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Microbial Recovery of Gold from Mahd El-Dahab Mine Kingdom of Saudi Arabia

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Abstract

This paper considers the use of the yeast *Saccharomyces cerevisiae* as a biosorbent for the recovery of gold from Mahd El-Dahhab gold mine in Kingdom of Saudi Arabia. Gold is a noble metal which was seldom used in electroplating, hydrogenation catalyst, etc. Heterogeneous composition of samples and low concentration offer renewed interest in the selective extraction of gold using various extractants. In the present work, *Saccharomyces cerevisiae*, is used to study the preconcentration of gold, which in the original sample was 0.44 ppm, while after treatment with *Saccharomyces cerevisiae* it raises to 7 ppm with efficiency reaching about 1600%.

Keywords

Recovery, *Saccharomyces cerevisiea*, Gold

Subject Areas: **Biotechnology, Microbiology**

1. Introduction

Gold, the noblest one of metals, has been used by man for more than 5000 years. Its extreme softness or malleability, and resistance to tarnish (oxidation), led to its earliest use in art and currency. Gold is the metal of choice for jewelers, and is often used in dentistry. Gold has been successfully used in many modern technological applications. It is used as the electrical contacts of computer chips. Minute quantities of gold (less than 3 micrograms) are vaporized to mirror lens surfaces [1].

Minor concentrations of gold occur in most natural substances. In seawater, for example, there is approximately 0.012 parts per billion (ppb) of gold and in fresh water it is slightly higher at 0.02 ppb. Its average concentration in the Earth's crust or lithosphere is approximately 5 ppb, and in certain sedimentary rocks it may achieve concentrations of up to 2100 ppb or 2.1 parts per million (ppm).

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1.1. Geology of Mahd El Dahab

Mahd El Dahab is an epithermal, low sulphidation, adularia type, polymetallic deposit. The total production has been estimated at over 100 tons of gold [2]. The reserves before the last stage of exploitation were around 1.2 Mt at 24 g/t Au, 92 g/t Ag, 0.65% Cu and 3.11% Zn.

Systematic rock geochemistry over a 50 m × 50 m grid discovered new reserves [3]. The meta-volcanics are rich in quartz veins. The polymetallic mineralization of chalcopyrite, sphalerite, galena, gold and silver tellurides, and rare electrum occurs in a dense vein network. the quartz veins contain cockade breccias , felsic agglomerate and crystal tuffites. The mineralization is older than 709 Ma, the age of the intersecting dikes [4]. Zinc grades increase to the North and at depth but Au grades decrease commensurately. In Nov 1999, the remaining 0.65 Mt were estimated to contain 12 g/t Au and 3.11% Zn.

Four successive alteration stages are observed: early alteration with quartz, sericite, pyrite; an intermediate stage with chlorite, sericite, microcline, sphalerite, pyrite; the gold bearing stage with quartz, chlorite, sphalerite, galena, pyrite, gold, tellurides, and accessory siderite-calcite-hematite and a final stage with quartz, calcite, and barite. Minor (apparently) gold indications are known from near Mahd Adh Dhahab, as Lahuf [5] (**Figure 1**).

1.2. Previous Work of Leaching

Extraction of gold from solutions is under active investigation by using a variety of physical, chemical, and biological processes. Recovery of ionic gold from dilute solutions usually involves either precipitation by zinc dust, carbon adsorption, solvent extraction, or ion exchange resins. All of these are of low selectivity and comparatively expensive [6] [7]. Chemical methods for the recovery of gold from ores include cyanidation and thiourea leaching, which present environmental and health risks [8] [9].

Figure 1. Location map of Mahd El Dahab, Kingdom of Saudi Arabia.

Biorecovery of dissolved gold from solution presents fewer environmental risks than chemical methods, and is documented for microorganisms [7] [10] [11], algae [7] [9], yeast [12] and fungi [7] [13]-[15].

The ore contained 8.2% Fe, 0.78% Cu, 0.88% As, and 0.035% with pyrite, hematite, arsenopyrite, and chalcopyrite as the main metal-bearing minerals. Initial gold recovery by conventional cyanidation on a crushed ore sample was 54%; concentration by flotation improved recovery to 56%. Concentrated samples (17.0 g Au/ton) were leached in reactors at pH 1.8. In the presence of bacteria, all dissolved iron was presented as ferric ion; gold recovery by cyanidation increased from 13% for the initial concentrate to 97% after 10 days of bacterial leaching. To further increase gold recovery, flotation tailings were submitted to cyanidation [16].

Algal cells, alive or dead, rapidly accumulated Au^{3+} and begin to reduce it to Au^{0} and Au^{+} within 2 days [17]. Four species of ground dried seaweeds *(Sargassum sp*., *Gracilaria sp*., *Eisenia sp*., and *Ulva sp*.) [18] treated seaweeds removed 75% - 90% of the gold within 60 min at pH 2 from solutions containing 5.0 mg Au^{3+}/L.

Gold (Au^{3+}) can be sequestered from acid solutions by dead biomass of a brown alga, *Sargassum natans* (Linnaeus), and deposited in its elemental form, Au^{0}.

The cell wall of Sargassum was the major locale for gold deposition, with carbonyl groups (C=O) playing a major role in binding, and N-containing groups have lesser role. Like activated carbon, the biomass of *Sargassum natans* is extremely porous, reportedly more than most biomaterials, and accounts, in part for its ability to accumulate gold [19].

The bioreduction of Au^{+3} to Au^{0} using biomass of the brown alga *Fucus vesiculosus* was investigated. The recovery and reduction process took place in two stages with an optimum pH range of 4 - 9 with a maximum uptake obtained at pH 7 [20].

The effects of ionic gold on *Saccharomyces cerevisiae*, extension of preincubation time of yeast cells in gold-containing medium resulted in a decreasing proton efflux rate and colloidal phase formation in the cell suspensions. The time between gold addition and the beginning of colloidal phase formation depended on the gold concentration used. Both Ca and Mg enhanced the inhibitory effect of gold on the yeast cells with Ca showing a stronger inhibitory effect than Mg [21].

2. Methodology

- Mahd El Dahab sampling:

Sample in this work was kindly obtained from Mahd El Dahab mine, Kingdom of Saudi Arabia.

- The organisms used in this work.

Saccharomyces cerevisiae used in this work was provided by EL-Hwamdiyah Company of Yeast and Sugar, Giza, Egypt. It is cultivated in (glucose-peptone-yeast extracts broth) medium which composition is

Yeast extracts	3 g L
Peptone	3 g L
Glucose	10 g L
Agar	15 g L
Dist. water	1000 ml.

The obtained glucose-peptone-yeast-extracts agar media was melted and poured into Petri dishes and then left to solidify, by using a sterile inoculation needle a loop full of one of colonies was taken and streaked on the surface of the solidified agar plates, followed by incubation for 72 hours at 30˚C. Sub culture was prepared by addition of pure *Saccharomyces* colonies on slants of glucose-peptone yeast agar.

- Preparation of liquid media:

An amount of 100 ml of glucose-peptone yeast extract previously prepared medium were added to 100 gm of Mahd Ad Dahab gold sample and let for incubation for 10 day at 30˚C. All flasks were sealed with removable cotton. Temperature was kept in the incubator at 28˚C - 30˚C during the experiment.

After separation the media, the result of Mahd Ad Dahab gold sample was dried and weighted then subjected to different analysis to identify the elements and groups which were affected by *Saccharomyces cerevisiae*.

- Sample analysis

The collected sample was identified through four collaborative techniques.

2.1. Fire Assay

Fire assay has been in use for thousands of years and still stands the test of time as the standard method to value

noble metals. A lot of attention are given to “micron gold” or “colloidal gold”. A cold hard fact of this type of gold requires a lot of material to make even one ounce. Generally, much more ore than an independent operator is equipped to move. No machine, recovery process, or chemical method will produce any gold if there are not enough values to collect it from the ore.

The ore must, in all case, be reduced to a state of fine powder so as to pass through at least a 60 mesh sieve. The metallic residues they must be carefully collected and cupelled separately with lead.

Pyritic ores: they are treated either by preliminary roasting at a low temperature until sweep and then treated just as the ores with basic gangues, or they may be fused at once with a heavy charge of red lead, and little or no charcoal.

A suitable charge would be:

Ore	50 gram
Red lead	60 gram
Borax	20 gram
Sodium carbonate	40 gram

A little sand may be added if silica is not already presented, and care must be taken to thoroughly saturate the charge with metallic iron. The precautions already referred to as to fritting of the charge, and the treatment of the button must be carefully absorbed. The sample was treated with fire assay before and after treatment with *Saccharomyces cerevisiae*

2.2. Environmental Scanning Electron Microscope

Environmental scanning electron microscope (ESEM), Philips XL 30 was applied for brief qualitative investigation of minerals in each sample for quick semi quantitative of the obtained products under the usual conditions at low vacuum. And voltage from 5 - 30 KV, the back scatter electron detector (BSE), and occasionally gas scatter electron detector (GSE) were used for photomicrographs. The attached unit of energy dispersive X-ray (EDX) was used to obtain the semi quantitative chemical analysis.

2.3. Infrared Spectroscopy

The samples are subjected to Infrared spectroscopy in order to determine the function groups of the samples. The used instrument is Naxux 670 FTIR, Company name (Nicollet) USA. The range of spectrum is usually performed from 4000 to 400 Cm^{-1}, with resolution 4 Cm^{-1}.

2.4. X-Ray Diffraction

XRD was used to identify the unknown minerals using instrument Philips PW3710/31 diffract meter with automatic sample changer PW1775, (21 positions) scintillation counter, Cu-target tube and Ni filter at 40 KV and MA. This instrument is connected to a computer system using X-40 diffraction program and ASTM cards for mineral identification in NMA.

3. Result and Discussion

The *Saccharomyces sp.* is one of the best studied and commercially exploited of all microorganisms [22]. *Saccharomyces cerevisiae* is a by-product of large scale fermentation and its annual production is in the order of millions of tons, thus it has the potential to provide large quantities of cheap biomass [23]. Yeasts require a minimum number of inorganic ions in micro and mill molar.

The roles of the ions in yeast cells are twofold:

1) Enzymatic functioning as part of the catalytic centre of an enzymes, as activators of enzyme activity and as physiological regulators. Zinc, cobalt, manganese and copper are common in catalytic centres; magnesium acts as an enzyme activator and potassium commonly functions as a metal co-enzyme;

2) A structural function. Potassium and magnesium have a role in neutralizing electrostatic interactions between cellular anionic units of polyphosphates, DNA, RNA and proteins [24]. Ca^{+}, Mi^{+} and Zn^{+} are often complexes to membrane phospholipids and cell wall phosphomannans.

The presence of cations in yeast is significant with respect to its exploitation as a metal biosorbent. Yeast cells require metals for normal metabolic processes and readily accumulate a range of metals. The second point re-

lates to the delicate intracellular ionic balance, with imbalances being reflected as alterations in metabolic processes and growth characteristics. Exposure of yeast from malt fermentation to high levels of metals in a bioremediation system would possibly lead to cell death and further metal removal would be by metabolism independent mechanisms.

The cell wall and plasma membrane are the most important components of the yeast cell in metal removal from aqueous solutions. The cell wall maintains structure and rigidity and is permeable to solutes smaller than 600 Dalton. It serves as the initial contact point and provides the negative functional groups for cation binding. The yeast cell wall consists of an intermesh of polysaccharide microfibrils in a matrix of various polysaccharides, proteins and lipids. The major constituent (~60%) of the cell wall is the polysaccharide mixture of glucans, phosphomannans and chitin [25] [26].

Chemical and enzymatic isolation of the cell wall components of *Saccharomyces cerevisiae* indicates metal binding to each of the cell wall components. The isolated components accumulate greater quantities of cations than intact cells, with the outer mannan-protein layer being identified as more important for metal binding than the inner chitin layer [27].

In contrast to bacteria with a single plasma membrane, *Saccharomyces cerevisiae* cells contain a number of specialized membranes. Most important to metal transport are the outer membrane separating the cell components from the external medium and the vacuolar membrane which compartmentalizes different metabolically important compounds [28]. The plasma membrane is composed of a lipid bilayer containing assymetrically located proteins. These proteins mediate the selective uptake and/or secretion of solutes.

Transport of solutes through the membrane is mediated by primary and secondary transport proteins. Primary transport is the active process in which chemical energy is converted to electrochemical energy [29].

Fire assay is used for the determination of gold before and after treatment with *Saccharomyces cerevisiae* the original gold content in the sample is 0.44 ppm and reach a higher value 7 ppm as shown in **Table 1** and **Figure 2**. BSE image of ESEM shows several images before starting treatment and **Figure 3** shows mineralized

Table 1. Fire assay analysis for gold.

Au Before (ppm)	Au After (ppm)
0.44	7

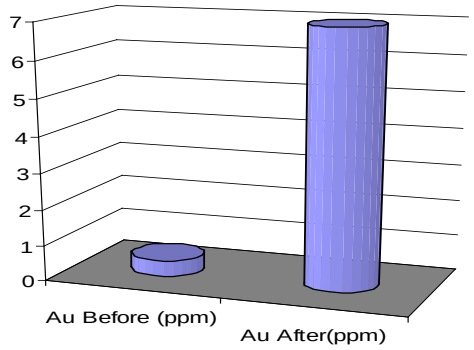

Figure 2. Fire assay analysis for gold.

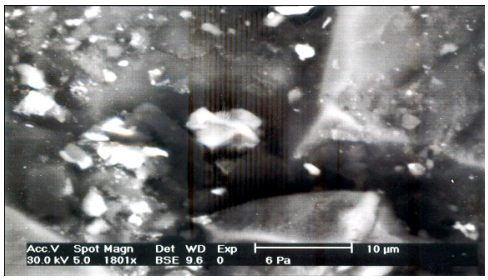

Figure 3. BSE image for mineralized volcanic rocks shows gold and sulphides.

volcanic rocks with gold and sulphides, while **Figure 4** shows gold crystal with chalcopyrite, and elongated gold crystal with high brightness in **Figure 5**. After treatment with *Saccharomyces cerevisiae* gold crystal appears associated with chalcopyrite in **Figure 6** while gold crystal only in **Figure 7**, finally **Figure 8** shows image showing a large agglomeration mode up of individual micro particles of gold due to microbial activity.

The cell wall of *Saccharomyces cerevisiae* was the major locale for gold accumulation. Treatment with *Saccharomyces cerevisiae* on Mahd Ad Dahab gold sample in infra red instrument notified the presence of functional group OH, Ch aliphatic, Co acidic and C-O-C, after action with *Saccharomyces cerevisiae* we notice the appearance of two function groups (CO imide & SO_2), with the disappearance of C=C after microbial action as appeared in **Figure 9**. Using XRD analysis data is to determine the difference of the mineralized rock before and after treatment. Chalcopyrite appears alone before using microbial activity and after treatment it shows low amount of chalcopyrite and quartz. The decrease of chalcopyrite amount in the analyzed sample indicated that gold was included in this minerals and was extracted by the action of *Saccharomyces cerevisiae*. The chalcopyrite has been completely dissolved with the appearance of quartz in low concentration as shown in **Figure 10**.

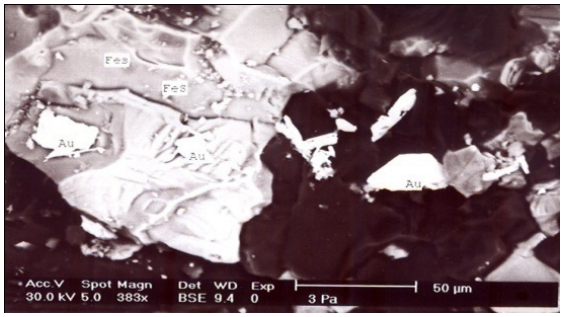

Figure 4. BSE image shows gold crystal with chalcopyrite.

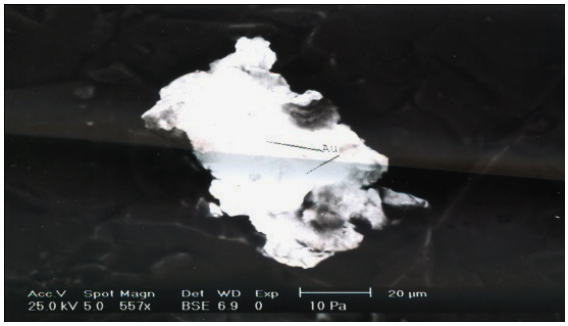

Figure 5. BSE image shows elongated gold crystal with high brightness.

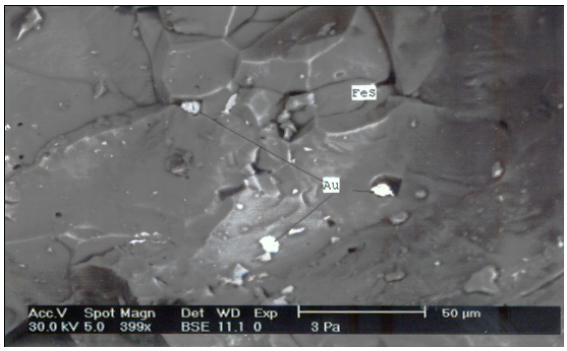

Figure 6. BSE image for gold crystal associated with chalcopyrite.

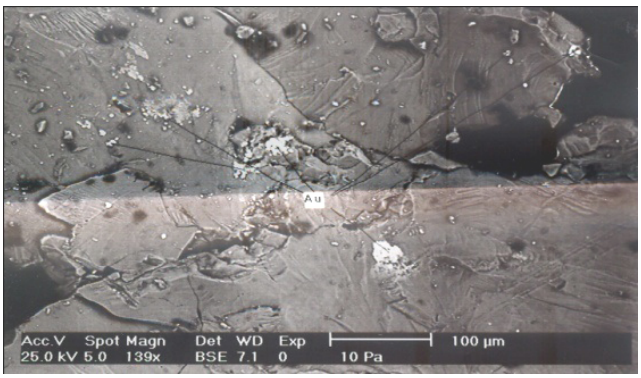

Figure 7. BSE image for gold crystal.

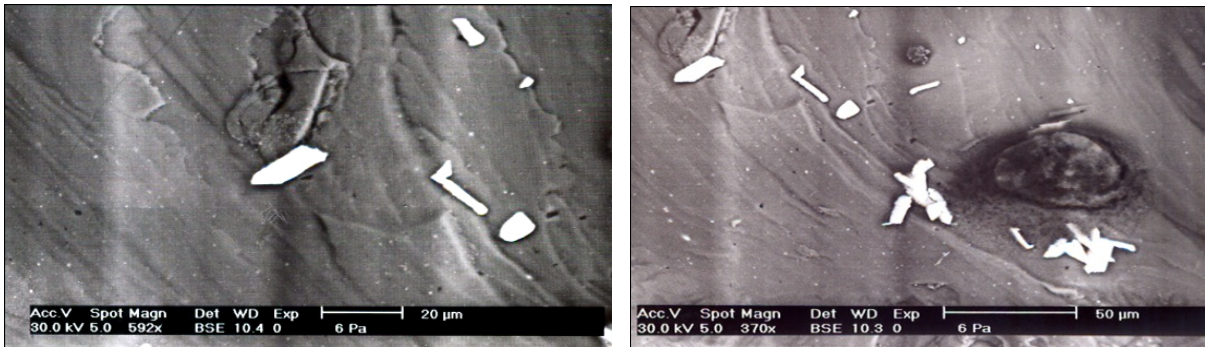

Figure 8. ESEM image shows a large agglomeration mode up of individual micro particles of gold.

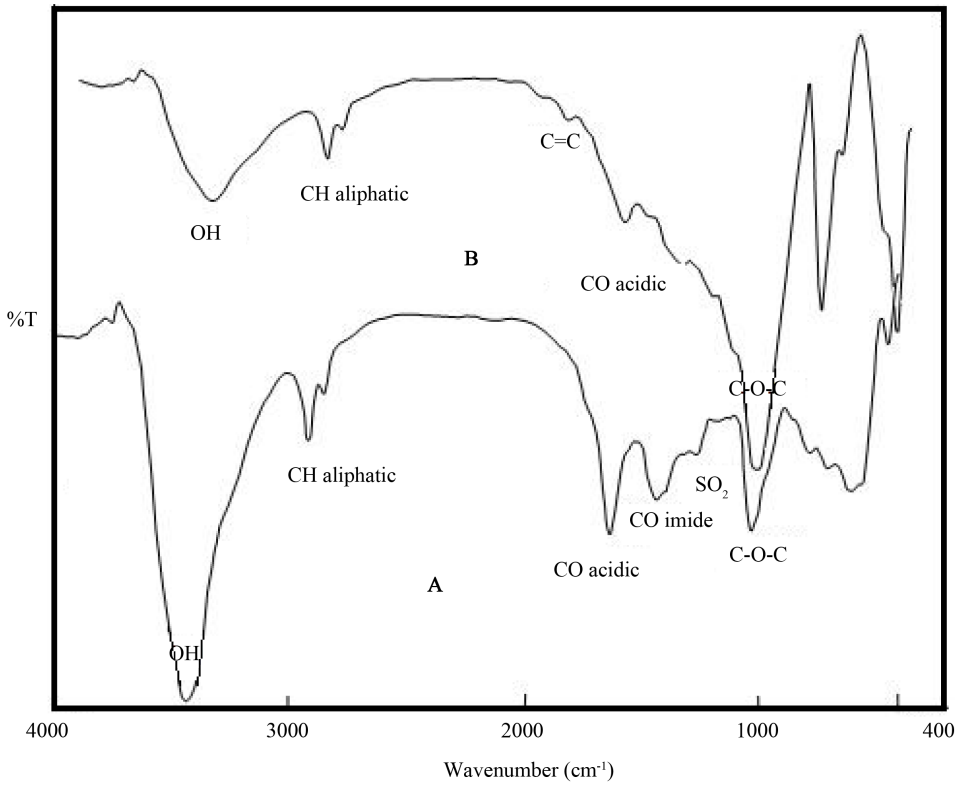

Figure 9. Infra red analysis of Mahd Ad Dahab Gold before and after treatment. A = after, B = before.

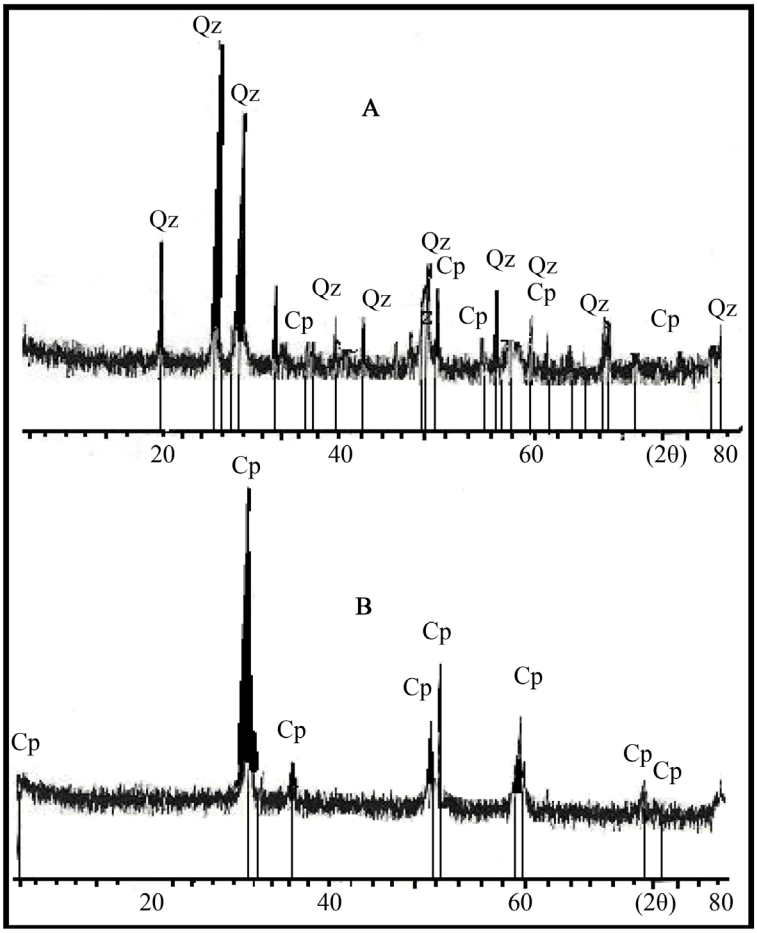

Figure 10. XRD pattern of Mahd Ad Dahab gold sample before and after treatment. A = after, B = before.

4. Conclusion

Gold is a noble metal, the term "noble metal" means that the metal will not oxidize or react with most acids. Mahd adh Dhabab is one of the largest gold mine in the Arabian world, the estimated reserve are around 1.2 MT of gold with an average grade of 24 g/t. The sample provided from the mine is taken at one of the richest part of the mine, it is noticed that the abundance of chalcopyrite in the original sample decreases after treatment in big amount. *Saccharomyces cerevisiae* action on the sample leads to recovery of gold from 0.44 ppm to 7 ppm with efficiency reaching about 1600% from the original sample.

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