Referee comment #1 – Dominika Lewicka-Szczebak

Dear authors, this is a very important and interesting study and I've read your manuscript with high interest. It is written precisely and clearly with very well-prepared figures. All the data are easily to follow, and your interpretations are clear. You present a very interesting new theory on the asymmetric intermediate in hybrid N2O production, which is well documented with your data and very well presented in your Figure 8.

We sincerely thank you Dominika Lewicka-Szczebak for the careful reading of our manuscript and for the constructive and detailed comments. We appreciate the positive evaluation of the data presentation and the interpretation, as well as the interest expressed in the proposed asymmetric intermediate for hybrid N_2O formation. In the following, we address the major concern regarding low in situ N_2O concentrations and respond to the individual comments point by point.

I have only one major concern about your data, because you have calculated the in-situ N2O isotopic signatures from very low N2O increase in concentration, as low as 20ppb. From my experience such low increases are not possible to be precisely determined for their isotopic signatures because of accumulation of all the uncertainties in the calculations, associated with isotopic signature of the background N2O and analytical errors. While analysing carefully all your data presented for me it is clear, that these lowest N2O samples may be biased, which I point out in below specific comments. This makes your interpretations less robust. I think if you disregard these lowest N2O samples (20ppb) you would get much clearer and reliable picture (see in the comments below).

We fully agree that the uncertainties for the Taylor Glacier samples with low in situ N_2O production (~20 ppb) are large. Still, we want to mention that all sources of uncertainty, including those related to atmospheric N_2O estimates and analytical precision, are already incorporated in the error estimates through a Monte Carlo simulation for error propagation.

It is important to note that although in situ N_2O production in ice cores is generally small in absolute concentration, it can lead to significant deviations in the measured N_2O isotopic composition because the precursor nitrate can be highly fractionated. The latter is especially pronounced for low accumulation sites such as Dome C considered in this study. As even small amounts of in situ production can significantly alter the measured N_2O isotopic composition, a characterization of such production is essential for ice core studies. For this reason, mapping the isotopic signatures of in situ N_2O across different ice cores is also a key objective of our work. For Taylor Glacier, the only affected samples exhibit low in situ production, so excluding them would prevent us from characterizing this site. As mentioned above, the uncertainties for Talos Dome ice are fully included in the analysis through stringent error propagation.

As a result, the calculated isotopic composition carry large uncertainties, but the general isotopic patterns (high $\delta^{18}O$ and low SP) are already apparent directly in the measured N_2O data, thus even before applying a mass balance approach. Although we cannot yet fully explain these patterns, we believe it is important to present all available data (together with transparent uncertainty estimates) because they contribute to the broader picture of in situ N_2O production in Antarctic ice.

Changes in the text (in **bold**):

L300: "3.2.4 Uncertainties – There are several sources of error in our approach, including measurement uncertainties in N_2O mixing ratios and isotopic composition, uncertainties in the TALDICE N_2O spline interpolation, uncertainties in gas-age estimates – which in turn impact the chronological alignment between ice cores **– and potential small in situ production in TALDICE.** We used a Monte Carlo method for error propagation by running 1000 simulations to determine the mixing ratio and isotopic composition of in situ N_2O. We varied the variables of Eq. (3) and (4) within their uncertainty ranges (Table B1 in Appendix B). The derived uncertainties were calculated as the standard deviation of the results of the 1000 simulations.

As expected, the uncertainty increases for low in situ N_2O mixing ratios (see Eq. 4). To avoid too large uncertainties, we excluded samples with calculated in situ N_2O mixing ratios below 20 ppb. This threshold was chosen because a large part of the dataset exhibit in situ mixing ratios below 50 ppb; applying a higher cutoff would remove a substantial number of samples and significantly limit our ability to investigate in situ N_2O production processes. For samples with in situ N_2O mixing ratios close to 20 ppb, the propagated uncertainties are large (up to several tens of per mil). Nevertheless, these samples were included in this study because in situ N_2O production in ice, although small in absolute mixing ratio, can significantly alter the measured N_2O isotopic composition (Fig. 3a) and these isotopic deviations should be documented."

L374: "**We stress that the mass balance approach used to calculate the isotopic signature of in situ N_2O leads to substantial uncertainties due to error propagation from atmospheric N_2O estimates and analytical precision. These uncertainties are not shown in Fig. 3 for readability but are reported in Figs. 4 – 7 and are explicitly taken into account in the following comparisons between $\delta^{15}N(N_2O_{\text{in situ}})$ and $\delta^{15}N(NO_3^-)$, including the regression analyses**."

L 74 (δ15Nα) (δ15Nβ) alpha and beta should be in uppercase, not lowercase

We have changed this as proposed.

L133-135 – what are the products of the NO3 photolysis, can N2O be also produced in this process? Can the gaseous products be trapped in snow layers? Or is NO2- one of the photolysis products? NO2- could be then the precursor for further chemodenitrification.

Thank you for this comment. Nitrate photolysis in snow produces mainly NO_2(g) and NO_2^-, with NO_2^- accounting for roughly 10% of the photolysis products (Meusinger et al., 2014; Warneck and Wurzinger, 1988). Photochemically driven reactions at the snow surface can, in principle, lead to N_2O formation (e.g., hybrid N_2O from reactions involving NO_3^- and NH_4^+) (Rubasinghege et al., 2011). However, such processes can only occur at the surface of the ice sheet where sunlight penetrates the snow, and where air is not yet enclosed in bubbles, i.e., where air exchange with the atmosphere still takes place. As a result, any N_2O produced by photolysis-driven reactions in this zone would be released to the atmosphere and not preserved in the ice core.

Regarding the role of NO_2^- as a precursor for in situ N_2O, this is a plausible pathway in principle. However, our isotopic results do not support a contribution from photolysis-derived NO_2^-. After NO_3^- deposition, photolysis enriches the remaining NO_3^- in ^{15}N, while the produced NO_2^- is depleted in ^{15}N, with its isotopic composition depending on the fraction of NO_3^- photolyzed. In that case, the $\delta^{15}N$ signature of in situ N_2O would depend on both the initial $\delta^{15}N(NO_3^-)$ and the extent of photolysis, and would therefore not be directly proportional to the $\delta^{15}N$ values of NO_3^- archived in the ice.

In contrast, our data show a direct proportionality between $\delta^{15}N(NO_3^-)$ and $\delta^{15}N(N_2O_{\text{in situ}})$, which suggests that the precursor of in situ N_2O forms deeper in the ice from archived NO_3^- that already carries its final, post-photolysis isotopic $\delta^{15}N$ signature, rather than from NO_2^- produced in the near-surface snowpack.

Changes in the text (in **bold**):

L506: "Source of central nitrogen atom N^{α} – In the case of in situ N_2O produced by N-nitrosation, the central nitrogen atom (N^{α}) may originate from NO_2^- after reduction of NO_3^-. However, it is unlikely that this NO_2^- derives directly from NO_3^- photolysis in the near-surface snowpack. NO_3^- photolysis produces both gaseous NO_2 and NO_2^- ion, with NO_2^- accounting for ~10% of the photolysis products (Meusinger et al., 2014; Warneck and Wurzinger, 1988). Photolysis occurs at the snow surface only, where sunlight penetrates and where air exchange with the atmosphere is still active. Any N_2O produced through photolysis-driven pathways in this zone would therefore be largely released to the atmosphere and not preserved in the ice core record. In addition, photolysis enriches the remaining NO_3^- in ^{15}N while producing NO_2^- that is depleted in ^{15}N, with its isotopic composition depending on the extent of photolysis. If photolysis-derived NO_2^- were a precursor of in situ N_2O, the $\delta^{15}N^{\alpha}$ signature of in situ N_2O would reflect both the initial $\delta^{15}N(NO_3^-)$ and the extent of photolysis, and would not be directly proportional to the $\delta^{15}N$ values of NO_3^- archived in the ice. Instead, our data show a strong proportionality between

$\delta^{15}N(NO_3^-)$ and $\delta^{15}N(N_2O_{in\ situ})$, indicating that the NO_2^- precursor must form deeper in the ice from archived NO_3^- that already carries its final, post-photolysis isotopic signature, rather than from photolysis-derived NO_2^- produced in the surface snowpack. Therefore, the reduction of NO_3^- to NO_2^- occurring within the ice requires a reducing agent. Such a reducing agent is likely associated with mineral dust, which is consistent with the observation that in situ N_2O production occurs only in dust-rich ice. Iron II (Fe^{2+}) is likely involved in this reduction step (see Sect. 5.2).”

L221 add basic details on the instruments used for N2O preparation and isotope measurements, also for SP analysis L227 and for NO3 analysis L305.

Changes in the text (in **bold**):

For N_2O bulk analysis:

L229: “The mixing ratio and bulk isotopic composition of N_2O were measured at the University of Bern, Switzerland, using the method described in detail in Schmitt et al. (2014). **Briefly, the air was extracted by melting the ice core samples with infrared light in a glass vessel under high vacuum. Water vapor was removed from the air sample with a cold trap, CO_2 was removed using Ascarite™, and N_2O, CH_4, and other trace gases were separated from the bulk air components (N_2, O_2, and Ar) using a cold trap filled with activated carbon. N_2O and CH_4 were then separated on a gas chromatography column, and N_2O was analyzed with an IsoPrime isotope ratio mass spectrometer (IRMS). The results were converted to international isotope scales.”**

For SP analysis:

L238: **“We analyzed the position-specific isotopic composition of N_2O from ice core samples at Oregon State University following gas extraction, N_2O purification, IRMS analysis, and data reduction methods described in detail in Menking et al. (2025). Briefly, the air was extracted by grating the ice core samples at -60°C to open the enclosed air bubbles (Bauska et al., 2016), i.e., using a so-called dry extraction device, without melting the ice. N_2O was purified and pre-concentrated using a series of progressively smaller-volume cold traps and gas chromatography separation of the trapped N_2O from residual CO_2. Isotopic measurements were performed using a Thermo Delta V Plus IRMS, where m/z 44, 45, and 46 and m/z 30 and 31 were monitored simultaneously for N_2O isotopes and N_2O fragment (NO) isotopes, respectively.”**

For NO_3^- analysis:

L324: “The method used for nitrate isotope analysis is described in detail in previous publications (Erbland et al., 2013; Kaiser et al., 2007; Morin et al., 2009). **Briefly, the NO_3^- concentration was measured by ion chromatography using a Dionex Integrion system; NO_3^-**

in the samples was preconcentrated using an AG 1-X8 anion exchange resin in the chloride form. After the sample was drained and the NO_3^- ions were quantitatively trapped onto the resin, NO_3^- was eluted from the resin with 6 mL of 1M NaCl solution in three portions of 2 mL. NO_3^- was then converted to N_2O through bacterial denitrification, using a strain of *Pseudomonas aureofaciens*. The bacteria were injected in 2 mL aliquots into 20 mL headspace vials. To remove air and dissolved N_2O, the vials were purged for 3 h with pure helium. The concentrated NO_3^- samples were added to the vials in volumes adjusted to obtain 100 nmol of NO_3^-, and were allowed to denitrify overnight. For isotope analysis, a continuous He flow was used to transfer the produced N_2O from the headspace vial and carry it through a purification line. The N_2O sample was passed through columns of perchlorate, Ascarite, and Supelco Purge Trap type F to remove water, CO_2, and VOCs, respectively. Following this purification step, the purified N_2O was decomposed to N_2 and O_2 on a gold catalyst kept at 850 °C, and the isotopic compositions of the obtained N_2 and O_2 were measured with a Thermo Fischer MAT 253 IRMS."

L310 Fig.3 You calculated and showed isotope values for N2O in situ as low as 20 ppb. These values are probably very unsure due to very small difference between atmospheric background N2O concentration and measured N2O concentration. Taking into account all the uncertainties associated with background N2O values (which are major, as you discuss a lot in the manuscript, which assumptions should be accepted for these!) and measurement errors, these values are associated with large uncertainties. For soil N2O emissions we accept at least 65ppb increase in N2O concentration to calculate the isotopic signatures for in-situ production (for lower increases the calculated propagated error was too large).

(e.g. Buchen et al.,2018, https://doi.org/10.1002/rcm.8132)

Please check such calculations of error propagation for your data and decide which minimal N2O in situ makes sense in your case. I expect not lower than 50 ppb. It is also visible on your graph 3B that the lowest in situ N2O concentrations are showing most extreme unrealistic isotope values.

Thank you for this thoughtful comment. We agree, and show in our manuscript, that the lowest in situ N_2O concentrations carry larger uncertainties. As noted in the manuscript (L263), we already quantified these uncertainties using a Monte Carlo simulation for error propagation. While we did not show the uncertainties in Fig. 3 for readability, they are shown in Figs. 4-7. They are also incorporated into all regression analyses comparing in situ N_2O with NO_3^-, including calculation of the slope and its uncertainty, using the regression method by York et al. (2004).

In our study, the aim is not to determine highly precise isotopic signatures for individual samples with low in situ production, but rather to capture a broad range of environmental conditions and nitrate isotopic compositions to evaluate the resulting patterns in in situ N_2O production. For this purpose, keeping a large number of data points across multiple ice cores, even with uncertainties of a few tens of per mil, provides valuable information on the potential production mechanisms. For our analysis, 20 ppb was therefore chosen as a threshold, which is lower than for typical analyses of modern N_2O isotope studies, because many samples in our dataset fall below 50 ppb. Using a higher cutoff would therefore remove a substantial part of the data and limit our ability to investigate the production processes. This does result in larger uncertainties for the small enhancements, but as mentioned above we do propagate all these uncertainties into our final results.

Changes in the text (in **bold**):

L300: "3.2.4 Uncertainties – There are several sources of error in our approach, including measurement uncertainties in N_2O mixing ratios and isotopic composition, uncertainties in the TALDICE N_2O spline interpolation, uncertainties in gas-age estimates – which in turn impact the chronological alignment between ice cores **– and potential small in situ production in TALDICE.** We used a Monte Carlo method for error propagation by running 1000 simulations to determine the mixing ratio and isotopic composition of in situ N_2O. We varied the variables of Eq. (3) and (4) within their uncertainty ranges (Table B1 in Appendix B). The derived uncertainties were calculated as the standard deviation of the results of the 1000 simulations.

As expected, the uncertainty increases for low in situ N_2O mixing ratios (see Eq. 4). To avoid too large uncertainties, we excluded samples with calculated in situ N_2O mixing ratios below 20 ppb. This threshold was chosen because a large part of the dataset exhibit in situ mixing ratios below 50 ppb; applying a higher cutoff would remove a substantial number of samples and significantly limit our ability to investigate in situ N_2O production processes. For samples with in situ N_2O mixing ratios close to 20 ppb, the propagated uncertainties are large (up to several tens of per mil). Nevertheless, these samples were included in this study because in situ N_2O production in ice, although small in absolute mixing ratio, can significantly alter the measured N_2O isotopic composition (Fig. 3a) and these isotopic deviations should be documented."

L374: "**We stress that the mass balance approach used to calculate the isotopic signature of in situ N_2O leads to substantial uncertainties due to error propagation from atmospheric N_2O estimates and analytical precision. These uncertainties are not shown in Fig. 3 for readability but are reported in Figs. 4 – 7 and are explicitly taken into account in the following comparisons between $\delta^{15}N(N_2O_{in\ situ})$ and $\delta^{15}N(NO_3^-)$, including the regression analyses**."

L344 "N2O analyzed in the extracted air" or the calculated in situ N2O isotopic signature? – Fig. 4 shows "N2O in situ" – ok – but be precise in distinguishing these terms in the text

Thank you for pointing out the ambiguity. The original wording was indeed confusing. We have revised the text to clearly distinguish between the N_2O measured in the extracted air and the calculated in situ N_2O isotopic signature.

Changes in the text (in bold):

L379: “In this section, we compare the **measured** isotopic composition of NO_3^- and the **calculated isotopic composition of** in situ N_2O to test our hypothesis that NO_3^- is a precursor for in situ produced N_2O. **The analyses come from the same ice samples: N_2O was measured in the extracted air and subsequently used to calculate the in situ N_2O isotopic signature, while NO_3^- was measured in the sample meltwater collected after air extraction**.”

L375, Fig 6b and 6d clearly supports my assumption that for low in-situ N2O you get biased results, your SP values below 0 for very low N2O amount of 20ppm are just calculation artefacts

Thank you for this comment. We understand the concern regarding the larger uncertainties at low in situ N_2O concentrations. However, we respectfully disagree that the low SP values in the Taylor Glacier samples are calculation artefacts. Although the uncertainties are indeed substantial – and we explicitly quantify them – the SP values for Taylor Glacier remain below zero within these uncertainties.

As a simple sanity check, which demonstrates that this is a real signal, low SP values are also visible directly in the measured (total) SP data (Fig. 6c), independent of any mass-balance calculation. While Vostok samples affected by in situ production show total SP values higher than the atmospheric signature, Taylor Glacier samples show the opposite pattern, with total SP values lower than the atmospheric signature. This strongly suggests that the in situ N_2O at Taylor Glacier has indeed low SP values.

For this reason, we believe it is valuable to keep these data, as they highlight a difference between the two ice core sites and point to different isotopic signatures of the N_2O precursors at Vostok and Taylor Glacier.

Changes in the text (in **bold**):

L434: **“Interestingly, the SP($N_2O_{\text{in situ}}$) values at TG are negative. Although the low amounts of in situ N_2O at TG result in large uncertainties in the calculated in situ signatures, the low SP signal is already visible in the measured (total) N_2O data (Fig. 6c), independent of any mass-balance calculation. While Vostok samples affected by in situ production show total SP values higher than the atmospheric signature, TG samples show the opposite pattern, with total SP values lower than the atmospheric signature. This opposite deviation indicates that the low SP values observed at TG reflect a real feature of in situ N_2O production rather than an artefact of the calculation.”**

L385-387 SP values are independent of bulk 15N for microbial N2O production processes, but such relation is typical for N2O reduction where preferentially N-O bonds of light isotopes are broken which results in enrichment in bulk 15N and on alpha position (=increase in SP). The reported slope (Fig. 6d) is within the typical slopes observed for N2O reduction (see Yu et al., 2020, Lewicka-Szczebak et al., 2020).

L391, Fig. 7a – OK – this Figure clearly indicates that N2O reduction plays no role, because due the partial N2O reduction the alpha position would be strongly affected and the slope would be much higher than 1. But I think it would be worth to add a short discussion on N2O reduction in the manuscript, you may indicate that you disregarded this process based on this strong relationship, Fig 7a.

(Comments on L385-387 and L391) Thank you for this very helpful comment. We agree that N_2O reduction should be addressed explicitly, and we have added a short discussion in the revised manuscript. This new section incorporates your argument based on the $\delta^{15}N_{\alpha}(N_2O_{in\ situ})$-$\delta^{15}N(NO_3^-)$ relationship (Fig. 7a), as well as Reinhard Well's comment regarding the SP-$\delta^{18}O$ relationship. These two lines of evidence both show that our data are inconsistent with N_2O reduction. We also added the suggested SP-$\delta^{18}O$ plot to support this conclusion. The revised paragraph is provided below.

Changes in the text:

L465: **"To assess whether microbial N_2O reduction to N_2 could provide an alternative explanation for the observed isotopic patterns of in situ N_2O – such as high SP values correlated with $\delta^{15}N_{bulk}$ or high $\delta^{18}O$ values – we examined the relationship between SP and $\delta^{18}O$ in our samples (Fig. 8). Previous studies showed that N_2O reduction produces a characteristic increase in both SP and $\delta^{18}O$ in the residual N_2O, with data falling along a "reduction line" defined by the ratio of the fractionation factors (median slope ≈ 0.36; Lewicka-Szczebak et al., 2015; Yu et al., 2020). In contrast, in both the Vostok and Taylor Glacier ice cores, SP increases while $\delta^{18}O$ decreases, and the data clearly do not fall along the expected reduction line (Fig. 8). This dual isotope plot for in situ N_2O is therefore incompatible with N_2O reduction. We note that published reduction lines were derived from studies conducted at warmer temperatures than those in ice cores (Yu et al., 2020). Very low temperatures could modify the fractionation factors; however, colder conditions would be expected to increase the fractionation for both SP and $\delta^{18}O$, still resulting in a positive slope. The mismatch between our data and the reduction line therefore remains regardless of potential temperature effects. A second line of evidence comes from the correlation between $\delta^{15}N(NO_3^-)$ and $\delta^{15}N_{\alpha}(N_2O_{in\ situ})$, which shows a slope of ~1. Because N_2O reduction preferentially breaks N-O bonds of light isotopes, the residual N_2O becomes enriched in ^{15}N at the α position, resulting in a steeper slope than observed here. Taken together, these observations show that microbial N_2O reduction cannot explain the isotopic**

signatures in the ice cores, and that N_2O consumption does not occur in the ice. The isotopic signature of in situ N_2O is therefore explained by hybrid N_2O production.”

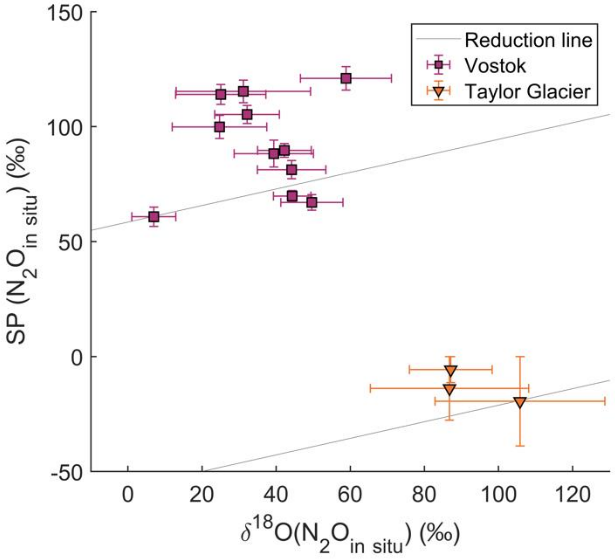


Figure 8. Dual isotope plot of $\delta^{18}O$ and site preference (SP) values of in situ N_2O. Theoretical N_2O reduction lines (thin black lines) are shown for comparison, with intercepts calculated assuming that samples with the lowest SP values are unaffected by N_2O reduction. The slope of the reduction line corresponds to the median of published fractionation ratios for SP versus $\delta^{18}O$ during N_2O reduction, as reported by Yu et al. (2020).

L395 ‘This correlation is robust when excluding the TG data.’ – because TG data are calculated for very low N2O concentration and are simply biased. I believe they should be excluded from any further interpretations.

Thank you for this comment. We agree that our original wording was unclear. The linear regression in Fig. 7 was calculated using both the Vostok and Taylor Glacier data, and a strong correlation is obtained when all data points are included. What we intended to convey is that this correlation also holds when the Taylor Glacier data are excluded: the Vostok data alone still show a significant positive relationship between $\delta^{15}N^{\alpha}(N_2O_{\text{in situ}})$ and $\delta^{15}N(NO_3^-)$, even though the isotopic range within a single core is smaller.

We have revised the text to make this point clearer: L445 **“In Fig. 7, the $\delta^{15}N^{\alpha}$ and $\delta^{15}N^{\beta}$ values of in situ N_2O are compared with $\delta^{15}N$ values of NO_3^- measured in the TG and Vostok ice cores. When considering both TG and Vostok samples, $\delta^{15}N^{\alpha}(N_2O_{\text{in situ}})$ shows a strong positive correlation with $\delta^{15}N(NO_3^-)$ (slope = 1.0 ± 0.1, $R^2 = 0.97$). This relationship also holds when considering the Vostok data alone, indicating that the correlation is robust even**

within a single ice core. In contrast, $\delta^{15}N^{\beta}(N_2O_{\text{in situ}})$ does not show a statistically significant correlation with $\delta^{15}N(NO_3^-)$ for either site".

L430 The d18O values are not really explained, for EDML exchange with water could explain the observed low d18O-N2O values, but for Taylor Glacier the very high d18O-N2O values, largely exceeding the d18O-NO3 values are not plausible – again in my opinion these values are biased and should be excluded from any further interpretations.

Thank you for this comment. We see two separate issues raised here:

(1) the accuracy of the mass-balance calculation for Taylor Glacier, given the low in situ N_2O concentrations and the resulting larger uncertainties. Large uncertainties do not imply a systematic bias toward high $\delta^{18}O$ values, but rather increased scatter around the true value.

(2) whether the measured N_2O at Taylor Glacier truly shows elevated $\delta^{18}O$ values relative to atmospheric N_2O.

Without applying the mass-balance calculation, the measured total N_2O at Taylor Glacier (Fig. 3a) already shows $\delta^{18}O$ values higher than the atmospheric signature in samples affected by in situ production. This indicates that the in situ N_2O at Taylor Glacier is indeed enriched in $\delta^{18}O$. A similar pattern is observed in the NGRIP ice core. Importantly, these elevated $\delta^{18}O$ values were measured using two different extraction methods and two different IRMS instruments at the University of Bern and Oregon State University, which strengthens our confidence that the signal is real and not an artefact of a single analytical setup.

Excluding these data because the $\delta^{18}O$ values are high would therefore require dismissing the measured values themselves, not just the mass balance results. We believe it is important to keep these data points, as they reveal meaningful differences among the ice cores.

Changes in the text:

L401: **"The very high $\delta^{18}O(N_2O_{\text{in situ}})$ values observed at TG may appear anomalous compared to other sites, but we are confident that they do not result from a bias in the measurements or calculation. Even without applying the mass balance calculation, the measured N_2O at TG already shows $\delta^{18}O$ values higher than the atmospheric signature in samples affected by in situ production (Fig. 3a). This indicates that the in situ N_2O at TG is indeed enriched in $\delta^{18}O$. A similar enrichment is observed in the NGRIP ice core. Importantly, these elevated $\delta^{18}O$ values were measured using two different extraction methods and two different IRMS instruments at the University of Bern and Oregon State University, which strengthens our confidence that the signal is robust and not an artefact of a specific analytical setup."**

L460, Table2: I do not see the point of referring here all the isotope effects for the processes which you do not deal here with (single precursor N2O) after previous review papers, I would only give the data for hybrid N2O.

L465 you mean Table 2 here, I think your description of values for the different pathways in text is enough with respective citations, as suggested above I would shorten the table 2 only to values directly found in your study

(Comments on L460 and L465) We agree that the original version of Table 2 included information not directly relevant to our analysis. Following your recommendation, we have shortened Table 2 to include only the information for hybrid N2O. We have also ensured that the text refers specifically to the revised table.

L467-468 – a citation for this theory of the common intermediate and the mechanism of N2O formation is missing

We have added the appropriate citations: (Heil et al., 2015; Toyoda et al., 2002, 2005, 2017)

L470 – as above – a citation for denitrification N2O formation mechanism is missing

We have added the appropriate citations: (Fehling, 2012; Toyoda et al., 2017)

L492 – citation needed

We have added the appropriate citations: (Frame et al., 2017; Spott et al., 2011; Stieglmeier et al., 2014; Terada et al., 2017)

L505 – very good and helpful Figure, your explanation and visualisation of the idea of asymmetric intermediate in hybrid N2O production is very convincing

Thank you for this positive feedback. We are pleased that the figure and its explanation were found helpful and convincing.

L542 – I agree that pH probably has the large impact on the extend of O-exchange, do you have data of pH for your samples? If we deal with very high pH it could have completely blocked the O-exchange and would support your theory. The lack of pH values is an important drawback, should be added if available.

Otherwise, without the known differences in pH, I do not believe that the mechanism could be so different to give totally opposite effects, from strongly negative to extremely positive. The story of O-exchange between nitrite and water is very convincing and the reported values are very plausible. However, the very high values in TK are suspicious, even with maximal branching effect they are a bit too high. Importantly, these values are based on 3 points calculated with in-situ N2O production of 20ppb, which is in my opinion to low to calculate representative and true isotope values. I strongly encourage you to critically evaluate this data. I would reduce the calculated in-situ N2O to the higher production only and present and further discuss only the proper values, not associated with possibly large errors. This would vastly improve your manuscript because you wouldn't need to find theories for the values which are very unsure.

Unfortunately, we do not have pH data for these specific samples. Glacial Antarctic ice is generally acidic, but without direct measurements we cannot assess potential pH differences between sites. Moreover, even if pH were measured in the liquid phase (meltwater), it would be difficult to directly infer the pH of the solid ice matrix, as pH is formally defined only for liquid solutions. In addition, the relevant reactions likely occur at the surfaces of dust particles (where the reduction of NO_3^- to NO_2^- may take place) rather than in the bulk ice, and the chemical conditions at these micro-environments may differ from those of the surrounding ice.

We agree that, as you point out, we currently lack sufficient information to draw firm conclusions about the mechanism responsible for the $\delta^{18}O(N_2O_{\text{in situ}})$ values. We will revise Section 5.4 to make it clear that we are listing possible mechanisms that could explain why the $\delta^{18}O(NO_3^-)$ signature is not transferred to in situ N_2O, but that the mechanism at play at Taylor Glacier (and in other ice cores) remains uncertain.

As noted in our responses to other comments, questioning the high $\delta^{18}O$ values at Taylor Glacier would imply questioning the measured total N_2O data themselves, not only the mass balance calculations. We therefore believe it is more appropriate to keep the TG data and adopt a cautious interpretation, acknowledging that the mechanism is not yet fully understood, rather than excluding data that do not fit the simplest explanation. This approach allows us to provide a more complete picture of in situ N_2O production across the different ice cores. We provide the uncertainty of the derived in situ signals in all figures to corroborate our conclusions.

Changes in the text:

L651: **"However, measurement constraints of pH remain limited. Although glacial Antarctic ice is generally acidic and Greenland ice is typically more alkaline, pH measurements are not available for the specific samples investigated here, making it difficult to interpret the differences in $\delta^{18}O(N_2O_{\text{in situ}})$ among Antarctic sites in terms of pH effects. Moreover, even if pH were measured in the liquid phase (meltwater), it would be difficult to directly infer the pH of the solid ice matrix, as pH is formally defined only for liquid solutions. In addition, the relevant reactions likely occur at the surfaces of dust particles (where the reduction of NO_3^- to NO_2^- may take place) rather than in the bulk ice,**

and the chemical conditions at these micro-environments may differ from those of the surrounding ice."

Referee comment #2

This manuscript investigated the origin and isotopic compositions of in-situ N2O in Antarctic glacial ice using site-specific 15N isotopomer analysis, combined with d18O of N2O and nitrate. The authors proposed a novel hypothesis of an asymmetric hybrid intermediate mechanism that forms N2O, with α-N derived from nitrate/nitrite and β-N from another unidentified N species likely under low-pH, Fe2+ available conditions.

The manuscript addresses an important topic that may have significant implications for the Earth's nitrogen cycle, isotope geochemistry, and paleoclimatology. It is well written, logically structured, and generally easy to follow. Although some figure legends are a bit confusing (see minor comments below), the figures clearly present the data. I have one major comment and a few minor suggestions.

We thank you for the careful and constructive review of our manuscript and for the positive assessment of its scientific relevance, clarity, and structure. Below, we address the major comment and the minor suggestions in detail.

Major comment:

I believe that under very low temperature conditions such as those in this study; all reactions occur extremely slowly, which could significantly affect isotopic fractionation. The authors cited several papers to compare SP values (Table 2); however, these references are all based on findings under ambient temperature and pressure conditions. Therefore, I question whether such comparisons are valid for this system.

I recommend including studies that are performed under more relevant temperature conditions, if available. If not, provide a brief theoretical discussion to justify the comparison between the SP values obtained in this study (low temperature, extremely slow reactions) and those of the cited studies (ambient temperature).

We thank the reviewer for this important comment. We agree that very low temperatures imply extremely slow reaction rates, which can significantly influence isotopic fractionation and complicate direct comparison with published SP values measured at ambient temperature conditions. To our knowledge, no SP measurements exist under temperature conditions relevant for Antarctic ice. We have therefore added a brief theoretical discussion in the revised manuscript explaining why our comparison focuses on the constancy or variability of SP with respect to the precursor $\delta^{15}N$ signature rather than on absolute SP values, and why this comparison remains valid despite the temperature difference.

Changes in the text (in **bold**):

L581: **"We note that all published SP datasets used for comparison were obtained at ambient temperatures, whereas in situ N_2O production in Antarctic ice occurs at very low temperatures. Such low temperatures imply extremely slow reaction rates, which may alter the magnitude of isotope fractionation and make direct comparison of absolute SP values with ambient-temperature experiments difficult. To our knowledge, no SP measurements exist under such cold conditions. However, our interpretation does not rely on comparing absolute SP values. Instead, we focus on whether SP is constant or variable with respect to the $\delta^{15}N$ signature of the precursor; this property should be independent of the absolute magnitude of isotope fractionation. Although lower temperatures may increase kinetic isotope effects and shift absolute SP values, they do not change whether SP remains constant (as in reactions involving symmetrical intermediates) or varies with the precursor isotopic composition (as expected when an asymmetrical intermediate forms). Thus, the comparison of the mechanisms remains valid even without low-temperature experimental data from previous studies.**

Several mechanisms could potentially explain why the intermediate of in situ N_2O production is asymmetric, even though its exact chemical structure remains unknown. Firstly, the precursor of the β-position N atom could be different from NH_2OH. Although most studies on hybrid N_2O production report a reaction between NH_2OH and NO_2^- (Frame et al., 2017; Spott et al., 2011; Stieglmeier et al., 2014; Terada et al., 2017), other nucleophilic precursors have been reported as precursors of hybrid N_2O. Hydrazine (N_2H_4), for example, forms the asymmetrical intermediate $HO\text{-}N{=}N\text{-}NH_2$ (Perron et al., 1976). **A second possibility is that very low temperatures modify the structure or stability of the intermediate normally formed from NH_2OH and NO_2^- at ambient temperature conditions, favoring an asymmetrical species and thereby generating the observed dependence of SP on the precursor $\delta^{15}N$ values."**

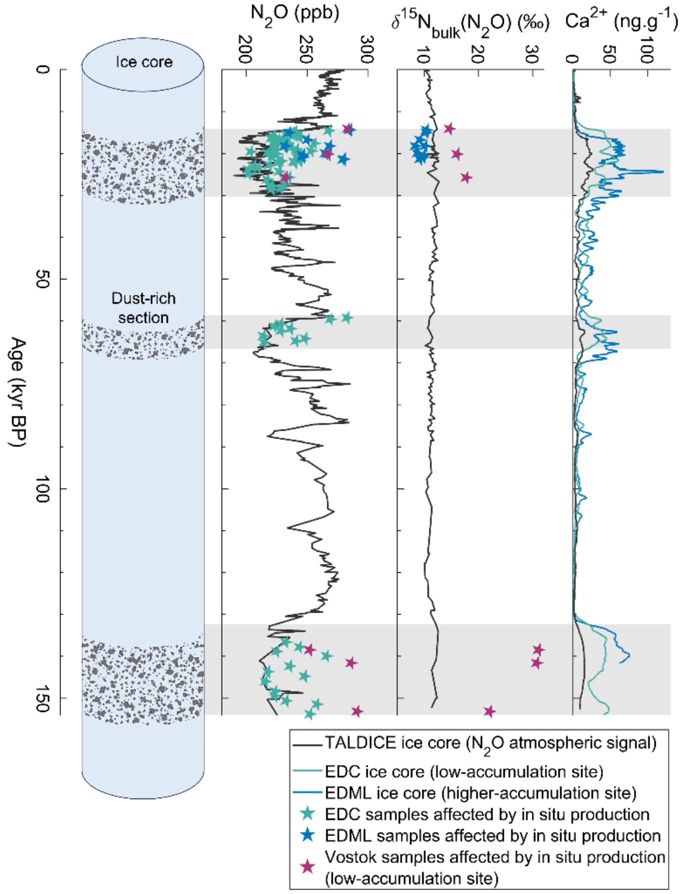
Minor comments:

Figure 1. TALDICE ice core values are shown as a solid black line in the figure. So, in the legend, change the black dot to a solid black line.

And are there samples unaffected by in-situ production shown as well? It is difficult to distinguish them. The figure is a bit confusing because EDC, Vostok and EDML are shown as colored dots while the samples affected by in-situ production are marked as stars. If the figure only displays the affected samples, consider using star markers for all three ice cores and deleting the last line in the legend.

We have updated the figure legend to use a solid black line for the TALDICE ice core, as recommended. The figure only displays samples affected by in situ N_2O production (with the exception of the TALDICE record), and these samples are shown using star markers. We have now adjusted the legend to make this clearer and to avoid any confusion between dots and stars.

Changes in the figure:



Line 71-75: Since SP is a key element of this manuscript, the introduction should provide more background on SP analysis and explain what information it conveys.

Changes in the text:

L71: **"Beyond bulk isotope analyses (δ^{15}N and δ^{18}O), position-specific measurements provide even more detailed insights into N_2O production mechanisms. Because N_2O is an asymmetric molecule, the nitrogen isotopic composition of the two N atoms can deviate from each other, and this difference can be measured separately at the central N atom (N^{α},**

bonded to oxygen) and the terminal N atom (N^β). From these values, the site preference (SP = $\delta^{15}N^\alpha - \delta^{15}N^\beta$) can be calculated. Because SP reflects intramolecular isotope partitioning during N-N bond formation, it is primarily controlled by the reaction mechanism and the structure of the last intermediate rather than by the isotopic composition of the precursor (Frame and Casciotti, 2010; Sutka et al., 2003, 2006; Toyoda et al., 2005). As a result, SP values are often characteristic of specific N_2O formation pathways and can remain constant even when precursor $\delta^{15}N$ values vary widely. In contrast to bulk $\delta^{15}N$, which integrates source and fractionation effects, SP provides mechanistic information on how N_2O is formed and is therefore widely used to discriminate between N_2O production pathways; SP values are typically negative for bacterial denitrification and positive for nitrification (Toyoda et al., 2017). Using this tool, Prokopiou et al. (2018) showed that SP values increased since preindustrial times, pointing to a relative shift from denitrification to nitrification, consistent with agricultural emissions playing a major role in the N_2O increase. Similarly, Menking et al. (2025) demonstrated that the increase in N_2O concentrations during the transition from the Last Glacial Maximum to the Holocene reflected contributions from both nitrification and denitrification, whereas the N_2O decrease during the Younger Dryas was driven by reduced nitrification.”

Line 106: The main hypothesis should be stated more explicitly.

Thank you for this suggestion. We have revised the last paragraph of the introduction:

L115: “This study uses isotope analysis to characterize in situ N_2O in **various** ice **cores**. **The background of the study, the extreme environmental conditions in the polar environment, and the potential consequences for the reactions involved are presented in Sect. 2. Based on the strong enrichment in ^{15}N observed in some samples affected by in situ N_2O production (Fig. 1), we hypothesize that NO_3^-, which can also be highly enriched in ^{15}N in ice, is one of the nitrogen precursors for in situ N_2O. To test this hypothesis, we measured the isotopic composition of N_2O and NO_3^- in the same ice core samples and calculated the isotopic signature of in situ N_2O (Sects. 3 and 4). Position-specific nitrogen isotope analysis of N_2O was carried out to further constrain the reaction pathway(s) involved. The potential mechanisms for in situ N_2O production are discussed in Sect. 5.”**

Line 166: Ca2+ concentrations are cited from different studies. Were they measured using the same analytical methods? Also isn’t there existing data on Fe content? If so, it would be valuable to include them in here.

Yes, the Ca^{2+} concentrations cited for the different ice cores were all measured using the same analytical technique (continuous flow analysis; Röthlisberger et al., 2000). We have clarified this explicitly in Table 1.

Regarding Fe content, existing Fe^{2+} data are indeed available for the EDC and TALDICE ice cores (Spolaor et al., 2013; Traversi et al., 2004). We have incorporated these values into Table 1 in the revised manuscript.

Line 252: Write Fig. A1 “in Appendix A”. Similarly, for the rest of the manuscript, specify when figures or tables are located in the appendix.

We have changed this as proposed.

Line 283: should this refer to Section 3.2.2?

Thank you for pointing this out. Yes, the reference should be to Section 3.2.2. We have corrected this in the revised manuscript.

Figure 3b. Include a gray cross in the legend.

We have changed this as proposed.

Line 465: Add the references for the “several studies” mentioned in here.

We have added the appropriate citations: (Heil et al., 2015; Toyoda et al., 2002, 2005, 2017).

References

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