



## The effect of simvastatin on soil ammonia oxidisers

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### Abstract

#### Keywords:

Simvastatin,  
ammonia  
oxidising archaea,  
ammonia oxidising  
bacteria, inhibition,  
competition.

*The quest to understand and distinguish the contributions of ammonia oxidising archaea (AOA) and ammonia oxidising bacteria (AOB) to nitrification in the soil has increased the search for compounds that can selectively inhibit either of them. This study is aimed to investigate selective inhibition of AOA, in pure cultures and soil, by simvastatin and to determine the effect of selective inhibition of AOA and AOB on NH<sub>3</sub> oxidation activity and growth of AOA and AOB in the soil. In pure cultures, simvastatin selectively inhibited all tested AOA isolates at concentrations between 8 and 100 μM. In soil microcosms incubated with 1 mM of simvastatin at low and high NH<sub>3</sub> concentrations for 28 days, simvastatin selectively inhibited AOA but not AOB in pH 6.5 soil. More so, simvastatin is a selective AOA inhibitor in culture and soil. AOA and AOB growth in soil were detected at low and high NH<sub>4</sub><sup>+</sup> concentration. Therefore, AOA and AOB are more active when competition for NH<sub>4</sub><sup>+</sup> in the soil is relieved using selective inhibitors*

Accepted: 27 June 2019

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### INTRODUCTION

Both ammonia oxidising bacteria (AOB) and ammonia oxidising archaea (AOA) are involved in ammonia oxidation (Prosser, 2011; Nicol *et al.*, 2011) and there is evidence for niche differentiation associated with ammonium (NH<sub>4</sub><sup>+</sup>) concentration and supply (Di *et al.*, 2009; 2010; Hofferle *et al.*, 2010; Stopnisek *et al.*, 2010; Verhamme *et al.*, 2011). The initial belief that AOB prefer highly fertilised soils, while AOA prefer soils receiving low rates of ammonia supply was questioned by recent discoveries on the isolation of *Candidatus* (Ca.) Nitrosocosmicus genus of AOA (Lehtovirta-Morley *et al.*, 2016; Jung *et al.*, 2016; Sauder *et al.*, 2017) and the detection of ammonia oxidation and growth of AOA in soil amended with low and high NH<sub>4</sub><sup>+</sup> concentration (Hink *et al.*, 2016; 2017). This latter discovery clearly suggests that differences in competition for ammonia by AOA and AOB in soil supplied with different concentrations of NH<sub>4</sub><sup>+</sup> are the differentiating factors between AOA and AOB rather than the differential inhibition of AOA and AOB by high ammonia concentration. Gubry-Rangin *et al.* (2010) detected ammonia oxidation activity and growth of AOA with no

recorded growth of AOB even though their presence was detected in two soils with different pH (4.5 and 6.0) and low ammonia concentration. The presence of AOB in acidic soil was supported by the detection of AOB phylotypes (Onodera *et al.*, 2010; Stopnisek *et al.*, 2010; Prosser and Nicol, 2012) and isolation of Gamma-proteobacteria AOB (strain TAO100) from acidic soil (Hayatsu *et al.*, 2017). However, information on the contribution of AOB to nitrification in acidic soil and in soil with low ammonia concentration is limited due to lack of a potent selective AOA inhibitor. The need to study the contributions of different groups of ammonia oxidisers (AOA and AOB) to ammonia oxidation in soil at different ammonia concentrations and pH lead to the use of compounds that can selectively inhibit ammonia oxidation activity of AOA or AOB. Although several chemicals inhibitors such as nitrapyrin, dicyandiamide, allylthiourea, carbon disulphide-based inhibitors, 3,4-dimethylpyrazol-phosphate, 2-amino-4-chloro-6-methylpyrimidine and acetylene have been characterised and used as inhibitors of soil nitrification with a view to their commercial use to increase the efficiency of ammonia-based fertilisers (Subbarao *et al.*, 2006; Singh and Verma,