

Reply on RC3:

General comments:

The paper is concerned with a problem which is certainly of environmental interest, but which nevertheless touches problems in concentrated solutions, which are beyond the reach of classical thermodynamic models. Under conditions of such low solvent availability, the activity coefficients can explode to values >1000, making the reactivity analysis very complex. I am personally in favor of making experiments other than the classic ones, such as in this paper. However, precisely because of this, interpretation can be difficult.

Authors' Response: Thank you so much for your constructive comments. The reactivity analysis in concentrated systems is complex, and our experimental results can provide valuable data to parameterize the complex thermodynamics in future studies. We also agreed with the reviewer that the interpretation of our findings should be further improved. Kindly please find our responses to the comments accordingly below:

Amino acids in real conditions (in aqueous solutions and in crystalline form) are separate charge systems ($\text{NH}_3^+\text{-CH}_2\text{-COO}^-$). Several possible zwitterionic conformers of glycine have been calculated with the addition of 1-3 water molecules [Opt. Spectrosc. 128, 1598–1601 (2020). <https://doi.org/10.1134/S0030400X20100161>]. In both low water content and crystalline form GLY is ZW ($\text{NH}_3^+\text{-CH}_2\text{-COO}^-$), as in water, always (and pH only changes the fractional amount of charges from positive to negative). In several parts of the article GLY is referred to as neutral/non-ionized/less zwitterionic. This is a confusion, because the form $\text{NH}_2\text{-CH}_2\text{-COOH}$ does not exist. Thus, the paper's key conclusion given on line 220 is inconsistent. Also, on the same lines, it is stated that "The -NH_2 was unprotonated...and more susceptible to oxidation". This is only possible at pH above 10, so the observed reactivity must have other explanations. In such a concentrated solution the pH is also difficult to define, and no idea of the actual protonation constants is reported. Thus, if it is true that the reactivity of OH is greater for anionic GLY, the "formal" pH would be basic. Are there any hypotheses about it?

Authors' Response: Aerosol pH was defined as the activity of hydrogen ions (H^+) in aqueous aerosol particles (Buck et al., 2002; Li et al., 2022; Peng et al., 2019). The pHs of the mixed GC nitrate particles at 80% RH were around 6 according to the pH paper measurements (Craig et al., 2018), at which zwitterionic glycine is the most abundant form. Without any information of possible unknown molecular interactions that affect the protonation equilibrium (i.e., protonation constants), we agree with the reviewer that glycine is mostly in the zwitterionic form and have removed the terminology of neutral and non-ionized to avoid confusion. We also added further information on zwitterion as the reviewer suggested.

Line 221-224: For instance, glycine can be ionized into different forms according to the local conditions, including cationic, zwitterionic, and anionic of different reactivities (Aikens et al., 2006). Zwitterionic denotes the charge-separated form amino acids in aqueous solutions and in crystalline states (e.g., $\text{NH}_3^+\text{-CH}_2\text{-COO}^-$). Several possible

zwitterionic conformers of glycine have been proposed with the addition of 1-3 water molecules (Krauklis et al., 2020).

As shown in Figure 4b, the strong linear correlation between nitrate photolysis rate constant and percentage glycine decay suggests that the efficiency of nitrate photolysis is the main factor for the glycine decay, which is modulated by the interaction between nitrate and glycine. We mentioned the protonation and configuration of glycine as a possible minor factor that could affect the photochemistry, while also acknowledging the difficulty of determining the protonation constants of glycine in concentrated solutions, as pointed out by the reviewer. Therefore, we have added this information to the revised manuscript with more explanation.

Line 216-229: The apparent nitrate photolysis rate constant J shows good correlation with the percentage GC decay ($R^2 = 0.99$, Figure 4b), which suggests that nitrate photolysis is the key driver for the glycine decay.

The different reactivity of glycine between SN+GC and AN+GC particles may also minorly contribute to the distinct photochemistry. For instance, glycine can be ionized into different forms according to the local conditions, including cationic, zwitterionic, and anionic of different reactivities (Aikens et al., 2006). Zwitterionic denotes the charge-separated form amino acids in aqueous solutions and in crystalline states (e.g., $\text{NH}_3^+-\text{CH}_2-\text{COO}^-$). Several possible zwitterionic conformers of glycine have been proposed with the addition of 1-3 water molecules (Krauklis et al., 2020). The rate of anionic glycine reacting with OH radicals is 2-orders of magnitude higher than that of zwitterionic glycine (Berger et al., 1999; Buxton et al., 1988), while that of zwitterionic glycine is several times higher than cationic glycine. These differences were due to the increased energy barriers for oxidation upon protonation (Wen et al., 2022). However, the protonation constants of glycine in concentrated solution were difficult to define. Qualitatively, a possible lower degree of glycine protonation in SN+GC particles than AN+GC particles might enhance the reactivity of glycine.

From the GLY ZW nature it follows that the addition of SN or AN could change:

1) The availability of free water, as part of it can be solvated by sodium or ammonium cations. This was deduced at line 219, as water to GLY ratio of 6 for AN and 2 for SN. Although it is reasonable as sodium is solvated more than ammonium, these ratios must be further supported by the authors.

2) the configuration of the GLY dimer or trimer as some possible complexation or charge interaction is possible both with the anion carboxylate, and the cationic protonated $-\text{NH}_3^+$ group. This last would change the photoreactivity due to the nitrate, the only absorbing species in the system (the absorption of GLY is below 260 nm, and not involved in the experiments). In Figure 3a the complexation of sodium is depicted, leaving free nitrate supposed to form reactive species. But also a weaker bond with ammonium would lead to the same configuration of free nitrate. Then, what is the ultimate explanation? Overall the paper need a strong revision.

Authors' Response: We observed that the nitrate photolysis rate constant was linearly correlated with the percentage glycine decay, indicating that the efficiency of nitrate photolysis was a key factor for glycine degradation. Moreover, the nitrate photolysis rate constant was significantly larger in SN+GC particles than in AN+GC particles, attributable to the different molecular interactions. Based on the literature, we illustrated the possible differences of interactions of nitrate salts and glycine between AN+GC and SN+GC particles. If the bonds between ammonium or GC and nitrate in AN+GC particles are weaker than those between Na^+ or GC and nitrate in SN+GC particles, there will be more free nitrates in AN+GC particles for photolysis than SN+GC particles. However, our Raman data show only one single symmetric nitrate peak in AN+GC particles, which was likely from inactive bonded nitrate, given the small nitrate photolysis rate constant and minor glycine decay. Therefore, the bonds between ammonium or GC with nitrate in AN+GC particulate matrix were likely stronger than that of sodium or GC with nitrate in SN+GC particles. This may explain the suppressed nitrate photolysis in AN+GC particles, compared with SN+GC particles.

The interaction-dependent nitrate photolysis was likely the key driver for glycine decay, but the exact molecular configurations of such interactions still require further work. We also agree with the reviewer that the availability of free water and the configuration of the glycine dimer or trimer may change the photo-reactivity of glycine under nitrate photochemistry. However, we hesitate to speculate too much on the exact molecular configuration since the detailed investigations of quantum chemistry and molecular dynamic simulation with appropriate parameterization for non-ideal solutions is beyond the focus of this paper. We view that such work is better suited for a physical chemistry journal than ACP. We hope that our experimental observations can stimulate researchers to look into the complex interactions described above and outlined by the reviewer.

Line 197-235: For instance, amino acid nitrate can form hydrogen bonding between nitrate from AN and the protonated amino group of glycine (Figure 3a) (Wang et al., 2022; Ashraf et al., 2021). As a result, the amino acids and nitrate ions in the droplet are bounded in an extensive three-dimensional hydrogen-bonded matrix (Wang et al., 2022), in which nitrate photolysis could be hindered (Vimalan et al., 2010). On the other hand, the COO^- of glycine can bind with SN via Na^+ directly to form a bidentate complex (Figure 3a) (Moision et al., 2002; Aziz et al., 2008; Selvarani et al., 2022), leaving nitrate unbonded. Nitrate in SN+GC particles has two Raman peaks (Figure 3b). One had the same Raman shift as nitrate in AN+GC, likely bonded nitrate, while the other peak at 1046 cm^{-1} was attributable to unbonded aqueous nitrate (Liang et al., 2022), which can undergo photolysis to form a wealth of oxidants that lead to glycine decay (Figure 3a). **The single symmetric nitrate peak, small J, and minor glycine decay of AN+GC particles suggested a negligible fraction of unbonded nitrate. However, we also note that the exact molecular configuration in concentrated particles can be much more complicated than the illustrative example shown in Figure 3a. Detailed investigations of quantum chemistry and molecular dynamic simulation with appropriate parameterization for non-ideal solutions are required.**

One would expect that the molecular interactions are more evident in droplets at lower RH, with higher solute concentrations and fewer water molecules. Figure 4a shows the percentage GC decay after irradiation as a function of the initial solute concentrations. At

0.01M, the percentage GC decay is approximately 5% in both AN+GC and SN+GC solutions. However, as the initial solute concentration increased from 0.01 M to 7.6 M, the percentage GC decay in SN+GC particles increased by more than one order of magnitude to 70%, while that of the AN+GC particles remained small. The apparent nitrate photolysis rate constant J shows a good correlation with the percentage GC decay ($R^2 = 0.99$, Figure 4b), which suggests that nitrate photolysis is the key driver for the glycine decay.

The different reactivity of glycine between SN+GC and AN+GC particles may also contribute to the distinct photochemistry. For instance, glycine can be ionized into different forms according to the local conditions, including cationic, zwitterionic, and anionic of different reactivities (Aikens et al., 2006). Zwitterionic denotes the charge-separated form amino acids in aqueous solutions and in crystalline states (e.g., $\text{NH}_3^+ - \text{CH}_2 - \text{COO}^-$). Several possible zwitterionic conformers of glycine have been proposed with the addition of 1-3 water molecules (Krauklis et al., 2020). The rate of anionic glycine reacting with OH radicals is 2-orders of magnitude higher than that of zwitterionic glycine (Berger et al., 1999; Buxton et al., 1988), while that of zwitterionic glycine is several times higher than cationic glycine. These differences were due to the increased energy barriers for oxidation upon protonation (Wen et al., 2022). However, the protonation constants of glycine in concentrated solution were difficult to define. Qualitatively, a possible lower degree of glycine protonation in SN+GC particles than AN+GC particles might enhance the reactivity of glycine.

We also note that the initial water-to-glycine mole ratios were higher for AN+GC particles (6) than SN+GC particles (2), and sodium has higher hydration number (6) (Medoš et al., 2019) than ammonium (4) (Guo et al., 2020). Therefore, the availability of free water in AN+GC particles is likely higher than in SN+GC particles. This could affect the configuration of glycine dimers or trimers, such as the possible complexation or charge interactions between the anionic carboxylate and the cationic $-\text{NH}_3^+$ groups. These factors could also modulate the photo-reactivity of glycine.

Line 273-280: Overall, our work sheds light on the potential role of particulate nitrate photolysis in the sink of the atmospheric FAAs, which impacts the cycling of atmospheric organic nitrogen. The reaction rate constants between FAAs and different oxidants from nitrate photolysis can further help quantify the contribution of nitrate photolysis in FAA degradation and improve the prediction of the atmospheric lifetime of FAAs. The reactivity analysis in concentrated systems is complex, and our experimental results can provide valuable data to parameterize the complex thermodynamics in future studies. Systematic studies of the detailed molecular mechanism and the factors influencing nitrate photochemistry and FAA decay, such as molecular configuration, alkalinity, and solvation are recommended. Quantum chemical and molecular dynamic simulations with appropriate parameters would be useful tools for this purpose.

Reference

Aikens, C. M. and Gordon, M. S.: Incremental Solvation of Nonionized and Zwitterionic Glycine, *Journal of the American Chemical Society*, 128, 12835-12850, 10.1021/ja062842p, 2006.

Ashraf, H., Guo, Y., Wang, N., Pang, S., and Zhang, Y.-H.: Hygroscopicity of Hofmeister Salts and Glycine Aerosols–Salt Specific Interactions, *The Journal of Physical Chemistry A*, 125, 1589-1597, 10.1021/acs.jpca.0c10710, 2021.

Aziz, E. F., Ottosson, N., Eisebitt, S., Eberhardt, W., Jagoda-Cwiklik, B., Vácha, R., Jungwirth, P., and Winter, B.: Cation-Specific Interactions with Carboxylate in Amino Acid and Acetate Aqueous Solutions: X-ray Absorption and ab initio Calculations, *The Journal of Physical Chemistry B*, 112, 12567-12570, 10.1021/jp805177v, 2008.

Berger, P., Karpel Vel Leitner, N., Doré, M., and Legube, B.: Ozone and hydroxyl radicals induced oxidation of glycine, *Water Research*, 33, 433-441, [https://doi.org/10.1016/S0043-1354\(98\)00230-9](https://doi.org/10.1016/S0043-1354(98)00230-9), 1999.

Buck, R. P., Rondinini, S., Covington, A. K., Baucke, F. G. K., Brett, C. M. A., Camoes, M. F., Milton, M. J. T., Mussini, T., Naumann, R., Pratt, K. W., Spitzer, P., and Wilson, G. S.: Measurement of pH. Definition, standards, and procedures (IUPAC Recommendations 2002), *Pure and Applied Chemistry*, 74, 2169-2200, doi:10.1351/pac200274112169, 2002.

Buxton, G. V., Greenstock, C. L., Helman, W. P., and Ross, A. B.: Critical Review of rate constants for reactions of hydrated electrons, hydrogen atoms and hydroxyl radicals ($\cdot\text{OH}/\cdot\text{O}^-$ in Aqueous Solution), *Journal of Physical and Chemical Reference Data*, 17, 513-886, 10.1063/1.555805, 1988.

Craig, R. L., Peterson, P. K., Nandy, L., Lei, Z., Hossain, M. A., Camarena, S., Dodson, R. A., Cook, R. D., Dutcher, C. S., and Ault, A. P.: Direct determination of aerosol pH: Size-resolved measurements of submicrometer and supermicrometer aqueous particles, *Analytical chemistry*, 90, 11232-11239, 2018.

Guo, J., Zhou, L., Zen, A., Michaelides, A., Wu, X., Wang, E., Xu, L., and Chen, J.: Hydration of $\{\text{NH}\}_4^+$ in Water: Bifurcated Hydrogen Bonding Structures and Fast Rotational Dynamics, *Physical Review Letters*, 125, 106001, 10.1103/PhysRevLett.125.106001, 2020.

Krauklis, I. V., Tulub, A. V., Golovin, A. V., and Chelibanov, V. P.: Raman Spectra of Glycine and Their Modeling in Terms of the Discrete–Continuum Model of Their Water Solvation Shell, *Optics and Spectroscopy*, 128, 1598-1601, 10.1134/S0030400X20100161, 2020.

Li, M., Su, H., Zheng, G., Kuhn, U., Kim, N., Li, G., Ma, N., Pöschl, U., and Cheng, Y.: Aerosol pH and Ion Activities of HSO_4^- and SO_4^{2-} in Supersaturated Single Droplets, *Environmental Science & Technology*, 56, 12863-12872, 10.1021/acs.est.2c01378, 2022.

Liang, Z., Chu, Y., Gen, M., and Chan, C. K.: Single-particle Raman spectroscopy for studying physical and chemical processes of atmospheric particles, *Atmos. Chem. Phys.*, 22, 3017-3044, 10.5194/acp-22-3017-2022, 2022.

Medoš, Ž., Plechkova, N. V., Friesen, S., Buchner, R., and Bešter-Rogač, M.: Insight into the Hydration of Cationic Surfactants: A Thermodynamic and Dielectric Study of Functionalized Quaternary Ammonium Chlorides, *Langmuir*, 35, 3759-3772, 10.1021/acs.langmuir.8b03993, 2019.

Moision, R. M. and Armentrout, P. B.: Experimental and Theoretical Dissection of Sodium Cation/Glycine Interactions, *The Journal of Physical Chemistry A*, 106, 10350-10362, 10.1021/jp0216373, 2002.

Peng, X., Vasilakos, P., Nenes, A., Shi, G., Qian, Y., Shi, X., Xiao, Z., Chen, K., Feng, Y., and Russell, A. G.: Detailed Analysis of Estimated pH, Activity Coefficients, and Ion Concentrations between the Three Aerosol Thermodynamic Models, *Environ Sci Technol*, 53, 8903-8913, 10.1021/acs.est.9b00181, 2019.

Selvarani, K., Mahalakshmi, R., and Srinivasan, N.: Growth and characterization of nonlinear optical crystal glycine sodium nitrate and its biological activity, *Journal of Materials Science: Materials in Electronics*, 33, 13408-13417, 2022.

Vimalan, M., Flora, X. H., Tamilselvan, S., Jeyasekaran, R., Sagayaraj, P., and Mahadevan, C.: Optical, thermal, mechanical and electrical properties of a new NLO material: Mono-L-alaninium nitrate (MAN), *Arch Phys Res*, 1, 44-53, 2010.

Wang, N., Guo, Y., Li, J., Pang, S., and Zhang, Y.: Hygroscopic behavior and phase state of mixed NH_4NO_3 /amino acids particles by microscopy and IR technology, *Atmospheric Environment*, 273, 118951, <https://doi.org/10.1016/j.atmosenv.2022.118951>, 2022.

Wen, L., Schaefer, T., Zhang, Y., He, L., Ventura, O. N., and Herrmann, H.: T- and pH-dependent OH radical reaction kinetics with glycine, alanine, serine, and threonine in the aqueous phase, *Physical Chemistry Chemical Physics*, 24, 11054-11065, 10.1039/D1CP05186E, 2022.