CONTRIBUTION OF PLANTS AND MICROBIOTA TO THE BIOLOGICAL TURNOVER OF GOLD AT THE TOMAKIN PARK GOLD MINE, NEW SOUTH WALES, AUSTRALIA.

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INTRODUCTION

Plants have been shown to accumulate heavy metals and metalloids such as Pb, Ni, Cu, Cd or As (e.g., Yang *et al.* in press). A number of studies with plants from different geographical regions in Europe, North America and Asia have shown that some plants are also capable of accumulating Au (e.g., Warren & Devault 1950, McInnes *et al.* 1996, Pyatt 1999). Because these plants accumulate Au from underlying mineralised zones, they can be used as sampling media for biogeochemical exploration (e.g., Boyle 1979, Erdman & Olson 1985). This method of bio-exploration has been used in North America and Europe for several decades (Boyle 1979, Erdman & Olson 1985) and has recently been recognised by the exploration industry in Australia. A number of studies conducted in Australia have shown that several native Australian plants are able to accumulate Au and may indicate the location of buried mineralisation (e.g., Lintern *et al.* 1997, Arne *et al.* 1999, Hulme & Hill 2003 Roach 2004). In samples of bluebush (*Maireana*) elevated Au concentrations were detected and its use as an indicator for underlying deposits in geochemical exploration in Australia was proposed (Lintern *et al.* 1997). *Eucalyptus sp.* bark samples from the Ballart goldfields in Victoria, contained up to 150 ppb of Au (Arne *et al.* 1999). In two recent studies Hill & Hulme (2003) and Roach (2004) have reported Au elevated concentrations in *Eucaplytus sp.* and *Casuarina sp.* growing in auriferous soils overlying mineralisation in arid and semi-arid areas of South Australia and New South Wales. By promoting the mobilisation and transport of Au, i.e., uptake in the root zone, transport against gravity from the roots to the leaves, and accumulation in the leaves, bark, twigs and finally the litter, plants may be important contributors to a biologically mediated cycle of Au in the regolith. Recent studies conducted with auriferous and non-auriferous soils from the Tomakin Park Gold Mine have shown that the indigenous bacterial microflora of auriferous soils is also capable of mediating the solubilisation of Au and may also contribute to the dispersion, transport and concentration of Au (Reith & McPhail, in review); in microcosm experiments up to 80 wt.% of the finely disseminated Au contained in the soil was solubilized within 70 days of incubation. However, the contribution of plants to the biological turnover of Au has not been quantified nor has it been compared to the microbially-mediated turnover. Thus, the aims of this study are to: (i) assess the bio-accumulation of Au by plants growing in auriferous soils; and, (ii) develop a conceptual model to compare the contribution of plants and microorganisms to the total biological turnover of Au in the regolith.

THE TOMAKIN PARK GOLD MINE

The Tomakin Park Gold Mine is located 2 km west of the coastal village of Tomakin in south-eastern New South Wales, Australia (Figure 1), at S 35° 48' 51.9" and E 150° 10' 26.4". The mine is located in the Molong-South Coast Anticlinorial Zone, which is a structural subdivision of the Lachlan Fold Belt. The regional geology consists of an anticlinorial zone, which displays a large, north-south elongated, cratonised block of Ordovician flysch sediments overlain by Middle to Late Devonian rift volcanics and Late Devonian transitional and cratonic sediments. The primary ore consists of massive and disseminated arsenopyrite and pyrite that contain Au in solid solution or as small inclusions (Bowman 1979). The climate in the region is temperate and is controlled by the proximity to the Pacific

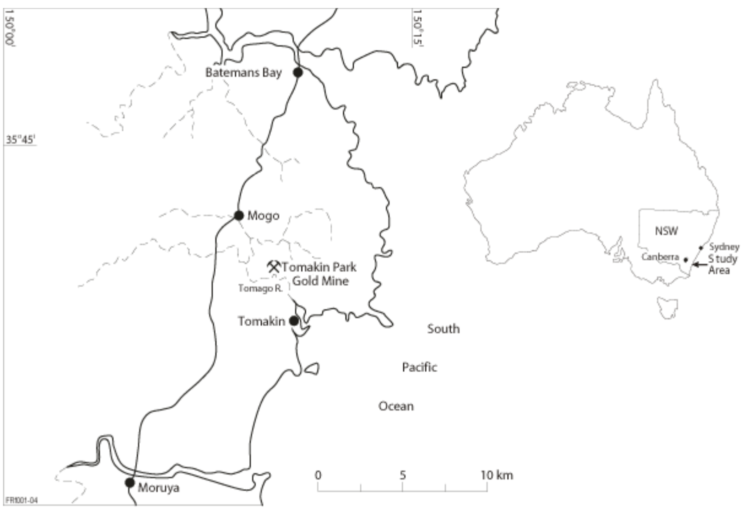

Figure 1: Location map of the Tomakin Park Gold Mine in south-eastern New South Wales, Australia.

In: Roach I.C. ed. 2005. *Regolith 2005 – Ten Years of CRC LEME*. CRC LEME, pp. 260-264.

Ocean. Average annual temperatures and rainfall approximate 15ºC and 1000 mm, respectively. The study area lies between 10 to 60 m above sea level and the landscape is characterised by narrow ridgelines with gentle to very steep side slopes. The regolith consists of moderately to intensely weathered materials to depths of up to several meters. Field analyses of soils show that the soils are colluvial cambisols. The soil textures are predominantly sandy or silty clay loams with gravel contents of up to 30 vol.% and the soil pH is between 5.5 and 6.0. The study area is covered by an open eucalypt forest consisting of *Eucalyptus maculata* (Spotted Gum), *Eucalyptus paniculata* (Grey Ironbark), *Eucalyptus pilularis* (Blackbutt), *Callitris columellaris* (Cypress pine), *Eucalyptus gummifera* (Red bloodwood) and *Eucalyptus sieberi* (Silvertop ash), with patches of rainforest in the gullies.

SAMPLING, SAMPLE PREPARATION AND CHEMICAL ANALYSIS

Vegetation sampling was conducted in February 2003. Leaf samples were collected by hand wearing powder-free nitrile gloves. Gloves were also worn during all preparation steps in the laboratory. The samples were packed in paper bags and transported to the laboratory, where leaves were separated from remaining twigs and other plant materials. The leaves were washed in 0.5 % HCl for 10 min and then 3 times 10 min each with double-deionised water in order to minimise the influence of airborne dust contamination. The washed samples were dried for 48 hours at 60ºC and then hand crushed. Five grams of the crushed leaves were placed into glass beakers, which were covered with glass petri-dishes and ashed at 600ºC for 20 h. Prior to use all glassware had been cleaned in an acid bath and repeatedly rinsed with double-deionised water. The ash was dissolved in concentrated aqua regia and diluted double-deionised water. A control blank using aqua regia and water was used to measure the concentrations of trace elements in the reagents, and the results reported in Table 1 were corrected by the values measured in the negative control. A depth profile in the auriferous soils overlying the mineralisation was taken in February 2003 using a soil corer. A mixed sample for every 5 cm of profile was taken, samples were sieved to a < 2 mm fraction and milled in a tungsten carbide mill to < 200 µm size. The total concentrations of Au in the soil samples were determined by digesting 3 g of sample for 24 h in concentrated aqua regia at 25°C. The supernatant was filtered through a No. 4 Whatman paper filter and diluted with double-deionised water to final HNO_3 and HCl concentrations of approximately 2 vol. % and 0.7 vol. %, respectively. Trace elements in the digests were measured using a Varian Ultramass Inductively Coupled Plasma-Mass Spectrometer (ICP-MS) at the Department of Earth and Marine Sciences, ANU for (detection limits in ppb): As (3.7), Au (2.8), Ce (0.1), Co (0.1), Cr (0.5), Cu (1), La (0.1), Ni (0.4), Pb (0.3), Sb (0.1), Se (0.1), Sn (1.7), Te (0.1), Y (0.1), Zn (2). The analytical precision was within 5-%.

TRACE ELEMENT CONTENT OF LEAVES AND LEAF LITTER

The trace element concentrations of the leaves and leaf litter digests from the Tomakin Park Gold Mine are given in Table 1. Leaves and leaf litter from plants growing in auriferous soils overlying the mineralisation at the Tomakin Park Gold Mine displayed measurable concentration of all trace elements analysed, including elevated Au concentrations. Trace element concentrations ranged from 0.8 to 108 ng g^{-1} (dry weight (d.w.) plant material). The concentrations of Au in the leaves ranged from 3.0 to 27.5 ng g^{-1} (d.w. plant material). In the dominant *Eucalyptus sp*. at the site the Au concentration was 6.8 ng g^{-1} (d.w. plant material). In leaf litter, which consisted mostly of *Eucalyptus sp.* leaves 18.2 ng g^{-1} (d.w. plant material) were detected indicating a possible enrichment of Au in the leaf litter compared to the leaves. Most other trace element concentrations in the litter samples were lower compared to concentrations in the leaves from the *Eucalyptus sp*., which may indicate that Au is strongly bound in leaf litter. By using the washing procedure described above the concentrations of trace elements in the plants materials may have been underestimated due to leaching in HCl.

PLANT- AND MICROBIALLY-MEDIATED TURNOVER OF GOLD – A MODEL

The results show that plants accumulate Au at the Tomakin Park Gold Mine and thus mediate its transport through the regolith. However, the extent of plant-mediated compared with the microbially-mediated turnover of Au is not known. To compare the plant- and microbially-mediated turnover of Au a quantitative model is developed. A number of parameters had to be set (see below) and are based on the following assumptions. The set parameters are then used to determine the calculated parameters. The Au content of leaves and leaf litter combined with the annual litter fall was used to estimate the plant-mediated Au turnover. It was assumed that the annual litter fall at the site was similar to the litter fall measured in at Kioloa State Forest, which is dominated by *Eucalyptus maculata* and located approximately 35 km from the Tomakin Park Gold Mine (Pook *et al.* 1997). The enrichment zone of Au in the soil at Tomakin Park was estimated to extend 10 m on either side of the Au-bearing-quartz vein and the total Au content of the top 30 cm of soil in the enrichment zone is approximately 500 ng g^{-1} (d.w. soil), based on Au concentration measured in samples from a traverse published by Reith *et al.* (2005) and in a depth profile in soil overlying the mineralisation (Figure 2). In the top 10 cm of the depth profile Au concentration approximated 1200 ng g^{-1} (d.w. soil), Au concentration in the soils from 11 to 42 cm ranged from 100 to 600 ng g^{-1} (d.w. soil). As an indicator for microbially-mediated Au turnover the percentage of finely disseminated Au that was released from the solid soil fractions during in the biologically active microcosms, i.e., approximately 80 wt. %, was used. However, the microbial activity is highly variable within soils and often confined to certain hot spots of activity, where conditions for the parts of the microflora are met that may mediate the mobilisation of Au. To account for the variability within the soil, the calculations were made for a range of scenarios. The soil mass in which the microbially-mediated solubilisation of Au may occur was varied between 10, 20, 30, 40, 50, 100 wt. %, and the percentage of Au that is mobilised from the soil was adjusted to 10, 20, 30, 40, 50, 80, and 90 wt. % of the total Au contained in the soil. The result of the calculations shown in Table 2 are based on the occurrence of 1 and 4 microbially mediated Au turnover events per year.

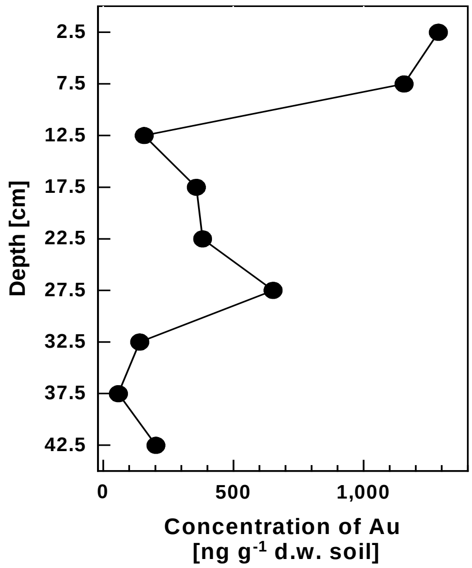

Figure 2: Concentration of Au in soil profile overlying the mineralisation at the Tomakin Park Gold Mine, New South

Table 1: Trace element concentrations in leaves and leaf litter of plants growing in auriferous soil overlying the vein-quartz deposit at the Tomakin Park Gold Mine in New South Wales, Australia. Concentrations are given in ng g^{-1} (d.w. plant material).

Scientific Name	Common Name	As	Au	Ce	Co	Cr	Cu	La	Ni	Pb	Sb	Sn	Y
Upper storey trees													
Casurina littoralis	Black She-oak	7.9	**3.0**	10.8	12.0	6.4	9.2	9.2	3.7	2.4	2.0	16.1	n.d.
Eucalyptus sp.	n.a.[a]	32.9	**6.8**	32.6	33.9	26.9	38.4	32.6	25.1	25.2	19.2	54.0	26.1
Middle storey trees													
Acacia implexa	Lightwood	n.d.[b]	**4.4**	0.5	23.4	108.3	n.d.	0.6	3.4	n.d.	0.8	1.5	1.3
Understorey shrubs													
Acacia myrtifolia	Myrtle Wattle	6.2	**6.2**	12.4	8.7	4.4	19.0	10.8	2.4	1.8	1.9	30.8	37.2
Coprosma quadrifida	Prickly currant-bush	34.0	**27.5**	64.1	69.0	37.7	53.4	63.3	56.6	37.4	16.1	32.4	62.5
Mixed leaf litter	n.a.	5.3	**18.2**	n.d.[b]	0.8	n.d.	15.6	0.3	5.8	5.9	n.d.	n.d.	n.d.

[a] n.a.: not applicable. [b] n.d.: not detected

Set parameters:

Length of the outcropping mineralisation [m] (Reith *et al.*, 2005): **152**

Width of the soil enrichment zone [m] (Reith *et al.*, 2005): **20**

Litter fall by *Eucalyptus maculata* (Spotted Gum) [t ha^{-1} a^{-1}] (Pook *et al.*, 1997):

3.1 (minimum), 5.7 (average), 7.5 (maximum)

Concentration of Au in plant materials [ng g^{-1} d.w. material]

Eucalyptus sp. leaves*:* **6.8**

Mixed leaf litter: **18.2**

Depth of the enrichment in the soil [m]: 0.3

Soil density (Scheffer *et al.*, 1992) **2.65**

Concentration of Au in the soil [ng g^{-1} d.w. soil] (this study; Reith *et al.*, 2005, Reith and McPhail, *in review*): **500** (range 150 – 1500)

Measured microbial turnover of Au in 70 days [wt.%] (Reith and McPhail, *in review*): **80**

(to be able to calculate a total Au dissolution range we assume 10, 20, 30, 40, 50, 100 wt.% for the volume of soil in which Au solubilisation is active and Au dissolution rates of 10, 20, 30, 40, 50, 80, and 90 wt.%)

Occurrence of microbial turnover of Au [events a^{-1}]: **1** and **4**

Calculated Parameters:

Area of the Au-enrichment zone [m^2]: **3040**

Litter fall in Au in the enrichment zone [t]: **1.73** (range: 0.94 – 2.28)

Total volume of the soil [m^3]: **912**

Total weight of the soil in the enrichment zone [t]: **2416.8**

Based on these parameters and assumptions the resulting plant-mediated turnover of Au was calculated. The deposition of Au by plants in the enrichment area was:
based on leaf data [mg a^{-1}]: **11.8 (range 6.4 – 15.5)**
based on leaf litter data [mg a^{-1}]: **31.5 (range 17.1 –155.0)**

Table 2: Calculated scenarios of a microbially-mediated turnover of Au in auriferous soil from the Tomakin Park Gold Mine.

Au-solubilisation [wt. %]	**Mass of soil displaying Au-solubilisation [wt. %]**					
	10	**20**	**30**	**40**	**50**	**100**
	Mass of Au solubilized *per annum* by microorganisms in the mineralized zone [g a^{-1}]					
1 Event a^{-1}						
10	12.1	24.2	36.2	48.3	60.4	120.8
20	24.2	48.3	72.5	96.6	120.8	241.6
30	36.2	72.5	108.7	145.0	181.2	362.4
40	48.3	96.6	145.0	193.3	241.6	483.2
50	60.4	120.8	181.2	241.6	302.0	604.0
80	96.6	193.3	290.0	386.6	483.2	**966.4**
90	108.7	217.4	326.2	434.9	543.6	1087.2
4 Events a^{-1}						
10	48.3	96.6	145.0	193.3	241.6	483.2
20	96.6	193.3	290.0	386.6	483.2	966.4
30	145.0	290.0	434.9	579.9	724.8	1449.6
40	193.3	386.6	578.0	773.1	966.4	1932.8
50	241.6	483.2	724.8	966.4	1208.0	2416.0
80	386.6	773.1	1159.7	1546.2	1932.8	3865.6
90	434.9	869.8	1304.6	1739.5	2174.4	4348.8

According to the calculations the plant-mediated turnover of Au is in the range of 6.4 to 155 mg a^{-1} in the enrichment zone. The model calculations conducted to assess the microbially-mediated Au turnover suggest that it may be several orders of magnitude higher, as shown in Table 2. The calculated range for the microbially mediated Au solubilisation in the enrichment zone was 12.1 to 4,348.8 g a^{-1}. Assuming Au solubilisation in the field would equal the mobilisation measured in the earlier microcosm study, i.e., 80 wt. % of Au dissolved in 100 wt. % of soil (Reith & McPhail, in review), the microbially-mediated turnover in the mineralised zone of Au is 966.4 g a^{-1}. Assuming that only 10 wt. % of the total Au was solubilized and that the solubilisation occurs in only 10 wt. % of the soil once per year the calculations suggest a microbially-mediated Au turnover of 12.1 g a^{-1}. This value still exceeds the maximum value calculated for plant-mediated Au turnover by a factor of 100. Thus, these results may indicate that microbially-mediated processes may dominate the turnover of Au in the auriferous top-soil at the Tomakin Park Gold Mine. The main contribution of plants to the biologically mediated turnover of Au appears to be the transport of Au from deeper zones within the regolith into the top-soils. Especially *Eucalyptus sp.*, with their extensive root systems, appear to be important in mediating the uptake and transport of Au from the deeper zones of the regolith to the surface. The obvious enrichment of Au in the O- and Ah-horizon at the Tomakin Park Gold Mine compared to the underlying B-horizon supports this suggestion.

CONCLUSIONS

This study has shown that plants growing in auriferous soils overlying mineralized zones in temperate New South Wales, Australia, are able to accumulate Au and thus contribute to the biologically mediated turnover of Au in the regolith. In addition, the results of this study suggest that *Eucalypt sp.* and *Acacia sp.* may be trialled as useful sampling media for exploration biogeochemistry in temperate regions in New South Wales. The results of the model scenarios indicate that the contribution of plants compared to the microbially-mediated turnover of Au in the top-soils may be limited. However, plants appear to be important in transporting Au from the deeper zones of the regolith or the even primary mineralization to the top of the profile by taking up Au with their roots, transporting into the leaves, and depositing it in the top-soils with the falling litter. Here the activity and cell numbers of the soil bacteria is particularly high, which might lead to constant micro-scale mobilization, transport and re-adsorption, i.e., turnover, of Au in the top-soils (Reith & McPhail, in review).

Acknowledgements: Thanks to CRC LEME for funding this project; Andy G. Christy at the Department of Earth and Marine Sciences, ANU, for help with the ICP-MS analysis; Peter and Kinuyo Wyatt at Tomakin Park Gold Mine for access to the site and help during my stay.

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