Menten kinetics based on 2 DOM pools, but this would not have any effect on the parameterization or modelling of algae physiology, which is the main goal of the paper, while increasing the number of parameters and computational costs, which we 790 tried to keep low. Since, the aim of the model is not to model bacteria growth, but algae growth and intracellular C:N:Chl ratios we do not see that a more accurate and more complex model of bacteria growth would improve the manuscript.”

However, we acknowledge that the fit of the logistic growth curve may be of interest and added the plot to the supplement.

- chlorophyll doesn't decline here but it should; and it does in a later supplementary plot (B1c); something doesn't seem quite right.

We thank the reviewer for the observation. In fact, the formulation of a stop in Chl production after Si limitation was the problem. In the previous version the formulation was  $dChl/dt = 0$ , while it should be  $dChl/dt = -RChl Chl$  (maintenance loss of Chl). We corrected the corresponding equation in the manuscript and modelling code and rerun the fitting routine for EXT.

Figure 6

- why do the model lines start at day = 1 and not day = 0?; the observations seem to start at day = 0 by contrast (and unlike in other plots) also, in the BAC- experiment, the model seems to start at a lower value than it did in the experiments

See comments above: "the model starts at day 1 in order to avoid artifacts caused by the stress of transfer. For POC,Chl,PON and NH<sub>4</sub> the values were measured at day 1 and this was straightforward. NO<sub>3</sub> values were measured at day 0 and day 2, but not directly at day 1. Hence, we used the mean between the values of day 0 and 2 as start NO<sub>3</sub> values for the model. Fig 6 still shows the values at day 0 of the measured data. We added the information (including why we start the model at day 1) to chapter 2.3."

Fig 6 in the previous version also included only NH<sub>4</sub> data for the days where NO<sub>3</sub> data are available, which lets the start of the model appear out of place. We now added all measured NH<sub>4</sub> data points to the plot for clarification.

We also double-checked all starting values with our measured data.

- the behaviour of modelled ammonium is still confusing; it seems to have very little to do with what the experiments did;

Considering the overall poor fit to measured NH<sub>4</sub> values, we discussed the effect of NH<sub>4</sub> immobilization and release via filtration in detail in the responses above. In summary, we suggest NH<sub>4</sub> immobilization in the biofilm (unavailable for diatoms) and release during filtration (available for the nutrient analyzer). We added this information as described above (including a straight forward acknowledgement of the data limitations).

Table A2

- what does "mio." mean?; does it mean "million"?; it's not an abbreviation I'm familiar with; and, if so, it looks like bacteria reach concentrations of 60 million cells per mL, but the value for bact\_max ranges 0.005-0.1 million cells per mL; this is confusing

The reviewer pointed out an error in the units that we missed and now corrected in the manuscript. We also double-checked all other units for consistency and solvability in the given equations. For consistency, we now adjusted the bacteria cell numbers, and carrying capacity for the model into carbon units (mgC L<sup>-1</sup>) in order to make them comparable to the C,N, and Chl units of the diatoms (20 fg C per cell). We did following changes in the manuscript:

- All units (except inorganic nutrients) are now given in mg L<sup>-1</sup>.
- We add a figure of the bacterial growth curve fit to the supplement
- We add a methods description of the bacteria cell to carbon conversion
- We adjusted the remineralization rates which are now based on bacterial C instead of cells.

We also double checked all equations in the manuscript and modelling code for consistent units and repeated the fitting routing of the EXT model (Now using bacteria converted to bacterial carbon, and as mentioned above the corrected formulations of labile and refractory DON remineralization).