May 2012

The Weathering of Platinum from Nuggets and Platinum Immobilisation by Cupriavidus metallidurans

Sean Gordon Campbell
The University of Western Ontario

Supervisor
Dr. Gordon Southam
The University of Western Ontario

Graduate Program in Geology

A thesis submitted in partial fulfillment of the requirements for the degree in Master of Science

THE WEATHERING OF PLATINUM FROM NUGGETS AND P
IMMOBILISATION BY CUPRIAVIDUS METALLIDURANS

(Spine title: WEATHERING AND IMMOBILISATION OF PLATIN

(Thesis format: Integrated Article)

by

Sean Gordon Campbell

Graduate Program in Department of Earth Sciences

A thesis submitted in partial fulfillment
of the requirements for the degree of
Master of Science

The School of Graduate and Postdoctoral Studies
The University of Western Ontario
London, Ontario, Canada

THE UNIVERSITY OF WESTERN ONTARIO
School of Graduate and Postdoctoral Studies

CERTIFICATE OF EXAMINATION

Supervisor	Examiners
_____	_____
Dr. Gordon Southam	Dr. Norman Duke
	_____
	Dr. Desmond Moser
	_____
	Dr. Richard Gardiner

The thesis by

Sean Gordon Campbell

entitled:

The Weathering of Platinum from Nuggets and P
Immobilisation by Cupriavidus metallidurans

is accepted in partial fulfillment of the
requirements for the degree of
Master of Science

_____ _____
Date Chair of the Thesis Examination Boa

Abstract

Two studies were conducted to investigate the biogeochemistry of
the first project, nuggets of platinum were examined to evaluate mine
precipitation processes occurring at the nugget-soil interface. The pr
dissolution pits, cavities, as well as acicular, iron ox
grain surface evidence that this interface is an importa
chemical weathering. Element mapping revealed that struc
can be linked to morphological features on the grain surface. In the s
soil bacterium, *Cupriavidus metallidurans*, was reacted with aqueous platinu
and rapidly immobilised platinum. XANES/EXAFS analysis
demonstrated that platinum bound to the bacteria and platinum
from chloride to carboxyl functional groups. TEM revealed
able to precipitate colloidal platinum. Both of these stu
importance of nano- and micro-scale platinum compounds in natural syste

Keywords: platinum, surficial weathering, Fifield Platinum Pro
Cupriavidus metallidurans, biomineralisation, secondary mineralisation

Co-Authorship Statement

Chapter 2, entitled Surficial weathering of platinum grains, has been prepared as a manuscript for submission for publication. Campbell performed the microscopy and analyses, drafted and prepared the figures and wrote the manuscript. Gordon, Reith and Southam provided technical support at Argonne National Laboratory, provided the platinum grains and Reith and Southam provided the funding to support this research and advised on all aspects of this study.

Chapter 3, titled Immobilisation of platinum group minerals in slag and refractory, was authored by S.G. Campbell, L. Maclean, D. Brewe, F. Gervais and G. Southam, and has been prepared as a manuscript for submission for publication. Campbell designed and executed the experiments, microscopy and analyses and wrote the manuscript and prepared the tables and figures. Maclean provided the synchrotron data, Brewe provided technical support at Argonne National Laboratory, and Reith and Southam provided the funding to support this research and advised on all aspects of this study.

Acknowledgments

No man is an island[1], nor is a thesis one man. I have to admit, I words& I hope I didn t miss anyone, but in case I did, than

There are many people who make a thesis come to fruition financial, motivational and emotional support make it po happen in every way possible. You re simply wonderful! M been a great support, especially my pal Tommy Campbell. Thank y much. Dr. Southam, thank you for believing in me, a scien just two years ago. You have the uncanny knack for turnin way of inspiring all us. And to you lablings& Jer, Jenine, Bird, Andrea, Nahed, Alex and Jie, I wish you the best friendship and support. I would be remiss if I didn t mer Maclean, and our postdoc, Chris Omelon, who provided instrumental su

There are many people who have made me the scienti to Dr. Stephen Altaner. If I hadn t sat in his class a deca a geologist. A special thanks to my family who took me to the mountains as especially to my Grandpa Havlena who taught me to love t me wonder as a boy. I owe my formal geology education Gordon Southam, Stephen Altaner, Dr. Fred Walker, Clark, Nesbitt, Guy

[1] Donne, J., 1624, Meditation XVII.

Cam Tsujita and Burns Cheadle. You have all provided

nurtured my enthusiasm.

I would also like to thank my defence committee and

Moser, Richard Gardiner and Steve Hicock. The staff at the Nanofab, ZEBL, [illegible]otron

and the beamline scientists at the Advanced Photon So

Laboratory have helped make this all happen.

I would like to thank a number of groups for funding

Ontario Graduate Scholarship and the Department of Earth Science

Table of Contents

CERTIFICATE OF EXAMINATION i

Abstract ii

Co-Authorship Statement iv

Acknowledgements v

Table of Contents vi

List of Tables ix

List of Figures x

List of Abbreviations and Symbols xi

Chapter 1 Introduction

1.1 Platinum enrichment in the crust 1

1.2 Platinum placer deposits formation 3

1.3 Platinum in supergene systems 3

1.4 Aqueous platinum geochemistry 5

1.5 Formation of secondary platinum and platinum grains 10

1.6 Geomicrobiology of platinum 11

1.7 Hypothesis and study objectives 15

References 17

Chapter 2 Surficial weathering of platinum grains

2.1 Introduction 22

2.2 Material and Methods 25

2.3 Results and discussion … 28

Reference … 36

Chapter 3. Immobilisation of platinum by Cupriavidus metallidurans

3.1 Introduction … 39

3.2. Materials and methods … 42

3.3. Results … 51

3.4. Discussion … 67

Reference … 72

Chapter 4. Conclusions … 77

Reference … 82

Appendix to Chapter 2 … 85

Reference … 88

Curriculum vitae … 89

List of Tables

Chapter III: Immobilisation of platinum by Cu(II) mediated metallidurans

3.1 Dilution factors and Pt^{2+} and Pt^{4+} supernatant standards 46

3.2. Toxicity effects of aqueous Pt^{2+} ions on metallidurans 52

3.3. Toxicity effects of aqueous Pt^{4+} ions on metallidurans 52

3.4. Immobilisation of Pt^{2+} by Cu(II) mediated metallidurans 54

3.5. Immobilisation of Pt^{4+} by Cu(II) mediated metallidurans 54

List Of figures

Chapter 2. Surficial weathering of platinum grains

2.1. SEM micrographs of the surfaces of two platinum grain

2.2. SEM micrographs, EDS and XANES spectra of surficial weathering features on the surface of a platinum grain

2.3. SEM micrographs, EDS spectra and element mapping of surficial weathering features on the surface of a platin

Chapter 3. Immobilisation of platinum by *Cupriavidus metallidurans*

3.1. TEM micrographs of *C. metallidurans* phase

3.2. TEM micrographs of cells exposed to 500 µM ($PtCl_6^{2-}$) (100 µg/mL)

3.3. TEM micrographs of cells exposed to 500 µM ($PtCl_4^{2-}$) (100 µg/mL)

3.4. Ultra-thin section TEM micrographs of *C. metallidurans* exposed to 500 µM ($PtCl_6^{2-}$) (100 µg/mL)

3.5. TEM micrographs of cells exposed to 500 µM (Pt^{4+}) (100 µg/mL)

3.6. TEM micrographs of cells exposed to 5000 µM (Pt^{4+}) (1000 µg/mL)

3.7. XANES spectra of bacteria exposed to 5000 µM ($PtCl_6^{2-}$) (1000 µg/mL) and Pt^{4+} solutions

3.8. XANES/EXAFS spectra of bacteria exposed to 5000 µM ($PtCl_6^{2-}$) (1000 µg/mL) solutions

3.9. EXAFS spectra of bacteria exposed to 5000 µM ($PtCl_6^{2-}$) (1000 µg/mL) solutions

Appendix to Chapter 2

A.1 Maps of Fifield Platinum Province 85

A.2 Schematic profile of Fifield Platinum Province ..... 86 laterite

A.3. µXRD of platinum grain in Figure 2.3A .....

List of Abbreviations Symbols and

ºC	Celsius
µg	Microgram
µL	Microlitre
µm	Micron or micrometre
µM	Micromolar
µXRD	Micro-X-ray Diffraction
aq	Aqueous
ATP	Adenosine triphosphate
CFU	Colony forming unit
DDI water	Distilled, deionised water
EDS	Energy dispersive spectroscopy
EMS	Electron Microscopy Sciences
eV	Electron volt
EXAFS	Extended X-ray Absorption Fine Structure
FEG	Field emission gun
hr	Hour
ICP-AES	Inductively coupled plasma atomic emission spectroscop
keV	Kiloelectron volt
km	Kilometre
L	Litre
M	Molarity
mg	Milligram

min	Minutes
mL	Millilitre
mm	Millimetre
mM	Millimolar
nm	Nanometre
g	Gram
PGE	Platinum group element
PGM	Platinum group mineral
pH	$-\log[H^+]$
ppb	Parts per billion
ppm	Parts per million
SEM	Scanning electron microscope
SRB	Sulphate-reducing bacteria
t	Time
TEM	Transmission electron microscope
XANES	X-ray Absorption Near Edge Structure
XAS	X-ray Absorption Spectroscopy

Chapter 1

Introduction

Platinum, one of the six platinum group elements (PG… osmium, iridium, palladium, ruthenium, and rhodium), is highly sought after beca… its physical and chemical properties (Macdonald 1987; N… 1991). Platinum is resistant to oxidation and corrosion and it is … and industrial processes (K… 2011). Platinum based chemotherapy drugs like cisplatin are also used in the treatment of various cancers (Alderden et al. 2006). The price of this metal has increased over time because the s… pace with growing demand. Platinum is among the rares… concentration in the lithosphere typically ranges from 0.4-5 ppb. To be mined profitably, platinum deposits require 100-1,000-fold enrichment over ba… level concentrations (Mungall and Naldrett 2008; Koek et al. 2010). There has been … increase in platinum deposit exploration and research over … 0 years … understand the mineral processes to discover new deposits, but the genesis of many platinum deposits remains unclear (Maier 2005).

1.1 Platinum enrichment in the crust

Principal PGE ore deposits are formed by magmatic, hydrothermal, alluvial and … processes (Macdonald 1987). Igneous bodies host most of the world-class economic deposits with an average grade of 5-20 g/t (5-20 ppm) (Nixon and Hammack 1991). The genesis of platinum deposits in mafic-ultramafic complexes, including the…

primary deposits that, unlike alluvial placers, are inextricably tied to sul chemistry and the processes that transport platinum and s crust (Macdonald 1987; Maier 2005; Mungall and Naldrett 2008).

Platinum-rich deposits are generated when sulphur concentrat the available platinum from the silicate portion of a melt (Maier 2005; Mungall and Naldrett 2008). During a metallogenic event, platinum-enriched sulphu ascends from the mantle to the crust where it becomes trapped. O sulphur saturation in the magma occurs and an immiscibl accumulates at the bottom of the magma chamber where it separates the highly chalcophile platinum into the sulphide horizon (Maier 2005; Mungall and Naldrett 2008; Koek et al. 2010). Platinum sulphides form during this crystallization step (Mungall and Naldrett 2008). These magmatic processes often for platinum-bearing reefs within large mafic-ultramafic intrusions (Ma 2005). Although poorly understood, hydrothermal brines can cause subsequent redistribution of PGE (Mungall and Naldrett 2008).

Alluvial PGE placers are often weathered from concentrically-zoned ma ultramafic 'Alaskan-type' or 'Ural-type' (and to a lesser extent 'ophiolitic-type') primary magmatic intrusions that were formed by processe above (Macdonald 1987; Nixon and Hammack 1991; Tolstykh 2005; Koek et al. 2010). Alaskan-Ural-type complexes intrude into fold belts, mo craton-platform environments and transitional oceanic-continental (Nixon and Hammack 1991; Tolstykh 2005).

1.2. Platinum placer deposit formation

An important source of platinum is from placer deposits (Grant and Bassett 2005). There are several types of deposits but alluvial placers provide the majority of placer platinum (Mertie 1969). The formation of an alluvial platinum pl begins with the extraction of platinum-bearing minerals from source rocks at expos or near Earth's surface. Kock et al. (2010) describes a process where minerals from primary magmatic deposits (e.g. platinum minerals) within supergene and lateritic profiles (the enrichment of secondary minerals) experience chemical and physical weathering. Over time, the platiniferous materials are physically and chemically mobilised from the source depos and washed into streams where it is concentrated, deposited and tra gravitational sorting and the water action. In some cases, platinum-bearing material is weathered in situ and lighter mineral fractions are p eroded away. Although Alaskan-Ural type magmatic intrusions are the primary sources of placer platinum, other types of magmatic intrusion can generate pl Platinum minerals have been documented in the alluvial sedim reefs of South Africa's Bushveld Igneous Complex where the world's largest reserve PGEs are hosted (Melcher et al. 2005).

1.3 Platinum in supergene systems

In alluvial placers and other surficial weathering profiles the proper conditions for platinum to be mobile exist. Platinum can be remob from exposed primary magmatic deposits and form significant secondary

enrichment (Bowles 1986; Boyle et al. 2001; Hanley 2005). Supergene e
and laterization are processes that concentrate gold in primary ore
deposits containing subeconomic sulphide mineralisation (Guilbert 1986
These regions commonly occur in tropical and semiarid climates where
temperatures and humidity promote chemical oxidation (
waters above the water table are able to leach a large vo
redeposit them into smaller volumes of high grade mate
(Guilbert 1986).

Pyrite (FeS_2) is the most common hypogene sulphide and
enrichment that takes place in the supergene environment with
metal sulphides (Guilbert 1986). Weathering exposes oxygen and iron/sulphur
oxidising bacteria above the water table, producing sulphuric acid (H_2SO_4)
and ferric sulphate ($Fe_2(SO_4)_3$), which in turn act as solvents for o
sulphides, including copper, (Mann 1984; Bowles 1986; Guilbert
1986; Enders 2000). Ferric sulphate is reduced during the
oxidation of other metallic sulphides. These will either be ca
from the site of oxidation by meteoric waters or will pre
(Anderson 1982; Enders 2000). Silver and gold have similar chemical
properties, is reasonable that processes responsible for the
gold are comparable for the dissolution and migration of
more easily oxidised and more soluble than gold, and
favourable for gold dissolution and mobility, are favourable for
platinum (Bowles 1986; Oberthür 2000; Reith 2007).

Acidified and oxidised waters descend and leave behind a leached cap that lacks the target metal. An insoluble mixture of hydrous iron-sulphates including jarosite ($KFe_3(OH)_6(SO_4)_2$), iron-oxyhydroxides such as goethite (and hematite (Fe_2O_3) are also formed during the oxidation of pyrite in th leached cap, giving it the characteristic rust appearance (Anderson 1982; E 2000; Suárez et al. 2008). As evidence of this oxidising pro oxyhydroxide coatings have been found on platinum n (Ottemann and Augustithis 1967; Bowles 1986; Suárez et al. 2008).

Descending surficial waters oxidise ore metal below the water t where redox conditions favour reduction and precipitatio mediates deposition of the ores as it readily transfers its other metals. These metals usually have a stronger affin strip pyrite of its sulphur. Much like the surface, metallic sulphides are fo however, they are more common in the supergene zone. The acidic wa neutralised when they come into contact with carbonates This process is cyclical and ongoing. Fluctuations in the water table alter the physical location of the redox boundary. Over time, thick blankets of ore metal can form and be of economic value (Anderson 1982; E[illegible] 2000).

1.4. Aqueous platinum geochemistry

Platinum is mobile under supergene conditions (Fuchs and Rose 1974; Bowles 1986; Azaroual et al. 2001). The platinum content of natural waters is 3 to 6 ord magnitude less concentrated than in the lithosphere; higher

concentrations are dependent on the mode of (Barref 1997; Mungall and Naldrett 2008; Locmelis et al. 2010; Kubrak et al. 2011). Surficial water flowing through areas with elevated levels of platinum group minerals and/or their mine tailings, would presumably have more concentration of platinum than waters draining areas lacking platinum enrichment. The solubility of platinum is also controlled by its aqueous chemistry. The presence of an aqueous medium as well as platinum's ability to form stable complexes with ligands commonly found in this solution, transportation and sequestration (Brownlee 1986; Colombo et al. 2008; Kubrak et al. 2011). Furthermore, hydrodynamic characteristics will also influence migration of dissolved material (Kubrak et al. 2011).

At standard surface water temperatures (25 °C) and pressure (1 atm), platinum as simple aqueous ions except at extremely oxidizing conditions, and a must complex with other dissolved ligands to be stable (Wood 1988). Complexation is an important process by which cations bind to one or more negatively charged anions or complexing species, concentrations of simple metal ions in solutions would remain too low to form economic deposits. Ions complexed to anions in solution, however, can be concentrated to generate profitable grades of metal (Hanley 2005).

The literature lacks reliable experimental data on the stability of platinum species and complexes found in natural waters (Mountain and Wood 1988; Colombo et al. 2008). The predicted oxidation state for platinum

Pt^{2+} as shown in Equation (Hanley 2005). Very oxidizing conditions, platinum ca
exist as Pt^{4+} ion, but Pt^{2+} is the more common aqueous species. A
experimental data relating to the thermodynamic stabilit
systems relate to this particular oxidation state (Hanley 2005; Mountain and Wood 2
Platinum prefers to form covalent interactions such as with chlorine,
cyanide, ammonia, oxides, sulphides and tellurides (Mountain and Wood 19
Vlassopoulos et al. 1990). Complexation, however, depends on
platinum's preference for various ligands; it ultimately de
the transporting fluids and the availability of ligands in
Rose 1974).

$$Pt_{(s)} + \tfrac{1}{2}O_2 + 2H^+_{(aq)} \rightarrow Pt^{2+}_{(aq)} + H_2O \qquad (1)$$

In oxidizing waters that are acidic, common chloride anions
stable complexes with Pt^{2+} and Pt^{4+} (Fuchs and Rose 1974; Bowles 1986; M
Wood 1988). These reactions are given in Equations 2 and
al 2005). The aqueous Pt^{2+} compounds, $PtCl^+$, $PtCl_2$, $PtCl_3^-$ are known to exist, bu
$PtCl_4^{2-}$ is the predominant anion at 25°C (Mountain and Wood
2001).

$$Pt^{2+}_{(aq)} + 4Cl^-_{(aq)} \rightarrow PtCl_4^{2-}{}_{(aq)} \qquad (2)$$

$$Pt^{4+}_{(aq)} + 6Cl^-_{(aq)} \rightarrow PtCl_6^{2-}{}_{(aq)} \qquad (3)$$

In oxidizing environments with more basic (less acidic)
complexation with hydroxides is favored (Xu et al. 2001; Colombo et al. 2008).
Equation 4 shows a simplified hydrolysis reaction (Wood 1991). Pt always pr
in water, especially in neutral and basic waters, and plati

predominate in platinum chlorocomplexes when chloride is not limiting (Mountain and Wood 1988; Wood et al. 1992; Azaroual et al. 2001). Aqueous Pt^{2+} hydroxide compounds, including $PtOH^{+}$, $Pt(OH)_2$ and $Pt(OH)_4^{2-}$. As pH increases, the predominance of these complexes shifts to hydroxylated species (Moun Wood 1988; Wood 1991; Azaroual et al. 2001). Mixed chloro-hydroxy complexes are considered to be unimportant for platinum mobility (Mounta

$$Pt^{2+}_{(aq)} + nOH^{-}_{(aq)} \rightarrow Pt(OH)_n^{2-n}{}_{(aq)} \qquad (4)$$

Depending on the concentration of other anions that Pt^{2+} cations, aqueous organic platinum carboxylic acids can mobilise p (Wood 1990; Hanley 2005). Humic substances are components of normally found in soil and water. Carboxyl functional groups in humic acid abundant oxygen and when dissolved in neutral waters, the carboxylic acids become negatively charged and can react with platinum to form soluble platinum organo complexes, stable platinum-organic colloids (Vlassopoulos et al. 1990; Wood 1990; Wood et al. 1992; Kubrakova et al. 2011). Humic substances have also been shown to increase the solubility of amorphous $Pt(OH)_2$ (Wood 1990; Hanley 2005 Despite the fact that ammonia and cyanides form stable complexes with plat not abundant in natural waters. Furthermore, their solubility with platin in surficial waters by hydroxides (Fuchs and Rose 1974; Mountain and Wood 198

Bisulphide would be expected to represent a significant portion of complexes in sulphur-rich supergene environments, however platin when conditions are favourable for bisulphide formation (Mountain and Wood There are, however, metastable thiosulphate aqueous platinum complexes (Pt(II)) tha

form in oxidised and acidic sulphate environments (Anthony and Williams 199 Thiosulphate complexation appears to be important during the weat bearing sulphide ores; in the reaction of platinum with sulphides in alluvial sediments draining the Bushveld complex could be a cause of significant platinu mobilisation within these deposits (Cousins 1983; Wood et al. 1992; Anthony and Williams 1994; Mechelen et al. 2005). Equation 5 depicts this proposed process (A Williams 1994).

$$Pt^{2+}_{(aq)} + 4(S_2O_3)^{2-}_{(aq)} \rightarrow [Pt(S_2O_3)_4]^{6-}_{(aq)} \quad (5)$$

These aqueous platinum complexes are transported by water until the solution encounters an environment that favours chemica precipitation. Subsequent deposition can occur (Bowles 1986; Wood 1990). Chemical transformation involves the formation metastable dissolved platinum complexes, e.g. platinum chloride compound transformed to a platinum hydroxide species when in acidic, m basic lakes, rivers or ground water (Wood 1990). If humic aqueous system, an organic matter protected platinum col 1990; Wood et al. 1992). If dissolved platinum is unstable over time, it ma chemically reduced to metallic platinum and precipitate out of solution (Bowles 1986). It is worth noting that while a number of aqueous possible, the kinetics of the reactions are generally too slow to occ unless catalysed (Westland 1981; Wood 1990).

15. Formation of secondary platinum and platinum grains

Placer platinum commonly occurs as platinum-iron alloy, e.g. platinum grains from Alaskan-Ural type complexes and lateritic weathering profiles comm occur as platinum-iron alloys (Bowles 1986; Cabri et al. 1996; Tolstykh et al. 2005). These grains are often found in deposits where no nuggets of that s in the ultramafic source rock. PGE grains in the source rocks are commonly < 5 but placer grains can be up to several centimetres (Bowles 1986; Melcher et al. 2005). This increase in platinum nugget size from the source to evidence that platinum nuggets can grow and are "secondary" in origin or mineraliza (Cousins and Kinloch 1976; Bowles et al. 1994, 1986; Cabri et al. 1996). It is thought that elemental and alloyed platinum nanoparticles grow through the continual accretion of other platinum or platinum-metal alloy grains (Bowles 1986; Melcher et al. 2005; Cabral et al. 2011). As iron is freely available in the lateritic zone, platinum in solution could form Pt-Fe alloys (Bowles 1986). It may be fair to speculate that copper in laterit could combine with Pt-Fe alloys to form some of the Pt-Fe-Cu grains found in placers (Cab et al. 1996). Primary high temperature magmatic origins and hydrotherm alteration, however, cannot be ruled out as the source of platinum grains (Slansky et al. 1991; Cabri et al. 1996, Tolstykh et al. 2001; Melcher et al. 2005).

Mineralogical and petrologic studies of platinum-metal alloys have shown accretionary zones at the periphery of some nuggets. Stumpfl and Tarkian (1976) noted the presence of platinum growth halos aroun proof that this process occurs. Likewise, Bowles (1986) also reports

overgrowth of PGE crystal faces on older, weathered grai… et al. (2011) finds that iron-oxyhydroxides formed in supergene environments from the … iron sulphides could preferentially scavenge inorganic PG… or adsorb them to their positively charged oxyhydroxide surfaces. Mix… platinum species could be transported as colloids in the en… sediments.

On the other hand, some palladium-platinum alloy grains show altera… suggesting that the nuggets are "shrinking" and undergo… particular grains, the periphery has experienced selectiv… platinum. In essence, the palladium-platinum grains are becoming enriche… as they are altered and weathered … et al. 2007; Tormé … (2008). Suárez et al. (2008) described highly irregular and fractured shapes of s… supergene platinum nuggets as further evidence of surficial alteration in weatherin… profiles.

The alteration of placer platinum, i.e., dissolution, precipitation and growth of grains, is the subject of ongoing controversy in the lite… Traoré et al. 2008). Further characterisation of the mineral dissolution-precipitation processes occu… the nugget-soil solution interface needs to be undertaken to fully understand the geochemical cycling of platiniferous material in surficial w…

1.6 Geomicrobiology of platinum

Changing chemical conditions within surficial weathering con… placers or supergene/lateritic deposits (i.e., environmental facto…

promote the abiotic dissolution and precipitation of compl role the biosphere plays in platinum mobility. Platinum reactivity with bacteria is largely unknown because few experiments have been con study this interaction. As such, any future work to elucidate platin biogeochemistry should be done in relation to our understanding of other metal-bacteria reaction systems.

Microbes surviving in environments with high metal concentrations have the ability to either use these metal compounds as part of their metabolis sources of nutrition and energy or mitigate the toxic effect (Reith et al. 2009). Metal resistance is genetically encoded and involves detoxification mechanisms (Silver 1996; Reith et al. 2009). To have any toxic effec metal cations must first be internalised. They can diffuse into the cell passively if dr a chemiosmotic gradient or actively if the energy molecule adenosin (ATP) is expended during transport. Once internalised, th necessary for normal cell activity or compete with ot physiological activity (Nies 1999). Metal detoxification requires that the cations are neutralised and/or expelled from the bacteria. Neutralisation requires that the toxic chemically reduced and/or complexed to other ligands at t Commonly, these products are insoluble or toxic in themselves so subsequ may still be required. Efflux systems pump out ions at the expense cation/proton antiporter proteins within the cell envelope energy gradient that exists across the cell to pump out metal cations and pump in protons (Silver 1996; Nies 1999; Reith et al. 2009).

The ability to metabolise or detoxify metallic compounds allows

participate in a number of reactions. For example, a well known

catalyze reactions in supergene systems (Singer and Stumm 1970; Enders et al.

2006). Acidithiobacillus thiooxidans, a sulphur oxidising bacterium and

Acidithiobacillus ferrooxidans, an iron and sulphur oxidiser, have been shown to

contribute to the formation of acid mine drainage by gener

oxidising pyrite faster than the abiotic rate (Singer and

Suzuki 1989; Enders et al. 2006). Similar aerobic, chemolithoautotro

bacteria have also contributed to the leaching of copper in the Moren

Porphyry Copper Deposit in Arizona, USA. Calculations suggest a few kilograms

of bacteria active in 100 m^3 of weathering rock within the deposit could mobilise

between 0.01 and 0.47 moles of copper each year as part of their metabolism (Enders et

al. 2006). Anaerobic sulphate reducing bacteria found in acid mine drainage sit

sulphate compounds as part of their metabolism, forming metal sulph

(Donald and Southam 1999; Enders et al. 2006). Enders et al. (2006) calculated tha

approximately 1.2% of all copper (both leached chemically and biologically) from the

Morenci deposit could be fixed by sulphate. Therefore, microbial reduction coupled with

inorganic processes contribute to supergene copper enrichment.

Microorganisms are involved in the biogeochemical c

metals, such as gold, at surface conditions (Reith et al. 2009; Southam

et al. 2009). The formation of gold in Australia, Alaska and the famous

Witwatersrand in South Africa have been attributed to bact (Enders et al. 2006 and

references therein). Because gold and platinum share a very similar aqueou

systems, understanding the interactions between bacteria and gold could guide work into platinum-microbe interactions (Bowles et al. 1986, 1990; Vlassopo Cabral et al. 2007).

Numerous laboratory-based experiments have shown that bacteria are precipitate gold from solution (Southam and Beveridge 1994, 1996; Reith et al. 2006; Lengke et al. 2006b; Lengke and Southam 2007). Southam and Beveridge (1994) related the formation of octahedral gold by bacteria to octahed gold found in placer deposits of Western Australia and the Witwatersrand (Watterson 1984; Minter et al. 1993). Filamentous cyanobacteria have been used to precip gold and the morphology of the micro-fossil shaped gold grains found in the Witwatersrand basin (Mossman and Dyer 1985; Lengke et al. 2006b). This morphology is bacteriform and scanning electron microscopy (SEM) of small nuggets from gold deposits have revealed grains coated particles that resemble gold bacteria (Reith et al. 2006; Southam et al. 2009). Sulphate reducing bacteria have been shown chemically to reduce aqueous to elemental gold nanoparticles. Further diagenesis of the scale octahedral gold (Lengke and Southam 2007). These experimental studies provide evidence that bacteria can participate in the biogeo gold and could contribute to the formation of secondary g (Reith et al. 2006; Southam et al. 2009).

Research on bacteria-mediated platinum immobilisation and placer formation is in its infancy. Pioneering laboratory studies by Lengke et al. (2006a) showed that upon exposure to aqueous platinum chloride species,

platinum, producing spherical platinum (I-) organic precipitate which was later reduced to crystalline elemental platinum. A recent finding by Cabral et al. (2011) supports experimental evidence that bacteria-mediated precipitation is a comp platinum nugget formation in placers. Backscattered electron scanning electron microscopy (BSE-SEM) of small alluvial platinum grains revealed that has morphologies that resemble bacteria. This is one of the first evidence of bacterioform platinum in the literature. The authors propose that n growth started with bioreduction of platinum on a covered decaying root. As the nugget grew and microbial activity increased, microbial activity concentrated biotic precipitated dissolved platinum and subsequent electrochemical accretion of elementa growing grain likely worked in conjunction with bioreduct and microbe-mediated precipitation were not mutually exclusive. These mechanisms were complementary and both contribute to platinum formation in placer deposits.

1.7 Hypothesis and study objectives

In order to improve our understanding of the biogeoch studies were conducted as part of this thesis. As platinum mobility in a supergene environment, signs of chemical and physical weathering should be identifiable on the surface of placer grains. Also, as microorganisms have proven successful in immobilising aqueous platinum, they should be able to play a role in the accumulation of weathered platinum material. For example, Cupriavidus metallidurans

negative, gold-precipitating, rod-shaped bacterium found to be living on the
grains obtained from auriferous soils in northern Queensl
Gold Mine and the Hit or Miss Gold Mine), was cultured
platinum-chloride solutions (Reith et al. 2006; 2009)

References

Alderden, R.A., Hall, M.D., and Hambley, T.W., 2006, The discovery and develop of cisplatin: Journal of Chemical Education, v. 83, p. 728-734.

Anderson, J.A., 1982, Characteristics of leached capping and techniques of appraisal, in Titley, S.R., ed., Advances in geology of the porphyry copper dep North America: Tucson, AZ, University of Arizona Press, p. 275-295.

Anthony, E.Y., and Williams, P.A., 1994, Thiosulfate complexing of platinum group elements: implications for supergene geochemistry, in Alpers, C.N., and Blowes, D.W., American Chemical Society Environmental geochemistry of sulfide oxida Washington, DC, ACS Books, p. 551-560.

Azaroual, M., Romand, B., Freyssinet, P., and Disnar, J.-R., 2001, Solubility of platinum aqueous solutions at 25°C and pHs to 10 under oxidizing conditions: Geochimica et Cosmochimica Acta, v. 65, p. 4453-4466.

Barefoot, R.R., 1997, Determination of platinum at trace levels in environmental and biological materials: Environmental Science and Technology, v. 31, p. 309-314.

Bowles, J.F.W., 1986, The development of platinum-group minerals in lateri Economic Geology, v. 81, p. 1278-1285.

Bowles, J.F.W., Gize, A.P., , 1994, The mobility of the platinum-group elements in the soils of the Freetown Peninsula, Sier The Canadian Mineralogist, v. 32, p. 957-967.

Cabral, A.R., Beaudoin, G., Choquette, M., Lehmann, B., 2007, Supergene leaching and formation of platinum in alluvium: Evidence from Serro, Minas Gerais, Brazil: Mineralogy and Petrology, v. 90, p. 141-150.

Cabral, A.R., Radtke, M., Munnik, F., Lehmann, B., Rein Tupinambá, M., and Kwitko-Ribeiro, R., 2011, Iodine in alluvial platinum-palladium nuggets: Evidence for biogenic precious-metal fixation: Chemical Geology, v. 281, p. 125-132.

Cabri, L.J., Harris, D.C., and Weiser, T.W., 1996, Mineralogy and distribu platinum-group mineral (PGM) placer deposits: Exploration and Mining Geology, v. 5, p. 73-167.

Colombo, C., Oates, C.J., Monhemius, A.J., and Plant, J.A., 2008, Complexation of platinum, palladium and rhodium with inorganic ligands in the environ Geochemistry: Exploration, Environment, Analysis, v. 8, p. 91-101.

Cousins, C.A., and Kinloch, E.D., 1976, Some observations on textures and
in alluvial platinoids: Economic Geology, v. 71, p. 1377-1398.

Donald, R., and Southam, G., 1999, Low temperature anaerobic bacterial
ferrous monosulfide to pyrite: Geochimica et Cosmochimica Acta, v. 63, p. 2019-2023.

Enders, M.S., 2000, The evolution of supergene enrichme
Copper Deposit, Greenlee County, Arizona [Ph.D. thesis]:

Enders, M.S., Knickerbocker, C., Titley, S.R., and Southam, G., 2006, The role of
bacteria in the supergene environment of the Morenci Porphyry Copper Deposit,
Greenlee County, Arizona: Economic Geology, v. 101, p. 59-70.

Fuchs, W.A., and Rose, A.W., 1974, The geochemical behavior of plati
palladium in the weathering cycle in the Stillwater Complex, M
Geology, v. 69, p. 332-346.

Garnett, R.H.T., and Bassett, N.C., 2005, Placer deposits, in Hedenquist, J.W.,
Thompson, J.F.H., Goldfarb, R.J., and Richards, J.P., eds., Econo
hundredth anniversary volume: 1905-2005: Littleton, CO, Society of Econo
Geologists, p. 813-843.

Goldhaber, M.B., 1983, Experimental study of metastable
during pyrite oxidation at pH 6-9 and 30°C: American Journal of Science,
193-217.

Guilbert, J.M., 1986, The geology of ore deposits: New York, W.H. Freeman and
Company, 985 p.

Hanley, J.J., 2005, The aqueous geochemistry of the platinum-group elements (PGE
surficial, low-T hydrothermal and high-T magmatic hydrothermal environments
Mungall, J.E., ed., Exploration for platinum-group element deposits: Mineralogical
Association of Canada Short Course 35: Ottawa, Mineralogical
Association of Canada, p. 35-56.

Koek, M., Kreuzer, O.P., Maier, W.D., Porwal, A.K., 2010, Thomp
A review of the PGM industry, deposit models and explora
for Australia's PGM potential: Resources Policy, v. 35, p. 20-35.

Kubrakova, I.V., Fortygin, A.V., Lobov, S.G., Koshcheeva
and Mironenko, M.V., 2011, Migration of platinum, pallad
systems of placer deposits: Geochemistry International, v. 49, p. 1072-1084.

Lengke, M.F., and Southam, G., 2007, The deposition of elemental gold fr
thiosulfate complexes mediated by sulfate-reducing bacterial conditions: Economic
Geology, v. 102, p. 109-126.

Lengke, M.F., Fleet, M.E., and Southam, G., 2006a, Synthesis of platinum nanoparticles by reaction of filamentous cyanobacteria with platinum(IV)-chloride complex: Langmuir, v. 22, p. 7318-7323.

___2006b, Bioaccumulation of gold by filamentous cyanobacteria between 25 a 200°C: Geomicrobiology Journal, v. 23, p. 591-597.

Lizama, H.M., and Suzuki, I., 1989, Rate equations and reactions involved in pyrite oxidation by Thiobacillus ferrooxidans: Applied and Environmental Microbiology, v. 55, p. 2918-2923.

Macdonald, A.J., 1987, Ore deposit models #12. The platinum group element d classification and genesis: Geoscience Canada, v. 14, p. 155-166.

Maier, W.D., 2005, Platinum-group element (PGE) deposits and o Mineralization styles, genetic concepts, and exploration: Journal of African Earth Sciences, v. 41, p. 165-191.

Mann, A.W., 1984, Mobility of gold and silver in lateritic weathe; some observations from Western Australia: Economic Geology, v. 79, p. 38-49.

Melcher, F., Oberthur, T., and Lodziak, J., 2005, Modification of detrital plati group minerals from the eastern Bushveld Complex, South Africa: Canadian Mineralogist, v. 43, p. 1711-1734.

Mertie, J.B., 1969, Economic geology of the platinum metals: U.S. Geological Survey Professional Paper 630, 120 p.

Minter, W.E.L., Goedhart, M., Knight, J., and Frimmel, H.E., 1993, Morphology of Witwatersrand gold grains from the Basal Reef: evidence for their detrital or Economic Geology, v. 88, p. 237-248.

Mossman, D.J., and Dyer, B.D., 1985, The geochemistry of Witwatersrand-type gold deposits and the possible influence of ancient prokaryo d dissolution and precipitation: Precambrian Research, v. 30, p. 303-319.

Mountain, B.W., and Wood, S.A., 1988, Chemical controls on the solubility, transport and deposition of platinum and palladium in hydrothermal solutions; a thermodynamic approach: Economic Geology, v. 83, p. 492-510.

Mungall, J.E., and Naldrett, A.J., 2008, Ore deposits of the platinum-group elements: Elements, v. 4, p. 253-258.

Nies, D.H., 1999, Microbial heavy-metal resistance: Applied Microbio Biotechnology, v. 51, p. 730-750.

Nixon, G.T., and Hammack, J.L., 1991, Metallogeny of ultramafic-mafic rocks in British Columbia with emphasis on the platinum group elements, in McMillan, W.J., ed., Ore deposits, tectonics and metallogeny in the Canadian Cordillera: British Columbia Ministry of Energy, Mines and Petroleum Resources Paper 1991-4, p. 125-161.

Otteman, J., and Augustithis, S.S., 1967, Geochemistry and origin of "platinum nuggets" in lateritic covers from ultrabasic rocks and birbirites of W. Ethiopia: Mineralium Deposita, v. 1, p. 269-277.

Reith, F., Etschmann, B., Grosse, C., Moors, H., Benotmane, M.A., Monsieurs, P., Grass, G., Doonan, C., Vogt, S., Lai, B., Martinez-Criado, G., George, G.N., Nies, D.H., Mergeay, M., Pring, A., Southam, G., and Brugger, J., 2009, Mechanisms of gold biomineralization in the bacterium Cupriavidus metallidurans: Proceedings of the National Academy of Sciences, v. 106, p. 17757-17762.

Reith, F., Lengke, M.F., Falconer, D., Craw, D., and Southam, G., 2007, The geomicrobiology of gold: International Society of Microbial Ecology Journal, v. 1, p. 567-584.

Reith, F., Rogers, S.L., McPhail, D.C., and Webb, D., 2006, Biomineralization of gold: Biofilms on bacterioform gold: Science, v. 313, p. 233-235.

Silver, S., 1996, Bacterial resistances to toxic metal ions - a review: Gene, v. 179, p. 9-19.

Singer, P.C., and Stumm, W., 1970, Acidic mine drainage: The rate-determining step: Science, v. 167, p. 1121-1123.

Slansky, E., Johan, Z., Ohnenstetter, M., Barron, L.M., and Suppel, D., 1991, Platinum mineralization in the Alaskan-type intrusive complexes near Fifield, N.S.W., Australia. Part 2. Platinum-group minerals in placer deposits at Fifield: Mineralogy and Petrology, v. 43, p. 161-180.

Southam, G., and Beveridge, T.J., 1994, The in vitro formation of placer gold by bacteria: Geochimica et Cosmochimica Acta, v. 58, p. 4527-4530.

____ 1996, The occurrence of sulfur and phosphorus within bacterially derived crystalline and pseudocrystalline octahedral gold formed in vitro: Geochimica et Cosmochimica Acta, v. 60, p. 4369-4376.

Southam, G., Lengke, M.F., Fairbrother, L., and Reith, F., 2009, The biogeochemistry of gold: Elements, v. 5, p. 303-307.

Stumpfl, E.F., and Tarkian, M., 1976, Platinum genesis: New mineralogical evidence: Economic Geology, v. 71, p. 1451-1460.

Suárez, S., Prichard, H.M., Velasco, F., Fisher, P.C.,
Weathering of the platinum group minerals in the Aguablanca Ni-Cu gossan (SW Spain): Revista de la Sociedad Española de Mineralogía, v. 9, p. 237-238.

Teluk, A.J., 2001, Fifield Platinum Project, NSW, Aust
Technical Report (electronic copy available from <htt
PDF/Geodyne%20report%20complete%20with%20figs.pdf.>
March 2012), 71 p.

Tolstykh, N.D., Sidorov, E.G., and Krivenko, A.P., 2005, Platinum-group element placers associated with Ural-Alaska type complexes, in Mungall, J.E., ed., Exploration for platinum-group element deposits: Mineralogical Association of Canada Series Volume 35: Ottawa, Mineralogical Association of Canada, p. 113-144.

Traoré, D., Beauvais, A., Auge, T., Parisot, J.C., Colin, F., 2008, Chemical and physical transfers in an ultramafic rock w
Dissolution vs. accumulation of platinum group minerals: American Mineralogist, v. 93, p. 31-38.

Vlassopoulos, D., Wood, S.A., 1990, Gold speciation in natural waters: II. the importance of organic complexing - experiments with some simple model lig
Geochimica et Cosmochimica Acta, v. 54, p. 1575-1586.

Westland, A.D., 1981, Inorganic chemistry of the platinum-group elements, in Cabri, L.J., ed., Platinum-Group Elements: Mineralogy, Geology, Recov
Institute of Mining and Metallurgy, Special Volume 23, p. 5-18.

Wilson, A.F., 1984, Origin of quartz-free gold nuggets and supergene go
laterites and soils - a review and some new observations: Australian Journal of Eart
Sciences, v. 31, p. 303-316.

Wood, S.A., 1990, The interaction of dissolved platinum
organic acid analogues in aqueous solutions: The Canadian Mineralogist, v. 28, p. 665-673.

____, 1991, Experimental determination of the hydrolysis constants of Pt^{2+} and Pd^{2+} at 25°C from the solubility of Pt and Pd in aqueous hydroxide solutions: Geochimica et Cosmochimica Acta, v. 55, p. 1759-1767.

Wood, S.A., Mountain, B.W., and Pan, P., 1992, The aqueous geochemistry of platinum, palladium and gold; recent experimental constraints and a re-evaluation of theoretical predictions: The Canadian Mineralogist, v. 30, p. 955-982.

Chapter 2

Surficial weathering of platinum grains

2.1. Introduction

Platinum, for the six platinum group elements (PGEs), is resistance to oxidation and corrosion has made it an ideal and automobile catalytic converters; yet, platinum is not 1987, Nixon and Hammack 1991). Platinum is mobile under s environments. It can be oxidised, dissolved, complexed ligands, transported, precipitated and deposited (Fuchs a Azaroual 2001; Hay 2005). Proper conditions for platinum re commonly occur in alluvial systems and supergene environments (Bo 1986; Azaroual 2001; Freyssinet 2005; Hanley 2005)

Alluvial platinum grains typically occur as platinum alloys and are associated with ultramafic to mafic primary magmatic intrusions (B Macdonald 1987; Nixon and Hammack 1991; Teluk 2001; Cabri 2005; Koekkoek 2010). These usually exceed 10 km in diameter and are often found in orogenic arc sutures (Nixon Hammack 1991; Teluk 2001; Cabri 2005). When platinum bearing h weathered, platiniferous material is physically and chemi and transported in fluvial depositional environments. Alternatively, in some cases, platinum material is weathered out of the host rock fractio eroded away, leaving a residual concentration of PGEs (Koekkoek 2010).

Favourable chemical and biological conditions for meta
weathering commonly prevail in arid to tropical climates (Anderson 1
Sillitoe 2005). Supergene mobilisation can occur when the chemical proc
enrichment processes that extract metal from source rock expo
surface. Pyrite (FeS_2), a common mineral in supergene environm
exposed to oxygen, water and sulphur oxidising bacteria, it ca
acid (H_2SO_4) and ferric sulphate ($Fe_2(SO_4)_3$), the latter of which mobilises o
sulphides including copper, silver and gold (Mann 1984; B
Enders 2000; Sillitoe 2005). Surficial waters dissolve su
metal and carry it with the water table where it is subsequently
economic concentrations (Guilbert 1986).

The acidic and oxidising supergene waters that mobili
could presumably solubilise platinum by a similar mechanism
more soluble than gold (Bowles 1986; Testa and Cabri 2007). To date,
however, mineralogical and petrologic analyses of platinum gr
explained the alteration processes affecting placer platinum
commonly larger than the source rock they are found with
platinum grains do undergo secondary mineralisation (Cou
1976; Stumpfl and Tarkian 1976; Bowles 1986; Cabri 1992; Cabri et al. 1996).
Conversely, other studies indicate that platinum is susceptible to
dissolution and in weathering environments (Cabri et al. 2005; Testa and Cabri
2008).

In order to fully understand the cycling of sulfur in bacterial weathering environments, further characterization of mineral precipitation processes occurring at the weathering interface needs to chemical evidence of these processes should be identifiable and detectable using electron microscopy and x-ray spectroscopy.

2.2. Material and Methods

Platinum nuggets and gravity separated platiniferous and auriferous soils near Fifield, New South Wales, Australia examined to characterise surface weathering processes. Upon collection, grains were immersed in 2% glutaraldehyde (aq) to chemically fix any biological

Description of Fifield, New South Wales, Australia

The Fifield Platinum (Figure 1) is an area of past and current exploration and excavation located 380 km WNW of Sydney, Australia (Johan 1989; Andrew 1995, Teluk 2001). Alluvial and eluvial platinum and gold are considered to be genetically linked to a 500 Devonian ultramafic-mafic Alaskan/Uralian intrusions that intruded into Ordovician Cambrian metasedimentary rocks. An Ordovician paleosuture reopened as a result of crustal extension in the Lachlan Orogen may have placed platinum Alaska/Ural dunites and peridotites (Johan 1989, Andrew 1995; Teluk 2001; Gray and Frost 2001; Gray et al. 2004). Subsequent periods of eluvial weathering (Figure 2) and fluvial erosion have rework platiniferous material throughout the Cenozoic (Teluk 2001). The thought to be primary, having been eroded directly from However, the primary source of mineralisation has not been found (Slansky et al. 1991; Teluk 2001).

Scanning electron microscopy

Platinum grains were dehydrated in 50% (aq), 75% (aq) and 25 3% X 100% (aq) ethanol series for 15 minutes at each step. Dried grains were using a Tousimis® Samdri® PVT-3B critical point drier and placed onto 12 mm carbon (Electron Microscopy Sciences [EMS]). Samples were osmium coated 5 nm with a OPC60 Filgen® osmium plasma coater. Osmium coating reduces sample charging. A LEO 1530 field emission scanning electron microscope (FESEM) (University of Western Ontario, Western Facility) a Zeiss 1540-XB SEM (University of Western Ontario, Nanofabrication Facility) and a Hitachi SU 6600 Analytical VP-FESEM (University of Western Ontario, Department of Earth Sciences) were used to collect platinum grains and reproduction photographs. Seven platinum grains representing the diversity of morphologies and features observed in SEM, were selected for further analysis. Oxford Instruments INCA sight energy dispersive spectrophotometer (EDS) on the Zeiss Xmax Silicon Drift Detector EDS on the Hitachi SU 6600 were used for identification.

X-ray emission spectroscopy (XES) and X-ray Absorption Near Edge Structure (XANES) spectroscopy data collection and analysis

Two platinum grains scanned by SEM-EDS were selected for further analysis at the Pacific Northwest Consortium/X-ray Science Division (PNC/XSD) Sector-20 Insertion Device beamline at the Advanced Photon Source, Argonne National Laboratory, Argonne, USA. X-ray emission spectroscopy

conducted to map the element distribution of the surface. X-ray emission X energy reference values from Kortright and Thompson (20 elements present in the scan. X-ray Absorption Near Edge Structure (XAN measurements were conducted at spots to determine the oxidatio iron. Triplicate XANES energy measurements of the Fe K-edge were collected fro each spot and were compared to the inflection of an iron (Williams 2001). Beam energy was set to a maximum of 7100 eV so that os would not be excited.

Element maps and X-ray emission spectra were analysed u Instruments LabVIEW™ 2-D Scan Plot v. 4 and National Instruments LabVIEW™ 2-D Scan Plot v. 3. XANES data was processed using the ATHENA software (Ravel and Newville 2005). The Fe K-edge of the samples was compared to the Fe K-edge of the reference foil. Sample edge energies higher than that of the foil (at 71 eV) indicate oxidised iron

Micro X-Ray Diffraction (µXRD)

Micro X-ray diffraction (µXRD) data on a platinum grain was collected using the Bruker AXS D8 Discover microdiffractometer in the De at the University of Western Ontario. This instrument uses a Co Kα radiation source. An omega scan was performed from 7.5 to 45 degrees, with and a beam size of 300 µm. Data was collected using a STAR-D2 detector and a associated General Area Detector Diffraction System (GADDS) software and processed using Bruker's DIFFRAC.plus EVA software. Grain was intact for analys

2.3. Results and discussion

Examination of the rough to rugged morphology and weathering textures around the periphery of the quartz grains (see Figure 2.3) was generally in good agreement with Sallstedt (1991), who concluded that placer gold from Fifield are primary (magmatic) in nature.

The overall morphology of these rugged grains considerably. This heterogeneity suggests that the grains were subjected to differing degrees of chemical weathering. A number of fluvial and alluvial deposits of the Fifield Palaeo-River Province throughout the Cenozoic era and the diverse morphology of this and previous work indicative of those conditions (Telus grains in Figure 2.1A and Figure 2.2A exhibit more rounding and higher while the grain in Figure 2.1C is well rounded but has low sphericity. Ro projections dominate of the grain in Figure 2.2A. Figure 2.2A shows a sub angular rounded grain with low sphericity, a rough crystal face or a plane of weakness. In Figure 2.2A indicate impression of micro-scale crystals that have been chipped away from the gra

Pitting, cavities and striations are common on the s demonstrate that solution is an important site of mecha and chemical weathering. Features from micro-scale solution pits and cavities observed in soil bed with sand clays from the environment (Figures 2.1D and 2.3A). Long parallel scars on the grain surface may relate to

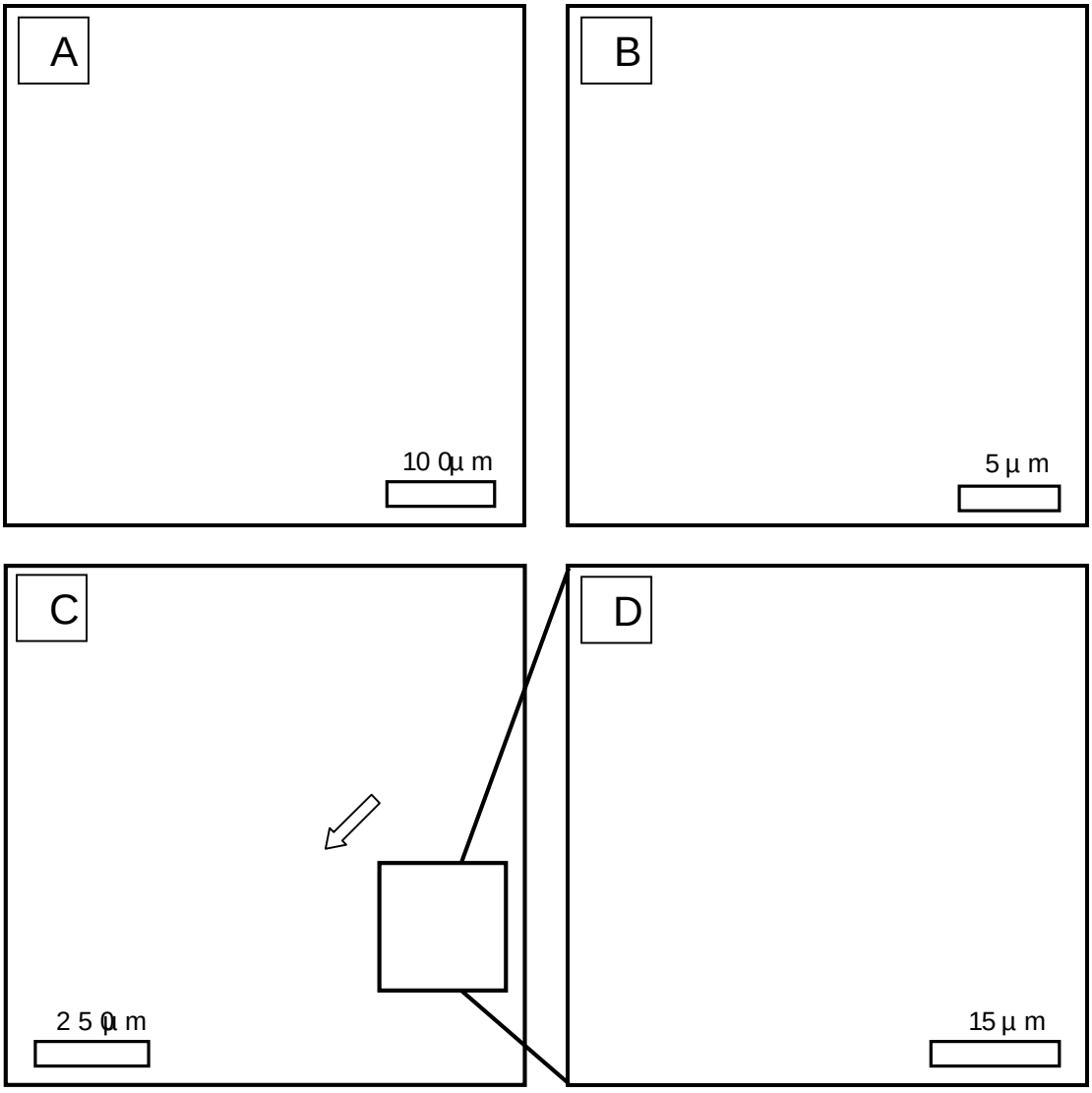

Figure 21. A: SEM micrograph of a polished platinum-coated, Nugget, scratched surface infilled with clay and organics (darker C: SEM micrograph of a depression within the Nugget, magnification SEM micrograph of a region from within th relatively deep crevice with secondary mineral grains from

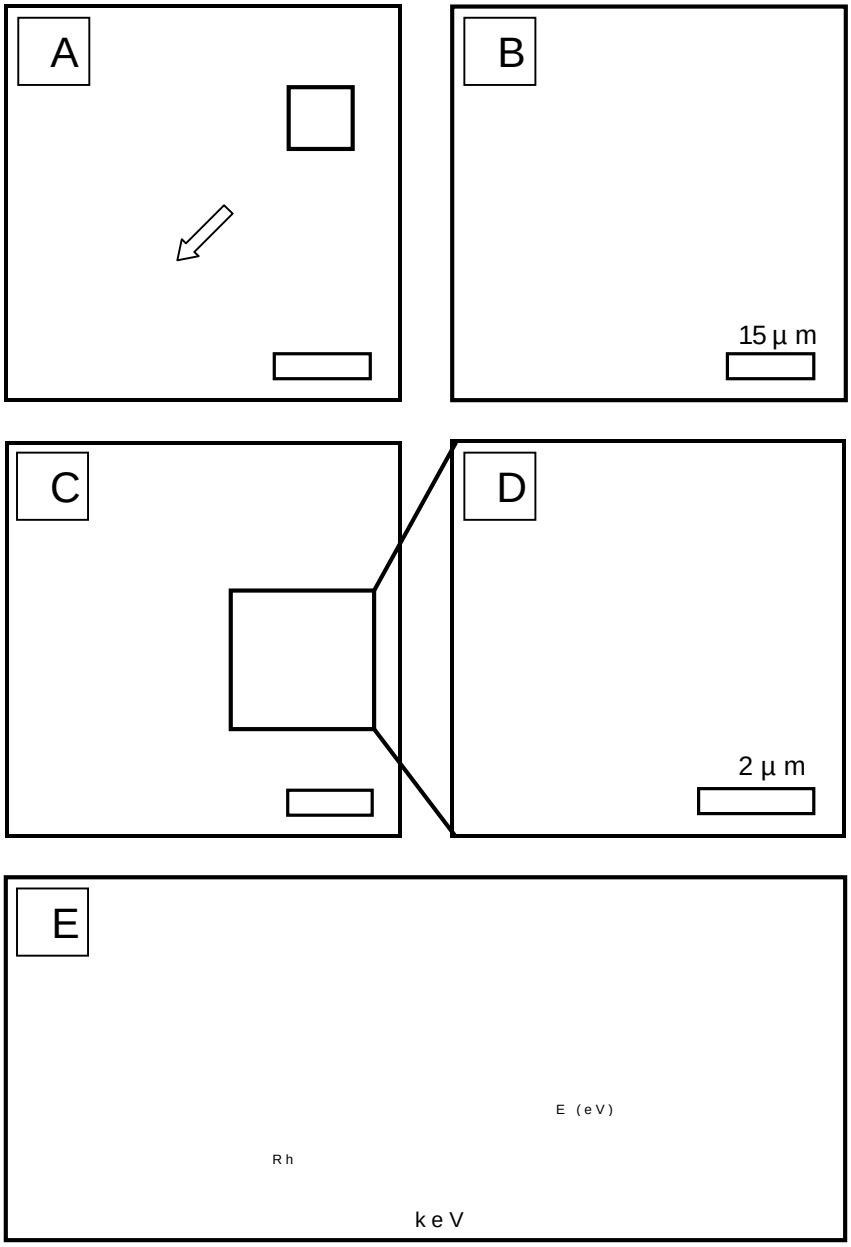

Figure 2. A: SEM micrograph of a heavily weathered platinu
micrograph of alloyed grain. Note the absence of deep e
oxides and cubic Cr oxides. C: SEM micrograph of squared area in A. A
acicular, iron oxides coats the surface of the grain; area not
C. The spectrum of the acicular, Fe oxide is presented in th
samples. The inset is the XANES spectrum of oxidised Fe^{3+}
indicates this nugget was in an aerobic weathering environ

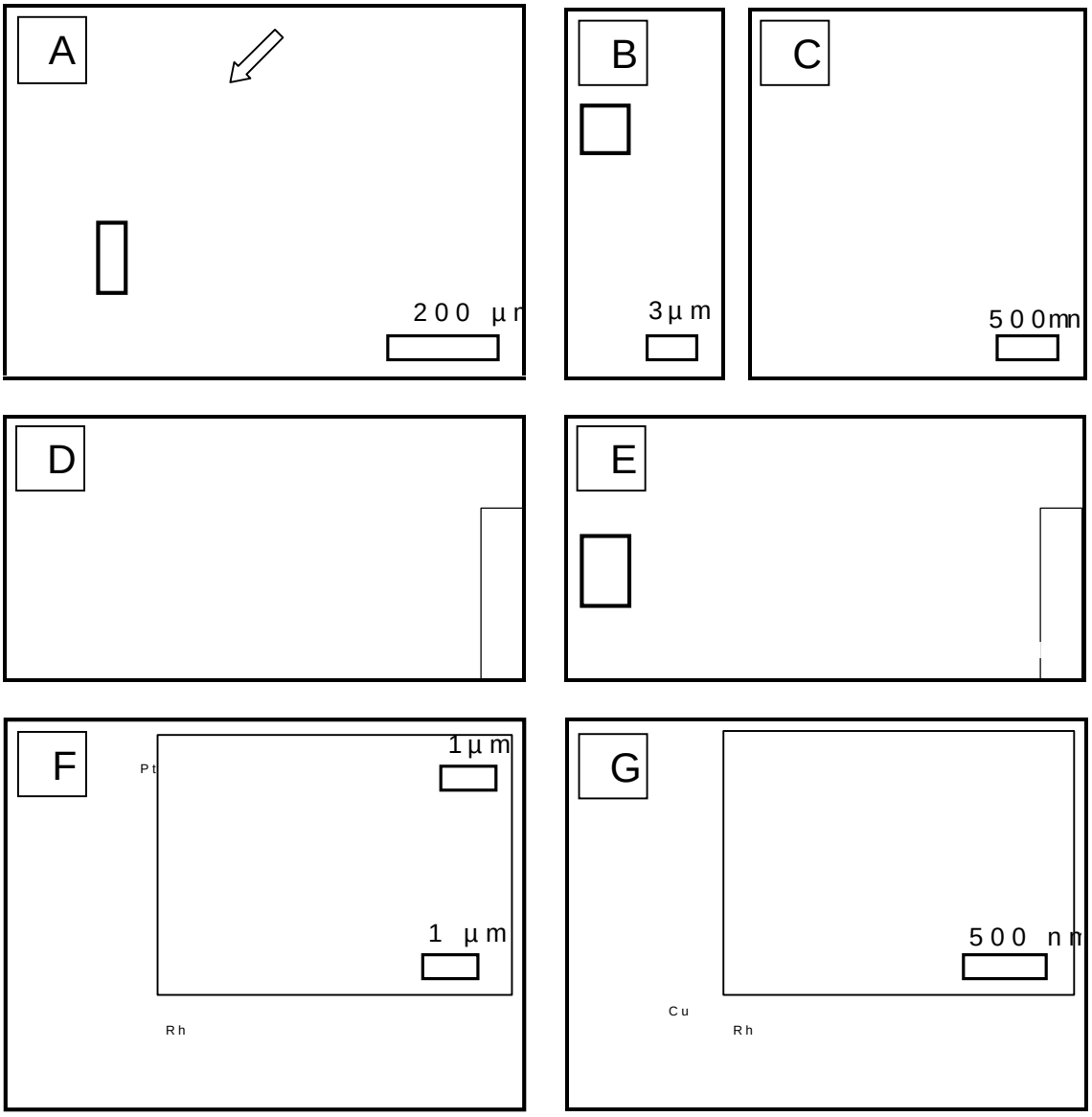

Figure 23. A: SEM micrograph of a weathered Bushveld magnetite grain. B: SEM micrograph of area noted in A. C: SEM micrograph of squared area in B. Note mineral in D and E. D and E: Iron and copper element map of grain in A, re rectangular area in E, which shows a streak of copper rich cubic mineral. F: SEM micrograph of cubic pits and E comparing the elemental composition of the bulk grain ar The bulk nugget is a Pt-Fe alloy (with trace Cu, Ni, and Rh) while t show elevated Cu. G: SEM micrograph and EDS of the acicular, iron oxides. of Ce. Scale bars in D and E show relative element co upward from low concentration (black) to high concentratio

the preferential weathering of osmiridium from the exsolution of osmiridium isoferroplatinum, as observed by Slansky et al. (1991) (Figures 2.3B and 2.3C). SEM shows 150 nm-sized cubic minerals embedded within the surface of the grain. The presence of these cubic minerals extending out from the surface of the platinum with similar sized holes in the surface (Figure 2.3F), suggests that they likely out from the grain, releasing Pt-Cu into the environment. Eventually, the removal of these nanophase minerals would occur over greater distances than grains and may contribute to the development of a larger-grade halo from these grains

Red-orange coatings found around the periphery of the grains were determined to be iron. Using SEM, acicular iron oxides were primarily found within the clay patches and directly affixed to the grain surface (Figures 2.2C and 2.2D and Figure 2.3G). The morphology of the iron oxides between grains is slightly differe suggests that an array of insoluble iron oxides possessing various hydration s these placer nuggets. Limonite, goethite and other iron oxides and found on platinum nuggets (Ottemann and Augustithis 19 al 2008). Energy dispersive spectroscopy (Figures 2.2E and 2.3E) and XANES spectra (inset Figure 2.2E) confirmed that iron existed in an oxidised Fe^{3+} state and was likely bound to oxygen. The K-edge occurred approximately 13 eV up en from the Fe^{0} reference standard, indicative of Fe^{3+} species (Das et al. 2004). The oxidation of iron promotes the formation of acid through t could contribute to platinum solubilisation, in particular (Gilbert 1986; the precipitation of acicular, iron oxides immediately on t

produced a local acidic interface that could have cont
dissolution and the release of additional iron from the gr
below) (Guilbert 1986).

The placer grains were determined to be a gold-silver alloy (Figure 2.3E) with
less amounts of copper (Figure 2.3F). An XRD analysis was performed on the gr
in Figure 2.3A in order to determine the mineralogy of the nugget, however
bulk nugget was sampled as a powder (Figure 2.3H), which is
consistent with Slansky (1991) and Gilbert et al. (1997). Trace amounts of nickel
and rhodium were also detected by EDS. Platinum was not detected by Slansk
al (1991) as being the most common mineral in the Fifie
mapping of the grain shown in Figure 2.3A was conducted. The Fe map in F
and the Cu map in Figure 2.3E clearly resemble the topography shown in the SE
micrograph. Element mapping of the grain shows that a
linked to morphological features on grains. The Cu map, i
copper highs (squared area) that correspond to minerals in the
Figure 2.3B and 2.3C. These cubic minerals have a higher coppe
bulk grain (Figure 2.3F) and their exposure at the nugget surface s
more resistant than the bulk grain as mostly selected by EDS
was found in small quantities using synchrotron radiation
found by EDS but not detected using synchrotron energy.
these elements cannot be definitively ruled out due to the 3
dimensional nature of the sample surface, which affects electron s
and detection. Nickel and copper, however, are known to r

iron in reducing environments (Melcher 2005). Levels of rhodium have been detected in Fifield placer grains (Slansky et al. 1991). Trace amounts of t earth element, cerium, were found to be associated with t reported by Andrew et al. (1995) as being present in some of the intrusive complexes.

Platinum-sulphide minerals were not observed in these gr consistent with prior studies (Slansky et al. 1991). The original magma m likely sulphide poor and any sulphur in the primary PGM s replaced during post-orogenic metasomatism or could have been weathered aw surface conditions (Johan et al. 1989; Andrew et al. 1995; Freyssinet et al. 2005; Sillitoe 2005). Hydrothermal alteration of primary platinum mi not explain why the platinum hosted in the Fifield source rock (undiscovered to date) micron sized while the alluvial placers are measured in millimetres or more thereo (Johan et al. 1989; Slansky et al. 1991; Teluk 2001). This increase of 1000 fold has been attributed to the dissolution and precipitation or subsequent accretion of pl alluvial and lateritic regimes by several groups, e.g., Cousins and Kinloch (19 Bowles (1986) and Bowles et al. (1994). However, the structural make-up and chem of these grains does not support the growth of platinum processes (Slansky et al. 1991). Only when the hard rock source of F is found will the diagenesis be better understood. For now, this study and similar studies clearly show signs of chemi at the nugget-solution interface. The grains are dissolving from the weathering products are being mobilised into the en where they can participate in a variety of abiotic, and potentially biotic Understanding

these reactions is key to improving our ability to recover and track its movement in nature, leading to more precise metal.

References

Anderson, J.A., 1982, Characteristics of leached capping
Titley, S.R., ed., Advances in geology of the porphyry co
North American: Tucson, University of Arizona Press, p. 275

Andrew, A.S., Hensen, B.J., Dunlop, A.C., and Agnew,
hydrogen isotope evidence for the origin of platinum mineralisati
Alaskan-type intrusions at Fifield, New South Wales, Australia: Geology, v. 19, p. 183

Azaroual, M., Romand, B., Freyssinet, P., and Disnar, J.,
aqueous solutions at 25°C and pHs 4 to 10 under oxidizin
Cosmochimica Acta, v. 65, p. 4545-4566.

Bowles, J.F.W, 1986, The development of platinum-group minerals
Economic Geology, v. 81, p. 1278-1285.

Bowles, J.F.W., Gize, A.P., and Cowden, A., 1994, The mo
elements in the soils of the Freetown Peninsula, Sierra
Mineralogy and Petrology, v. 52, p. 95-107.

Cabral, A.R., Beaudoin, G., Choquette, M., Lehmann, B.
Supergene leaching and formation of platinum in alluvium:
Gerais, Brazil: Mineralogy and Petrology, v. 90, p. 141-160.

Cabri, L.J., Harris, D.C., and Weiser, T.W., 1996, Mine
platinum-group mineral (PGM) placer deposits of the world:
Geology, v. 5, p. 147-167.

Cousins, C.A., and Kesler, S.E., 1976, Some observations on textures a
in alluvial platinoids: Economic Geology, v. 71, p. 1337-1338.

Enders, M.S., 2000, The evolution of supergene enrichme
Copper Deposit, Greenlee County, Arizona: Unpublished Ph.D. thesis, 516

Freyssinet, PH., Butt, C.R.M., and Morris, R.C., 2005, Ore
processes related to the latest Economic Geology 100th Anniversary
Volume, p. 267-281

Fuchs, W.A., and Rose, A.W., 1974, The geochemical behavior of platinum
palladium in the weathering cycle in the Stillwater Com
Geology, v. 69, p. 332-346.

Gray, D.R., and Foster, D.A., 2004, Tectonic evolution
southeast Australia: historical synthesis, and modern pers
Australian Journal of Earth Sciences, v. 51, p. 773

Guilbert, J.M., 1986, The geology of ore deposits: New
Company, 985 p.

Hanley, J.J., 2005, The aqueous geochemistry of the platinum
surficial, hydrothermal, and magmatic-hydrothermal environments
Mungall, J.E., ed., Exploration for platinum-group element deposits:
Association of Canada Short Course Series Volume 35
Association of Canada, p. 35

Johan, Z., Ohnenstetter, M., Slansky, E., Barron, L.M., an
mineralization in the Halleck Harbour ultramafic complexes near Fifield, Ne
Australia Part 1: Platinum-group minerals in the Kelvin
Prospect, Owendale Intrusion: Mineralogy and Petrology, v

Koek, M., Kreuzer, O.P., Maier, W.D., Porwal, A.K., Thomp
A review of the PGM industry, deposit models and exploration
for Australia's PGM potential: Resources Policy, v.

Kortright, J.B., and Thompson, A.C., 2001, The X-ray
A.C., and Vaughan, D., Eds., X-ray booklet: Lawrence Berkele
Laboratory, University of California, Section 1.2.

Macdonald, A.J., 1987, Ore deposit models #12: The plati
classification and genesis: Geoscience Canada, v.

Mann, A.W., 1984, Mobility of gold and silver in lateritic
observations from Western Australia: Economic Geology, v

Melcher, F., Oberthur, T., and Lodziak, J., 2005, Modifi
group minerals from the eastern Bushveld Complex, S
Mineralogy and Petrology, v.

Nixon, G.T., and Hammack, J.L., 1991, Metallogeny of u
British Columbia with emphasis upon the Alaskan-type mafic-ultramafic
ed., Ore deposits, tectonics, and metallogeny in the Canadian
British Columbia Ministry of Energy, Mines and Petroleum Resour
12-161.

O'Day, P.A., Rivera, N., Root, R., and Carroll, S.A.,
spectroscopic study of Fe reference compounds for the analysis of natural
American Mineralogist, v. 89, p. 572

Ottemann, J., and Augustithis, S., 1967, Geochemistry and origin of platinum-nuggets in lateritic covers from ultrabasic rocks and birbirites o... Deposita, v. 1, p. 269-277.

Ravel, B., and Newville, M., 2005, ATHENA, ARTEMIS, ... analysis for X-ray absorption spectroscopy using IFEFFIT: Jou... Radiation, v. 12, p. 537-541.

Reith, F., Lengke, M.F., Falconer, D., Craw, D., and S..., 2007, The geomicrobiology of gold: International Society of Microbia... 567-584.

Sillitoe, R., 2005, Supergene oxidized and enriched po... deposits: Economic Geology 100th Anniversary Volume, p. 723-768.

Slansky, E., Johan, Z., Ohnenstetter, M., Barron, L.M., an... mineralization in Alaskan-type intrusive complexes near Fifield, N.... Part 2. Platinum-group minerals in placer deposits at Fifield: Min... v. 43, p. 161-180.

Stumpfl, E.F., and Tarkian, M., 1976, Platinum genesis: N... Economic Geology, v. 71, p. 1451-1460.

Suárez, S., Prichard, H.M., Velasco, F., Fisher, P.C., ... Weathering of platinum-group minerals in the Aguablanca Ni-Cu (SW Spain): Revista de la Sociedad Española de Mineralogía, v... p. ...-238.

Teluk, A.J., 2001, Fifield Platinum Project, NSW, Aust... Technical Report (electronic copy available from <http://www.... PDF/Geodyne%20report%20complete%20with%20figs.pdf.> ... March 2012), 71 p.

Tolstykh, N.D., Sidorov, E.G., and Krivenko, A.P., 2005, Platinum-group element placers associated with Ural-Alaska type complexes, in Mungall, J.E., ed., Explorati... for platinum-group element deposits: Mineralogical Association of Canada ... Series Volume 35: Ottawa, Mineralogical Association of Ca..., p. ...-144.

Traoré, D., Beauvais, A., Auge, T., Parisot, J.C., Colin, F., ..., 2008, Chemical and physical transfers in an ultramafic rock w... Dissolution vs. accumulation of platinum group minerals: A... v. 93, p. 31-38.

Williams, G.P., 2001, Electron binding energies, in Thompson, A.C., and Vaughan, ... eds., X-ray Data Booklet: Lawrence Berkeley National Labo... California, Section 1.1.

Chapter 3

Immobilisation of palladium by metallidurans

3.1 Introduction

Microorganisms are able to thrive in a variety of extreme co
including strongly acidic and alkaline environments (Enders
Reith et al. 2009). Some metal tolerant bacteria in these environments
out a niche existence by using the accumulating compounds
sources of nutrition and energy (Reith et al. 2009). Alternatively, some bact
invoke genetically encoded detoxification mechanisms to
metal concentrations. These resistance mechanisms include reduction, comple
precipitation, efflux or a combination thereof (Silver 199
2009).

The biogeochemical cycling of metals and the formation
deposits can be attributed to microbial activity (e.g. Cloud
Mossman and Dyer 1985; Southam and Beveridge 1996; Reith
evidence from studies has shown that bacteria are able to precip
gold from solutions (Southam and Beveridge 1994, 1996; Reith et al. 2006, 2009;
Lengke et al. 2006c, d; Lengke and Southam 2007). The morph
precipitates so closely resemble the gold found in place
South Africa's Witwatersrand basin that the latter is now suspected to have a biogenic origin
(Wilson 1984; Mossman and Dyer 1985;

and Beveridge 1994; et al. 2006, 7). Reith The similar geochemical be
platinum and gold suggests that a comparable cycle for pla

Platinum, one of the six platinum group elements, is a
1987; Nixon and Hammack 1991). Its concentration in the
can reach economic concentrations in primary and secondar
Naldrett 2004; et al. 2010). Under surficial weathering conditi
mobile as Pt^{2+} or Pt^{4+}. It commonly forms complexes with chlorides
thiosulphates, but amorphous platinum oxide colloids
occur (Mountain and Wood 1988; Wood 1990; Wood 1990; Anthony
Williams 1994; Atz 2001; Hanley 2005; et al. 2006). Aqueous
platinum is transported in the surficial environment until
transformation or chemical precipitation. Subsequent deposition
platiferous material may occur (Bowles 1986; Wood 1990)
assumed primarily biotic, bacterially mediated platinum immobilisation
contribute to the formation of platinum placers (Reith et al. 2009a).

Lengke et al. (2006b) demonstrated that cyanobacteria are able to
aqueous Pt^{4+} species in a stepwise reaction that first produ
extracellular spherical platinum(IV) oxide nanoparticles. The amorphous p
(II) colloids experienced further diagenesis to produce cr
These experiments were recently supported by the disc
characteristic small alluvial platinum morphologies characteri
platinum-encrusted bacteria. Microbes attached to decaying organic
plant root) precipitated platinum from solution onto their

thought that continued bioreduction and subsequent electrochemical acc platiniferous nanoparticles likely contributed to the forma grains (Cabral et al. 2011).

Further investigation is still needed to understand t platinum biomineralisation. For this work, biogeochemically dive proven ability to immobilise gold should be reacted with characterise their response to platinum to determine wheth biogeochemical cycle of platinum complexes. This study will employ the use of Cupriavidus metallidurans CH34, formerly Wautersia metallidurans, Ralstonia metallidurans, Ralstonia eutropha and Alcaligenes eutrophus. C. metallidurans is an aerobic, gram-negative, facultative chemolithoautotrophic, rod-shaped proteobacterium known for being resistant to the toxic effects of a number of metal ions, e.g. Cu, Pb, Zn, Cd, Ag and Au (Mergeay et al. 1985, 2003; Reith et al. 2006, 2007 and Monchy et al. 2007). Resistance is primarily facilitated by metal-transporting ATPase efflux proteins in the cell envelope and cation reduction mechanisms in the cy, which have been shown to confer metal immobilisation (Nies 1999; Reith et al. 2005; Ledric 2007). This bacterium was found living on the surface of gold grain from auriferous soils in northern Australia (Reith et al. 2006, 2009). Reith cultured bacterium and reacted it with Au(III)-chloride solution. C. metallidurans rapidly precipitated gold within hours. This capacity of the bacterium to accumulate gold suggests that a multidisciplinary study of its Pt-chloride reactivity w species found in nature should produce comparable reactivi platinum.

3.2. Materials and methods

Cupriavidus metallidurans

Cupriavidus metallidurans strain CH34® (ATCC 43123™) acquired from the American Type Culture Collection in Manassas, Virginia ATCC® prescribed liquid medium containing 5 g/L peptone and 3 g/L beef extract (ingredients of Bacto® Nutrient Broth [Difco Laboratories; Detroit, Before experimentation, the culture was transferred (~10 stationary/early death phase) to 13 x 100 mm borosilicate (capped with plastic push caps to allow for free gas exchange and to prevent evaporation and contamination) and grown to early station maximise the amount of metabolically active biomass. The incubated at room temperature (21-23°C) under ambient O_2 conditions.

After incubation, separate batches in 50 mL Falcon tubes thoroughly vortexed (VWR Analog Vortex Mixer) to ensure a consis For the platinum immobilisation experiments, 1 mL aliquots of bacterial suspension were transferred to microcentrifuge tubes and harvested by centrifugation at 12,000 g for 5 min using a VWR Galaxy 16 microcentrifuge. After supernatant was decanted and cells were resuspended in 1 mL of filter-sterilised distilled, deionised (DDI) water. The cells were thoroughly vor remove any remaining culture medium. After vortexing, bacterial suspension centrifuged at 12,000 g for 5 min. The supernatant was discarded and the bacterial pellets resuspended in the experimental platinum solutions. The total number of bacteria in a washed sample was determined by the direct count

using a Petroff-Hausser Counting Chamber and a phase contrast light microscope (Zeiss microscop Fluorescent Z1 microscope).

C. metallidurans and aqueous platinum experiments

The bacterial experiments were conducted to exar C. metallidurans in the immobilisation of platinum from aqueous solutions of platin (II) chloride (K_2PtCl_4, Premion®, 99.99+% [metal basis], Pt 46.4% min; Alfa Aesar®, Ward Hill, Massachusetts USA) and platinum (IV) chloride ($PtCl_4$, Premion®, 99.99+% [metal basis], Pt 57% min; Alfa Aesar®). Platinum salts were dissolved in DDI at 16.2 MΩcm obtained from a Millipore system.

Washed C. metallidurans samples were suspended in 1 mL aqueous p solutions (from stock solutions of 0.5 µM [0.1 µg/mL], 5 µg/mL], 500 µM [100 µg/mL] and 5000 µM [1000 µg/mL] final Pt^{2+} or Pt^{4+} concentrations) at room temperature (21-23 °C) for 1 min, 1 h, 1 day, 2 weeks and weeks. Experiments were maintained in the dark because when exposed to light (Lengke et al. 2006). Microcentrifuge tubes contain bacteria-platinum mixture were only removed from darkness during longer exposure times. Reactions were performed in

Following exposure, reaction tubes were centrifuged X g for 5 min and the supernatant was removed for chemical analysis of residual platinum. The remaining bacterial pellets were washed 1 time by resuspension in filter-sterilised DD water to remove unreacted aqueous platinum and then centrifug X g for 5 min. Excess water was decanted and the pellet resuspended in 1 mL wa

filter-sterilised DDI water in preparation for whole mount microscopy or the examination of bacterial cells. Bacteria that were to be thin-sectioned were suspended in 2% glutaraldehyde rather than w

C. metallidurans aqueous Pt^{2+}-chloride and Pt^{4+}-chloride kill curves

The effect of aqueous Pt^{2+}-chloride and Pt^{4+}-chloride on the bacteria viability w determined using the spread plate method. Washed cells were resuspended in 1 mL of water and used in serial dilution with filter-sterilised DDI water. Each dilution was plated in duplicate onto beef extract-peptone agar plates that contain (in g/L): peptone, 5; beef extract, 3; agar, 15 (Difco Laborator for 4 days at room temperature (21-23°C) under ambient atmospheric conditions. Colony-forming units (CFU) were counted with the aid of a New Brunswick C-110 Colony Counter with digital counter and probe.

Chemical analyses

Platinum concentrations were measured over the cou using a Perkin-Elmer Optima 3000 DV System Inductively Coupled Pl Emission Spectrometer (ICP-AES). Instrumental uncertainty for measuring platinum was ≤5%, with a detection limit of 0.05 µM (0.01 µg/L). Samples were diluted as necessary with filter-sterilised DDI water (Table 3.1). Platinu calibration were prepared using the aqueous Pt^{2+} and Pt^{4+} stock solutions used in the laboratory-based C. metallidurans aqueous experiments. These calibr standards were diluted by the same factor as the corresp

(Table 3.1). The pH of the stock solutions was measured using a
Basic pH meter. The electrodes were calibrated in buffer
Analytical uncertainty of pH measurements is ±0.01 units
solutions after exposure to bacteria was monitored using the
ColorpHast® Bleeding Indicator strips.

Transmission electron microscopy (TEM)

Unstained whole sample mounts and thin sections (prior to
reaction with the Pt-tetracyanide) were examined using a
Phillips-10 CM transmission electron microscope (TEM) operated
ultra-thin sections). The whole mounts were prepared by flo
Sciences (EMS) Formvar coated 400 mesh copper grids on a drop of c
several minutes to allow the bacteria to adsorb to the grid
gentle dipping into a series of filtered deionized water. The grids were
completely air dry prior to microscopy.

Samples for thin sectioning were fixed overnight (4°C) in 2% glutaraldehyde
(EMS; Hatfield, Pennsylvania USA). Fixed cultures were s
14,000 X g for 1 min. The supernatant was discarded and
was enrobed in 2% (weight/volume) bacto-agar (Difco). Agar blocks were dehydr
using a 25%, 50% 75% and 3 X 100% acetone series (incu
The acetone was slowly replaced with a mixture of EMS Spurr epoxy resin
incubated at room temperature intervals in a 50%:50% [v/v], 25%:75% [v/v] a
acetone:epoxy resin series). Epoxy resin components were blended at a 2.5:2

Table 3. Dilution factors for Pt^{2+} and Pt^{4+} supernatants and standards. Dilutions were required because submitted samples contained more than 100 µg/mL (200 µg/mL), the linear portion of the standard curve.

Initial concentration of platinum	Dilution factor for ICP-AES
5000 µM or 1000 µg/mL	100x
500 µM or 100 µg/mL	10x
50 µM or 10 µg/mL	10x
5 µM or 1 µg/mL	10x

812, DDSA (Dodecenyl Succinic Anhydride) and NMA (Na
Samples were incubated overnight in 100% epoxy resin ar
with fresh epoxy resin containing 3% (2,4,6
tri(dimethylaminoethyl) phenol (DMP-30). Ratios by volume were 2.5:2:1:0
were cured in a 60°C oven (Blue M Electric Company Bac
Gravity Convection, 48 hrs.) until hard. Embedded shrimp eyes were ult
using a Reichert Ultracut E microtome and a Diatome" diamond
knife to a thickness of 70 nm and collected on 100 mesh copper ca
grids.

X-ray Absorption Spectroscopy (XAS)

XAS energy measurements from the platinum reference compounds
XANES (X-ray Absorption Near Edge Structure) and EXAFS
Absorption Fine Structure) were conducted at the Pacific Northwest
Science division (PNC-XSD) sector 20 beamline at the Advanced Photo
Argonne National Laboratory, Argonne, Illinois, USA. The oxidation state of plat
can be determined by XANES while information regarding
local coordination of platinum is provided by EXAFS data. D
state and ligand partners of platinum will shed light on how aqu
bacterial cells (approx. 2000 cells). XAS energy measurements were
collected from each sample and were calibrated to the in
11564 eV (Williams 2001).

Prior to any XAS measurements, C. metallidurans were reacted following the previously described "Cupriavidus metallidurans aqueous platinum experiments". For XAS, replicate reactions were harvested, i.e., collected by centrifuga and re-suspended in 200 µL water in order to concentrate the sample for detection. The reacted bacterial samples were then placed in acid-resistant teflon fluid cell with teflon-coated kapton film windows or pipetted directly onto Whatman® 42 Ashless Filter Papers. XAS measurements on the conducted in fluorescence mode.

Reference compounds were needed to determine the platinum in these reactions. Standards were selected to represent the most likely oxidation states and bonding partners of platinum expected in t reacted bacterial samples. The XAS spectra for model compounds inc (IV)-chloride ($PtCl_4$), platinum(IV)-cyanide ($K_2Pt(CN)_6$), platinum(II)-chloride (K_2PtCl_4), platinum(II)-cyanide ($K_2Pt(CN)_4$) and platinum(II)-tetraammine nitrate ($(NH_3)_4Pt(NO_3)_2$) were measured in aqueous form in liquid cells. Stocks (up to 65 mM) were prepared from solid compounds without pH XAS spectra for platinum(IV)-oxide hydrate ($PtO_2 \cdot xH_2O$), platinum-cis-(II) diamminedichloride (cisplatin; $Pt(NH_3)_2Cl_2$), platinum(II)-sulphide (PtS) and platin foil (Pt) were measured in solid form in transmission mode, as they are insoluble. With the exception of the foil and platinum(II)-sulphide, the standards were p teflon fluid cells with kapton film-windows. Platinum(II)-sulphide was and was concentrated onto Millipore™ 0.45 µm 13 mm membrane filters. compounds were commercially available from Alfa Aesar® and Sigma-Aldrich® (with

exception of platinum(II) chloride (PtCl$_2$) which was synthesized in the laborato
equal molar solutions of platinum(II) chloride (PtCl$_2$) with aqueous sodium
sulphide hydrate (Na$_2$S·9H$_2$O] (J.T. Baker®, Phillipsburg, New Jersey USA
produce black platinum(II) sulphide (PtS) precipitate. The EDTA reference spectrum o
platinum foil was simultaneously collected during measur
reacted, bacterial samples. Incident energy from the beam did not al
the standards or samples.

XAS data analysis

ATHENA analysis software was used to process and evaluate XAS
2005). Energy scans from samples and standards were calibrated
standard platinum foil reference energy. Multiple scans w
and were averaged to produce a single spectrum represen
and complexation state. XANES spectra were analyzed
comparing the energy position of the platinum edge in the
the energy location from the standards. Linear combination
mathematically identified which standard(s) aligned with
immobilized platinum. Energy position alignment correspon
the immobilized platinum (Lee and Korshin 2006). EXAFS data analysis was
in ARTEMIS, a XAS analysis software program complimen
and Newville 2005). Mathematical derivatives of post edge en
bound platinum in the bacterial samples were fitted to pos

platinum in the set alignment, normalized to platinum ch Peak align. standards,

indicated potential binding partners of the immobilized platinum (et al., 2006).

3.3 Results

Laboratory-based *C. metallidurans* aqueous platinum experiments

The addition of K_2PtCl_4 and $PtCl_4$ to filter-sterilised, DDI water promot hydrolysis of water, as indicated by a drop in the pH of the overall K_2PtCl_4 solutions was 5000 µM (1000 µg/mL; pH 4.0), 500 µM (100 µg/mL; pH 5.2), 50 µM (10 µg/mL; pH 5.8) and 5 µM (1 µg/mL; pH 6.1). The pH of $PtCl_4$ at 5000 µM (1000 µg/mL; pH 2.4), 500 µM (100 µg/mL; pH 3.0), 50 µM (10 µg/mL; pH 4.1) and 5 µM (1 µg/mL; pH 5.3). The pH of the platinum solution after reaction with b not change significantly from the pH of the solution. The addition of 5000 µM K_2PtCl_4 turned the pellet a grey-brown colour at 1 min, 1 hr, 1 day, 2 we exposure times. Similarly, the addition of 5000 µM $PtCl_4$ to the washed bacterial p immediately turned the white pellet yellow at all exposure were much fainter or not identifiable at lower concentrat provided macroscopic evidence that platinum was binding to the cells.

Table 3.2 and Table 3.3 were prepared by comparing count to the unreacted count for the appropriate exposure to a standard CFU count (5.0 x 10^8 CFU/mL) to allow comparison between exposur times and treatments. The experiments demonstrated that upon exposure of bacteria to 5000 µM Pt^{2+} and Pt^{4+}, no colonies grew. Platin toxicity was directly proportional to concentration and Pt^{2+} solutions being slightly less toxic than Pt^{4+} solutions for a given parti concentration.

Table 3.2. Toxicity study when *C. metallidurans* reacted with Pt^{2+} aqueous solutions. Increased exposure time and increased concent viable cells, as indicated by a decrease in CFU/mL.

Pt^{2+} concentrat	CFU/mL after 1 min expo	CFU/mL af 1 hr expos	CFU/mL af 1 day expo
0 µM or 0 µg/mL	$5.0 X 10^{8}$	$5.0 X 10^{8}$	$5.0 X 10^{8}$
0.5 µM or 0.1 µg/m	$4.9 X 10^{8}$	$2.7 X 10^{8}$	$4.6 X 10^{8}$
5 µM or 1 µg/mL	$3.7 X 10^{8}$	$3.2 X 10^{8}$	$4.3 X 10^{6}$
50 µM or 10 µg/mL	$4.1 X 10^{8}$	$1.3 X 10^{7}$	$2.1 X 10^{3}$
500 µM or 100 µg/m	$9.9 X 10^{6}$	0	0
5000 µM or 1000 µg/m	0	0	0

Table 3.3. Toxicity study when *C. metallidurans* reacted with Pt^{4+} aqueous solutions. Increased exposure time and increased concentration resu viable cells, as indicated by a decrease in CFU/mL.

Pt^{4+} concentrat	CFU/mL after 1 min expo	CFU/mL af 1 hr expos	CFU/mL af 1 day expo
0 µM or 0 µg/mL	$5.0 X 10^{8}$	$5.0 X 10^{8}$	$5.0 X 10^{8}$
0.5 µM or 0.1 µg/m	$4.5 X 10^{8}$	$3.2 X 10^{8}$	$3.3 X 10^{8}$
5 µM or 1 µg/mL	$4.9 X 10^{8}$	$2.5 X 10^{8}$	$7.6 X 10^{7}$
50 µM or 10 µg/mL	$1.1 X 10^{8}$	$6.4 X 10^{6}$	$4.8 X 10^{3}$
500 µM or 100 µg/m	$6.6 X 10^{2}$	0	0
5000 µM or 1000 µg/m	0	0	0

The amount of soluble platinum remaining in solution
AES. These results were used to determine the concentration of soluble platinum
mg dry weight C of metallic gold was averaged to determine the amount of platinum
immobilized. The mass of metallic gold was calculated using the mass
Escherichia coli (2.95×10^{-10} mg dry weight per cell), assuming
bacterium approximately ~23% larger by volume (Neu and Beveridge
al. 1990). Volume estimates of bacteria were based on diameter and
measurements of bacteria shown in the TEM micrographs
(Figures 3.1, 3.2, 3.3, 3.6). As rod shaped bacteria can be approximated by
bounded by two half spheres, the volume can be calculated
Bankston 1988).

$$V = 4/3(\pi r^3) + \pi r^2 h \qquad (1)$$

ICP-AES results are shown in Tables 3.4 and 3.5. I
immediately upon exposure to platinum. As exposure time incr
concentration, the amount immobilized also increased; how
at these longer exposure times the rate of platinum immobilization
effective cation exchange of platinum ions by bacteria, based on cell vo
exceed the T = 0 solution concentration in all reaction sys
C. metallidurans saturation was not reached except
bacteria ($5.000\ \mu M^2$) solutions.

Table 3.14: Immobilisation studies with live and dried *Cupriavidus metallidurans* CH34 exposed to Pt^{2+} solutions. Measured effective accumulation concentrations within the bacterial cells and ¼g Pt immobilised per mg dry weight of bacteria are shown. Increased exposure time resulted in increased immobilisation.

Pt^{2+} concentration	Immobilisation after 1 min exposure (mM; ¼g/mg)	Immobilisation after 1 h exposure (mM; ¼g/mg)	Immobilisation after 1 day exposure (mM; ¼g/mg)
5 µM or 1 µg/mL	1.80; 0.4	1.60; 0.4	153; 35
50 µM or 10 µg/mL	51; 13	44; 11	67; 16
500 µM or 100 µg/mL	120; 28	120; 29	410; 100
5000 µM or 1000 µg/mL	850; 210	800; 200	850; 210

Table 3.15: Immobilisation studies with live and dried *Cupriavidus metallidurans* CH34 exposed to Pt^{4+} solutions. Measured effective accumulation concentration within the bacterial cells and ¼g Pt immobilised per mg dry weight of bacteria are shown. Increased exposure time resulted in increased immobilisation.

Pt^{4+} concentration	Immobilisation after 1 min exposure (mM; ¼g/mg)	Immobilisation after 1 h exposure (mM; ¼g/mg)	Immobilisation after 1 day exposure (mM; ¼g/mg)
5 µM or 1 µg/mL	1.60; 0.4	2.60; 0.6	9.62; 2.4
50 µM or 10 µg/mL	112; 27	133; 32	409; 98
500 µM or 100 µg/mL	45; 11	160; 40	210; 51
5000 µM or 1000 µg/mL	310; 75	350; 84	870; 210

TEM

TEM micrographs of C. metallidurans exposed to platinum solution

presented in Figures 3.2 to 3.6. Micrographs of the cont

shown in Figure 3.1. Note that the cytoplasmic contents

3.1A. Furthermore, internal detail of the cell is lacking in thin sections as shown in

Figure 3.1B. The cell envelope is visible as a white halo

staining was applied to these bacteria; therefore, any dif

following micrographs are attributed to aqueous platinum.

Figure 3.2A demonstrates that bacterial staining oc

exposure to 5000 μM Pt^{2+}, 1 h. However, not all bacteria at 1 h were stained by

platinum. Generally, longer exposure times revealed more staining, especially a

poles (Figure 3.2B). Some bacteria produced nanometre-scale platinum colloids a

day (Figures 3.2C and D). Nanoparticles were not observed at short

Figure 3.3A shows that at 1 h, some bacterial cells did not bind platinum from

5000 μM Pt^{2+} solution. The majority of cells were stained wi

exposure time point. All bacteria at 1 h were observed to be cons

after 24 hours. Platinum colloid formation was observed at 1 day exposure

times (Figures 3.3C, E and F). Figure 3.3C shows a partially lysed

platinum nanoparticles bound in cytoplasmic material. Cell lysis is likely a response t

platinum toxicity and/or the pH of the solution. Thin section TEM micrographs o

bacteria exposed to 5000 μM Pt^{2+} at 1 h are shown in Figure 3.4. Platinum nanop

along the periphery of the cell wall, within the cytoplas

vesicles. C. metallidurans likely released membrane vesicles as a protective

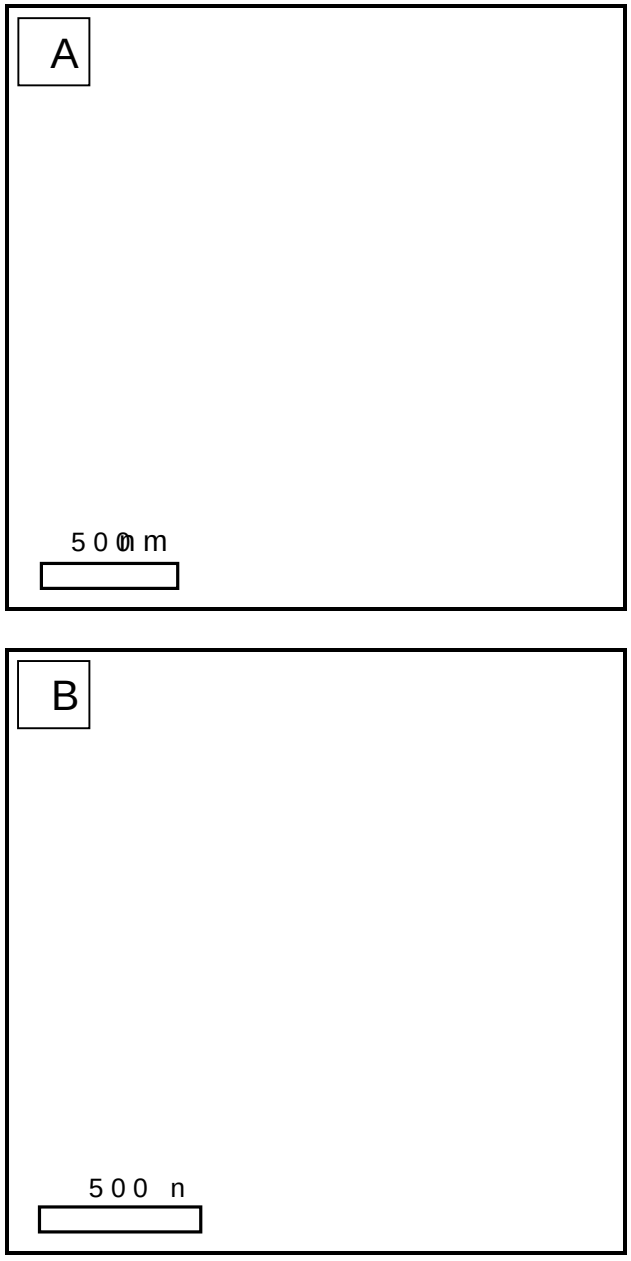

Figure 3A.1 Whole mount TEM micrograph of an unstained ultrathin
bacterium. Phosphate bodies within the cell are clearly v
generally electron translucent. B. Transverse ultrathin section TEM micrograp
unreacted metallic nitrates. Note the internal detail of cell is lacking
was applied.

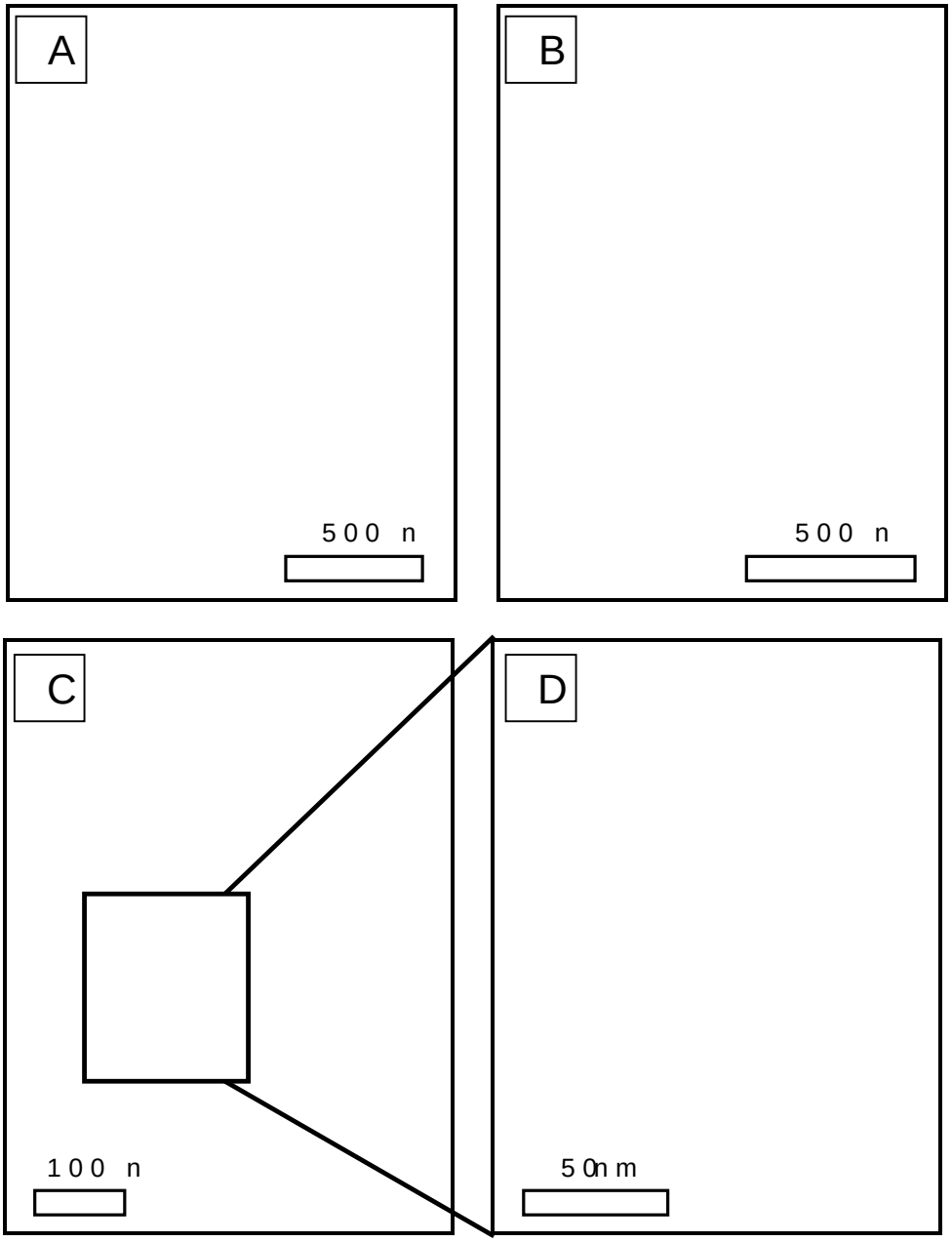

Figure 3.2 Whole mount TEM micrographs of cells treated with 500 µM $PtCl_4^{2+}$ (100 µg/mL Pt) for A: 1 min, B & C 1 day. D: Micrograph of squared area in C. At 1 day, all cells have internalized platinum nanoparticles (B) whereas some cells (C and D) do not.

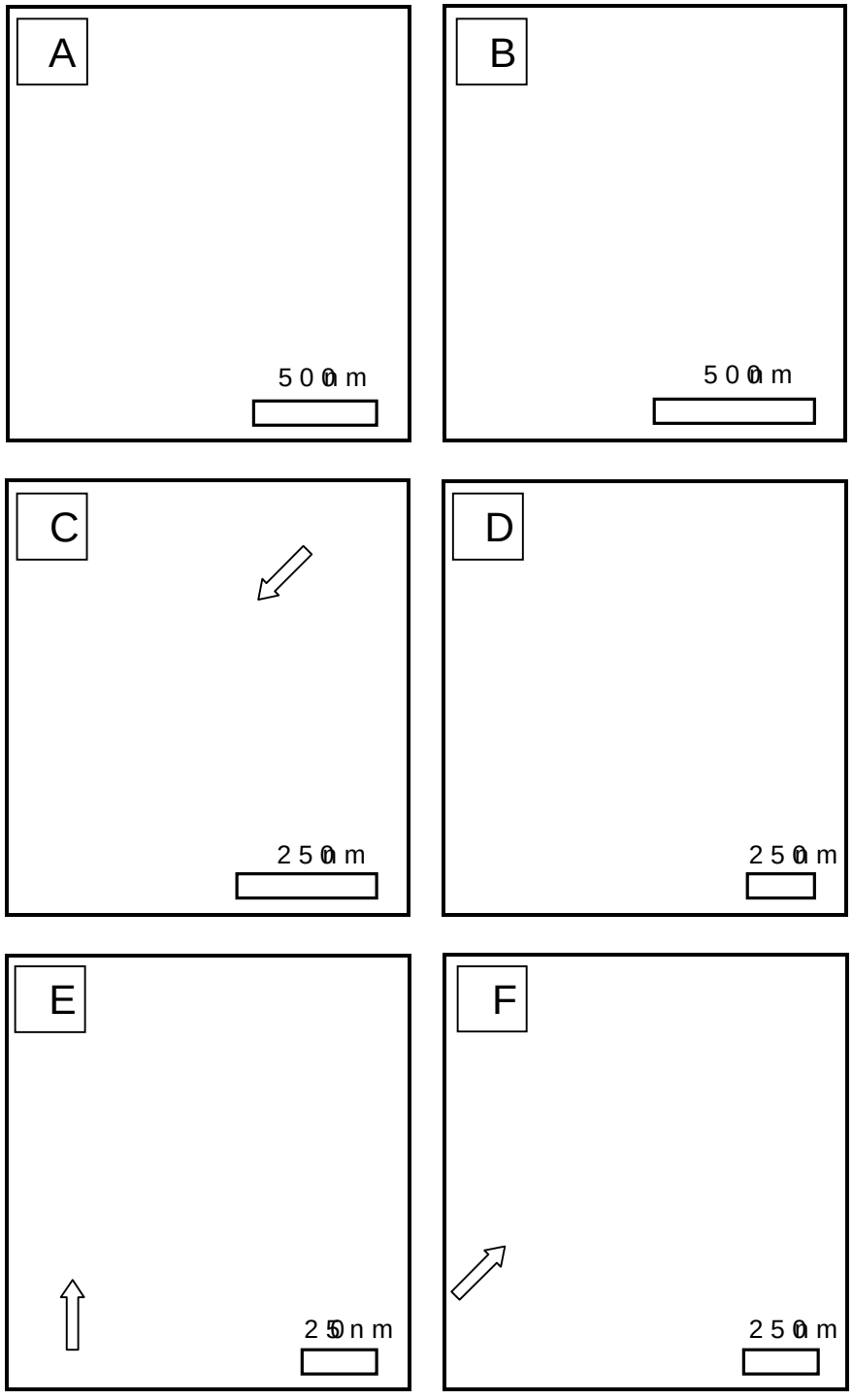

Figure 3. Whole mount TEM micrographs of cells exposed to 5000 µM Pt^{2+} (1000 µg Pt/mL) for A: 1 min. Binding of Pt is minimal as cell re electron parameters. B & C: 1 hr; D, E and F: 1 day. At longer incubation times cell staining is apparent in all cells. Some cells show nan indicated by arrows.

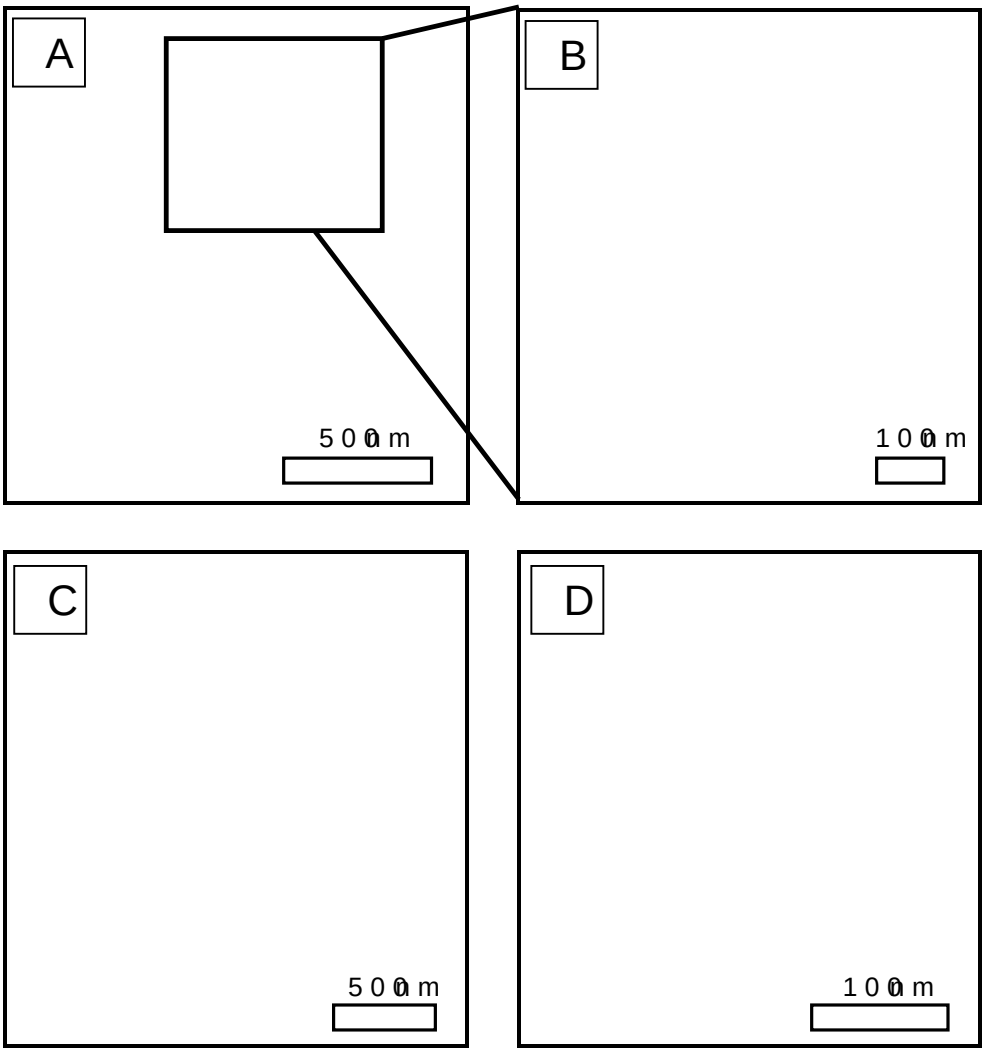

Figure 3.14: Ultrathin section TEM micrographs of a cell exposed to 5000 µM Pt^{2+} for 1 hr. A: Nanoparticle Pt along cell envelope. B: Micrograph of square area noted in A. C: Micrograph of immobilised Pt along cell envelope. D: Nanoparticle Pt immobilisation along the periphery of the membrane vesicle.

to keep toxic material away from the cell (Beveridge 2006, Beveridge 2007).

As indicated by the micrographs in Figure 3.5, neutralized and Pt^{4+} responded to solution much like it did to Pt^{2+} solution. Platinum appeared to be immediate but incomplete at (Figure 3.5A). Longer time exposure produced a darker stain and preferential at the bacterial cells pole nanoparticles were observed to precipitate within the cell (Figures 3.6A and B). Cell lysis was observed at 5.0 mM Pt^{4+} cytoplasmic material bound platinum-free. As with 5.0 mM Pt^{2+} solutions, cell lysis could have been due to the low pH of the solution, or a combination of both

It was noted that cells adhered to each other upon solution, perhaps as a result of neutralization of anionic charge groups envelope by palladinhydronium, allowing hydrophobicity to aggregation (Gilbert et al. 1990). This response could protect some interior by shielding them from the platinum exposure. Platinum immediate platinum was bound throughout cells.

XAS Spectra

XAS spectra in Figure 3.7 show the speciation and binding platinum immobilized by bacteria. Figure 3.7 is the XANES with 5000 µM (1000 mg/L) Pt^{2+} and Pt^{4+} for 4 hours. The peak edge energy of Pt^{2+} is slightly less than that of Pt^{4+}, indicating that electrons more tightly in the more oxidized platinum species. There

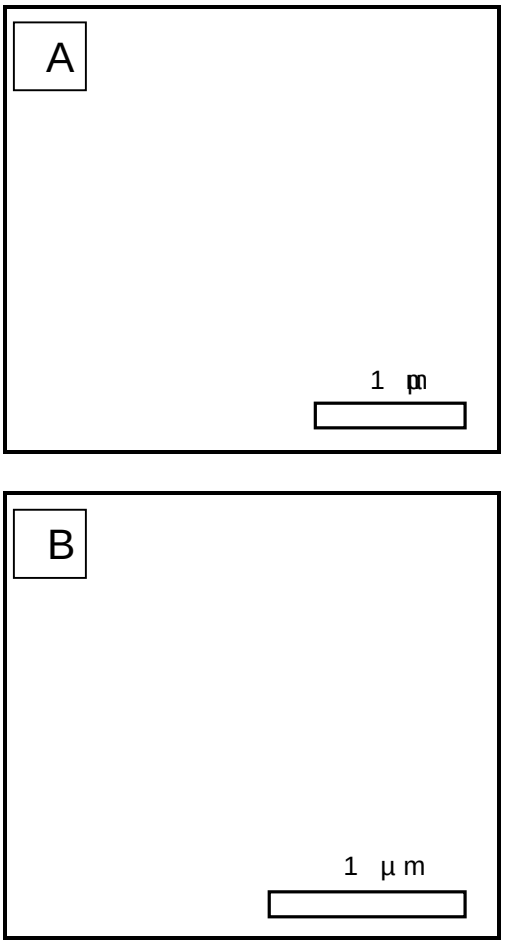

Figure 3.5 Whole mount TEM micrographs of cells treated with 500 µM Pt^{4+} (100 µg/ml) for A: 1 min; and B: 1 day. As incubation time increase

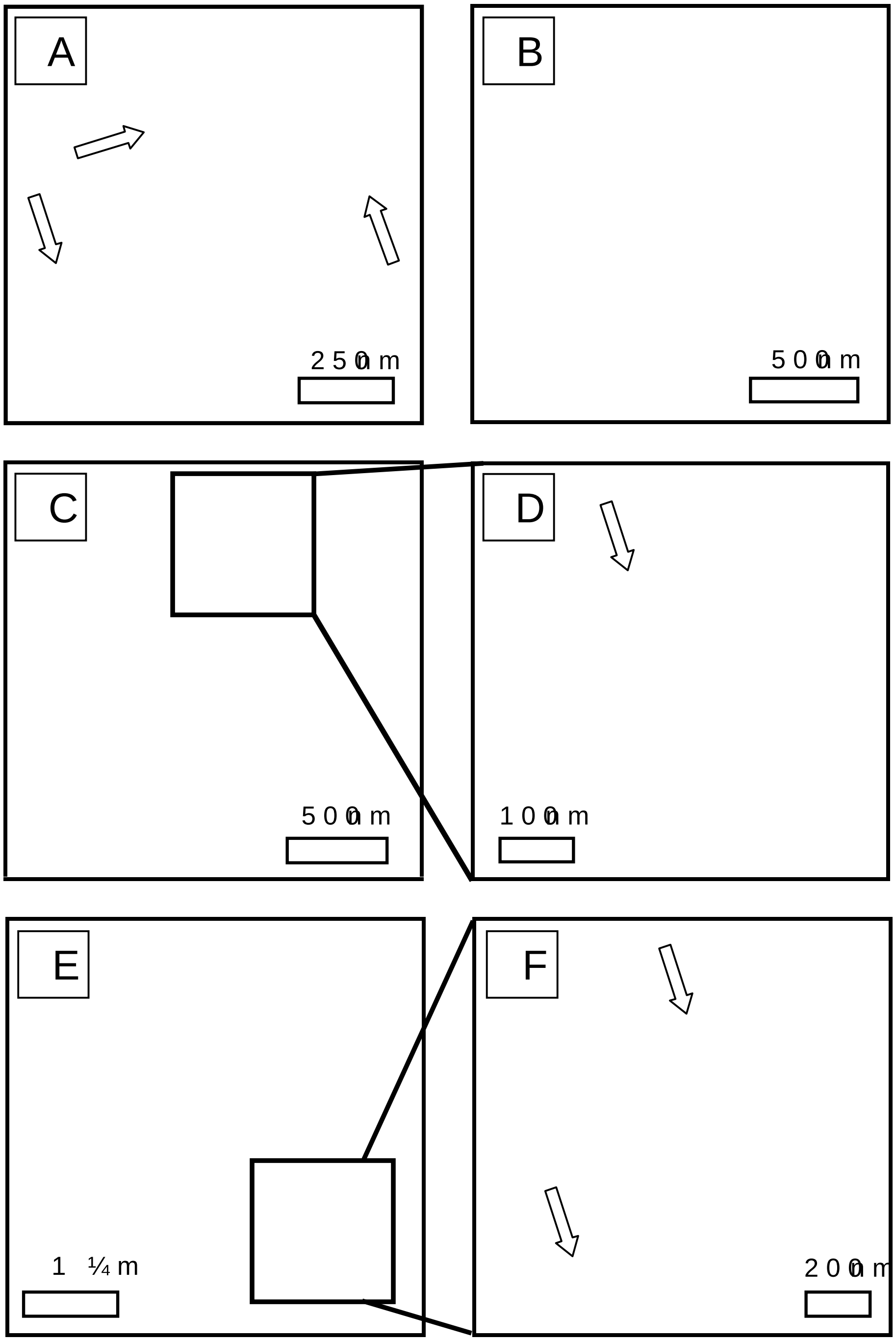

Figure 3.6: TEM micrographs of C. metallidurans treated with 5000 µM (1000 µg/mL) Pt^{4+}. A: 1 min (whole mount); B: 1 hr (thin section). Pt has been immobilised at the cell envelope. C & D: 1 hr (whole mount); E & F: 1 day (whole mount). Cells have lysed and cytoplasmic m possesses colloidal Pt. Arrows indicate immobilised Pt n becomes more pronounced at longer incubation times (C-F).

Figure 3.7: XANES spectra of bacteria reacted with 50 µM Pt^{2+} and Pt^{4+} (1000 µg/m) for 1 to ... days. The line corresponds to the Pt^{2+} energy. Note that the peak energy of Pt^{4+} is slightly higher energy than the Pt^{2+} peak due to the higher oxidation state. There is no downward shift of Pt^{2+} or Pt^{4+} over the 28 days, indicating that synchrotron energy did not

energy for either immobilised platinum species, indicatin did not detect a change in platinum oxidation over the experiments. It is important to note that at least 10% of the immobilised pla synchrotron radiation would detect the change in speciati are representative of the XANES spectra obtained at intervals (data not shown).

Figures 3.8 and 3.9 show the XANES and EXAFS spe with 5000 µM (1000 µg/mL) Pt^{2+} for 4 weeks. Similar spectra were o reactions with 5000 µM (1000 µg/mL) Pt^{4+} as well as with 500 µM Pt^{2+} and Pt^{4+} solutions (data not shown). Examination of the EXAFS demonstrated that the Pt–Cl fingerprint decreased in intensity over that the chlorine bound to Pt^{2+} was replaced by another binding partn indicates that the replacement was immediate, as the short between bonds occurred within 1 min; however, gradual i Pt^{2+} with substitution of chlorine continued over the 4 wee replacing chlorine is best matched to oxygen on a carboxyl grou indicative of an O–Pt bond on carboxyl groups is ~2.06Å, which com 2.07 Å and 2.08 Å bond distances (Alkhani 1983; Ackerman et al 2000; Moret et al 2009)

Pt-Cl fingerp

Figure 3.8. XANES and EXAFS spectra of bacteria reacted with Pt^{2+} solutions (1000 μg/mL) for 1
days. The arrow notes the Pt-Cl fingerprint, which decreases over time. There i
Pt^{2+} over the 28 days, indicating that synchrotron energy did not detec

Figure 3.9: EXAFS spectra of bacteria reacted with 5000 µM (1000 µg/ml) Pt^{2+} solution for up to 28 h. Arrows show the first shell binding of Pt-Cl and Pt-C/N/O. Note the Pt

3.4. Discussion

The interaction of C. metallidurans with $PtCl_2$ and $PtCl_4$ resulted in a variety of bacterial responses: cell death and cytolysis, plat nanoparticle formation, which were directly proportional to p time (Tables 3.2-3.5).

The literature lacks consistent experimental data on Pt^{2+} and Pt^{4+} complexation on Potential Pt^{2+} complexes present in the reactions system included $Pt(OH)_n^{2-n}$ and $PtCl_n^{2-n}$. Likely Pt^{4+} compounds include $Pt(OH)_n^{4-n}$ and $PtCl_n^{4-n}$, but very little data exists for these species (Bowles 1986, Mountain and Wood 1988; Wood et al. 1992; Azaroual et al. 2001; Colombo et al. 2008). EXAFS confirmed Pt-chloride to be the dominant aqueous species prior to reaction. The observed uptake of these Pt^{2+} and Pt^{4+} complexes from solution is consistent with the literature describing uptake of other metals, i.e., immobilisation by bacteria on the cell envelope a within the cytoplasm (Beveridge and Murray 1980; Nies 19

Platinum sorption occurred throughout the incubation and nanoparticle formation and immobilisation occurred within the cell. Cell surface reactivity controlled by the proton exchange capacity of carboxyl (R-COOH), phosphoryl (R-OPO_3H_2 and (R_2-PO(OH))), amine (R-NH_3^+) and hydroxyl (R-OH) functional groups on the membrane. In all but the 500 μM and 5000 μM Pt^{4+} reactions, most carbox groups would be deprotonated, negatively charged and avail reaction with oxidised platinum; phosphoryl, amine and hydroxyl groups would have remained mostly protonated and positively charged at the pH conditions consiste the acidity of platinum aqueous solutions used in this experiment (Beveridge and

Murray 1980; Fein et al. 1997; Guiné et al. 2006). XANES and EXAFS spectra cl
show the replacement of chlorine for oxygen on carboxyl
binding mechanism. It is worth noting that immobilised platinum occur
platinum-organo complexes at the cell surface and in the cy
organo-platinum complexes may be preferred to inorganic com
present. This process has been described in natural systems where carboxyl function
groups on soluble organic acids are able to react with pla
organo-complexes (Vlassopoulos et al. 1990; Wood 1990; Wood et al. 1992;
Kubrakova et al. 2011)

As suggested by Beveridge and Murray (1980), platinum bindir
envelope likely proceeded first as a stoichiometric event
platinum and then as a nucleation event, where nanopart
non-stoichiometric fashion, which continued at a slower pace. The fact it occur
progressive staining of the cells and formation of nanoparticles at inc
times and concentrations indicated that this process may
system. Platinum immobilisation was also observed in the cytoplasm. The rapid uptake
of platinum may have occurred because membrane transporter proteins
regulate the entry of these particular compounds, i.e. all complexes freely enter
cell driven by the osmotic ion gradient that existed across the en (Nies 1999)
Passive diffusion would have ceased once platinum nearly instanta
concentrations within the cell equal the platinum concentration within th
Aqueous platinum that entered the neutral pH of the cytoplasm

bound to deprotonated functional groups and complexed with carboxyl groups (Fein et al. 1997; Guiné et al. 2006).

Bacterial cells that remained viable following exposur[e to] platinum would have presumably initiated a biological resp[onse in] an attempt to survive. In metals inhibit the function of importa[nt] bind to important functional groups, or other important physi[ological] cations. Detoxification responses include chemical reduction or co[mplexation of the] metal to a less toxic state and/or the efflux of the metal out of the ce[ll]. These mitigation responses drain the cell of energy and can lead to oxidati[ve], causing continued cell death at longer exposure (Silver 1996; Nies 1999). XANES did not indicate platinum reduction; however, the appearance of nanometre-scale platinum colloids in the cytoplasm (and at the cell e[nvelope]) is evidence that some platinum reduction occurred.

The relationship between toxicity and immobilization [...] comparison of Tables 3.4 and 3.2 versus Tables 3.5 and 3.3. For a particular platinum species, immobilisation of platinum generally correlated with concomitant death of cells, e.g., the more platinum that was immobilised, the mo[re] [...] Resistance of C. metallidurans to pH 4.1, 50 µM Pt^{4+} solution demonstrates tha[t] toxicity experienced by the cells in the 2000 µM Pt^{2+} solution (pH 4.4) was primarily due to a platinum effect. Similarly, pH effect cell death was immedia[te] [...] µM and 5000 µM Pt^{4+} solutions, but C. metallidurans was exposed to pH [...] comparable solutions of HCl, corresponding [...] observed (data not shown). Intuitively, acidic solutions will induce some

results demonstrate that platinum immobilization trigger toxicity. It is important to note that dead cells cannot actively bind pl immobilisation occurred long after cells died; at the 5000 µM Pt^{4+} solution killed all bacteria within 1 minute and immobilization continued. Dead cells no longer metabolise and the proton motive force is lost, thus efflux pumps do not operate; consequently, there is less competition for platinum groups (Urrutia et al. 1992). The reactivity of metallic cations is known to be higher than the reactivity of the cells (Guiné et al. 2007). In this situation, the appearance of new reactive sites from denatured c cytoplasmic contents likely contributed to the continued even when cell death was instantaneous.

Further examination and comparison of Tables 3.2 to 3.5 revea differences between the Pt^{2+} reaction systems and Pt^{4+} systems. First, Pt^{2+} reactions were slightly less toxic than Pt^{4+}. While Pt^{2+}-chloride solutions were not as acidic as Pt^{4+}-chloride solutions, bacteria that remained viable following exp may have been able to neutralise and pump out Pt^{2+} cations easier than with Pt^{4+} via C. metallidurans divalent cation efflux ATPases likely in the (Liedrich 2005). bacterium could not the highly oxidised platinum, thus the attempt to neu it internally would have brought about a fatal oxidative stress (N et al. 2009). It is also important to note that the bacteria immobilize more platinum from Pt^{2+} solutions than from Pt^{4+} solutions. From a concentration and c character of platinum vs. hydronium, platinum in Pt^{2+} systems exceeded h by approximately 2 orders of magnitude and generally

concentration in the Pt^{4+} systems. Therefore, Pt^{2+} would have to compete with hydronium for carboxyl binding sites whereas Pt^{4+} would have more compe and therefore less opportunity to be bound and immobilise

References

Ankudinov, A.L., Ravel, B., Rehr, J.J., and Conradson, S.R., 2000, Hybridization p XANES: Chemical Physical Letters, v. 316, p. 495

Anthony, E.Y., and Williams, P.A., 1994, Thiosulfate com elements: Implications for supergene chemistry, in Alpers, C.N., and Blowes, D.W., and American Chemical Society, eds., Environmental geochem Washington, DC, ACS-Books, p. 551

Azaroual, M., Romand, B., Freyssinet, P., and Disnar, J., aqueous solutions at 25 °C and 1 bar under oxidizing conditions: Cosmochimica Acta, v. 61, p. 4-4566.

Baldwin, M., and Bankston, P., 1988, Measurement of li interference microscopy and steriologic methods applied to models: Applied and Environmental Microbiology, v. 54, p.

Beveridge, T.J., and Murray, R.G.E., 1980, Sites of metal Bacillus subtilis: Journal of Bacteriology, v. 141, p. 876

Bowles, J.F.W, 1986, The development of porous textures Economic Geology, v. 81, p. 1279-1285.

Cabral, A.R., Radtke, M., Munnik, F., Lehmann, B., Rein Tupinambá, M., and Kwitko, R., 2011, Iodine in alluvial plat nuggets: Evidence for biogenic fixation: Chemical Geology, 12-132.

Cloud, P., 1973, Paleoecological significance of the banded Geology, v. 1, p. 708-713.

Cloud, P., and Licari, G.R., 1968, Microbiotas of the sedi Proceedings of the National Academy of Sciences of the U 61, p. 779-786.

Colombo, C., Oates, C.J., Monhemius, A.J., and Plant, J platinum, palladium and rhodium and their distribution in the environment: Geochemistry: Exploration, Environment, Analysis,

Enders, M.S., Knickerbocker, C., Titley, S.R., and South bacteria in the supergene environment of the Morenci P Greenlee County, Arizona: Econ. Geology,

Fein, J., Daughney, C., Yee, N., and Davis, T., 1997, A ch metal adsorption onto bacterial surfaces: Geochimica et C 3319-3328.

Goldberg, S., Doyle, J., and Rosenberg, M., 1990, Mechanism microbial cell hydrophobicity by cationic polymers: Journa 5650-5654.

Guiné, V., Martins, J.M.F., Causse, B., Durand, A., Gaude Efect of cultivation and experimental conditions on the su resistant bacteria Cupriavidus metallidurans CH34 to protons, cadmium and Chemical Geology, v. 236, p. 266-280.

Guiné, V., Spadini, L., Sarret, G., Muris, M., Delolme, C., Gaudet, J.P., and M J.M.F., 2006, Zinc sorption to three gram-negative bacteria: combined titration modeling, and EXAFS study: Environmental Science and Tech 1813.

Hanley, J.J., 2005, The aqueous geochemistry of the platinum-group elements (PGE) surficial, low-T hydrothermal and high-T magmatic-hydrothermal environments Mungall, J.E., ed., Exploration for platinum-group element deposits: Mineralogical Association of Canada Short Course Series Volume 35 Mineralogical Association of Canada, p. 35-56.

Koek, M., Kreuzer, O.P., Maier, W.D., Porwal, A.K., Thomp A review of the PGM industry, deposit models and explora for Australia's PGM potential: Resources Policy, v. 35, p. 20-35.

Kubrakova, I.V., Fortygin, A.V., Lobov, S.G., Koshcheeva, and Mironenko, M.V., 2011, Migration of platinum, palladiu systems of platinum deposits: Geochemistry International, p. 1072-1084.

Ledrich, M., Stemmler, S., Laval-Gilly, P., Foucaud, L., and Falla, Precipitation of silver-thiosulphate complex and immobilisation Cupriavidus metallidurans CH34: BioMetals, v. 18, p. 643-650.

Lengke, M.F., and Southam, G., 2007, The deposition of elemental g thiosulfate complexes mediated by sulfate-reducing bacterial conditions: E Geology, v. 102, p. 109-126.

Lengke, M.F., Fleet, M.E., and Southam, G., 2006a, Morph synthesized by filamentous cyanobacteria from gold(I)-thiosulfate and gold(III) chloride complexes: Langmuir, v. 22, p. 2780-2787.

Lengke, M.F., Fleet, M.E., and Southam, G., 2006b,
nanoparticles by reaction of filamentous cyanobacteria with platinum(IV)-chloride
complex: Langmuir, v. 22, p. 7318-7323.

_____ 2006c, Bioaccumulation of gold by filamentous cyan
200°C: Geomicrobiology Journal, v. 23, p. 591-597.

Lengke, M.F., Ravel, B., Fleet, M.E., Wanger, G., Gordon, R.A., and Southam, G.,
2006d, Mechanisms of gold bioaccumulation by filamentou
(III)-chloride complex: Environmental Science and Technolog ... -6309.

Macdonald, A.J., 1987, Ore deposit models #12: The platinum group element deposits: classification and genesis: Geoscience Canada, v. 14, p. 155-166.

Mergeay, M., Monchy, S., Vallaeys, T., Auquier, V., Be
Taghavi, S., Dunn, J., van der Lelie, D., and Wattiez, R., Ralstonia metallidurans,
a bacterium specifically adapted to toxic metals: towards
responsive genes: FEMS Microbiology Reviews, v. 27, p. 3 ... -410.

Mergeay, M., Nies, D., Schlegel, H.G., Gerits, J., Charle
1985, Alicaligenes eutrophus CH34 is a facultative chemolitho-troph wi
bound resistance to heavy metals: Journal of Bacteriology ... -334.

Minter, W.E.L., Goedhart, M., Knight, J., and Frimmel, H
Witwatersrand gold grains from the Basal reef: evidence for their detrital or
Economic Geology, v. 88, p. 237-248.

Moret, M.E., Keller, S.F., Slootweg, J.C., and Chen, P.,
(II) complexes incorporating ^{2}-carboxylate ligands: Synthesis, structur
Inorganic Chemistry, v. 48, p. 6972-6978.

Mossman, D.J., and Dyer, B.D., 1985, The geochemistry ... -type gold
deposits and the possible influence of ancient prokaryo
dissolution and precipitation: Precambrian Research, v. 30, p. 303-319.

Mountain, B.W., and Wood, S.A., 1988, Chemical controls
and deposition of platinum and palladium in hydrothermal
approach: Economic Geology, v. 83, p. 492-510.

Mungall, J.E., and Naldrett, A.J., 2008, Ore deposits of the platinum-group elements:
Elements, v. 4, p. 253-258.

Neidhardt, F., Ingraham, J., and Schaechter, M, 1990, Phy
molecular approach: Sunderland, Massachusetts, Sinauer A

Nies, D.H., 1999, Microbial heavy-metal resistance: Applied Microbio
Biotechnology, v. 51, p. 730-750.

Nixon, G.T., and Hammack, J.L., 1991, Metallogeny of ultramafic-mafic rocks in
British Columbia with emphasis on the platinum-group elements, in McMillan, W.J.,
ed., Ore deposits, tectonics, and metallogeny in the Can
British Columbia Ministry of Energy, Mines and Petroleum Resour -4, p.
125-161.

Ohba, S., Sato, S., and Saito, Y., 1983, Electron-density distribution in crystals o
potassium tetrachloroplatinate (II) at 120 K and the influence of X-ray A
Crystallographica Section B, v. 39, p. 49-53.

Ravel, B., and Newville, M., 2005, ATHENA, ARTEMIS,
analysis for X-ray absorption spectroscopy using IFEFFIT: Journal of
Radiation, v. 12, p. 537-541.

Reith, F., Etschmann, B., Grosse, C., Moors, H., Benotm
Grass, G., Doonan, C., Vogt, S., Lai, B., Martinez-Criado, G., George, G.N.,
D.H., Mergeay, M., Pring, A., Southam, G., and Brugger, J., 200
biomineralization in the bacterium Cupriavidus metallidurans: Proceedings of the
National Academy of Sciences, v. 106, p. 17757-17762.

Reith, F., Lengke, M.F., Falconer, D., Craw, D., and Southam, G., 2007, Winog
Review—The geomicrobiology of gold: International Society
Journal, v. 1, p. 567-584.

Reith, F., Rogers, S.L., McPhail, D.C., and Webb, D., 200
Biofilms on bacterioform gold: Science, v. 313, p. 233-235.

Silver, S., 1996, Bacterial resistance to toxic metal ions - a review: Gene, v. 179, p. 9-
19.

Southam, G., and Beveridge, T.J, 1994, The in vitro for
bacteria, Geochimica et Cosmochimica Acta, v. 58, p. 4527-4530.

____ 1996, The occurrence of sulfur and phosphorus w
crystalline and pseudocrystalline octahedral gold forme
Cosmochimica Acta, v. 60, p. 4369-4376.

Urrutia Mera, M., Kemper, M., Doyle, R., and Beveridge, T.J., 1992, The
induced proton motive force influences the metal binding a
Bacillus subtilis cell walls: Applied and Environmental Microbiology, v. 58, p. 3837-3844.

Vlassopoulos, D., Wood, S.A., and Mucci, A., 1990, Gold speciation in natural waters: I. Solubility and hydrolysis of gold in aqueous solutions: Geochimica et Cosmochimica Acta, v. 54, p. 1575–1586.

Williams, G.P., 2001, Electron binding energies, in Thompson, A.C., and Vaughan, D., eds., X-ray Data Booklet: Lawrence Berkeley National Laboratory, California, Section 1.1.

Wilson, A.F., 1984, Origin of quartz-free nuggets and supergene gold in lateritic regoliths: a review and some new observations from Australia: Australian Journal of Earth Sciences: An International Geoscience Journal of the Geological Society of Australia, v. 31, p. 303–323.

Wood, S.A., 1990, The interaction of dissolved platinum with simple organic acid analogues in aqueous solutions: Mineralogical Magazine, v. 54, p. 625–636.

Wood, S.A., Mountain, B.W., and Pan, P., 1992, The aqueous geochemistry of platinum, palladium and gold; recent experimental constraints and a re-evaluation of theoretical predictions: The Canadian Mineralogist, v. 30, p. 955–982.

Chapter 4

Conclusions

The traditional notion that platinum is inert has bee biogeochemical paradigm (Anthony and Williams 1994). transport of platinum from the mantle to the crust and subsequent weat of PGM-bearing host rocks is an incomplete metallogeny of Subsequent-occurring dissolution, transportation and precip biological and abiotic processes, potentially over geologic time, are now cons be key components in placer platinum formation (Fuchs and Rose 1974; Bowles Azaroual et al. 2001; Hanley 2005; Reith et al. 2009). The structural and c examination of platinum grains, revealing features, combined with biogeochemistry of candidate platinum compounds have de more to learn about the mobility of platinum in natural sy has highlighted the importance of organo-platinum compounds in platinum exploration, which could lead to applications for

With respect to the current controversy regarding precipitation and growth of platinum grains (Traoré et al. 2010), this highlighted work on Australian platinum grains, which provides clea platinum found in natural systems ultimately comes from material, supports the dissolution on a micrometre scale. The surficial feat highlight the importance of examining platinum grains in secondary imaging before, more classic, cross polishing to

mineral grains. Deep weathering scars, pits and cavities
of platinum grains are important sites of mechanical and
weathering. SEM micrographs show delicate, 10 micron scale
that could be lost by polishing and wouldn't be as obvious
imaging, which does not achieve the same resolution as
dissolution of the bulk nugget around these minerals demo
homogenous, reflecting the geological conditions of the
minerals once left in the regolith or the alluvium. The pre
oxides provides evidence that acidic conditions occur a
presents a possible mechanism for chemical dissolution
(Guilbert 1986, 2005).

For future work on this system, the next step in chara
from Fifield requires that these polished grains be examined using the same tech
as was used to study their surfaces. Any weathering link b
features and the bulk interior of the nugget needs to
heterogeneity, from the periphery of platinum grains to
preferential weathering of the bulk grain (Chaurand et al. 2007; cubic
Traore et al. 2008). Synchrotron element mapping would be a good method
interior morphological features noted in microscopy wit
Element mapping would likely require a synchrotron spot size of 100
detect the chemical gradient from surface to interior and
and precipitation processes affecting platinum grains.

A large number of microorganism have been implicate
cycling of metals at near surface or surface conditions (Southam and
Reith et al. 2009; Southam et al. 2009). The precipitation of secondary
bacteria has been well documented (Southam and Beverid
2006, 2009; Lengke et al. 2006c, Lengke and Southam 2007). A c
microbe-mediated immobilisation of platinum was hypothesize
chemical similarity of gold and platinum, but did not occur
et al. 1990; Cabral et al. 2007). Cupriavidus metallidurans, a known metal
resistant soil bacterium that has been implicated in the biochemical cycling of
(Reith et al. 2006; 2009), immobilised appreciable amounts, up to 1% of pla
quantities by mass (see Tables 3.4 and 3.5), transformed the platinum into
appreciable amounts of secondary platinum. TEM clearly d
immobilised at the cell envelope and within the cytoplasm
immobilisation was determined by synchrotron radiation
(existing as platinum chloride) preferentially bound to exposed carboxyl oxyge
groups on the cell wall and within the cytoplasm (Bowles, Beverid
1986, Mountain and Wood 1988; Fein et al. 1997; Azzaro et al.
2001; Guiné et al. 2006). Although platinum reduction was no
synchrotron methods, the reduction of at least some of th
particles of elemental platinum was observed. Due to the number
ubiquitous nature of bacteria in near-surface environments and the h
both Pt^{2+} and Pt^{4+} for organically derived carboxyl functional g
suggests that microorganisms must be important in natural platinum system

bearing materials are exposed to weathering conditions, platinum in association with organic acids and the biosphe

Work into understanding the biosphereise, the influence biogeochemical cycle, one needs to continue. Recommendatio experiment include Ce. exposing pure aqueous platinum species fo periods of time (months) to promote and examine platinum et al (2006a) exposed cyanobacteria to platinum for up to a mo reported minor carbon platinum particles. Given that consortia of other microbes should be tested for their ability to immob Likely, many bacteria are able to actively precipitate platinu combined presence in weathering profiles may provid transformation. Also, bacteria interactions are likely not re immobilisation of aqueous platinum species, analysis the weath platinum-bearing material in primary ore deposits. The sulph Acidithiobacillus thiooxidans and sulphur Acidithiobacillus ferrooxidans contribute to the supergene alteration of sulphides via me oxidation and sulphuric acid production (Stumm 1970; Lizama a 1985; Sillitoe 2005; Roberts et al 2006). As platinum-group minerals are the main PGE most world's platinum deposits, including those of the Southern African Bushveld Igneous Complex, bacteria may be able to enhan the host rocks (Macdonald 1987; Teluk 2001).

The current value of platinum has nearly matched gold highs over the last few years. Continued exploration and min

these prices and companies will look for more cost-effective recovery and processing techniques to boost their bottom lines. The use of microbes to recover once inaccessible metals dissolved in mine waste rock piles could increase efficiency for mining companies. Furthermore, along with metals li and precious metals, microbes may one day be used as bio platinum enrichment (Corbisier et al. 1997; Reith 2009).

References

Anthony, E.Y., and Williams, P.A., 1994, Thiosulfate com... elements: Implications for supergene chemistry, in Alpers, C.N., Blowes, D.W., eds., Environmental geochemistry of s... American Chemical Society, Washington, DC, ACS Books, p. 551-560.

Azaroual, M., Romand, B., Freyssinet, P., and Disnar, J., ... aqueous solutions at 25°C and pHs 4 to 10: ... Geochimica et Cosmochimica Acta, v. 65, p. 4453-4466.

Beveridge, T.J., and Murray, R.G.E., 1980, Sites of metal ... Bacillus subtilis: Journal of Bacteriology, v. 141, p. 876-887.

Bowles, J.F.W, 1986, The development of platinum-group minerals in lateri... Economic Geology, v. 81, p. 1278-1285.

Cabral, A.R., Beaudoin, G., Choquette, M., Lehmann, B. ... Supergene leaching and formation of platinum in alluvium: ... Gerais, Brazil: Mineralogy and Petrology, v. 90, p. 141-150.

Corbisier, P., 1997, Bacterial lux-sensors for a rapid determin... heavy metal bioavailability and toxicity in solid samples: ... 148, p. 533-534.

Enders, M.S., Knickerbocker, C., Titley, S.R., and Southam, G., 2... bacteria in the supergene environment of the Morenci P... Greenlee County, Arizona: Economic Geology, v. 101, p. 59-70.

Fein, J., Daughney, C., Yee, N., and Davis, T., 1997, A chemical equilibrium mode... metal adsorption onto bacterial surfaces: Geochimica et C... 3319-3328.

Guiné, V., Spadini, L., Sarret, G., Muris, M., Delolme, C. ... J.M.F., 2006, Zinc sorption to three gram-negative bacteria: combined t... modeling, and EXAFS study: Environmental Science and T... 1806-1813.

Fuchs, W.A., and Rose, A.W, 1974, The geochemical be... palladium in the weathering cycle of the Stillwater Complex, Montana: Economic Geology, v. 69, p. 332-346.

Guilbert, J.M., 1986, The geology of ore deposits: New ... Company, 985 p.

Hanley, J.J., 2005, The aqueous geochemistry of the platinum-group elements (PGE) i surficial, low-T hydrothermal and high-T magmatic-hydrothermal environments Mungall, J.E., ed., Exploration for platinum-group element deposits: Mineralogical Association of Canada Short Course Series Volume 35 Association of Canada, p. 35-56.

Lengke, M.F., and Southam, G., 2007, The deposition of elem thiosulfate complexes mediated by sulfate-reducing bacteria conditions: E Geology, v. 102, p. 109-126.

Lengke, M.F., Fleet, M.E., and Southam, G., 2006a, nanoparticles by reaction of filamentous cyanobacteria chloride complex: Langmuir, v. 22, p. 7318-7323.

____ 2006b, Bioaccumulation of gold by filamentous cyan 200°C: Geomicrobiology Journal, v. 23, p. 591-597.

Lengke, M.F., Ravel, B., Fleet, M.E., Wanger, G., Gordo 2006c, Mechanisms of gold bioaccumulation by filamentou (III)-chloride complex: Environmental Science and Technolog 6309.

Lizama, H.M., and Suzuki, I., 1989, Rate equations and kine reactions involved in pyrite oxidation by Thiobacillus ferrooxidans: Applied and Environmental Microbiology, v. 55, p. 2918-2923.

Macdonald, A.J., 1987, Ore deposit models #12. The platinum group element deposits: classification and genesis: Geoscience Canada, v. 14, p. 155-166.

Mountain, B.W., and Wood, S.A., 1988, Chemical controls and deposition of platinum and palladium in hydrothermal approach: Economic Geology, v. 83, p. 492-510.

Reith, F., Etschmann, B., Grosse, C., Moors, H., Benotm Grass, G., Doonan, C., Vogt, S., Lai, B., Martinez-Criado, G., George, G.N., D.H., Mergeay, M., Pring, A., Southam, G., and Brugger, J., 2009, Mecha biomineralization in the bacterium Cupriavidus metallidurans: Proceedings of the National Academy of Sciences, v. 106, p. 17757-17762.

Reith, F., Rogers, S.L., McPhail, D.C., and Webb, D., 2006, Biomineralization of gold: Biofilms on bacterioform Gold: Science, v. 313, p. 233-235.

Sillitoe, R., 2005, Supergene oxidized and enriched po deposits: Economic Geology, 100th Anniversary Volume, p. 723-768.

Singer, P.C., and Stumm, W., 1970, Acidic mine drainage: The rate-determining step: Science, v. 167, p. 1121-1123.

Southam, G., and Beveridge, T.J, 1994, The in vitro for
bacteria, Geochimica et Cosmochimica Acta, v. 58, p. 4527-4530.

_____ 1996, The occurrence and significance of phosphorus within bacteri
crystalline and pseudocrystalline octahedral gold forme
Cosmochimica Acta, v. 60, p. 4369-4376.

Southam, G., and Saunders, J.A., 2005, The geomicrobiolo
Geology, v. 100, p. 1067-1084.

Southam, G., Lengke, M.F., Fairbrother, L., and Reith, F.
of gold: Elements, v. 5, p. 303-307.

Teluk, A.J., 2001, Fifield Platinum Project, NSW, Aust
Technical Report (electronic copy available from <http://www.ri
PDF/Geodyne%20report%20complete%20with%20figs.pdf.>
March 2012), 71 p.

Traoré, D., Beauvais, A., Auge, T., Parisot, J.C., Colin, F
Chemical and physical transfers in an ultramafic rock weathering
Dissolution vs. accumulation of platinum group minerals: A
p. 303-318.

Vlassopoulos, D., Wood, S.A., and Mucci, A., 1990, Gold
II. The importance of organic complexing with some simple
ligands: Geochimica et Cosmochimica Acta, v. 54, p. 1575-1586.

Wood, S.A., Mountain, B.W., and Pan, P., 1992, The a
platinum, palladium and gold; recent experimental constraints
theoretical predictions: The Canadian Mineralogist, v. 30, p. 955-982.

Appendix Chapter 2

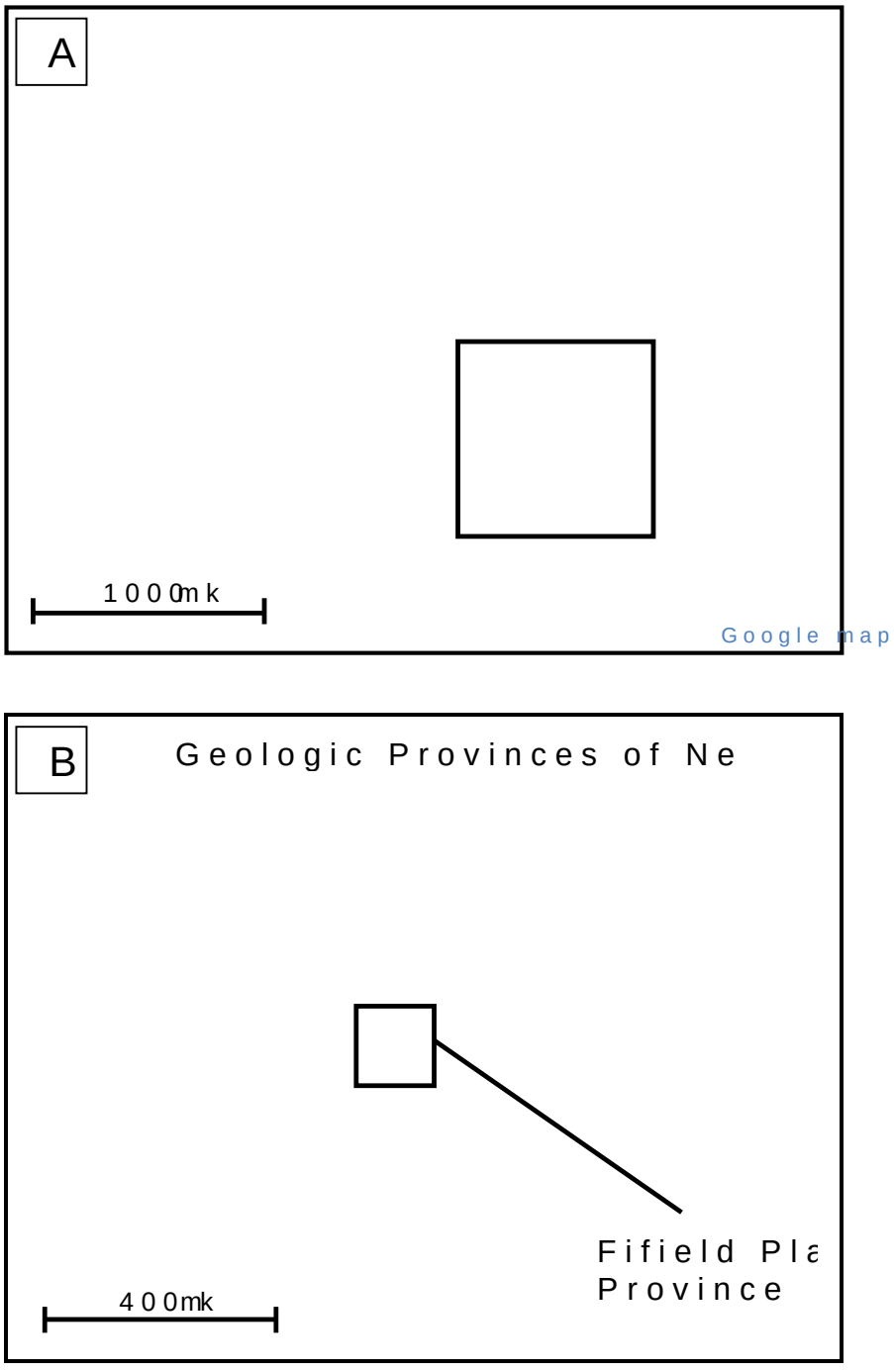

Figure A.1 A: Map of Australia with state of New South Wales B: South West Slopes and Central West provinces of New South Wales and squared area noting the Fifield Platform Province (Xie et al. 2009)

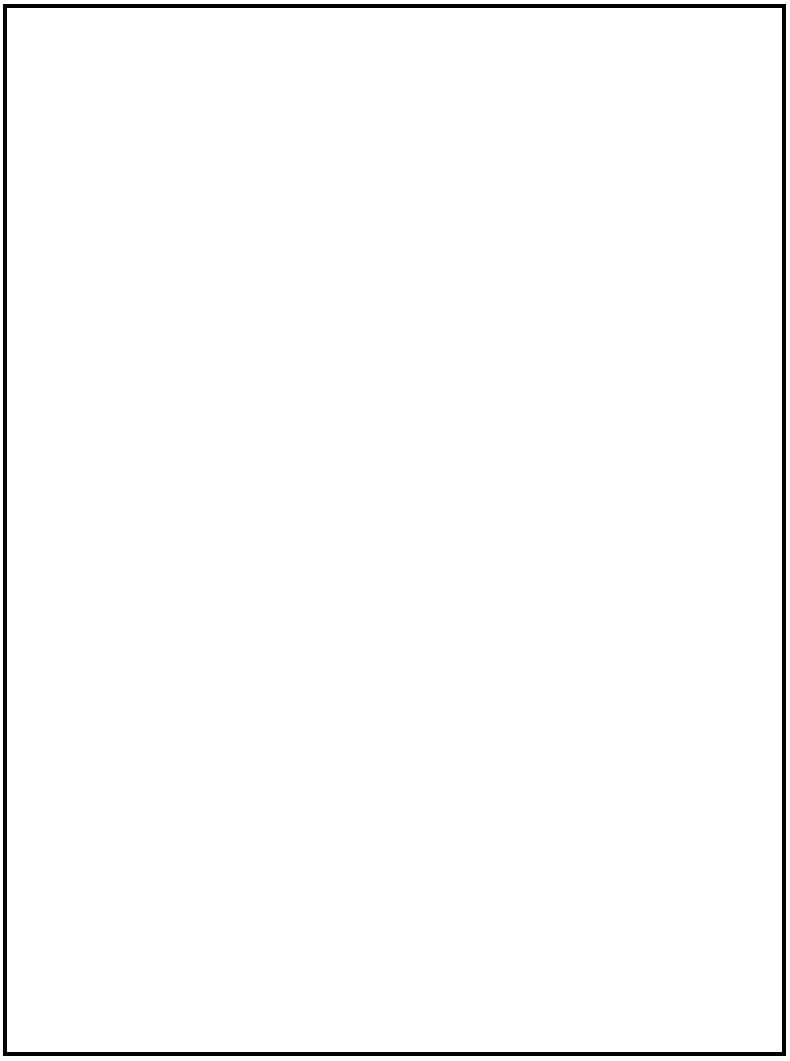

Figure A.2 Schematic profile of Fifield Platinum Province la
Platinum is found in high grade goethite zone.

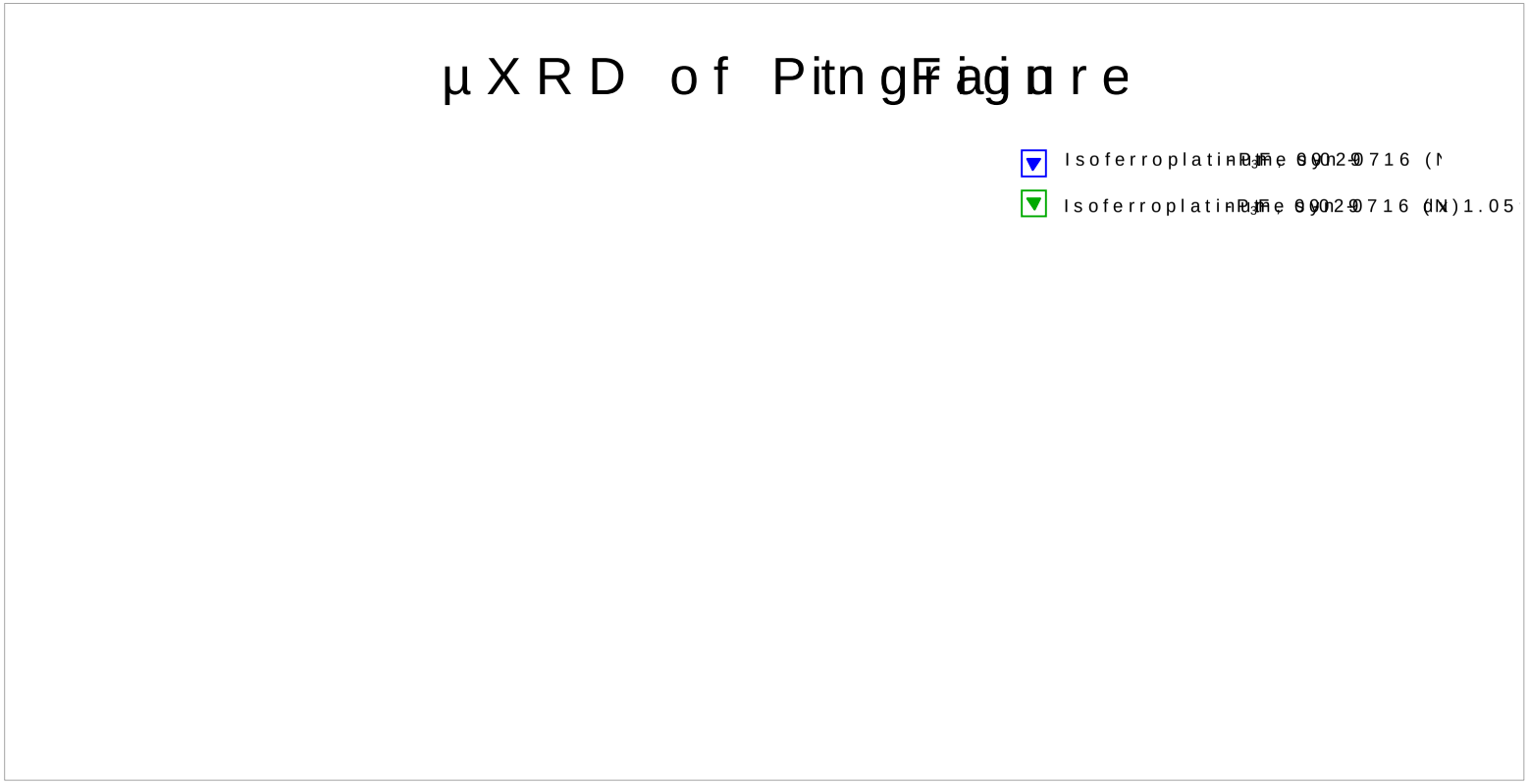

Figure 2.3 µXRD of platinum grain in Figure 2.3A. The alloy, called isoferroplatinum, is a rare mineral (Pt3Fe).

References

Teluk, A.J., 2001, Fifield Platinum Project, NSW, Aust
Technical Report (electronic copy available from <ht
PDF/Geodyne%20report%20complete%20with%20figs.pdf>
March 2012), 71 p.

Xie, J., Colquhoun, G.P., Raymond, O.L., Liu, S.F., Rette
A., Percival, D.S., and MacGregor, A.C., Surface geology of New
Wales: Geological Survey of New South Wales, 1 sheet.

Curriculum Vitae

S. Gordon Campbell

Postsecondary Education and Degrees:

2011-2012
University of Western Ontario,
London, Ontario
M.Sc. Geology

2006
University of Alberta,
Edmonton, Alberta
B.Comm. Finance

2001-2003
University of Illinois,
Champaign-Urbana, Illinois
B.Sc. Biology

Honours and Awards:

2011-2015
NSERC Canada Graduate Scholarship,

2011-2012
Ontario Graduate Scholarship,

2005
BP Canada Energy Scholarship in Business,

Related Work Experience

2011
Teaching Assistant,
University of Western Ontario, Department of

2007-2010
Senior Financial Analyst,
Sobeys Corporate Office, Edmonton, Alberta

2003-2006
Lab Technician,
University of Alberta, Plant Physiology & Hort

Scholarly Activity:

2011-2012
Outreach volunteer,
University of Western Ontario, Department of

Goldschmidt Geochemistry Conference
Prague, Czech Republic, August 2011: Poster