The Speciation of Gold in Mine Wastes and Natural Waters

A thesis submitted for fulfilment of the degree of
Doctor of Philosophy

Christine Ta
BTech (Forens&AnalytChem), BSc (Hons)

Flinders
UNIVERSITY

Faculty of Science and Engineering
School of Chemical and Physical Sciences

May 2013

1. Introduction

1.1 Gold

Highly valued by man since the earliest of time, gold is recognised for its world-wide use in monetary systems, jewellery and for decorative purposes [3]. Gold is the most inert noble metal (unaffected by water, acids, oxygen or sulfur) and has high malleability, ductility, and electrical conductivity [1]. Its electronic configuration, [Xe] $4f^{14}5d^{10}6s^{1}$, with the inner *s* shell electrons tightly bound and shielding the outer *d* shell from the nuclear charge, allows gold to both accept (into the *s* shell) and donate electrons (from the *d* shell). Therefore, gold is also a catalyst for reactions at C-C bonds. As a result of these unique properties, the use of gold is not restricted to coinage and jewellery, and it can be found in dental work, electronics and plating materials [4]. Gold's other properties – its relatively low melting and boiling points (m.p.1064.43 °C and b.p. 2860 °C) – can be attributed to its lattice type [3]. Gold (Au) is in the same group as silver and copper and has a face-centred cubic lattice, where each atom has 12 equidistant neighbours and Au–Au distances of 288.4 pm [3].

1.2 Gold in the environment

Gold is relatively rare with an average abundance in the Earth's crust of 0.005 ppm [1]. With an identical atomic radius to Ag (1.44 Å) [1], native gold is almost always alloyed with silver (0.1% to 50%) [5, 6]. The native elements, alloys and metallic compounds of gold are listed in Table 1-1. The most specific pathfinder elements of gold are Ag, As, Sb and Te [1]. "Non-metallic" gold also appears as "invisible gold", where particles less than 0.1 μm are found within grains of pyrite and As-rich and Fe-deficient arsenopyrite [7, 8]. The primary types of gold deposits includes auriferous porphyry dykes, sills and stocks, skarn-type deposits, disseminated deposits, auriferous and gold-silver veins in igneous, volcanic and sedimentary rocks and gold deposits in quartz-pebble conglomerates and quartzites [1].

Quartz pebble conglomerates (QPC) are the most significant source of the world's production of gold [9]. However gold does not usually originate as QPC deposits. According to Reith *et al.* [10], gold begins as a primary deposit (60 to 90 wt. % gold), usually in hydrothermal and deep subsurfaces. The gold then undergoes many transformations to form secondary gold (which is generally much purer than primary gold, up to 99.5 wt% gold), and eventually a QPC deposit [10]. The two possible origins of secondary gold: are detrital or chemical accretion [1]. Detrital gold is the physical formation of gold from the weathering of pre-existing rocks embedded with gold [11]. Evidence for detrital gold arises from the morphology of certain gold grains (round, toroidal grains) [12, 13], and the isotopic ages of the surrounding minerals of gold (which rule out formation by hydrothermal events) [14, 15], however there is evidence that the chemical mobilisation and re-concentration of gold also contributes to the cycle of gold in the environment, as depicted in Figure 1-1.

Table 1-1 Known gold minerals as listed by Boyle [1].

Mineral	Composition
Native gold	Au
Argentian gold (electrum)	(Au, Ag)
Cuprian gold (cuporauride)	(Au, Cu)
Palladian gold (porpezite)	(Au, Pd)
Rhodian gold (rhodite)	(Au, Rh)
Iridic gold	(Au, Ir)
Platinum gold	(Au, Pt)
Bismuthian gold	(Au, Bi)
Gold amalgam	Au_2Hg_3 (?)
Maldonite	Au_2Bi
Auricupride	$AuCu_3$
Palladium cuproauride	$(Cu, Pd)_3Au_2$
Uytenbogaardtite	Ag_3AuS_2
Calaverite	$AuTe_2$
Krennerite	$(Au, Ag)Te_2$
Montbrayite	$(Au, Sb)_2Te_3$
Petzite (antamokite)	Ag_3AuTe_2
Muthmannite	(Ag, Au)Te
Sylvanite	$(Au, Ag)Te_4$
Kostovite	$AuCuTe_4$
Nagyagite	$Pb_3Au(Te, Sb)_4S_{5-8}$
Aurostibite	$AuSb_2$
Fischesserite	Ag_3AuSe_2
Gold tellurate (?)	

The chemical mobilisation of gold in the environment is driven by organic matter, microorganisms, oxidising minerals and solubilising ligands available in the soil and groundwaters. Gold grains and nuggets are commonly coated by organic matter containing secondary gold in colloidal form, implying a role for organics in the colloidal gold formation [16]. It is known that microbes play a role in the dispersion and concentration of gold in the environment [10]. Southam *et al.* [16] suggests that the microbial solubilisation and precipitation of gold are responsible for the wide range of morphologies of some secondary gold (wire, dendritic, octahedral, porous and sponge gold [13, 17-21]). The role of organics and microbes are closely linked, where the concentration of organics in the soils can effect heterotroph activity in ores and therefore the migration of gold [22]. The interaction of gold with plants and organic matter and microbes are reviewed separately below.

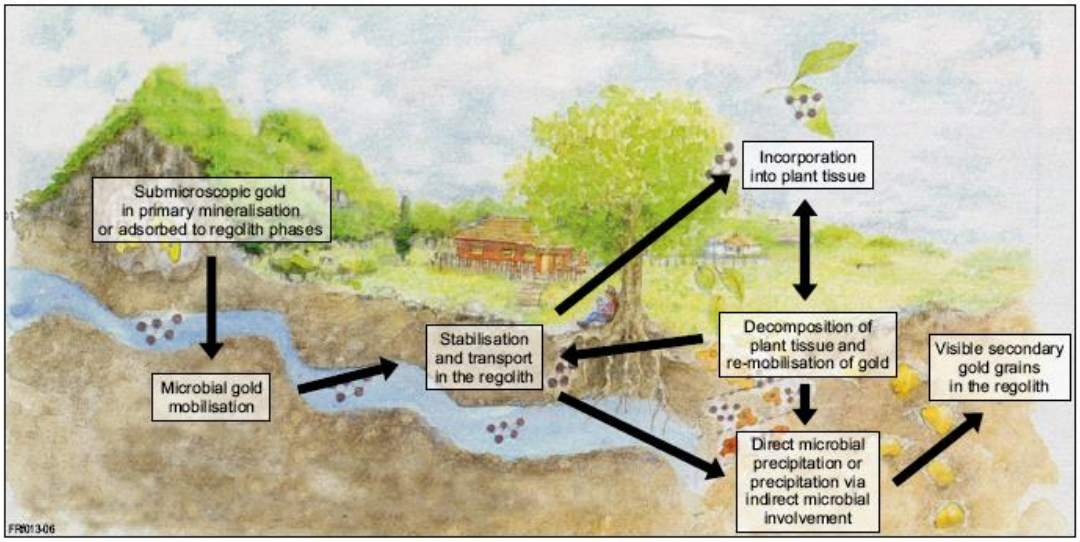

Figure 1-1 **The geomicrobiological cycle of gold in the environment.**
An illustration from taken from Küchler [23] of Reith's [24] conceptual model linking the processes of gold solubilisation, transport, precipitation and authigenic gold nugget formation.

1.2.1 Plants and organic matter

Plants

The ability of plants to uptake gold has been documented by many [25, 26], and plants such as wormwood (*Artemisia* sp.), Douglas fir (*Pseudotsuga menziesii*) have been used for biogeochemical prospecting [27]. It has been suggested that the ability of plants to uptake gold from the soil is dependent on plant secretions (especially cyanide secretions) [26, 28], however non-cyanogenic plants can be induced to hyperaccumulate gold by adding thiocyanate, cyanide, thiourea or thiosulfate solutions as a complexing agent to solubilise the gold in the soil [29-31], allowing the accumulation of gold to be independent of plant species [27]. There is much interest in using hyperaccumulator plants for gold phytomining (extracting gold from tailings and waste dumps) [31]. Plants also present an alternative method for the synthesis of gold nanoparticles, as certain plant matter are able to reduce Au(III) to Au(0) [32-34].

Jones and Peterson [35] suggested that the uptake of gold by plants may also be influenced by humic matter. The authors proposed that humic-bound gold may have limited bioavailability and mobility, as they found the perennial ryegrass (*Lolium perenne* L.) absorbed more gold as Au(III)-chloride over Au-humics. [35]. However there are other studies in which humics are shown to aid the dissolution of gold. The role of humics (and other organics) in the biogeochemical cycle of gold are further discussed below.

Humics

Humics (or natural organic acids) are hydrophilic colloids that have a particle size of 2.4 to 10 µm range and have negative charges due to dissociations of their carboxylic and phenolic groups [36]. They consist of substituted aromatic rings linked together by aliphatic chains and have anywhere from about 15 to 92% aromaticity [37]. Humics exist in three groups – humic acids (HA), fulvic acids and humin – which vary in molecular weight and solubility [37-39]. Most studies on the mobility of gold have been conducted with humic acids.

Friese [40] reported the dissolution of gold by humic acids and concluded that gold migrated chemically as a humate. This was supported by Boyle *et al.*'s [41] solubility experiments which indicated that gold was either chelated, organometallically bonded or adsorbed on to humic matter under supergene conditions. Fetzer's [42] attempt to duplicate Friese's [40] experiment was unsuccessful, even with a range of minerals. Fetzer [42] concluded that humic acids did not dissolve minerals and the solvent action of aqueous humic acid sols was no greater than that expected of water (equilibrated with carbon dioxide). Fezter [42] declared that HA-Au complexation – an oxidation process – was unlikely as Au prefers to retain its valence electrons and HA is a reducing agent. This was supported by later studies that reported the reduction of Au complexes by humic acids [43, 44].

Amino acids

Amino acids are a group of organic molecules – in the form of $H_2N–RCH–COOH$ with different side chains attached to the α carbon – which occur in natural waters and soils. Amino acids have been used in the synthesis of gold colloids or gold nanoparticles (GNPs). GNPs can be synthesised from $[AuCl_4]^-$ ion reduction with aspartic acid [45], citrate [46], tyrosine [47], or tryptophan. In the latter, Selvakannan *et al.* [48] identified the indole group as the reducing segment. Peptides containing such reducing amino acids are also capable of GNPs synthesis [49], whereas amino acid derivatives tend to form complex crystals [50, 51].

Like humic substances however, different conditions and amino acids will dissolve Au(0), including cysteine, glutathione, alanine, glycine, asparagine and histidine [52-54]. Under alkaline conditions, Korobushkina *et al*. [22] ranked complex-forming capacity of amino acids with Au(I) to be:

cysteine > histidine > asparagine > methionine > glycine, alanine and phenylalanine

Zhang *et al.* [54] found that gold was most soluble in histidine (compared to DL-aspartic amide, DL-alanine, glycine, L-galacystine, and DL-aspartate). With the exception of DL-aspartate, these amino acids extracted more gold from gold ores than from pure gold wire [54]. Zhang *et al.* [54] suggested that dispersed gold is more easily extracted by amino acid

solutions. Similarly Brown *et al.* [53] found histidine and glycine extracted more gold from 9 carat gold than 18 carat or 22 carat. As a gold-copper alloy was used, Brown *et al.* [53] suggested that the increased surface area – exposed by the dissolved copper – enhanced the solubility of gold. Amino acids may serve as complexing agents for the transport of gold in natural systems and – as some amino acids are produced or taken up by bacteria – promote the biogeochemical cycling of gold [52].

1.2.2 Microorganisms

Microorganisms play a vital role in the transportation of gold as they can accumulate gold or assist the leaching of gold. Sulfur-oxidising species such as *Acidithiobacillus ferooxidans*, *A. thiooxidans* and *A. caldus*, are known to promote gold leaching environments via production of sulfuric acid. The *Acidithiobacillus* species and *Leptospirillum ferrooxidans* are used to treat refractory gold ores, such as pyrites or arsenopyrites, to liberate gold within the sulfide matrix [55]. Bacteria are also able to produce metabolites capable of gold solubilisation. According to Southam *et al.* [16] microbially mediated gold solubilisation can occur from the production of thiosulfate (in poor organic environments), or amino acids and cyanide (organic-rich environments) in the presence of oxygen. Cyanogenic bacteria such as *Chromobacterium violaceum, Pseudomonas aeruginosa, P. Fluorescens* and *Bacilus megaterium* are known to form water-soluble metal cyanides, and to solubilise gold through cyanidation [56, 57].

The capacity of bacteria and other microorganisms (such as actinomycetes, fungi and yeasts) to accumulate gold has been widely demonstrated [22, 58, 59]. Gold bioaccumulation by bacteria is thought to explain the variety of secondary gold morphologies and why gold in supergene placer environments is larger than the potential source rocks [18]. *Acidithiobacillus thiooxidans* accumulates Au from the gold(I) thiosulfate complex, $Au(S_2O_3)_2^{3-}$ and precipitates elemental Au as a metabolic process [18, 19]. Lengke and Southam [18, 19] reported the eventual formation of octahedral crystals, gold wire and other irregular structures of gold, after the death phase and the release of gold nanoparticle from the cells. Similarly, *Bacillus subtilis* 168 and *Plectonema boryanum* UTEX 485 immobilise and reduces Au^{3+} to form octahedral gold [60, 61]. Some bacteria are able to accumulate specific organo-gold complexes, such as *Sporosarcina ureae* with L-asparagine-Au [52].

As a possible origin of secondary gold, biomineralisation can explain the lace-like surface structures (Figure 1-2) – known as “bacterioform” gold – common in QPC deposits [10, 62, 63], however, the origin of secondary gold cannot be based on observed morphologies, as extremely acidic, experimental conditions can produce artefacts resembling “bacterioform” Au [64].

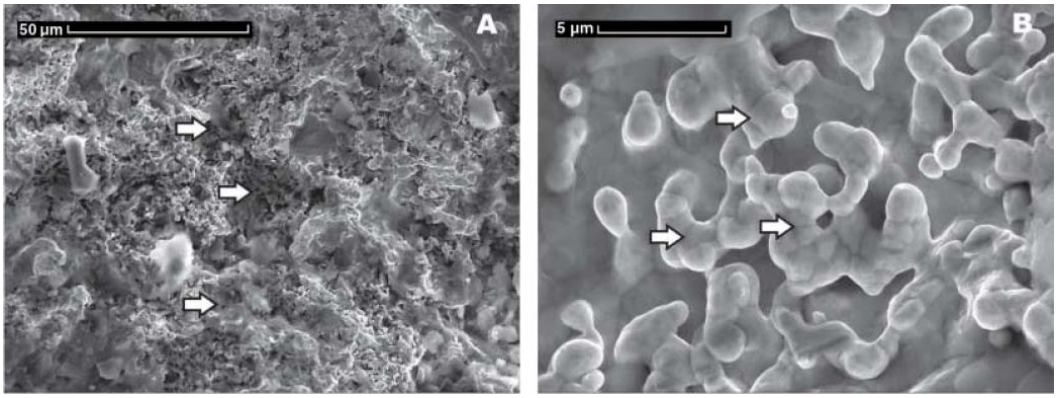

Figure 1-2 **Secondary electron micrographs of bacterioform gold.**

Micrographs show (A) possible biofilm exopolymers (white arrows) and (B) preserved cell wall structures (white arrows) within budding cell-like network, from biofilms on gold grains from the Hit or Miss Gold Mine in Queensland, Australia, taken from Reith et al. [65].

1.2.3 Aqueous forms of gold

While the geochemical cycle of gold includes organic and nanoparticulate gold, aqueous inorganic gold species are of particular interest to geochemists due to their importance in both the environment and the mining industry. Cyanide and sulfidic lixiviants are commonly used in gold leaching [66], but also naturally available in the environment from plants or the activity of microorganisms [18, 57, 67]. Halide complexes are also of interest in gold speciation, especially due to the abundance of chloride ions in waters and the slight solubility of gold in seawater (~30 pg L^{-1} [68]). Therefore, the following review will focus on gold-cyanide, -sulfur and –halide (and –hydroxide) complexes.

Overview

Gold exists in oxidation states from -I to +V [69], however, the most common oxidation states of Au in aqueous solutions are Au(I) and Au(III). As B-type metal ions (heavy transition metal cations), the stability of Au(I) and Au(III) complexes tends to decrease with increasing electronegativity of the ligand donor atom [70]. Unbound Au(I) and Au(III) cannot occur in aqueous solution because they are stronger oxidising agents than water [36]. Therefore, in aqueous solutions, Au(I) and Au(III) are coordinate complexes (usually linear two-coordinate complexes, and square planar complexes respectively).

In natural waters and in the absence of other ligands, the predominant form of Au(I) is the linear $Au(OH)(H_2O)$ [71, 72]. The absence of O_2 in subsurface or ground waters favours the formation of metallic gold, whereas ionic gold is more predominant in river waters [36]. In a review, Senanayake [66] summarised the stability of Au(I) complexes:

$$CN^- > HS^- > S_2O_3^{2-} > SC(NH_2)_2 > OH^- > I^- > SCN^- > SO_3^{2-} > NH_3 > Br^- > Cl^- > CH_3CN$$

and Au(III) complexes with different ligands:

$$CN^- > OH^- > SCN^- > Br^- > Cl^-$$

Due to the acidic oxidising conditions that Au(III) tends to exist in, potential complexing ligands are less stable and hence fewer in number.

Gold sulfur complexes

Depending on the bisulfide (HS^-) and thiosulfate ($S_2O_3^{2-}$) concentrations, Au(I) tends to form bisulfide complexes in reducing environments (i.e. hydrothermal conditions or anoxic lakes), or thiosulfate complexes in alkaline oxidising conditions (i.e. oxidising sulfide ores). In sulfidic solutions, the species [Au(HS)] and $[Au(HS)_2]^-$ are present at pH 2–10, as HS^- ions are more stable than OH^- ions [71]. In S-saturated solutions, polysulfide species ($[AuS_nS]^-$, n = 2–7) are dominant (i.e. S_3^{-2} and S_4^{-2} ligands will displace the SH^- ligand from $[Au(SH)_2]^-$ to form $[Au(S)_3]^-$ and $[Au(S)_4]^-$) [71]. Thiocyanates (SCN^-) – formed from glycosides – are not very abundant in soils and hence such natural gold complexes are scarce [73], however, thiocyanate and thiosulfate have been considered as an alternative to cyanide for gold recovery [74-76].

Gold cyanide complexes

The cyanide ion (CN^-) is the most significant carbon donor ligand and forms the most stable gold complex in solution [72]. In the environment, cyanides are produced by the hydrolysis of cyanogentic glycosides [73]. CN^- is found in various plant species and is released to the soil in order to solubilise metabolically required metals, or upon decay of the plant [73]. Another source, hydrocyanic acid (HCN), is produced by more than 1000 species of plants and a variety of microorganisms [57, 67, 73]. While the Au(III) square planar analogue exists, $[Au(CN)_4]^-$, it is not formed under typical conditions [77], and hence the most important form of gold cyanide complex in soils is $[Au(CN)_2]^-$ [78].

Gold halide and hydroxide complexes

As a soft metal, the stability of Au(I)–halide complexes increases along the series $Cl^- < Br^- < I^-$ ($[AuF_2]^-$ has not been identified). Excess halide is required to prevent the Au(I)-halide complexes $[AuCl_2]^-$ and $[AuBr_2]^-$ undergoing disproportionation in aqueous solution [79]:

$$AuX_2^- \rightarrow AuX_4^- + 2Au + 2X^-$$

Due to the abundance of Cl^- ions in natural waters, gold-chloride complexes feature in many studies [80-84]. Pan and Wood [82] found that at temperatures ≤100 °C, the square planar Au(III)-chloride complex $AuCl_4^-$, is predominant in acidic solution, but at temperatures >100 °C is partially transformed into the linear Au(I)-chloride complex, $AuCl_2^-$. This reaction was also favoured by a reduction of the oxygen fugacity and an increase of pH, and provides evidence for the existence of $AuCl_2^-$ in hydrothermal solutions [82].

Gammons *et al.* [85] found that native gold catalyses the disproportionation of Au(I)-chloride complexes and the gold crystals formed during the disproportionation at 25°C are of varying morphologies. They suggested that $AuCl_2^-$ is the dominant form of dissolved gold in brines at near-neutral pH (i.e. seawater). With the increase of temperature, the stability of $AuCl_2^-$ increases relative to $AuCl_4^-$, as does the solubility of Au [85]. These authors [85] theorised that Au is transported as $AuCl_2^-$ rather than $AuCl_4^-$, and the cooling of ore fluids could precipitate some of the dissolved metal via disproportionation.

When Au is dissolved in NaOH, $[Au(OH)(H_2O)]$ is the predominant species at pH 0–12 [71]. This changes at pH << 0, as $[Au(H_2O)_2]^+$ is much more acidic than H_2O [71]. Colin and Vieillard [86] calculated the Au(I) gold complex $AuCl(OH)^-$ to be most stable, in waters of pH 4–6 at Dondo Mobi, Gabon.

Au(III) forms stable square planar complexes with halides and hydroxides [84]. Usher *et al.* [84] found that the iodide ion (I^-) is unstable in the presence of Au(III) and oxidises rapidly to $I_{2(g)}$ and precipitates Au(III). Usher *et al.* [84] identified three intermediate complexes between $[AuCl_4]^-$ and $[AuBr_4]^-$ and found that Au(III)-hydroxide precipitates approximately between pH 8 and 13. So Au(III)-chloride-bromine complexes can be important in transporting gold in brines with high bromide-chloride ratios (e.g. > 0.05), under oxidising (atmospheric) and acidic (pH < 5) conditions [84]. This is in agreement with Gammons *et al.* [85], who proposed Au(III) is the dominant species in air-saturated brines for all temperatures up to 300 °C (pH < 3).

The most extensive studies on Au(III) complexes, however, have been on the hydrolysis of $[AuCl_4]^-$. Both Peck *et al.* [83] and Usher *et al.* [84] used UV-VIS spectroscopy to observe the successive replacement of chloride ligands in $[AuCl_4]^-$, with hydroxide ligands at increasing pH. Figure 1-3 and Figure 1-4 (A) are absorbance spectra displaying the pH effect on Au(III)-chloro-hydroxide speciation, from Peck *et al.* [83] and Usher *et al.* [84] respectively. Both authors report the peak at 314 nm, $[AuCl_4]^-$, decreasing in absorbance and shifting toward lower wavelengths as $[AuCl_4]^-$ undergoes hydrolysis.

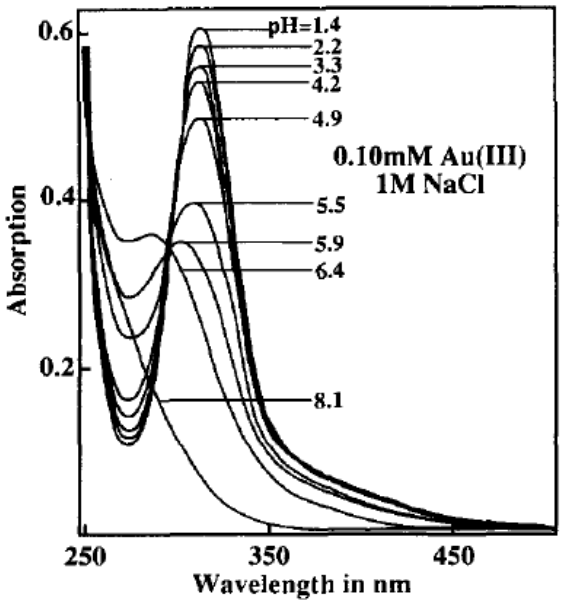

Figure 1-3 **Absorption spectra of 10^{-4} M Au(III) in 1 M NaCl solution at varying pH.**
Spectra are taken from Peck et al. [83] and show an isosbestic point at 300 nm between pH 5 and 6.

Peck *et al.* [83] observed an isosbestic point at 300 nm (in Figure 1-3). According to Rao [87] an isosbestic point is the criterion that two species (and only two) are in equilibrium. Hence Peck *et al.* [83] suggested there was a change in the relative abundances of $[AuCl_3(OH)]^-$ and $[AuCl_4]^-$ between pH 5 and 6. In contrast, Usher *et al.* [84] did not find an isosbestic point and due to changes in the absorbance at specific wavelengths (shown in Figure 1-4 (B)), proposed at least four Au(III)-Cl-OH species were present at pH 5–6. Both Peck *et al.* [83] and Usher *et al.* [84] allowed some time for their solutions to equilibrate after pH adjustment. The former allowed several hours for the solutions to equilibrate after pH adjustment, while the latter stirred the solution for 30 minutes before analysis. But it was Lee and Gavriilidis [88] who spectroscopically investigated the effects of ageing time on pH adjusted $HAuCl_4$ solutions.

Figure 1-5 shows the absorbance spectra of 2.5×10^{-3} M $HAuCl_4$ solutions ranging from pH 5 to 11 at 15–720 min. Similar to Peck *et al.*'s [83] work, the greatest change in absorbance over this time period was seen at the lower pH levels (i.e. pH 5 and 7), suggesting a change in the dominant species at that pH range. According to Lee and Gavriilidis [88], the expected dominant species at equilibrium for pH 5, 7, 9 and 11 are $[AuCl_2(OH)_2]^-$, $[AuCl(OH)_3]^-$ and $[Au(OH)_4]^-$ (at both pH 9 and pH 11) respectively. But at short ageing times, the dominant species are possibly $AuCl_2(H_2O)OH$, $[AuCl(OH)_2]^-$, $[AuCl(OH)_3]^-$ and $[Au(OH)_4]^-$ respectively [88]. Lee and Gavriilidis's [88] work indicates that the exchange of ligands on gold complexes can occur over hours, especially at lower pH's (i.e. pH 5 – 7).

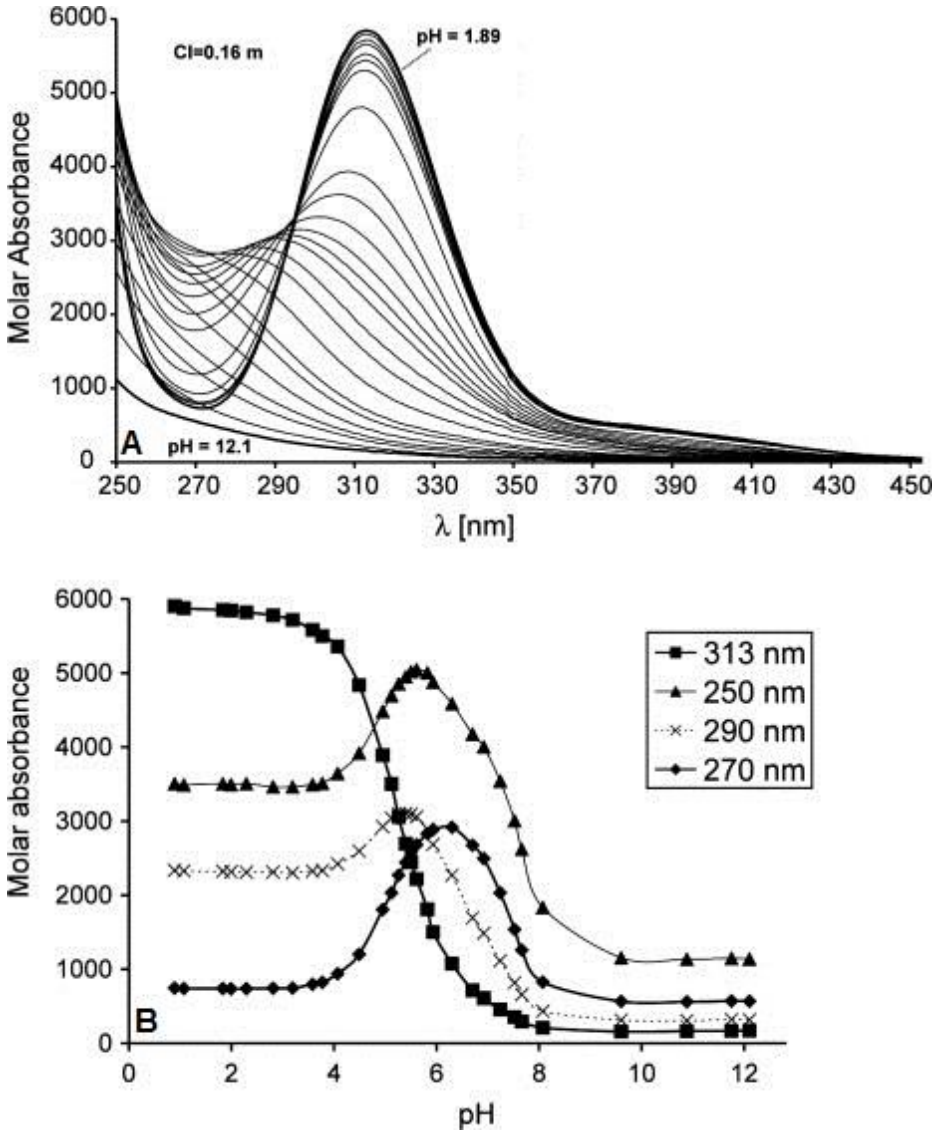

Figure 1-4 **Absorbance spectra of Au(III)-Cl-OH complexes as a function of pH.**

Spectra shows Au(III) (total concentration 1.4 m) in 0.16 m Cl^- as (A) variation of absorbance of with pH at various wavelengths; and (B) principal component analysis of the matrix of absorbances for the Au(III)-Cl-OH system. Spectra are taken from Usher et al. [84].

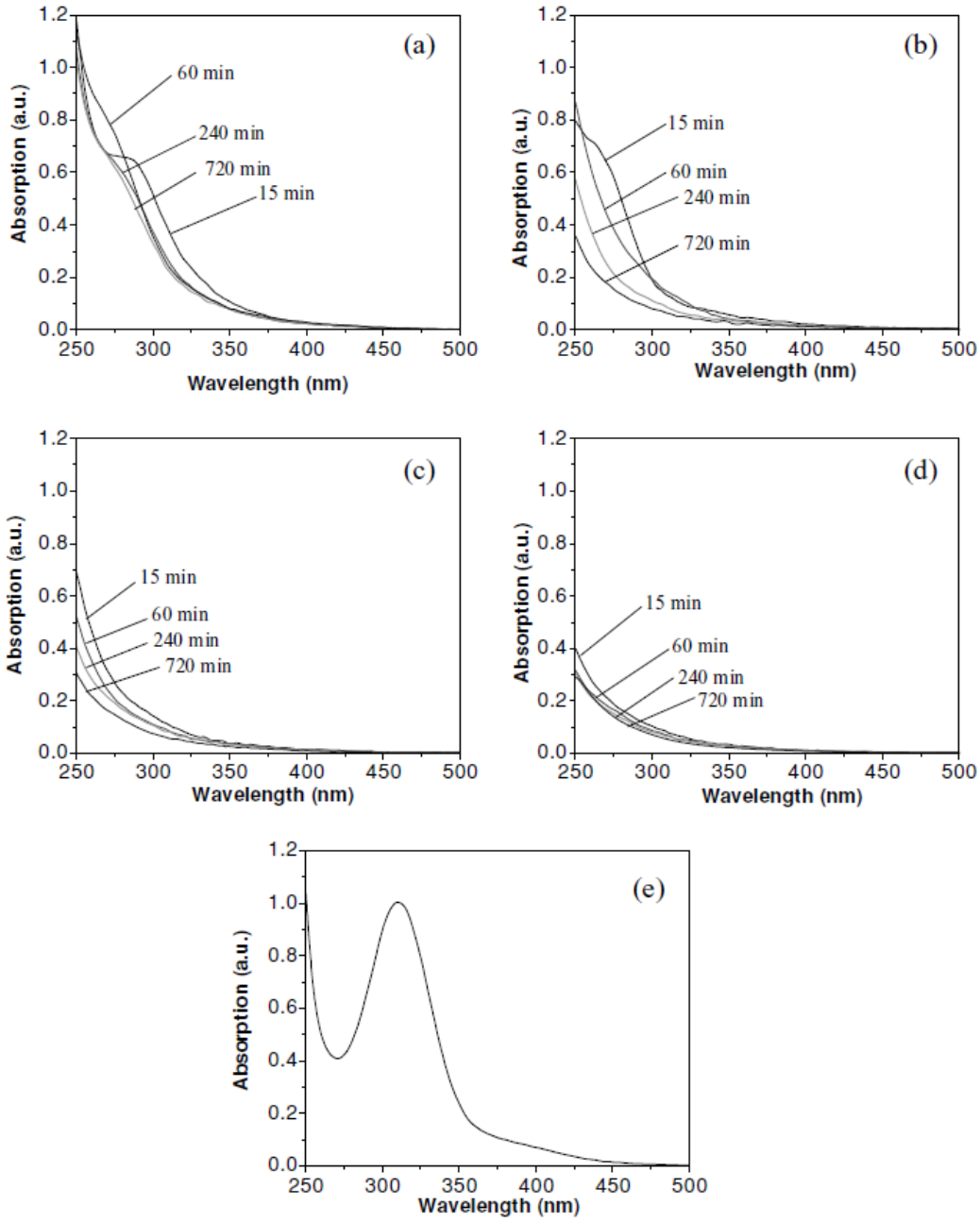

Figure 1-5 **Absorbance spectra of 2.5 x 10^{-3} M $HAuCl_4$ solutions.**

Spectra from Lee and Gavriilidis [88] show (a) pH 5, (b) pH 7, (c) pH 9, (d), pH 11 and (e) pH 2 without adding Na_2CO_3 solution.

The hydrolysis of $[AuCl_4]^-$ (as a function of pH) has also been monitored with Raman spectroscopy [81-83]. The Au-Cl and Au-OH stretches are represented by peaks in the 320-350 cm^{-1} and 560-580 cm^{-1} regions respectively, as shown in Figure 1-6, a series of Raman spectra of a Au(III)-chloride solution adjusted to pH 1-12 by Murphy and LaGrange [81]. It can be seen, that as the pH is increased and the chloride ligands are replaced with hydroxide groups, the peak area ratio of Au-OH to Au-Cl increases.

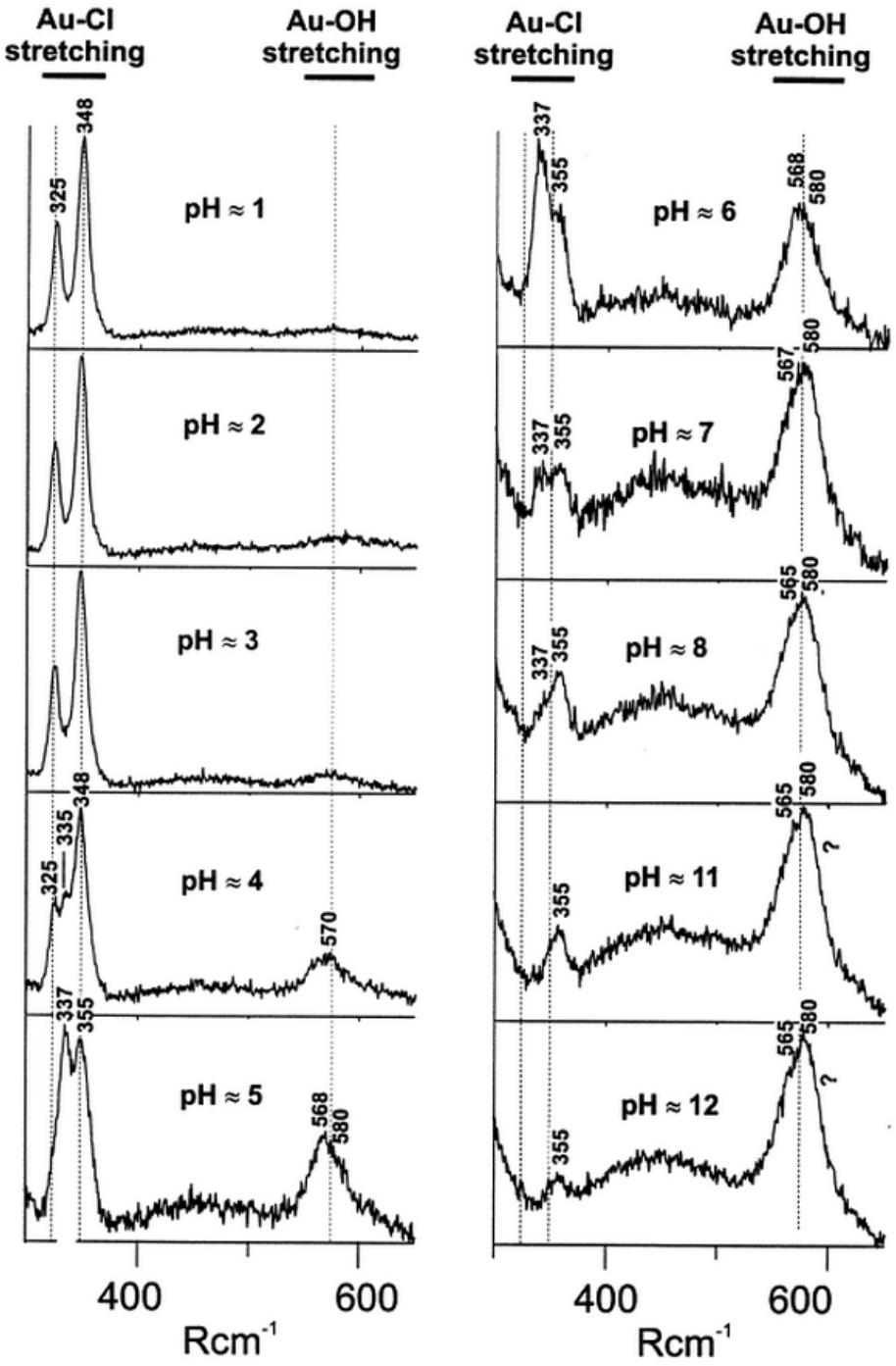

Figure 1-6 **Raman spectra of the Au-Cl stretching and Au-OH stretching ranges.**

Spectra of 0.02 M $HAuCl_4 \cdot 4H_2O$ solution at pH 1–12 taken from Murphy and LaGrange [81]. Vertical lines are shown for comparison of peak positions. Intensity is not necessarily to scale. The unit "Rcm^{-1}" denotes "relative cm^{-1}", meaning the frequency is measured relative to the frequency of the laser light [89].

Peck *et al.* [83] also observed the hydrolysis of the $[AuCl_4]^-$ complex with the increase of pH, however, they noted that hydrolysis began at pH ≈ 5.8, while Murphy and LaGrange [81] recorded hydrolysis beginning at pH = 3.8. Murphy and LaGrange [81] attributed the difference in results to variation in gold and chloride concentrations (the higher the gold concentration or chloride concentration, the higher the pH was for the start of hydrolysis), and variation in equilibrium times. As expected, there was no evidence of hydrolysis in Pan and Wood's [82] study, which were conducted under very acid conditions (m_{HCl} = 2-5).

In conclusion, an array of $[AuCl_4]^-$ hydrolysis studies have been conducted by UV-VIS [83, 84, 88, 90-92] and Raman spectroscopy [81-83]. These studies have found that the equilibrium time, the matrix and the concentration of gold and chloride in the solutions analysed played a significant role in the speciation of gold. Gold speciation techniques, UV-VIS, Raman spectroscopy and others are discussed in the following section.

1.3 Previous gold speciation techniques

1.3.1 Overview

As previously discussed (in Section 1.2), the transport and mobility of gold in the environment and its interaction with plants and microorganisms has been extensively studied. A majority of these studies were comprised of gold dissolution/precipitation experiments to hypothesise what conditions promote the mobility or formation of gold in the environment. Other studies have focussed on measuring the speciation of gold under aqueous conditions. Discussed in this section, are the various techniques used to determine the speciation of gold in those studies. It will be shown that most studies have only been on model solutions, due to the limitations of certain techniques. There are a couple of studies that have examined the speciation of gold in natural waters, yet only indirectly with chemical speciation modelling [93, 94]. Therefore developing a new method that is capable of determining the speciation of gold in natural waters is vital to furthering our understanding of the behaviour of gold in the environment.

1.3.2 Ultraviolet-Visible Spectroscopy (UV-VIS) studies

Au(III) like other d^8 transition metal ions forms square planar complexes with halide ions, $[AuX_4]^-$ (X= Cl, Br), which yield two intense bands in the U.V. region [95, 96]. Figure 1-7 is an absorbance spectra of $[AuCl_4]^-$ and $[AuBr_4]^-$ displaying these two bands. Gangopadhayay and Chakravorty [96] attributed the two peaks around 225 nm and 315 nm to ligand-to-metal charge-transfer transitions (LMCT), $p_\sigma \rightarrow dx^2–y^2$ and $p_\pi \rightarrow dx^2–y^2$ respectively. As a result of the LMCT absorptions, Au(III)-halides are coloured ($[AuCl_4]^-$ is yellow and $[AuBr_4]^-$ is orange) [84].

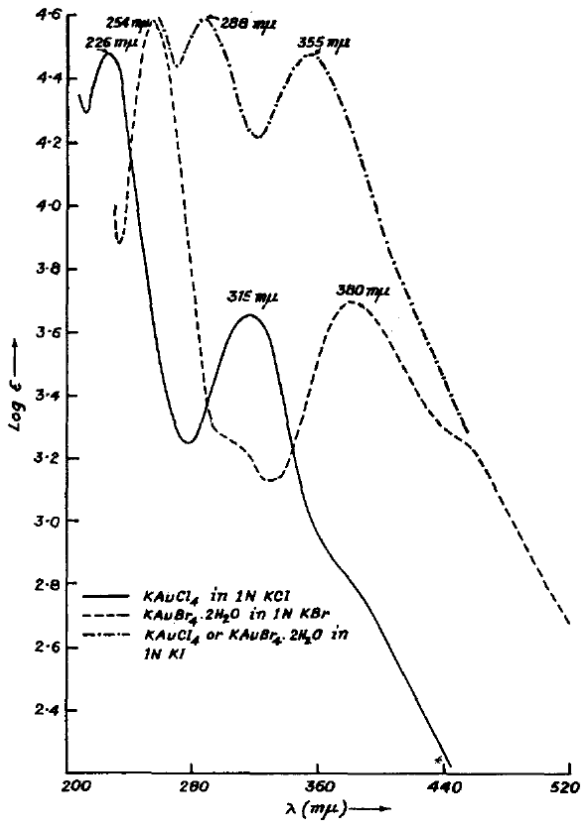

Figure 1-7 **Absorbance spectra of $[AuCl_4]^-$ and $[AuBr_4]^-$.**
Spectra taken from Gangopadhayay and Chakravorty [96].

In 1962, Lingane [97] found absorption measurements were an accurate means of determining the concentration of $AuCl_4^-$ in the presence of $AuCl_2^-$ since $AuCl_4^-$ solutions are yellow and $AuCl_2^-$ solutions are colourless. Gammons *et al.* [85] used this difference in light absorbance to determine the Au(I)/Au(III) ratio in mixed Au(I)/Au(III) solutions by measuring at 314 nm (a local maximum for $AuCl_4^-$), then analysing for total gold with AAS. The detection limits were ~0.1 ppm Au(III) for UV-VIS and 0.5 ppm for AAS.

Gold nanoparticles (GNPs) are also known to be a deep red colour (with absorbances around 525 nm) [98], although this can vary with size and shape [99]. Bohren and Huffman [99] compared the optical effects of gold particles of different radii in Figure 1-8. The absorption by gold particles is independent of size between about 26 and 100 Å, but broadens outside this range [99]. Gold particles larger than 100 Å change from a ruby red through to purple and violet to pale blue at 800 Å [99].

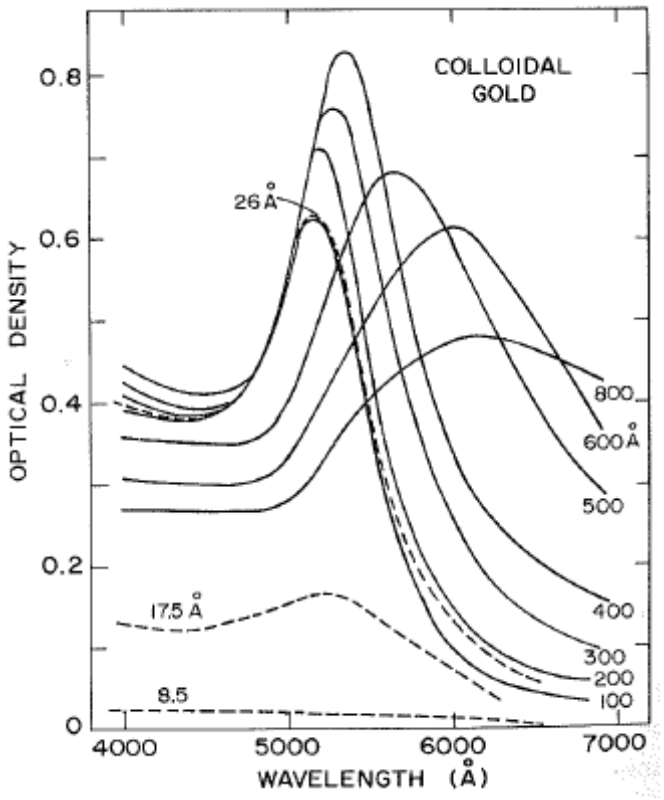

Figure 1-8 **Absorption spectra by gold particles of different radii.**

The solid curves are from Turkevich et al. [100] and the dashed curves are from Doremus [98], as shown in Bohren and Huffman [99].

While UV-VIS spectrophotometry can be used to detect different gold complexes, care must be taken as Au(I) and Au(III) complexes are light sensitive and photolysis commonly leads to the deposition of metallic gold [69]. Another disadvantage of UV-VIS analysis of natural waters is that groundwaters often contain colloids, bacteria, algae or suspended particles [101], which can absorb in regions of interest. Also UV-VIS spectroscopy is not sensitive enough to detect the typical low levels of gold found in groundwaters (ng L^{-1}) [50, 102-105].

1.3.3 Raman studies

The square planar $[AuCl_4]^-$ has 9 vibrational modes, three of which are Raman active: A_{1g} + B_{1g} + B_{2g} [82], as shown in Figure 1-9. The majority of Raman studies have been conducted on the $[AuCl_4]^-$ complex and the effect of pH on its hydrolysis products, $[AuCl_{4-n}(OH)_n]^-$ (where x = 0-4) [81-83]. Raman spectroscopy is ideal for aqueous speciation studies as the Raman spectrum of water is unlikely to interfere with analysis [81, 83]. Another advantage is that spectra with mixed complexes can be simplified through Resonance Raman (RR) scattering. RR scattering is achieved by tuning the excitation laser to the same frequency of the electronic absorption band of one complex. This can increase the scattering intensity by a factor of 10^4 – 10^6 and selectively enhance the one complex as shown in Figure 1-10 [83].

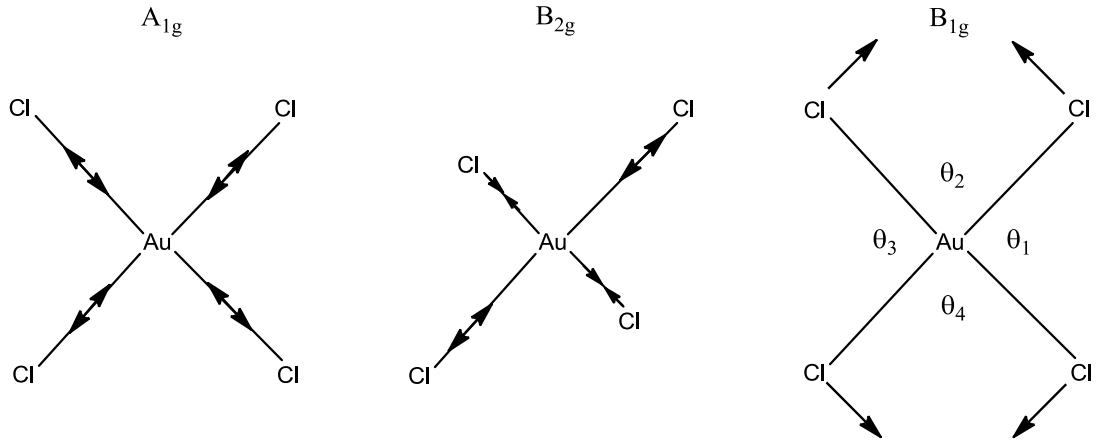

Figure 1-9 **Raman active vibrational modes of the $[AuCl_4]^-$ complex.**
Stretching (A_{1g}, B_{2g}) and bending (B_{1g}) modes.

A disadvantage in Raman is that although concentration is proportional to peak intensity a direct comparison of concentrations is not usually possible. However different bonds (such as Au-OH and Au-Cl) have different intensities and the Au-Cl stretching peak is more intense in $[AuCl_4]^-$ than in $[AuCl_3(OH)]^-$, (the former contains four Au-Cl bonds and the latter three) [81]. Therefore the detection limit for gold speciation in Raman is more dependent on the species present rather than the total gold concentration [81]. Typical concentrations in previous Raman studies range from ~10^{-2} to 10^{-3} M gold, (the latter in excess chloride solutions) [81-83].

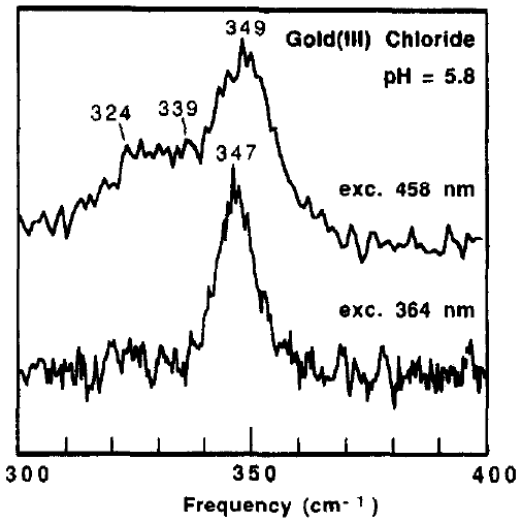

Figure 1-10 **Enhancement of the symmetric stretch of $[AuCl_4]^-$ at ~347 cm^{-1}.**
Spectra of normal (laser excitation at 458 nm) and resonance (364 nm) Raman of 10^{-2} M Au(III) in NaCl solution at 4ºC at pH = 5.8. Spectra taken from Peck et al. *[83].*

1.3.4 X-ray absorption spectroscopy (XAS)

The absorption of X-rays can excite the *1s* (K edge) or *2s,2p* (L edge) electrons of an element to empty orbitals or the continuum [106]. There are two regions in X-ray absorption spectroscopy: X-ray Absorption Near Edge Structure (XANES), which indicates the oxidation state of the metal ion and the coordination chemistry of the neighbouring atoms [107], and Extended X-ray Absorption Fine Structure (EXAFS), which provides information on interatomic distances for ligands and neighbouring atoms [108]. These two spectral regions are illustrated in a schematic of a XAS spectrum in Figure 1-11.

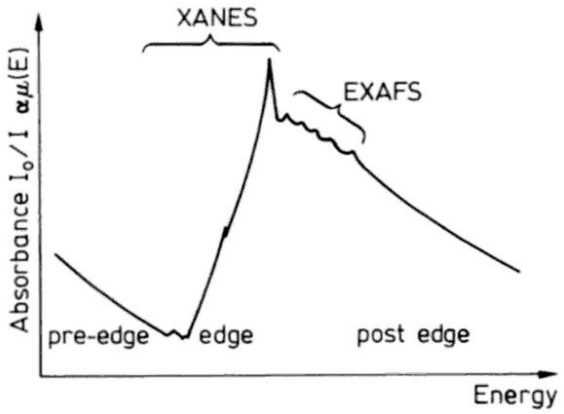

Figure 1-11 **The two spectral regions in XAS: XANES and EXAFS.**
Schematic is taken from Vainshtein [108].

For gold, the absorption of the *2p→5d* electronic transition at the Au-L_{III} edge gives rise to a peak (also known as a "white-line") on the edge of the XANES region [109]. The "white-line" intensity reflects the density of the unoccupied *d* states or the oxidation state of the metal ion [110]. Figure 1-12 (A) shows that the "white-line" intensity is highest for Au(III) and lowest for Au(0). The absorption spectrum of the metal ion is also affected by its neighbouring atoms, where the ejected photo-electron from the metal ion interacts and scatters from the surrounding atoms. This high energy region or EXAFS, is used to determine the distances or the coordination of the neighbouring atoms, as shown in Figure 1-13.

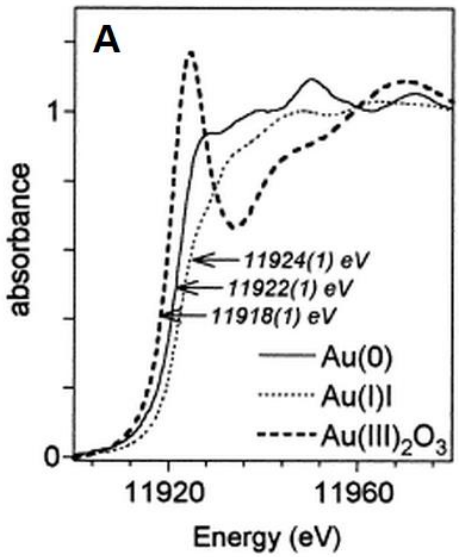

Figure 1-12 **Typical Au-L_{III} edge XANES spectra as taken from Berrodier *et al.* [111].**

Spectra for selected model compounds showing (A) the shift in the absorbance edge and "white-line" intensity for Au(0), Au(I) and Au(III).

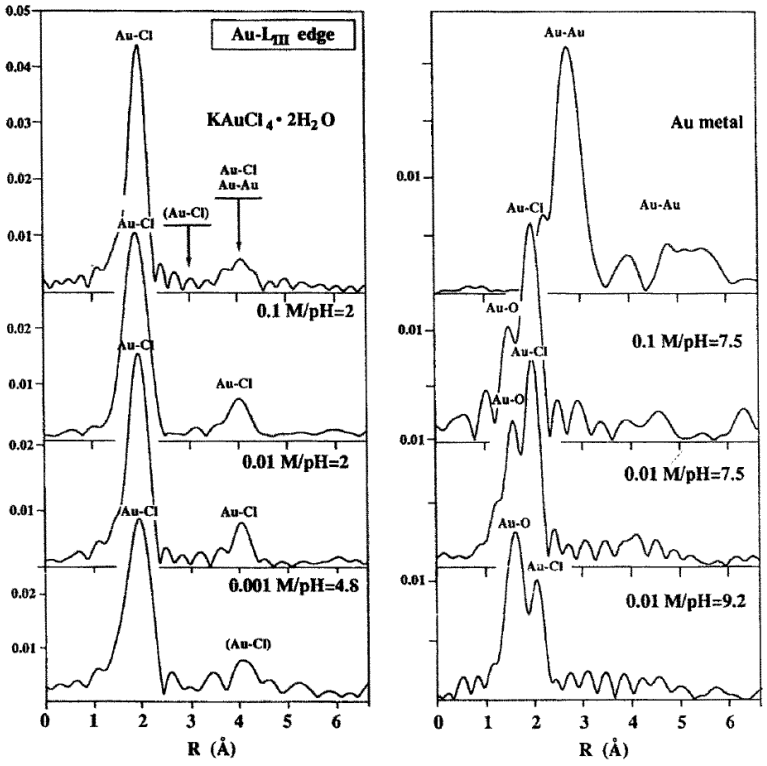

Figure 1-13 **Fourier transforms of EXAFS spectra of Au solutions.**

Spectra of 1 M NaCl, crystalline $KAuCl_4 \cdot 2H_2O$ and metallic gold (taken from Farges et al. [80]).

Previous XAS gold speciation studies have looked at both solid and aqueous samples. Solid studies included investigations into the geometry of potential antitumor gold drugs [112] and the determination of the chemical form of invisible gold in arsenopyrite [113, 114]. Farges *et al.* [80] used EXAFS to monitor the change in speciation of Au(III)-chloride with the change in pH. XANES has also been used to monitor changes in gold oxidation states, to show the *Shewenella algae* cells' ability to reduce $AuCl_4^-$ ions to Au(0) [115]. Berrodier *et al.* [111] used XAS to examine Au(III) speciation in solutions and on mineral surfaces, such as goethite, ferrihydrite and boehmite.

The advantage of using XAS for gold speciation is that the oxidation state of gold can easily be interpreted from the intensity of the "white-line", however, determination of distances and neighbouring atoms from the multiple scattering feature or the EXAFS region is more difficult to interpret. A high energy X-ray source is also necessary for XAS spectroscopy, requiring access to a synchrotron X-ray fluorescence beam line.

1.3.5 Extraction-photometric reagents

Extraction-photometric speciation methods rely on the formation of an ion associate between $[AuCl_4]^-$ and large organic cations such as rhodamine, tripheynlmethane, antipyrine, thiazine and oxazine dyes [116]. This is then usually followed by a liquid-liquid extraction step with an organic solvent and then photometric analysis with atomic absorption spectroscopy, fluorescence or UV-VIS etc. [116-119].

The gold can also be preconcentrated during the liquid-liquid extraction step, which is necessary for the sensitivity of photometric detections (spectrophotometric methods are limited to the molar absorptivity of the reagent). This is achieved by limiting the volume of the extractor solvent. Another method is the use of an adsorbant column to preconcentrate the gold. Examples of reported detection limits after preconcentration include:

- Serbin *et al*.'s [117] detection limits of 0.031 mg L^{-1} and 0.084 μg L^{-1} for spectrophotometric and atomic absorption analysis respectively.
- Tang *et al*. [118] achieved a detection limit of 0.16 μg L^{-1} by using a polyamide column for the adsorption and preconcentration of Au^{3+} ($AuCl_4^-$ can bind to the O and N in the polyamide by static effect) before fluorescence.
- Tsukahara [119] was able to detect 0.1 – 33 mg L^{-1} of gold with spectrophotometric analysis.

Yet despite these low detection limits for gold, most of the described extraction-photometric methods are more suited for the detection of gold – rather than its speciation – as most methods require a specific pH for the extraction, and for the gold to be in either the $AuCl_4^-$ or $AuBr_4^-$ form [116-120]. Even El-Shahawi *et al.*'s [121] liquid-liquid extraction separation of

Au(I) and Au(III) ions relied on the oxidation of Au(I) species to Au(III) species. Hence, there can be many extraction steps for these methods.

1.3.6 Chemical speciation modelling

Chemical speciation modelling is based on the presumption of a system reaching equilibrium and writing mass balance and mass action equations for the possible minerals and species in the system. The equilibrium constants required for this are determined experimentally by measuring the concentration of species under varying conditions (pH, temperature etc.), and can be stored in databases such as the IUPAC Critical Database and the NIST Standard Reference Database [122]. In previous gold speciation studies these equilibrium (or stability) constants are often used for comparing and verifying experimental results [72, 83, 84]. There are also thermodynamic modelling softwares that are able to access these databases of equilibrium constants for chemical speciation (i.e. HYDROGEOCHEM [123], PHREEQC [124] and The Geochemist's Workbench® [125]). (The latter (or GWB) was used in this thesis and modelling with GWB is explained in more detail in Section 1.5.1).

Speciation modelling is often paired with an analytical method that measures the metal ion or the total metal concentration [122]. Leybourne *et al*. [94] used inductively coupled plasma emission spectrometry (ICP-ES) and ICP-MS to determine the major and trace elements in creek water. These species were then modelled with the software PHREEQC to determine the ligands available for complexing with Au. Based on their modelling, Leybourne *et al.* [94] theorised that gold was being mobilised from the nearby tailings as $[Au(CN)_2]^-$. Gray and Pirlo [93] also analysed groundwaters in South Australia with ICP-ES and ICP-MS, and then constructed equilibrium activity diagrams, computed solution species and mineral saturation and modelled ion interaction with GWB [125], PHREEQE [126] and PHRQPITZ [127] respectively. Gold(I)-halides (i.e. $[AuCl_2]^-$) was expected to be the dominant complex in the saline regions [93].

A limitation of chemical speciation modelling is that it is an indirect method of speciation and there are potentially many sources of errors associated with modelling, arising from uncertainties in the chemical analyses, whether the correct model has been used, relying on assumption about the system and the depth and accuracy of the thermodynamic datasets [125]. Use of incorrect equilibrium constant values and the propagation of the errors can lead to significant uncertainties [122]

1.3.7 HPLC-ICP-MS studies

Unlike chemical speciation modelling, HPLC-ICP-MS is a direct speciation technique. This powerful speciation technique has been used to speciate many trace elements, particularly As, Se and Sb [128-130]. Gold possesses only one stable isotope, ^{197}Au, has a first

ionisation energy of 890.1 kJ mol^{-1} and can be analysed by ICP-MS with detection limits below the part-per-trillion (ppt) range [131]. Previous Au speciation work on HPLC-ICP-MS has arisen from pharmacokinetic studies on gold-based drugs. These methods include size-exclusion, weak anion-exchange and reversed-phase ion pairing chromatography (RP-IPC), where gold-based drugs and their metabolites were successfully separated [132-135].

Size exclusion chromatography with ICP-MS detection has been used to monitor the size distribution of protein-bound gold in blood plasma, serum and red blood cell lysate [133, 134]. Low molecular weight gold metabolites, however, were strongly retained on size exclusion columns. Hence gold metabolites were also studied by Elder and colleagues with weak anion-exchange chromatography and reversed phase-ion pair chromatography (RP-IPC) with ICP-MS detection [132-135]. Matz *et al.* [133] and Elder *et al.* [132] used weak anion-exchange chromatography with gradient elution to monitor changes to the gold distribution in patient urine and blood undergoing gold therapy (containing 0.20–1.45 mg L^{-1} of total gold). Quantification of the gold species was difficult, however, due to the gradient elution. Alternatively, RP-IPC can be used with an isocratic elution. Zhang *et al.* [134] used RP-IPC coupled with ICP-MS to detect Au-glutathione, $[Au(CN)_2]^-$ and other gold-complexes in red blood cell lysate containing approximate 0.1–2.5 µg L^{-1} of total gold. Zhao *et al*'s [135] RP-IPC method reported detection limits of 0.3 ng of drug using an injection volume of 200 µl for the gold drug auranofin. This equates to a detection limit of 1.5 µg L^{-1} using these conditions.

There are limitations to what samples can be analysed with ICP-MS, due to the sensitivity of the technique/instrumentation. Difficulties mainly arise from matrix issues such as:

- Drift in the signal caused by deposition of sample material on the interface cones and the ion optics [136],
- Suppression/enhancement of the signal from components in the matrix [137], and
- Plasma instability caused by introduction of high volumes of organic solvent commonly used in HPLC mobile phases [138].

Yet there are still many important advantages of HPLC-ICP-MS over other gold speciation techniques:

- It is a direct and sensitive speciation technique,
- Little sample preparation is required, and
- There is potential to differentiate between gold thiosulfate, cyanide and halide-hydroxide species.

Therefore, a new method for the speciation of gold complexes will be developed for HPLC-ICP-MS (with RP-IPC).

1.4 Instrumentation

1.4.1 Separation by HPLC

Reversed Phase Ion Pair Chromatography (RP-IPC) is a mode in High Performance Liquid Chromatography (HPLC) that allows the separation of ionic analytes. In reversed phase liquid chromatography (RPLC), the stationary phase is non-polar (hydrophobic) and the mobile phase is polar. Separation is based on the partitioning of analyte *X* between the mobile (X_m) and stationary phase (X_s), (i.e. the partition coefficient, *K*, governs the distribution of *X)* [139, 140]:

$$X_m \leftrightarrow X_s \qquad \text{Partition coefficient } K = \frac{[X_s]}{[X_m]}$$

In order to separate ionic species, an organic ion-pairing reagent (or counter-ion) is added to the mobile phase to form an ion pair with the sample component. Figure 1-14 shows the structure of tetrabutylammonium chloride (TBAC); an ion-pairing agent suitable for anionic species. The organic end reacts with the stationary phase, whereas the ionic end reacts with the analyte [140]. These “detergent” like molecules improve the retention of ionic analytes and retention is proportional to the concentration and hydrophobic chain length of the ion-pairing agent [141]. Once the sample has been separated on the HPLC, the analytes are then introduced into the ICP-MS for detection.

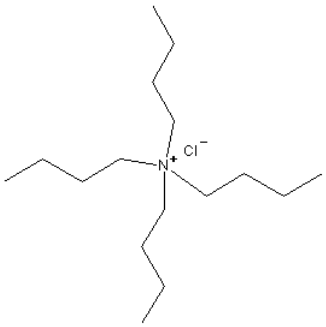

Figure 1-14 TBAC structure.

1.4.2 Detection with ICP-MS

As depicted in Figure 1-15, the ICP-MS instrument consists of several components, including the spray chamber, plasma, quadrupole and detector. The role of these components in the sample introduction and the production, focussing and detection of the ions are briefly discussed below.

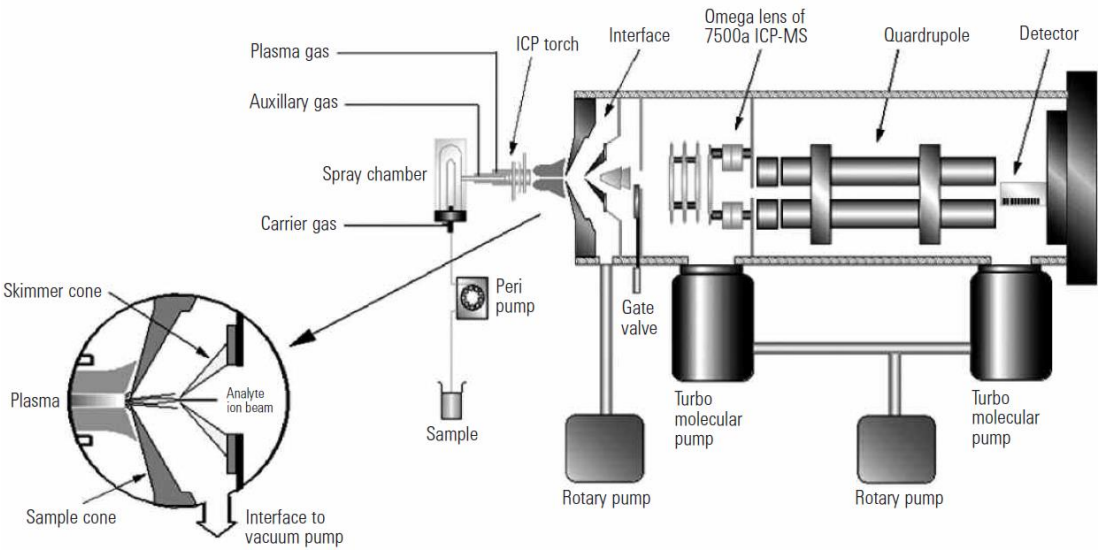

Figure 1-15 **Schematic of the Agilent ICP-MS set up.**
Schematic taken from Agilent Technologies [142].

Sample introduction

The liquid sample is introduced into the ICP-MS through the nebuliser. The purpose of the nebuliser is to convert the liquid sample into an aerosol for transport into the plasma. The large aerosol droplets are then removed in the spray chamber, allowing only small droplets to be introduced into the plasma.

Ion production and focusing

The plasma is the source of ion production, and at ~6500 K, has sufficient energy to dry, atomise and ionise the sample aerosol [142]. From the plasma, a sample of the ions enters the interface, which consists of the vacuum between the sample and skimmer cone (depicted in the inset in Figure 1-15). As only positive ions are produced in the plasma, the ion beam can be focussed by the electrostatic field generated by the following lenses (this also removes the neutral species and photons from the ions). The ion beam then passes through a collision/reaction cell where the ion beam reacts with the chosen gas (i.e. H_2, He) to remove interfering species (not required for HPLC-ICP-MS). The ions are then mass filtered by the electric field generated by the quadrupole (comprised of two pairs of cylindrical rods, with varying AC and DC fields), so only ions with the desired mass-to-charge ratio (m/z) reach the detector.

Detection

The most common detector in ICP-MS instruments is the electron multiplier [142]. As depicted in Figure 1-16, the electron multiplier is made up of a series of dynodes, which

release several electrons upon impact of an ion on the first dynode surface. The released electrons continue to impact on the dynodes located further down the detector, effectively amplifying the signal. This amplification is largely responsible for the high sensitivity associated with ICP-MS [142].

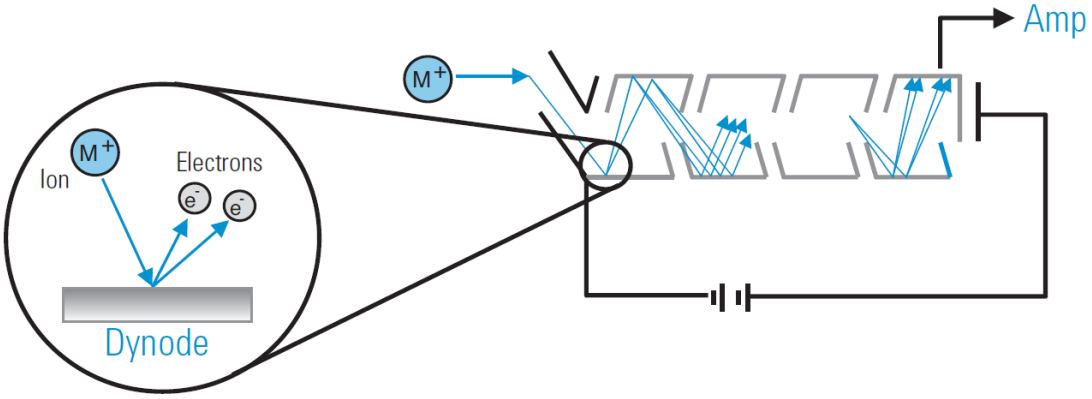

Figure 1-16 **Schematic of electron multiplier.**
Schematic taken from Agilent Technologies [142].

1.5 Software and modelling

1.5.1 The Geochemist's Workbench® (GWB)

The Geochemist's Workbench® (GWB) [125] is a geochemical modelling software package. The programs consist of RXN, ACT2, TACT, SPECE8, REACT, GTPLOT, X1T, X2T, XTPLOT. GWB allows users to balance chemical reactions, calculate equilibrium constants, trace reaction pathways and generate stability diagrams.

As detailed by Bethke [125], GWB modelling consists of:

- Defining the initial equilibrium system (i.e. the amounts of minerals in the system and dissolved components in the fluid, fugacities of gases, temperature and the pH).

- Writing a combination of mass action equations (relating to the species' activities to the reaction's equilibrium constant), mass balance equations (the concentrations of the system's species, minerals and gases which add up to give the system's bulk composition) and charge balance equations (where the ionic species in the system are balanced for an overall system electroneutrality).

These equations are then solved iteratively to obtain the distribution, concentration and activity of the species in the equilibrium state.

1.6 Conclusions & Project Premise

In view of the limited direct speciation techniques for aqueous gold in groundwaters, there is a need to develop a sensitive analytical method for the speciation of gold. A direct and sensitive speciation technique for gold in waters may lead to improved gold recovery and extraction processes, benefit gold mineral exploration and advance the understanding of the geological and geobiological cycle of gold. Gold complexes that are significant in the environment and in mining processes include gold cyanide, gold-sulfur, and gold-halide complexes. Hence, the research in this thesis is focussed on speciating complexes that are readily available and known to be soluble in water: Au(I)-thiosulfate, Au(I)-cyanide, Au(III)-chloro-hydroyl complexes and Au(III)-bromo-hydroxyl complexes. In order to conduct the studies in this project a HPLC-ICP-MS method will need to be developed for the speciation of gold.

Therefore the aims of this research are:

1. Systematically develop a HPLC-ICP-MS method for the speciation of Au(I)-thiosulfate, Au(I)-cyanide, Au(III)-chloro-hydroxyl complexes and Au(III)-bromo-hydroxyl complexes.

2. Determine the speciation of gold in various mine wastes and groundwaters samples.

3. Investigate possible mechanisms for the formation of Au(III) complexes in groundwaters.