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# ANALYSIS OF MOLYBDENITE BIOLEACHING BY THIOBACILLUS FERROOXIDANS IN THE ABSENCE OF IRON (II)

L.Pistaccio [*], G.Curutchet [*,#] and E.Donati [*]

* Facultad de Ciencias Exactas, Universidad Nacional de La Plata, 47 y 115, (1900) La Plata.
# Corresponding author, Comisión de Investigaciones Científicas, 11 y 526 (1900) La Plata, Argentina.

## SUMMARY

*Thiobacillus ferrooxidans* attachment on $MoS_2$ and Mo dissolution are increased by the addition of the tensioactive agent Tween 80 in absence of iron(II), which suggests that the poor bioleaching of $MoS_2$ is caused by its hydrophobic character. Additionally, inhibition of *Thiobacillus ferrooxidans* growth by the presence of $MoO_4^{2-}$ and the effect of variable amounts of Tween 80 on bacteria growth and on $MoS_2$ bioleaching are considered in this paper. Data confirm the need of bacterial attachment to insoluble substrate for bioleaching by the direct mechanism.

## INTRODUCTION

*Thiobacillus ferrooxidans* is a lithoautotrophic bacteria able to grow by the catalytic oxidation of reduced sulphur compounds and iron (II). Metals initially associated with sulphide ions are solubilized in this way, by conversion of sulphide to sulphate.

Two mechanisms of bacterial action are recognised:
1) <u>direct</u> oxidation of sulphide ion by the bacteria and
2) <u>indirect</u> oxidation of sulphide ion by the iron (III) obtained through bacterial action on iron (II).

In a previous paper we demonstrated the need of bacterial attachment to substrate in order to produce the direct mechanism, in the case of low solubility sulphides (Pogliani *et al*, 1990). Moreover, we found very low bacterial attachment to molybdenite ($MoS_2$) and, at the same time, almost negligible Mo extractions by the direct

mechanism in the absence of iron (II) (Donati *et al*, 1992).

Literature references to $MoS_2$ dissolution by *Thiobacillus ferrooxidans* and posible negative aspects are not clear (Torma, 1977; Brierley, 1974; Brierley, 1982; Sugio *et al*, 1992).

The strong hydrophobic character of the $MoS_2$ due to its structural characteristics may prevent bacterial attachment and, consequently, the direct bacterial mechanism. We have studied the attachment of *Thiobacillus ferrooxidans* to $MoS_2$ in the presence of the tensioactive agent Tween 80. Additionally, studies of *Thiobacillus ferrooxidans* growth in the presence of Tween 80 and variable amounts of $MoO_4^{2-}$, and of $MoS_2$ bioleaching in presence of variable amounts of Tween 80 were made. In order to compare other possible effects of Tween 80, experiments were also done with CuS instead of $MoS_2$ (CuS has low hydrophobic character).

## EXPERIMENTAL

Bacteria attachment experiments were made according to the technique previously used (Donati *et al*, 1990) with 0.2 g of $MoS_2$ (Strem Chemicals Inc.) in contact with 10 ml of a culture of *Thiobacillus ferrooxidans* in a 9 K medium (Silverman and Lundgren, 1959) without iron(II) and with 1000, 2500 and 5000 ppm of Tween 80 respectively. Blanks with the same composition but without $MoS_2$ were run. For all flasks bacterial counts were done and the percentages of bacteria attached to the $MoS_2$ were calculated discounting those attached to the glass (blanks). Similar experiments with CuS (covellite) were done.

Toxicity effects of soluble $MoO_4^{2-}$ on *Thiobacillus ferrooxidans* and bacterial growth in the presence of Tween 80 were studied in 9 K medium inoculated at 5 % v/v with a culture at exponential stage of growth. Toxicity experiments were made adding $MoO_4^{2-}$ at $10^{-4}$, $10^{-3}$ and $2\times10^{-3}$ mole.$l^{-1}$ and bacterial growth was studied in presence of 50, 100, 200, 500, 1000, 2500 and 5000 ppm of Tween 80.

Bioleaching experiments of $MoS_2$ and of CuS in 9 K medium without iron (II), inoculated at 10 % v/v with a culture in exponential growth and with a pulp density of 0.1 % p/v were made. Inoculated systems without Tween 80 and with 1000, 2500 and 5000 ppm of tensioactive agent and a sterile system (in which inoculum was replaced by a 2 % p/v of methanol solution of thymol), were studied.

Bacterial counts were done using a Petroff-Hauser camera and a Labophot microscope with a contrast phase

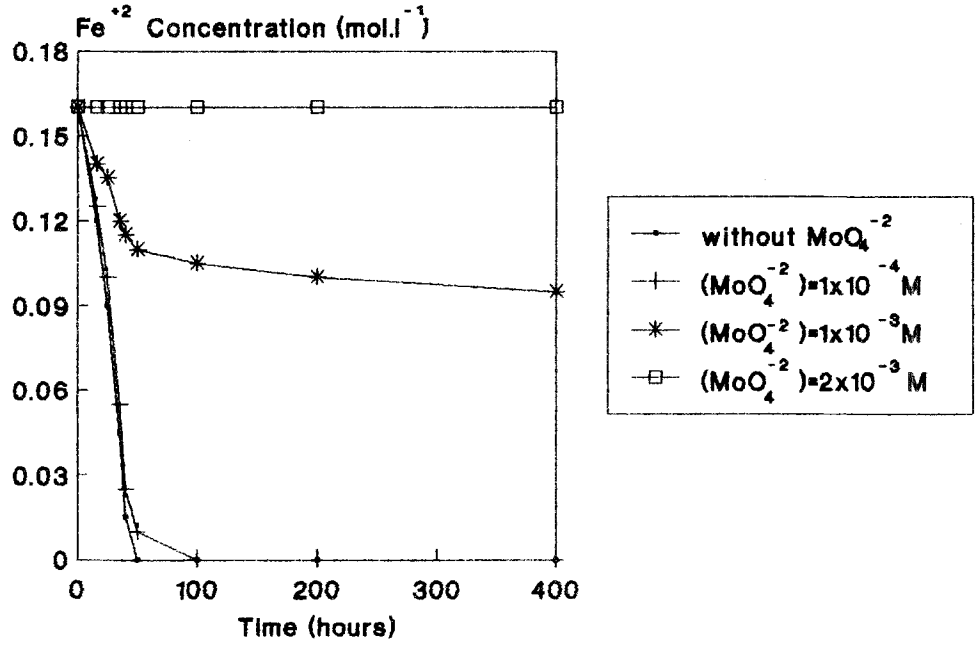
attachment. Triplicate countings were made in duplicate systems. Maxima dispersion was 10 % and mean dispersion was less than 5 %. Ions in solution were determined using atomic absorption spectrophotometry.

In all cases temperatures of 30 $\pm$ 1 C and initial pH of 1.8 were used.

## RESULTS AND DISCUSSIONS

In figure 1 *Thiobacillus ferrooxidans* growth measured through the iron (II) concentration ($[Fe^{2+}]$) in presence of several amounts of $MoO_4^{2-}$ is shown. An almost normal growth for $[MoO_4^{2-}] = 10^{-4}$ M, small initial growth for $[MoO_4^{2-}] = 10^{-3}$ M a complete inhibition within 400 hours for $[MoO_4^{2-}] = 2 \times 10^{-2}$ M are observed. At the same time experiments on *Thiobacillus ferrooxidans* growth in presence of iron(II) and increasing amounts of Tween 80 (data not shown) indicated that the rate of iron (II) oxidation and bacteria growth were independent of the presence of Tween 80. Only when the amounts of tensioactive agent was higher than 5000 ppm no iron(II) oxidation was observed within 200 hours (at these conditions normal growth is obtained within 48 hours).

**FIGURE 1: Molybdenum toxicity on Thiobacillus ferrooxidans**



In table 1 results of bacterial attachment experiments with and without Tween are shown. Percentage of bacterial attachment was calculated as $P(\%) = (\frac{n_o - n}{n}).100$, when $n_o$ and n are bacteria numbers in the filtrate of the system without and with substrate ($MoS_2$ or CuS) respectively, both in presence of same amounts of Tween 80.

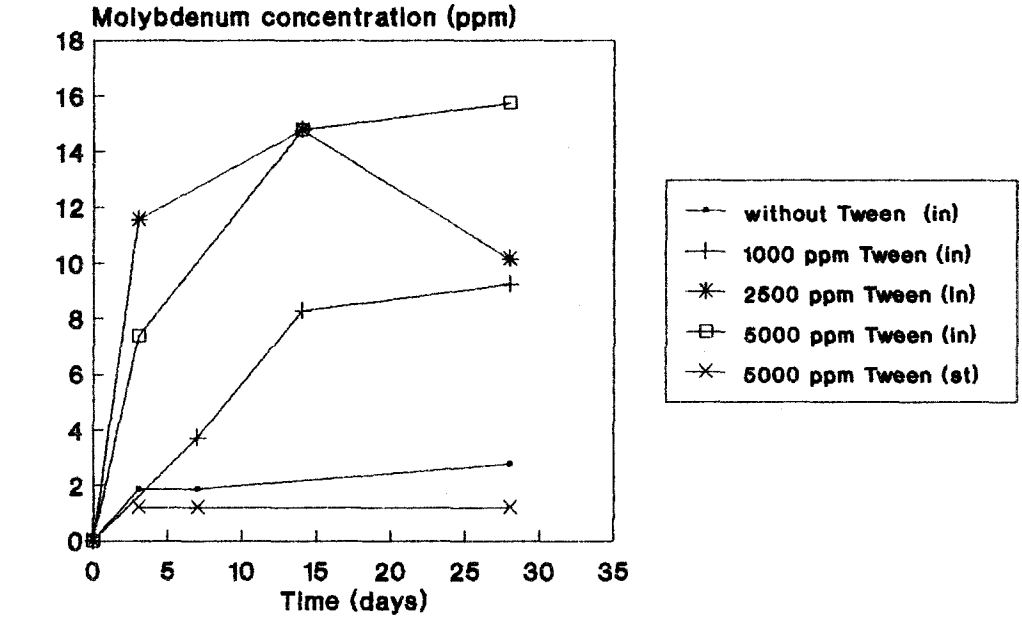
Table 1: Percentage of bacterial attachment

| | $MoS_2$ | CuS |
|---|---|---|
| Without Tween 80 | 17,9 % | 48,0 % |
| [Tween 80] = 1000 ppm | 78,4 % | 73,4 % |
| [Tween 80] = 2500 ppm | 86,3 % | 74,3 % |
| [Tween 80] = 5000 ppm | 87,0 % | 76,4 % |

As it is shown *Thiobacillus ferrooxidans* attachment on $MoS_2$ is very low and when Tween 80 is added it is considerably increased. Much higher bacteria attachment is observed on CuS and this is further increased with Tween 80.

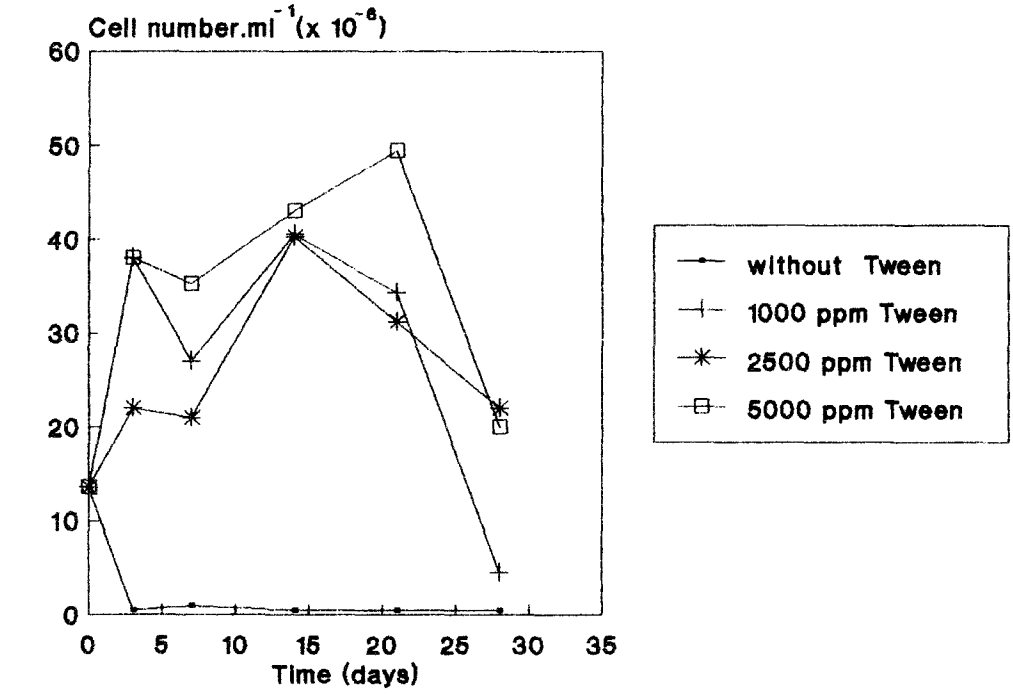
In figure 2a Mo concentration in solution as a function of bioleaching time is shown. It is seen that in the system without Tween 80 a very low extraction is produced although higher than in the sterile system (st) where the Tween 80 concentration was 5000 ppm (the highest concentration used). In the inoculated systems (in) Mo extraction was higher with higher Tween 80 concentration; this is confirmed in figure 2b where bacteria populations in the different systems are shown (it has to be remembered that in these systems bacterial growth only occurs through the direct bacteria mechanism, because no iron (II) is present).

## FIGURE 2: Molybdenite bioleaching
## Influence of tensioactive agent




in=inoculated; st=sterile

2a) Molybdenum concentration




2b) Bacterial population

Molybdenum extraction in all cases is stopped when its concentration in solution reaches the limit at which inhibition in bacterial growth is produced. In CuS bioleachings no effect is observed when Tween 80 is added (data not shown).

## CONCLUSIONS

*Thiobacillus ferrooxidans* attachment on $MoS_2$ and molybdenum dissolution are considerably increased by the addition of the tensioactive agent Tween 80, even in absence of iron (II). This means that the direct mechanism of substrate oxidation by the bacteria is increased by the addition of Tween 80. This is a proof that the difficulties in the direct bioleaching of $MoS_2$ are due to its hydrophobic character. Moreover, the obtained data confirms the fact that bacterial attachment on the substrate is necessary for the direct mechanism of bacterial attack of very insoluble sulphides.

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