

Bioaccumulation of platinum from waste

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Introduction

Platinum is a scarce metal, being one of the least abundant in the earth's crust. This research focusses on determining the optimum conditions for recovery of platinum from waste waters where it is found in its soluble, ionic form. Bacteria have been found to bioaccumulate metals using both active and passive methods referred to as biosorption and bioaccumulation respectively.

Literature review

Six unique bacterial species have been identified along with one mixed group of sulphate reducing bacteria;

Shewanella algae (Konishi, et al., 2007) formed 5nm nanoparticles (NP) in the periplasmic space within 60mins when reacted with aqueous $H_2PtCl_6^{2-}$ at 25°C and neutral pH.

Acinetobacter calcoaceticus (Gaidhani, et al., 2014) generated 2-3nm sized NPs intracellularly. Optimum conditions was a salt concentration of 1mM, temperature of 30°C, pH 7 and a 72hr incubation period.

Cyanobacterium *Plectonema boryanum* UTEX 485 (Lengke, et al., 2006) precipitated Pt(II) after up to 28 days with aqueous Pt(IV). The size ranged from 30-300nm.

Pseudomonas aeruginosa (Srivastava & Constanti, 2012) managed room temperature Pt NP synthesis without the addition of salts, electron donors etc.

Desulfovibrio desulfuricans showed equilibrium with 90% of total sorption achieved in the first 15 minutes (de Vargas, et al., 2004)

Bacillus megatherium D02 (Lin, et al., 2009) showed Pt(IV) was bio-reduced to elemental Pt(0).

Sulphate-reducing bacteria (SRB) as either a mixed culture or cell free extract or when mixed with industrial waste (Rahamuse, et al., 2008) (Rahamuse & Whiteley, 2007), (Riddin, et al., 2010) produced NPs between 200-1,000nm in size.

Method – chemical analysis

Initial preparation work involved determining growth curves for the bacterium used (not shown), absorption curves for hexachloroplatinic(IV) acid, the ideal initial platinum salt concentration and optical density of different initial cell concentrations.

A standard absorption curve for these dilutions were generated using a Jasco-V630 UV spectrophotometer. Using data from Konishi, 334nm was determined to be an appropriate wavelength (data not shown).

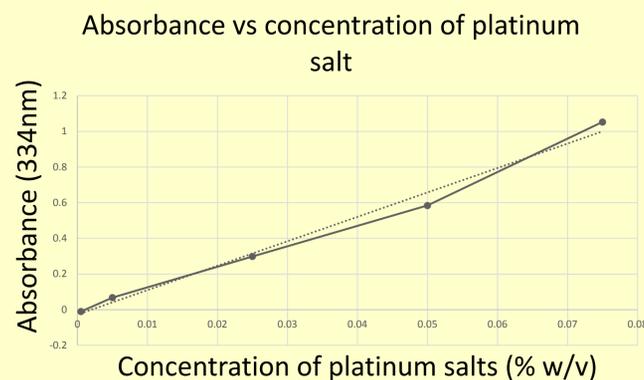


Fig 1. Absorption graph for aqueous $H_2PtCl_6^{2-}$ solution.



Fig 2. Decreasing concentrations of aqueous $H_2PtCl_6^{2-}$ solution. (Authors own, 2016)

Conclusion

From the combination of data produced, an optimum starting platinum salt concentration could be determined – 0.075% (OD 1.000 at 334nm); an optimum time after inoculation - 6 hours for *P. aeruginosa* to ensure logarithmic growth phase; and starting bacterial optical density with known cell numbers used – an OD of 0.6 at 450nm gave approximately 2×10^8 bacteria.

Method – microbial assay

Fresh normal media (50ml) was inoculated with 2.5ml *P. aeruginosa* from an overnight culture which was then incubated at 37°C for 6 hours. The optical density was read at 450nm to confirm cell numbers per ml.

A 20ml sample was centrifuged at 14,000 rpm for 10mins and cell pellet transferred to 10ml 2X fresh nutrient broth. To this was added 10ml 2X platinum salt solution – i.e. 0.15%. Overall this gave 20ml of 1X nutrient broth and platinum salt solution. This was repeated in triplicate and labelled A, B and C. The analysis was alongside a sterile control mixes of 10ml 'double strength' broth with 10ml 'double strength' platinum salt solution labelled as X and Y.

After one hour contact time at room temperature, three 2ml samples were taken from each of the five experimental tubes and centrifuged for 10mins at 14,000 rpm to remove any cells. The supernate of each sample was analysed at 334nm and the readings averaged.

Table 1 – Average absorbance at 334nm after 1 hour for broths A, B and C and controls

	Initial bacterial optical density (450nm)	Absorbance of supernate at 334nm after 1 hour and centrifuging	Standard deviation
Broth A	0.737	0.653	0.00528
Broth B	0.724	0.668	0.00698
Broth C	0.725	0.623	0.02986
Control 1		0.685	0.13674
Control 2		0.692	0.01662

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Results

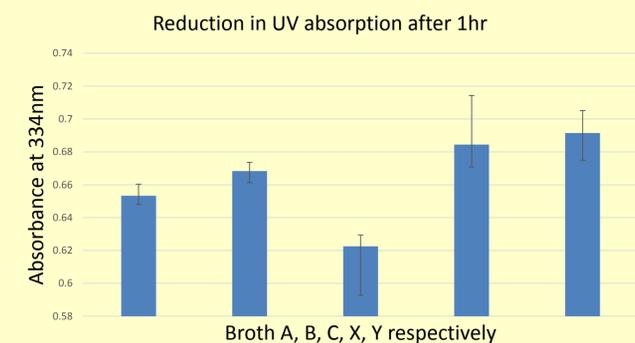


Fig 3. Graph of broth A, B, C and controls showing reduction in absorbance at 334nm

Preliminary results show a reduction in optical density of the platinum solution when with *P. aeruginosa* over control (sterile) broth. Using fig 1., the initial concentration of platinum was 0.075% and the readings in the *P. aeruginosa* are calculated to be 0.049%, suggesting bioabsorption by the bacteria in the order of about 34%.

Further experimentation is planned to confirm these results and analyse the bacteria for NP production.

References

- Konishi, Y. et al., 2007. Bioreductive deposition of platinum nanoparticles on the bacterium *Shewanella algae*. *Journal of Biotechnology*, pp. 648-653.
- Gaidhani, S. et al., 2014. Bioreduction of hexachloroplatinic acid to platinum nanoparticles employing *Acinetobacter calcoaceticus*. *Process Biochemistry*, pp. 2313-2319.
- de Vargas, I., Macaskie, L. & Guibal, E., 2004. Biosorption of palladium and platinum by sulphate-reducing bacteria. *Journal of Chemical Technology and Biotechnology*, pp. 49-56.
- Srivastava, S. & Constanti, M., 2012. Room temperature biogenic synthesis of multiple nanoparticles (Ag, Pd, Fe, Rh, Ni, Ru, Pt, Co, and Li) by *Pseudomonas aeruginosa* SM1. *J Nanopart Res*, pp. 831-841.
- Lin, Z. et al., 2009. A further insight into the biosorption mechanism of Pt(IV) by infrared spectrometry. *BMC Biotechnology*, Volume 9, p. 62.
- Lengke, M., Fleet, M. & Southam, G., 2006. Synthesis of platinum nanoparticles by reaction of filamentous cyanobacteria with platinum (IV)-chloride complex. *Langmuir*, pp. 7318-7323.
- Rahamuse, K., Mutambenengwe, C. & Whiteley, C., 2008. Enzymatic recovery of platinum(IV) from industrial wastewater using a biosulphidogenic hydrogenase. *African Journal of Biotechnology*, Volume 7, pp. 1087-1095.
- Rahamuse, K. & Whiteley, C., 2007. Bioreduction of Pt(IV) from aqueous solution using sulphate-reducing bacteria. *Appl Microbiol Biotechnol*, Volume 75, pp. 1429-1435.
- Riddin, T., Gericke, M. & Whiteley, C., 2010. Biological synthesis of platinum nanoparticles: Effect of initial metal concentration. *Enzyme and Microbial Technology*, pp. 501-505.