Interactive comment on “Role of extracellular polymeric substances (EPS) from *Pseudomonas putida* strain MnB1 in dissolution of natural rhodochrosite” by H. Wang and X. Pan

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By L. Wang (Referee #3): “In this manuscript, an interesting laboratory study was carried out to investigate role of extracellular polymeric substances (EPS) from *P. putida* strain MnB1 in enhancing dissolution of natural rhodochrosite. The results showed that EPS were found to play an important role in increasing dissolution of natural rhodochrosite. To my opinion, the study was innovative and the manuscript should be accepted for publication after minor revision. Some of my suggestions are as follows:” 1. In the part of introduction in pages 7274 and 7275, the author mentioned that “Oxidative dissolution of rhodochrosite leads to produce dissociative Mn(II) and Mn oxides.” What kinds of Mn oxides are formed in the process of the oxidative dissolution of rhodochrosite? And what kinds of Mn oxides were formed through the dissolution of microorganisms? Reply: These questions were resolved in the revised manuscript. “XRD and SEM results indicated that biogenic Mn oxides were composed of poorly order, poorly crystalline phyllomanganate, similar to δ-MnO2, but varied in morphology (Figs. 2 and 3). The SEM graphs showed that a number of cells (the arrows) were adhered to the surface of biogenic Mn oxides (Fig. 2a-d). EDS analysis showed that the biogenic and synthetic Mn oxides were mainly composed of O and Mn, and other elements such as Fe, P and Mg.” 2. The role of EPS in oxidative dissolution of natural rhodochrosite was investigated using a Mn oxidizing bacterium, *Pseudomonas putida* MnB1. Dose the bacteria itself have an effect on the oxidative dissolution of natural rhodochrosite? Reply: In this work, *Pseudomonas putida* MnB1 played an important role in the oxidative dissolution of natural and rhodochrosite. First of all, living cells can effectively dissolve natural and synthetic rhodochrosite and subsequently oxidize liberated Mn(II) ions to form bacterial Mn oxides. Moreover, bacterial EPS from *P. putida* strain MnB1 played an important role in enhancing the dissolution of natural rhodochrosite. 4. In the part of discussion in page 7281 line 14 to 16, these results suggested that the functional groups of N-H in proteins, C=O in COOH or amide I and C-H or C-O-C in
polysaccharides were directly involved in the dissolution of natural rhodochrosite. Why these functional groups are considered to play an important effect on the dissolution of natural rhodochrosite? Reply: In this study, functional groups of bacterial EPS involved in dissolution of natural rhodochrosite were investigated by FTIR analysis. As shown from Fig. 6, we compared with FTIR spectra of EPS before and after reaction with natural rhodochrosite. Several absorption peaks (e.g., 2391 cm⁻¹, 1400 cm⁻¹ and 1083 cm⁻¹) were changed significantly after the reaction between EPS and natural rhodochrosite, and these results were mainly attributed to the involvement of functional groups in bacterial EPS.

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