Author response to comments by Referee #2

All referee comments are shown in black, our author responses in blue; suggested new manuscript text is indicated in red with text suggested to be removed in *red italics*.

General comments:

Ervens et al. presents a thoughtful, well-written piece summarizing previous, and motivating future, research on microorganisms in the atmosphere. The detailed figures were especially informative and effectively conveyed the concepts discussed throughout the article. While some considerations are not wholly original, they are clearly and concisely encapsulated here.

Author response: We thank the referee for their positive and constructive comments. We agree that not all concepts presented in our article are completely new. However, it was not the primary motivation of this 'Ideas & Perspectives' article to present entirely new findings but instead to synthesize current knowledge from the intersections of atmospheric chemistry, microphysics and biology. With the recent growth of interest in atmospheric biology - both in the fields of atmospheric sciences and biogeosciences - we seek with this article to put (more or less) well-known facts into a broader context.

Specific comments:

Page 3, Lines 57-59: The sentence discussing settling velocity is a bit unclear. Consider replacing "particle size" with "particle diameter". Are you assuming that doubling the number of cells would double the particle diameter? What about in the case of high RH or a cloud droplet where a second cell may just displace water (cf. Figure 3)?

Author response: We agree with the referee that this sentence may have oversimplified the relationship between number of cells and particle size or even surface $(\propto v_t)$. Two cells (even if of identical sizes) may not double the surface of the particle due to more compact geometric arrangement. It may lead, however, to more water uptake since more hygroscopic mass will lead to more water uptake. We modified the sentence as follows:

The settling velocity of particles approximately scales with the square of particle size. thus, doubling the number of cells (of same size) in a single particle may decrease their settling time by a factor of 4 (Section *S1.1, supplemental information).*

The presence of more than a single cell in a particle leads to a larger particle. However, the resulting total particle surface area might not scale proportionally with the number of cells since the particle shape and total volume might be mostly determined by the hydration shell.

Page 3, Lines 63-64. Please provide a reference for these statements. It may be appropriate to cite Fankhauser et al. (2019) who were among the first to suggest that microbes were physically isolated from one another in the atmosphere.

Author response: We realized that the underlying assumptions for the second sentence were not fully clear. We added appropriate references and an expanded text how to derive the fraction of bacteria in CCN populations. We would like to point out that these conclusions were not unique to Fankhauser et al. Instead, we cite Ervens and Amato (2020) where it is explicitly stated that "bacteria are unevenly distributed among cloud drop populations as statistically only 1 in ∼10 000 droplets may contain a single bacterial cell", together with some basic numbers on particle concentrations and sizes.

Fewer than 1 out of 1000 atmospheric aerosol particles contain a bacteria cell. Accordingly, it may be concluded that the number fraction of bacteria-containing droplets is on a similar order of magnitude.

Atmospheric concentrations of bacteria cells are typically in the range of 0.001 - 0.1 cells cm[−]³ *air* [\(Burrows](#page-1-0) [et al., 2009;](#page-1-0) [Després et al., 2012\)](#page-1-1) with typical sizes on the order of 100 nm - 1 *µ*m [\(Sattler et al., 2001;](#page-2-0) [Pöschl and Shiraiwa, 2015\)](#page-2-1). The total atmospheric number concentration of aerosol particles of such sizes ('fine particles') ranges from $10^3 \t- 10^5$ particles cm_{air}^{-3} [\(Seinfeld and Pandis, 2006\)](#page-2-2). The comparison of these numbers reveals that bacteria comprise $\ll 1\%$ of all atmospheric aerosol particles. A cloud droplet forms by water vapor condensation on an individual particle, i.e. on a cloud condensation nucleus (CCN) that is typically in the size range of fine particles. The fact that the bacteria number concentration is much smaller than the total CCN concentration in the atmosphere led [Ervens and Amato](#page-1-2) [\(2020\)](#page-1-2) to conclude that only 1 out of ∼10000 cloud droplets contains a bacteria cell.

Page 6, Section 2.3: This section assumes that microorganisms are metabolically active in the atmosphere. The article would benefit from a brief discussion of dormancy, in relation to this and other stressors.

Author response: We thank the referee for this suggestion. Indeed, we imply that bacteria are metabolically active in this section. To clarify this caveat, we add the following text at the beginning of this section to point out the different levels of activity, despite very little data on this on atmospheric microorganisms:

In the atmosphere, bacteria cells may exhibit different levels of metabolic activity, which range from mere survival strategies, i.e., activity focused solely on repairing cellular damage, to dormancy, during which cells sustain their essential biological functions, to growth and multiplication as the most energy-intensive activities [\(Price and Sowers, 2004\)](#page-2-3). Cells may become dormant under water-limited conditions [\(Haddrell and Thomas,](#page-1-3) [2017;](#page-1-3) [Smets et al., 2016\)](#page-2-4) or due to other stressors [\(Šantl-Temkiv et al., 2022\)](#page-2-5). In cloud water, [Sattler et al.](#page-2-0) [\(2001\)](#page-2-0) observed cell activity at 0◦C compatible with cell growth, whereas dormancy was observed outside clouds [\(Smets et al., 2016\)](#page-2-4). Given that particles (including bacteria calls) only spend a fraction of their time inside clouds ([\(Ervens and Amato, 2020\)](#page-1-2)), it can be, thus, expected that many bacteria may be dormant for long period of their atmospheric residence time. Dormancy has been shown in other environments to be an efficient response to harsh conditions and ultimately being beneficial for survival [\(Jones and Lennon, 2010\)](#page-1-4).

In the conclusion section, we modified the following sentence:

Similarly, the rationale for exploring biodegradation rates in cloud water could be extended from focusing on potential impacts on chemical budgets to consequences of limited nutrient availability on **levels of metabolic activity, including dormancy**,*microorganism* starvation and survival.

pH response (Author Response to Referee #1): The inclusion of a new subsection on effect of pH is appreciated. It is suggested to add additional commentary in light of work by Liu et al. (2023, ACP) whose laboratory experiments reported on the effects of pH (in combination with light exposure) on bacterial survival.

Author response: We thank the referee for reminding us of the study by [Liu et al.](#page-1-5) [\(2023\)](#page-1-5). In addition, to the text we suggested in our response to Referee 1, we will add:

[Liu et al.](#page-1-5) [\(2023\)](#page-1-5) found different trends when they examined the pH dependence of the survival and biodegradation rates of two strains of *Enterobacter* bacteria isolated from ambient air in a polluted environment: The showed that in the presence of light, the survival rate decreased in particular at $pH \leq 5$. These trends may point to different sensitivities of this particular bacteria type to pH, as compared to the responses by bacteria in cloud water [\(Vaïtilingom, 2013\)](#page-2-6). The concurrent responses to low pH and the presence of sunlight may suggest some photolytic or photochemical mechanism that influences the biodegradation activity.

Technical corrections:

Page 2, Line 29: The word "role" is written twice.

Page 4, Line 79: Missing period after closed parenthesis and "Novel".

Page 4, Line 88: Extraneous closed parenthesis before comma.

Page 5, Line 108: Extraneous period between times and during.

Author response: Thank for pointing out these typos. They will be all corrected in the revised manuscript.

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