ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Evaluation of Fungal Growth (Penicillium sp. and Trichoderma sp.) using Cobalt Ferrite ($Co_xFe_{3-x}O_4$) Magnetic Nanoparticles.

R Vidya[1]*, B Keerthika[1], K Divya[1], K Venkatesan[2], and D Rajan Babu[2].

[1]School of Biosciences and Technology, VIT University, Vellore – 632 014, Tamil Nadu, India.
[2]Advanced Materials Research Centre, School of Advanced Sciences, VIT University, Vellore – 632 014, Tamil Nadu, India.

ABSTRACT

The cobalt ferrite ($Co_xFe_{3-x}O_4$) magnetic nanoparticles were prepared by solution combustion synthesis route. Thermodynamic modeling of the combustion reaction showed that, while varying the fuel ratios, they produce fewer amounts of gases and final adiabatic temperature when increased to fuel rich condition resulted in single phase of $CoFe_2O_4$. The fungal activity on the prepared cobalt ferrite magnetic nanoparticles was investigated against Trichoderma viride and Penicillium antarticum. The results revealed that the cobalt ferrite magnetic nanoparticles induced the growth of the fungi when the concentration of prepared nanoparticles was increased.

Keywords: Solution combustion, Metal-Oxide NPs, fungal growth

Corresponding author

ISSN: 0975-8585

INTRODUCTION

Nanomaterials are having high surface to volume ratio, which makes them more active in surface related phenomena (e.g. adsorption, reaction rates, electronic conductivity, etc.) [1]. Metal oxide nanoparticles are being recently manufactured at the industrial level and have tremendous applications in water treatment, medicine and cosmetics to name a few. These materials are present in a number of commercially available products including fillers, catalysts and many other industrial applications. Microbes are more unlikely to develop resistance against nanoparticles since they attack a broad range of targets which requires the microorganism to simultaneously undergo a series of mutations in order to protect themselves. Metal oxide nanoparticles have immense applications in numerous fields ranging from water treatment, cosmetics, medicine, toothpaste, beauty products, sunscreens, textiles and engineering [2-10].

Nanoparticles can be prepared using various methods likeMicro emulsion [11], hydrolysis [12], redox process [13], solid state reaction [14], Polyol [15], co-precipitation [16] and combustion method [17,18]. Among the different methods, solution combustion synthesis route was adopted because it produces, uniform distribution of the metal ions, good chemical stability, better crystalline nature and better size of the particles in the as prepared condition. The solution combustion synthesis route is simple, involves low cost and without further heat treatment the single phase of proposed materials could be yielded. When increasing the concentration of Fe^{3+} ions on $CoFe_2O_4$ ($Co_xFe_{3-x}O_4$) an increase in the fungal growth was observed when $CoFe_2O_4$ nanoparticles were treated with fungus. The fungal activity has been investigated in this paper [19].

EXPERIMENTAL

The nanoparticles of $CoFe_2O_4$ were prepared by self-propagated combustion route. The metal nitrates and fuel were dissolved in double distilled water to get the precursor solution. The preparation of precursor solution was taken with different concentration of oxidizers and fuels. The solution was kept into the preheated furnace for the ignition process. At the end of the combustion process the fluffy foam of the final product was obtained without further calcinations steps.

Fungal Assay

The nanoparticle activity was tested against *Penicillium sp* and *Trichoderma viride sp.* Dimethyl sulfoxide (DMSO) was used as solvents to prepare the colloidal solution of cobalt ferrite nanoparticles at different concentrations of 100, 200, 300, 400 and 500 mg/L.

The cultures were transferred to sterile Yeast extract broth, incubated for 7 days at room temperature and stored for further use. The nanoparticles were suspended in sterile water at different concentrations to make the colloidal solution. The Martin Rose Bengal Agar (MRBA) medium was prepared, poured on sterile plates and cooled down to reach a semi solid form. A well was made at the center of MRBA medium using sterile cork borer. The spore suspension of 20 µl was added to the well. The plates containing the nanoparticles and MRBA medium were incubated at room temperature for 7 days. The fungal growth was noted and the mean diameter of the colony was calculated every day. The results is shown in figure. 1 .

RESULTS AND DISCUSSION

The prepared nanomaterial when added to the media provided a rich source of iron to the fungi and helped in its luxuriant growth. A fungal organism needs iron for its metabolism, growth and secondary metabolite production. Fungi have a remarkable capacity to take up iron which is present in a wide variety of forms, which include free iron ions, low – affinity iron chelates, siderophores, etc. Pathogenic fungi [20] have evolved a strategy for obtaining iron from transferrin and hemoglobin. Most fungi synthesize and secrete siderophores which are small organic compounds that binds ferric ion which has a tremendous affinity and specificity [23]. *Sacromyces cerevisiae* retains a significant quality of iron binding molecules called siderophores in the cell wall. The cell wall mannoproteins contribute to the retention of siderophores to chelate iron in the cell wall. The uptake of iron is enhanced when bound to certain siderophores [21, 22]. Several fungal species which includes *Candida albicans, Aspergillus nidulans, Aspergillus fumigatus, Aspergillus oryzae, Ustilago mayadis* and *Schizosaccharomyces pombe* synthesize or secrete siderophores to cleave iron from the environment [24-26]. According to the earlier reports iron is an important element for the fungal

metabolism. Fungi need iron for the growth and metabolism. In this study, the fungal species namely *Penicillium and Trichoderma* utilizes Fe^{3+} source from nanoparticles for its growth and iron requirement [27]. Due to the presence of Fe^{3+} ions. *Penicilliumsp* showed luxuriant growth compared to the *Trichoderma sp*.

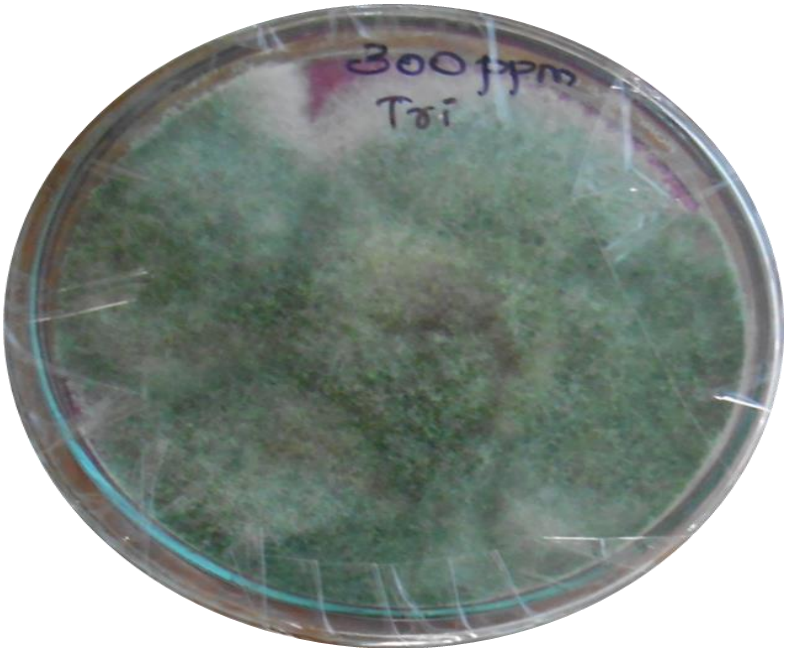

Figure 1: Effect of cobalt ferrite magnetic nanoparticles on fungal growth

The fungal colony growth measured after 72 hours showed that the nanoparticles were capable of attaching to the membrane of the microbe which helped in the fungal growth at a faster rate. When increasing the concentration of the nanoparticles the growth level also increased. So, the prepared nanomaterial acts as a nutrient for the fungal activity and also for its luxuriant growth. The secondary metabolite Penicillin is produced from *Penicillium sp.* and acts as an effective anti-biotic and *Trichoderma sp* produces trichodermin as a secondary metabolite which is a mycotoxin and it inhibits a number of pathogenic fungi. Trichodermin has the following applications. It is used as a

- Disease control agent
- Plant growth promoter
- Biochemical elicitor of disease
- Bioremediation agent

It produces antibiotics and toxins such as trichothecin and sesquiterpine which causes the direct effect on other organisms. Since the nanomaterial favours the growth of fungi in an enhanced way, the fungal secondary metabolites production (Penicillin and Trichodermin) could be targeted in futuristic studies using nanomaterials.

CONCLUSION

$CoFe_2O_4$ magnetic nanoparticles were successfully prepared by solution combustion synthesis route. It can be concluded that the cobalt ferrite magnetic nanoparticles material acted as nutrients so it was used to support the growth of fungi. Future studies needs to be targeted to focus on secondary metabolite production of penicillin and trichodermin. Since the nanoparticles favour the growth of metabolite producing fungi and the secondary metabolites also have a large number of industrial applications, studies should be focused on the growth of fungi to grow in the presence of magnetic nanoparticles and also to target the secondary metabolite production which can be for industrial application. *In vitro* and *in vivo* studies should be conducted on the toxicity of nanoparticles towards living organisms and the applicability of the fungi towards secondary metabolite production should be also taken in consideration in future research activity.

ACKNOWLEDGEMENTS

We acknowledge the management of VIT University, Vellore, Tamil Nadu, India, for their great support and encouragement. The authors K. Divya, B. Keerthika and K.Venkatesan extend their sincere thanks to the VIT management for providing the Research Associateship during the course of study.

REFERENCES

[1] Giovanni De Filpo et al. Int Biodeterior Biodegrad 2013; 85:217 – 222.
[2] Amitava Mukherjee et al, Science against microbial pathogens: communicating current research and technological advances, A.Méndez-Vilas (Ed.), 245 – 251.
[3] Pal S, Tak YK, Song JM. App Environ Microbiol 2007; 73:1712–1720.
[4] Gupta AK, Gupta M. Biomater 2005; 26:3995–4021.
[5] Applerot G, Lipovsky A, Dror R, Perkas N, Nitzan Y, Lubart R and Gedanken A. Adv Funct Mater 2009; 19:842–52.
[6] Nair S, Sasidharan A, Divya Rani V V, Menon D, Nair S, Manzoor K and Raina S. J Mater Sci Mater Med 2008; 20:S235–41.
[7] Hanley C, Layne J, Punnoose A, Reddy K M, Coombs I, Coombs A, Feris K and Wingett D, Nanotechnol 2008; 19:295103.
[8] Le Lay C, Akerey B, Fliss I, Subirade M and Rouabhia M. J Appl Microbiol 2008; 105:1630–9.
[9] Thevenot P, Cho J, Wavhal D, Timmons R B and Tang L. Nanomed 2008; 4:226–36.
[10] Anagnostakos K, Hitzler P, Pape D, Kohn D and Kelm J. Acta Orthop 2008; 79:302–7.
[11] Choi E. J, Ahn Y, Kim S, An DH, Kang KU, Lee BG. J Magn Magn Mater 2003;262:L198 – 202.
[12] Duong GV, Hanh N, Linh DV, Groessinger R, Weinberger P, Schafler E, Zehetbauer M. J Magn Magn Mater 2007;311:46–50.
[13] Rajendran M, Pullar RC, Bhattacharya AK, Das D, Chintalapudi SN, Majumdar CK. J Magn Magn Mater 2001;232:71 – 83.
[14] Rafferty A, Prescott T, Brabazon D. Ceram Ind 2008;34:15–21.
[15] Baldi G, Bonacchi D, Innocenti C, Lorenzi G, Sangregorio C. J Magn Magn Mater 2007;311:10 – 16.
[16] Millot N, Gallet SL, Aymes D, Bernard F, Grin Y. J Eur Ceram Soc 2007;27:921–926.
[17] Tamara Slatineanu, Eliano Diana et.al. Cent Eur J Chem 2012;10(6):1799 – 1807.
[18] Salunkhe AB, Khot VM, Phadatare MR, Pawar SH. J Alloys Compd 2012;514:91-96.
[19] Nasrollahi A, Pourshamsian Kh, Mansourkiaee, Int J Nano Dim winter 2011; 1(3): 233-239.
[20] Aisen P, Leibman A, Zweier J. J Biol Chem 1978;253:1930–1937.
[21] Lesuisse E, Raguzzi F, Crichton R. R. J Gen Microbiol 1987; 133:3229–3236.
[22] Protchenko O, Ferea T, Rashford J, Tiedeman J, Brown PO, Botstein D, Philpott CC. J Biol Chem 2001; 276; 49244–49250.
[23] Neilands JB. J Biol Chem 1995; 270:26723–26726.
[24] Mei B, Budde AD, Leong SA. Proc Natl Acad Sci USA 1993; 90:903–907.
[25] Schrettl M, Bignell E, Kragl C, Joechl C, Rogers T, Arst Jr. H. N, Haynes K, Haas H. J Exp Med 2004; 200: 1213–1219.
[26] Yamada O, Na Nan S, Akao T, Tominaga M, Watanabe H, Satoh T, Enei H, Akita O. J Biosci Bioeng 2003; 95: 82–88.
[27] Eisendle M, Oberegger H, Zadra I, Haas H. Mol Microbiol 2003; 49:359–375.