Fungal-Transformation of Surrogate Sulphides and Carbonaceous Matter in Refractory Gold Ores: Revisited*

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Abstract

In the recovery of gold from refractory gold ores, pretreatment is required to decompose sulphides and liberate occluded gold before cyanidation, and to deactivate carbonaceous matter and prevent it from adsorbing dissolved gold. Until the past three decades, most commercial pretreatment processes had been by abiotic means. Biological pretreatment methods on commercial basis is therefore a recent phenomenon, and several researches are underway to assess the ability of different biological agents in the breakdown of sulphur and carbonaceous matter (CM) in gold ores. This paper, which is a revisit of an earlier one, presents an overview of on-going research aimed at assessing the capability of the fungus, *Phanerochaete chrysosporium*, to degrade sulphides and CM. Surrogate carbonaceous materials (lignite, bituminous and anthracite coals) and pure sulphides (pyrite and arsenopyrite) were used to model the behavior of CM and sulphides in refractory gold ores. To monitor the extent of biotransformation, preg-robbing test was conducted on the as-received and treated CM, and sulphide sulphur in the residual sulphidic materials was also determined. From an initial preg-robbing effect of 125 µg of gold per gram of CM, the ability of CM to preg-rob gold reduced by 70-95% in the order of lignite<bituminous <anthracite within 21 days of treatment, whereas there were 18% and 39% oxidation of sulphide sulphur in pyrite and arsenopyrite respectively. XRD examination of the treated anthracite confirmed reduction in the graphitic structure of carbon following fungal transformation. Similarly, there was a decline in the major sulphide peak after microbial pretreatment. The results indicate that the fungus biotransforms through destruction of the ordered structure, followed by introduction of oxygen groups. The amorphous nature, thus generated, inhibits the uptake of aurocyanide ions by CM, while enhancing the affinity of cyanide for the oxidised sulphide material in subsequent cyanidation treatment. The findings contribute to knowledge on novel and technically viable alternative methods for oxidative pretreatment of refractory gold ores.

Keywords: *Phanerochaete Chrysosporium*, Carbonaceous Matter, Metal Sulphides, Biotransformation

1 Introduction

1.1 Gold Ores and Refractoriness

Gold ores can be classified broadly as non-refractory (alluvial and free-milling) and refractory from a metallurgical standpoint, depending on the mineralogy and the ease of gold extraction. In alluvial ores, gold exists as discrete particles naturally liberated by weathering, and may be recovered by scrubbing and concentration processes, where the high specific gravity of gold is exploited for separation. Free-milling ores are those from which about 95% of the gold can be recovered by gravity concentration and/or simple cyanidation after milling to 80% passing 75 µm (Marsden and House, 2006).

Refractory gold ores are more difficult to treat, and depending on the degree of refractoriness, recovery could be below 50% (Vaughan, 2004). Since gold ores are fixed resources, the incessant mining and subsequent depletion of non-refractory gold ores continues to generate profound interest in research on gold recovery from refractory ores. These hard-to-treat ores occur in several gold mining regions throughout the world, and production of gold from refractory ores continues to increase with time (Boyle, 1979; Brierley, 1997; Adam *et al.*, 2017).

In refractory ores, gold usually exists as extremely fine particles (Marsden and House, 2006) in intimate association with gangue materials, making gold extraction by direct cyanidation and carbon adsorption processes inefficient and unattractive. The major problem of refractoriness centres on the presence of metal sulphides and/or carbonaceous matter (CM). In sulphidic refractory gold ores, tiny gold particles may be highly disseminated and locked up or occluded within the grain boundaries or fractures of sulphides, typically pyrite and arsenopyrite. For this reason, decomposition of the sulphides is required to liberate the gold for subsequent cyanidation (Boyle, 1979; Guay, 1981; Arriagada and Osseo-Asare, 1984; Ofori-Sarpong *et al.*, 2011; 2013b; Adam *et al.*, 2017). The presence of carbonaceous matter (CM) in gold ores leads to two main deleterious effects: (a) confinement of gold with attendant difficult release from the CM matrix and (b) loss of dissolved metal via the ability of CM to adsorb gold from gold-impregnated solution (Afenya, 1991; Pyke *et al.*, 1999; Helm *et al.*, 2009; Ofori-Sarpong *et al.*,

2010; Yang *et al.*, 2013; Liu *et al.*, 2014). Unlike activated carbon, adsorption of gold by CM leads to gold losses as the fine particles escape the openings of the typical screens used in the leaching circuit, and this phenomenon is termed as preg-robbing (Osseo-Asare *et al.*, 1984; Hausen and Bucknam, 1985). Preg-robbing of CM is generally investigated using various coal ranks as surrogates due to the similarities between CM and coal (Ibrado and Fuerstenau, 1992; Amankwah and Yen, 2006; Ofori-Sarpong *et al.*, 2010).

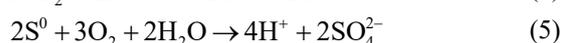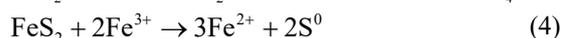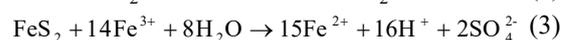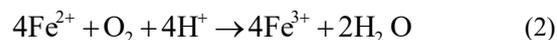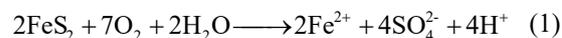## 1.2 Pretreatment of Refractory Gold Ores

For gold locked up in the refractory components, especially sulphides, to be amenable to cyanidation, pretreatment is required to decompose these shielding minerals, thus liberating the gold. The processes that have been used commercially to pretreat refractory gold ores include roasting, pressure oxidation and bacterial oxidation (Arriagada and Osseo-Asare, 1984; Berezowsky *et al.*, 1988; Marsden and House, 2006). Roasting involves the use of high temperature (450-750°C) to decompose sulphidic and carbonaceous materials in the presence of oxygen/air. The combustion reaction leads to generation of harmful gases such as sulphur dioxide and arsenic trioxide, which cause environmental problems if mishandled (Komnitsas and Pooley, 1989; Nyavor and Egiebor, 1992; Marsden and House, 2006). Pressure oxidation makes use of high temperature and pressure, typically 180-225°C and 1500-3200 kPa, to decompose metal sulphides in the presence of water. Though this process eliminates the harmful gaseous products, it is associated with operational difficulties such as safety due to high operating temperatures and pressures, higher rates of material corrosion, and higher equipment cost (Yannopoulos, 1991; Berezowsky *et al.*, 1988; Marsden and House, 2006).

In view of the above, the use of biooxidation which can be seen as the biological counterpart of pressure oxidation has become more prominent for the past two decades. Biooxidation partly reduces the high cost associated with roasting, and generally improves upon gold recovery and environmental/safety aspects of processing (Brierley, 1997; Hackl, 1997; Rawlings, 1998; Marsden and House, 2006).

1.2.1 Biooxidation of Sulphidic Gold Ores

Several bacteria are known to oxidize sulphides but the ones commonly used in commercial biooxidation of refractory gold ores are *Leptospirillum ferrooxidans*, *Acidithiobacillus caldus*, *Leptospirillum ferriphilum*, *Acidithiobacillus thiooxidans*, *Acidithiobacillus ferrooxidans* and *Sulfobacillus* spp (Rawlings, 1998; Amankwah *et al.*, 2005; Harneit *et al.*, 2006). The

bacteria catalyze the biooxidation reaction by oxidizing iron (II) and elemental sulphur respectively to iron (III) and sulphuric acid, which act as lixiviants for indirect oxidation of the sulphides (Holmes and Bonnefoy, 2006). The main reactions involved in pyrite oxidation can be summarized in Equations 1 to 5 (Bennett and Tributsch, 1978; Sand *et al.*, 1995).

$$2FeS_2 + 7O_2 + 2H_2O \longrightarrow 2Fe^{2+} + 4SO_4^{2-} + 4H^+ \quad (1)$$

$$4Fe^{2+} + O_2 + 4H^+ \rightarrow 4Fe^{3+} + 2H_2O \quad (2)$$

$$FeS_2 + 14Fe^{3+} + 8H_2O \rightarrow 15Fe^{2+} + 16H^+ + 2SO_4^{2-} \quad (3)$$

$$FeS_2 + 2Fe^{3+} \rightarrow 3Fe^{2+} + 2S^0 \quad (4)$$

$$2S^0 + 3O_2 + 2H_2O \rightarrow 4H^+ + 2SO_4^{2-} \quad (5)$$

Equation 1 is basically a chemical reaction, but it is reported to occur much faster in an environment of suitable bacteria whereas 2 and 5 do not proceed to any appreciable limit under atmospheric conditions and are said to rely entirely on bacterial catalysis. Though Equations 3 and 4 can proceed without microbial involvement, their products, Fe^{2+} and S^0, have to be oxidised by the bacteria to regenerate the lixiviants for ongoing oxidation of pyrite (Rawlings *et al*, 2003; Holmes and Bonnefoy, 2006; Marsden and House, 2006).

At the end of biooxidation, gold may be totally liberated or associated with relatively more porous iron oxides, and thus amenable to cyanidation (Marsden and House, 2006). Unfortunately, because most of the major bacterial strains are autotrophs, and thus meet their carbon requirement by synthesizing carbon dioxide, they do not metabolise organic carbon in the ore (Silver, 1970; Madigan and Martinko, 2006; Ofori-Sarpong and Osseo-Asare, 2013). Consequently, CM, if present, is not degraded, and thus routed into downstream cyanidation circuits where it preg-robs dissolved gold (Amankwah *et al*., 2005; Ofori-Sarpong *et al*., 2013b; Adam *et al*., 2017).

1.2.2 Biomodification of Carbonaceous Matter

Apart from roasting that aims at eliminating the organic carbon, passivation has been employed using carbon-blanking agents such as kerosene and other organic reagents (Abotsi and Osseo-Asare, 1987; Hutchins *et al*., 1988; Adams and Burger, 1998). Though yet to be applied commercially, biomodification of carbonaceous matter appears more attractive from economical and environmental points of view, as it occurs at relatively low temperatures (25-40 °C) and at atmospheric pressure. In addition, lixiviants are regenerated *in situ* due to the presence of microorganisms. Microorganisms employed in this manner include the bacteria; *Pseudomonas sp.*, *Achromobacter sp.*, *Streptomyces setonii*, and the
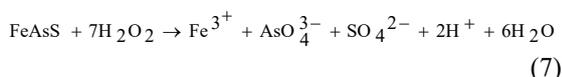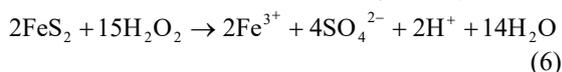
fungi; *Aspergillus bruneio, Penicillium citrinum, Trametes versicolor* and *Phanerochaete chrysosporium* (Brierley and Kulpa, 1993; Amankwah and Yen, 2006; Yen *et al*., 2008; Ofori-Sarpong *et al*., 2010; Konadu *et al*., 2016).

1.3 *Phanerochaete chrysosporium*

Phanerochaete chrysosporium is a white rot fungus which belongs to the basidiomycetous family; a family of filamentous fungi that exhibits mushroom-like growth with extensive network of cross-walled filaments (Madigan and Martinko, 2006; Ofori-Sarpong *et al*., 2013a). The filaments are able to penetrate wood and degrade lignin (the most recalcitrant component of wood) with the aim of getting access to cellulose and hemicellulose (Tien and Kirk, 1984), thus leaving specks of white delignified areas amidst thin areas of firm wood, a phenomenon referred to as 'white rotting'. The first lignin-degrading enzyme, lignin peroxidase (LiP), was isolated from *P. chrysosporium*, and characterized by Tien and Kirk (1984). The fungus has hence been studied as a model organism, since it grows quickly, degrades lignin rapidly, and has a relatively medium temperature optimum (37-39 ^{o}C) and low pH optimum (3.5-4.5), though it can grow over a wider range of temperature and pH (Ofori-Sarpong *et al*., 2010; 2011).

P. chrysosporium also secretes another enzyme, called manganese peroxidase (MnP). The oxidative enzymes of the fungus catalyze the biodegradation of lignin and a variety of aromatic carbonaceous materials including low rank coals, wood chips and environmental pollutants (Glenn *et al*., 1983; Bumpus and Aust, 1986; Ralph and Catcheside, 1997; Tunde and Tien, 2000; Martin and Petersen, 2001). Carbonaceous matter (CM) in refractory gold ores is characterized to be similar to coal, and thus potential substrates for the fungus (Ofori-Sarpong *et al*., 2013b).

Aside LiP and MnP, *P. chrysosporium* secretes a H_2O_2-generating enzyme, glyoxal oxidase (Kersten and Kirk, 1987), and hydrogen peroxide is known to solubilize pyrite and arsenopyrite as presented in Equations 6 and 7 (Antonijevic *et al.*, 1997; Jennings *et al.*, 2000). The iron (III) and sulphuric acid produced, constitute the reagents created by the sulphide-oxidising bacteria for the indirect biooxidation of sulphides (Sand *et al.*, 1995; Hackl, 1997; Holmes and Bonnefoy, 2006).

$$2FeS_2 + 15H_2O_2 \rightarrow 2Fe^{3+} + 4SO_4^{2-} + 2H^+ + 14H_2O$$
(6)

$$FeAsS + 7H_2O_2 \rightarrow Fe^{3+} + AsO_4^{3-} + SO_4^{2-} + 2H^+ + 6H_2O$$
(7)

The strong oxidizing environment produced by the oxidative enzymes of *P. chrysosporium* is also known to be responsible for the oxidative removal of sulphur from coal (Gonsalvesh *et al*., 2008). *P. chrysosporium* therefore has the potential to catalyze the biotransformation of carbonaceous matter and sulphides, and boost gold extraction in the subsequent cyanidation step (Ofori-Sarpong *et al*., 2010; 2011; 2013b; Liu *et al*., 2014; Konadu *et al*., 2016).

This paper presents a review of the authors' ongoing research that utilizes fungal-mediated processes to achieve decomposition of metal sulphides and deactivation of carbonaceous matter in refractory gold ores. Analysis of the data obtained from this study is a contribution to the pool of scientific knowledge regarding processing of refractory gold ores and hence is of immense benefit to academia. In addition, the results could find application in gold production communities.

2 Resources and Methods Used

2.1 Materials

Coal of various ranks was used as surrogate for carbonaceous matter whereas pure pyrite and arsenopyrite were used to represent sulphides in refractory gold ores. Pyrite and arsenopyrite samples were supplied by Ward's Natural Science Establishment in the USA. Coal (anthracite, bituminous, and lignite) samples were provided by the Coal Bank of the Earth and Mineral Science Energy Institute whereas fungal spores of *P. chrysosporium* ME446 were obtained from Prof. Ming Tien of the Department of Biochemistry and Molecular Biology, all of the Pennsylvania State University, USA. The sulphide and carbon materials were crushed and sieved to all passing 250 μm before incubation. Standard gold solution (50 mg/L in 1 g/L sodium hydroxide and 0.5 g/L sodium cyanide) was supplied by High Purity Standards, USA, and all other chemicals used were of reagent grade.

2.2 Medium Preparation, Incubation and Harvesting of Treated Material

The millet and wheat bran (MWB) medium was prepared by using 8 g millet plus 2 g wheat bran. Double-distilled (dd) H_2O used for the incubation was buffered with succinic acid and the pH adjusted to 4 using sodium hydroxide. The medium was moistened with water in Erlenmeyer flasks, in the ratio of 1 g medium: 1 mL dd H_2O, and autoclaved at 121 ^{o}C for 30 min. On cooling, the medium was inoculated with 1 mL of spore suspension of *P. chrysosporium* (made by suspending 1 vial of spores in 25 mL of dd H_2O). The optical density of the cell suspension used for inoculation was 0.3 relative to water, as measured with a Pharmacia LKB Ultraspec II UV-visible spectrometer at 600 nm. The samples were

incubated in triplicates at 60% solids and 37 °C for up to 21 days (Ofori-Sarpong *et al.*, 2010; 2011). Fig. 1 shows autoclaved MWB medium (a) and coal cultured with *P. chrysosporium* in MWB (Ofori-Sarpong *et al.*, 2010).
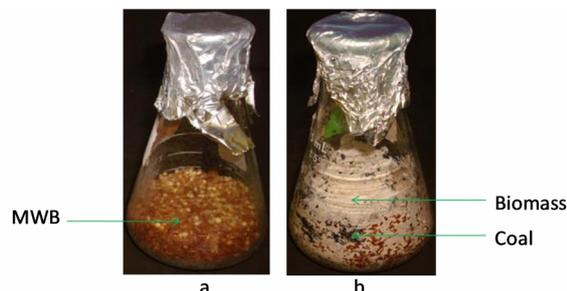

Fig. 1 Autoclaved MWB Medium (a) and Coal Cultured with *P. chrysosporium* in MWB (b) (After Ofori-Sarpong *et al.*, 2010)

Control experiments were also set up for 3-14 days under similar conditions as described above, except that there was no addition of fungus. This was necessary to establish the exclusive effect of the fungus on biotransformation of sulphides and CM. At the end of the incubation period, the samples were washed with water in the ratio of about 1 g sample to 200 ml H_2O to get rid of the media and fungal biomass, and then dried at 37 °C for 7 days.

2.3 Determination of the Extent of Biotransformation

Various examinations were performed on the as-received and treated carbonaceous and sulphidic materials to assess the extent of biotransformation. These include gold adsorption, sulphur speciation and x-ray diffraction studies.

2.3.1 Preg-robbing of Carbonaceous Materials

Triplicate samples of biomodified, control and as-received coals of various weights (0.1 g - 1 g) were contacted with 25 mL of 5 mg/L gold solution at pH 11.5 and agitated at 150 rpm for 24 hours. At the end of the contact time, the residual solution was filtered, and gold in the filtrate, determined using Perkin-Elmer Optima 5300 inductively coupled plasma – atomic emission spectrometer (ICP-AES). The difference between the concentration of gold in solution before and after the adsorption test was computed with respect to the amount of carbon-containing material used for the adsorption, as shown in Equation 8, to obtain the Preg-robbing Effect of Carbon (PEC) in g/t.

$$PEC\left(in\ \frac{gram\ of\ gold}{tonnes\ of\ carbon}\right) = 25\ mL\ x\left((IC_{Au} - FC_{Au})^{\mu g}/_{mL}\right)\left(\frac{1}{W_C\ g}\right) \quad (8)$$

In Equation 8, IC and FC are the initial and final concentrations of gold in solution, W_C is the weight of carbonaceous material used in the adsorption test, and 25 mL is the volume of gold solution used. The difference in PEC values between as-received and treated carbonaceous material gives an indication of the effect of fungal-biomodification on preg-robbing of gold by the carbon-containing materials.

2.3.2 Sulphide Sulphur Oxidation in Sulphide Materials

Sulphide sulphur in the residual sulphidic materials was determined in triplicates by the volumetric combustion technique using LECO Sulphur determiner SC-4444DR. These values compared with those obtained for the as-received gave the extent of biotransformation. Equation 9 depicts the Percentage Conversion of Sulphur (PCS) in the residual solids (Ofori-Sarpong *et al.*, 2011), where S_B and S_A are respective sulphide sulfur contents before and after fungal treatment.

$$PCS\ (wt\%) = \frac{(S_B - S_A)\ (g)}{S_B\ (g)}\ x100\% \quad (9)$$

2.3.3 X-ray Diffraction of Carbonaceous and Sulphidic Materials

The XRD analysis was carried out using PANalytical X'Pert Pro powder diffractometer with X'celerator detector. The samples were ground to fine powder in an agate mortar and then sprinkled on the surface of quartz zero-background sample holder. A Ni-filtered CuK_α radiation produced at 45 KV and 40 mA was used for the analysis. The scan was run at a scan speed of 2 deg/min from 20 to 34 deg and 25 to 50 deg 2-theta for anthracite and pyrite respectively. Data acquisition was done using MDI Jade 9 software.

3 Results and Discussion

This paper discusses the pretreatment of carbonaceous matter and sulphides generally associated with refractory gold ores. The transformation was done by incubating surrogate substrates with the fungus, *Phanerochaete chrysosporium* maintained on millet and wheat bran (MWB) medium. The results are presented and discussed in the ensuing sections.

3.1 Fungal-transformation of Carbonaceous Matter

Anthracite, bituminous, and lignite coals were used as surrogates to study the carbonaceous matter (CM) in gold ores, and the extent of biotransformation was measured primarily by the reduction in the ability of these coals to adsorb aurocyanide complex. To measure the degree of reduction in gold adsorption, it was necessary to

know the amount of gold that can be adsorbed by the various coals in their as-received states, and this is presented in Fig. 2. It is clear from the figure that only anthracite was able to adsorb a substantial amount of gold cyanide complex; about 750 g/t. In contrast, lignite and bituminous coals adsorbed less than 50 g/t, and this was attributed to their relatively immature nature (Ofori-Sarpong *et al.*, 2010).
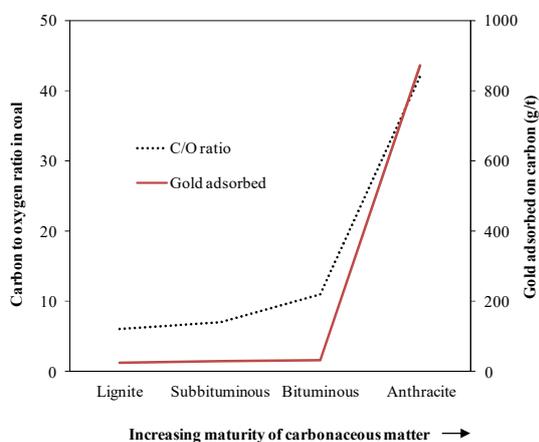

Fig. 2 Relationship of Coal Maturation to Gold Adsorption Ability

As coal matures from lignite to anthracite in the coalification process, there is cleavage of hydrogen and oxygen from the coal structure, leaving a skeletal carbon structure similar to that obtained in the production of activated carbon (Marsden and House, 2006). The maturity of coal is thus directly related to the degree of graphitization, which decreases in the order of anthracite > bituminous > sub-bituminous > lignite, with a corresponding decrease in the carbon to oxygen ratio (Van Krevelen *et al.*, 1993). The graphitic structure of carbon is known to be a major factor in the adsorption of gold cyanide onto carbon (Jones et al, 1989; Ibrado and Feurstenau, 1992; Helm *et al.*, 2009; Amankwah and Yen, 2006; Ofori-Sarpong *et al.*, 2010; 2013a). The preferential adsorption of gold cyanide on anthracite relative to the other coals is therefore due to better developed graphitic structure in anthracite. This is demonstrated in Fig. 2, which relates the carbon to oxygen ratios in the various coal to their gold adsorption capacity. Correlating the carbon-to-oxygen ratios in various coals with gold adsorption, it was inferred by Ofori-Sarpong *et al.* (2010) that the adsorption of gold cyanide on anthracite is due to its high carbon-to-oxygen ratio of about 42 compared to 5-11 for lignite, sub-bituminous and bituminous coals. Despite the relatively low degree of gold adsorption on the other coal ranks, Fig. 3 indicates that the ability to adsorb gold declined drastically for all the coals following fungal transformation. From an initial preg-robbing effect of 125 µg of gold per gram of CM, the ability of CM to preg-rob

gold reduced by 70-95% in the order of lignite<bituminous <anthracite within 21 days of treatment. This is possibly due to relative gold adsorption abilities of the various coals in their as-received states.
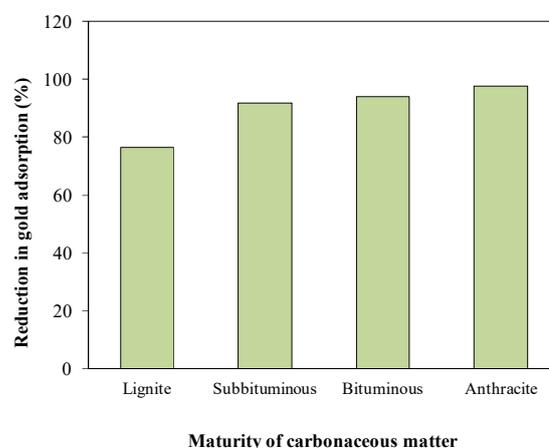

Fig. 3 Reduction in Gold Cyanide Adsorption on Biotransformed Coal

Since anthracite has the ability to adsorb more than 10 times that of the other coals, it was necessary to extend the investigation to cover other factors that may affect the degree of biotransformation of anthracite. The effect of fungal transformation time on gold adsorption as presented in Fig. 4, shows more than 90% decrease in preg-robbing after treatment for one week. Also, to demonstrate the exclusive effect of the fungus on the decline in gold adsorption by anthracite, control experiments were set up using the medium alone, and the result is depicted in Fig. 5.
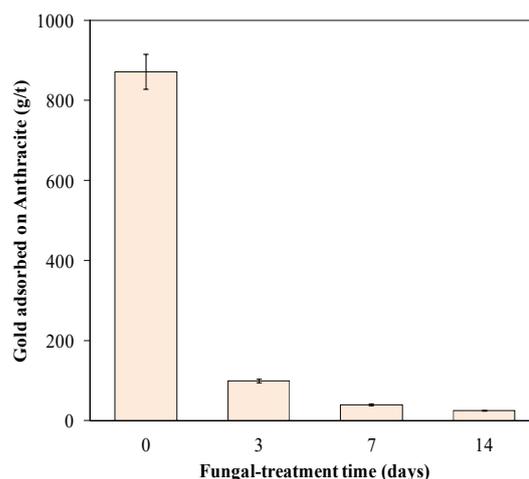

Fig. 4 Effect of Fungal Transformation Time on Gold Adsorption by Anthracite Coal
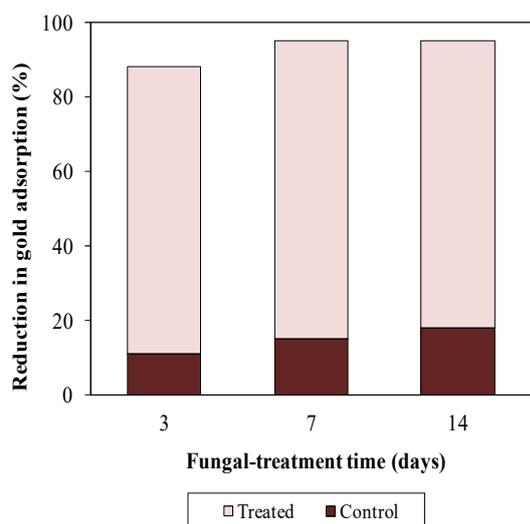
Fig. 5 The Exclusive Effect of *P. chrysosporium* on Biotransformation of Anthracite Coal

The lower portions of the bars represent percentages of the total transformation attributed to the medium alone whereas the upper portions show the additional reduction in preg-robbing capacity of anthracite, when the medium was cultured with *P. chrysosporium*. It is clear from the figure that, though the medium alone results in some blinding of adsorption sites (Ofori-Sarpong *et al*., 2010; 2013a), the presence of the fungus imparts about 400% more deactivation on the carbonaceous matter.

Characterization of anthracite using XRD as shown in Fig. 6, indicates a great reduction in the graphite peak at 27 deg on the 2-theta axis as a result of fungal-treatment. This peak is characteristic of very structured graphite. Thus a reduction in this peak indicates a decrease in the J-factor, which implies an increase in the degree of disorderliness/amorphousity (Gordon and Asiam, 2012) of the treated carbon relative to the as-received. Since gold adsorption is reported to occur mostly on the graphitic planes (Ibrado and Fuerstenau, 1992), destruction of the graphitic structure followed by introduction of oxygen groups will disrupt the continuous planes required for gold cyanide adsorption, and this in part, explains the reduction in gold adsorption (Ofor-Sarpong *et al*., 2013a; Liu *et al*., 2014). In work done by Konadu *et al*. (2016) on biotransformation of powdered activated carbon (PAC) using enzymes of *P. chrysosporium*, the authors recorded a decrease of 57% in post-treatment gold adsorption. Futher characterization of the solid residues of PAC by ^{13}C NMR revealed a decomposition of polyaromatic into aliphatic carbons, whereas the specific surface area also reduced by roughly 50%.
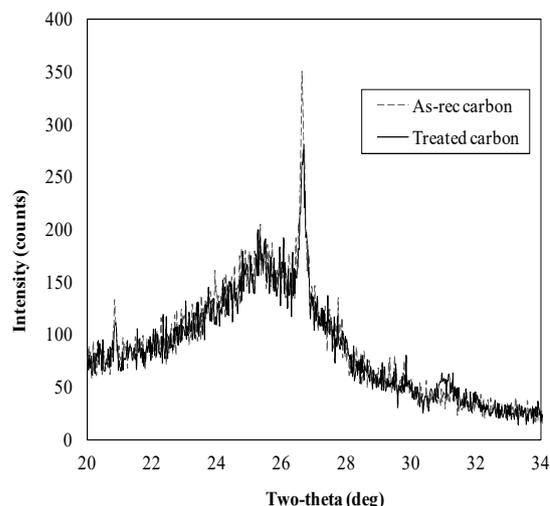

Fig. 6 X-ray Diffraction of Anthracite Coal before and after Fungal-treatment

3.2 Fungal-transformation of Metal Sulphides

The forms of sulphur in sulphur compounds are generally classified into sulphide sulphur, elemental sulphur and sulphate sulphur, with oxidation states of -2, 0 and +6 respectively. Sulphide (pyritic) sulphur, being the most reduced form, may be oxidised into several forms such as elemental sulphur, dithionate, thiosulphate and sulphate, some of which are unstable and may further transform into other products. The transformaton of sulphides may thus be monitored by the reduction in concentration of sulphide sulphur in the parent material. The transformation of pyrite and arsenopyrite were measured using the LECO sulphur titrator, and the resulting sulphide sulphur oxidation are depicted in Figs. 7 and 8 respectively, whereas Fig. 9 compares the control experiment with the fungal-treatment after a 2-week contact time.
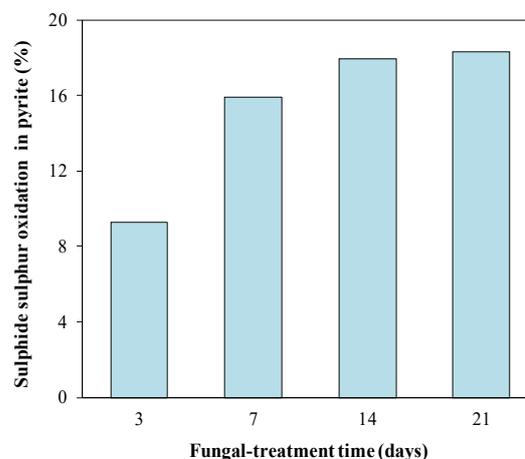

Fig. 7 Oxidation of Sulphide Sulphur in Pyrite following Pretreatment with *P. chrysosporium*
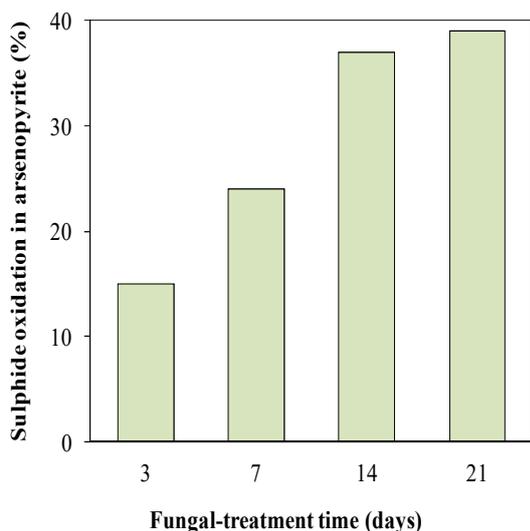
Fig. 8 Oxidation of Sulphide Sulphur in Arsenopyrite following Pretreatment with *P. chrysosporium*

Figs. 7 and 8 show time dependence of the sulphide transformation especially in the case of arsenopyrite. Pyrite realised about 10% transformation within 3 days of treatment, and this increased to about 16% in 7 days plateauing off at 18% in 21 days. In the case of arsenopyrite, there was about 15% transformation in 3 days, 24% in 7 days, 36% in 14 days and 39% in 21 days. Biotransformation in arsenopyrite was higher due to its less noble state. Relating the extent of fungal-transformation to the control experiments, it is clear from Fig. 9 that the transformation of sulphides were mainly due to the presence of the fungus, as more than 4-fold oxidation (400%) of sulphide sulphur was achieved.

X-ray diffraction studies of pyrite is presented in Fig. 10. The main sulphide peak occurs at 33 deg on the 2-theta axis, and the figure shows that following fungal transformation, the peak decreased by more than 50%, indicating a major degree of alteration. This implies a change in J-factor of the sulphides from an undisturbed lattice value of 1 to about 0.5 following fungal-transformation. Since the sulphides used in this study were the pure forms containing 52% and 20% sulphide sulphur respectively in pyrite and arsenopyrite, there is no doubt that a higher degree of transformation can be achieved when treating refractory gold ores and concentrates which usually contain less than 15% sulphide sulphur (Osseo-Asare *et al*., 1984; Ofori-Sarpong *et al*., 2013b).
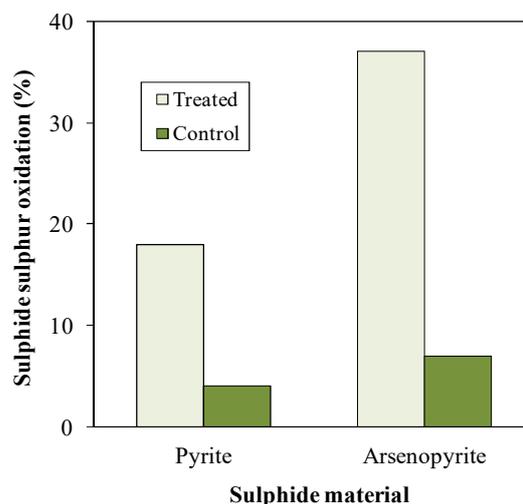

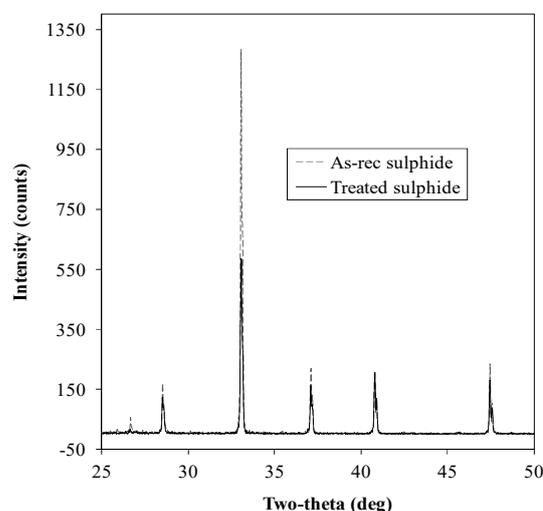**Fig. 9 The Exclusive Effect of *P. chrysosporium* on Biotransformation of Sulphides**

Fig. 10 X-ray Diffraction of Pyrite before and after Fungal-Treatment

4 Conclusion

This paper reports on an overview of the authors' research into the possibility of pretreating refractory gold ores using the fungus, *P. chrysosporium*. Coal of various ranks was used as surrogate for carbonaceous matter whereas pure pyrite and arsenopyrite were used to represent sulphides in refractory gold ores. The study revealed a reduction of more than 90% gold adsorption by anthracite coal, which has about 15 fold higher ability to adsorb aurocyanide than the other coals. The ability of anthracite to adsorb relatively higher levels of gold cyanide is due to the well developed graphitic structure which is important for aurocyanide adsorption. As a result of fungal treatment, there was disruption in the graphitc structure, and this accounted for the reduction in gold adsorption.

In the case of the pure sulphides, there was about 18% and 39% transformation of pyrite and arsenopyrite respectively by fungal-treatment. Comparative studies using XRD revealed a change in J-factor of the sulphides from an undisturbed lattice value of 1 to about 0.5 following fungal-transformation. The findings suggest a higher degree of transformation when treating refractory gold ores and concentrates which usually contain less than 15% sulphide sulphur.

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