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Magnetic resonance investigation of magnetic-labeled baker's yeast cells

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Abstract

In this study, the interaction of DMSA-coated magnetite nanoparticles (5 and 10 nm core-size) with *Saccharomyces cerevisiae* was investigated using magnetic resonance (MR) and transmission electron microscopy (TEM). The TEM micrographs revealed magnetite nanoparticles attached externally to the cell wall. The MR data support the strong interaction among the nanoparticles supported by the cells. A remarkable shift in the resonance field was used as signature of particle attachment to the cell wall.

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75.50.Mm; 76.50+g; 87.68.+z

Keywords: Magnetic fluid; Magnetic resonance; Transmission electron microscopy; Yeast cell

The use of nanosized magnetic materials in the development of field-gradient magnetic separation (FGMS) technologies has attracted much attention in recent years. Wastewater containing processed textile dyes, for instance, is an increasing source of environmental contamination. Recent experiments showed that the yeast cells (*Saccharomyces cerevisiae*) efficiently interact with magnetic nanoparticles dispersed as magnetic fluids (MFs), leading to the formation of magnetic-labeled cells which could be easily separated from the system using appropriated magnetic separators [1]. Also, yeast cells are good candidates to promote removal and degradation of dyes from wastewater [2]. In this study, transmission electron microscopy (TEM) and magnetic resonance (MR) were used to investigate the interaction of *S. cerevisiae* cells with magnetic nanoparticles (MNPs) after incubation with magnetite-based, DMSA-coated MF samples (DMSA-MF).

Baker's yeast (*S. cerevisiae*) was obtained locally. Magnetite-based (5 and 10 nm average diameter), DMSA-coated biocompatible MF samples were prepared as described in the literature [3]. Compressed baker's yeast (2 g) was suspended in saline (6 mL), centrifuged

and resuspended in 6 mL 0.1 M PBS (pH 7.4). After centrifugation the pellet was once again resuspended in PBS to obtain a 33% yeast suspension (v/v; yeast cells volume determined after sedimentation for 24 h at 1 g). Three mL of the yeast suspension was added to 1 mL of the DMSA-MF sample containing 1.2×10^{16} particle/cm³. Two distinct DMSA-MF samples (5 and 10 nm average diameter MNP's) were used in the experiments. The suspension was mixed and then incubated at room temperature for 1 h without mixing. After this time period most of the yeast cells were magnetically modified by the added DMSA-MF (the cells responded to external magnetic field gradients). Nonmagnetic yeast cells and residual MF were removed by repeated static magnetic separation using PBS and saline as washing liquids. After treatment cells were harvested, fixed, and processed for TEM analysis. Modified cells were also used for MR measurements. The TEM analysis (see Fig. 1) revealed that the 5 nm-size, DMSA-coated magnetite nanoparticles, was externally attached to the *S. cerevisiae* cell wall. Similar result was observed using the 10 nm-size, magnetite-based DMSA-MF sample. Incubation for 4 h instead of 1 h leads to similar observations.

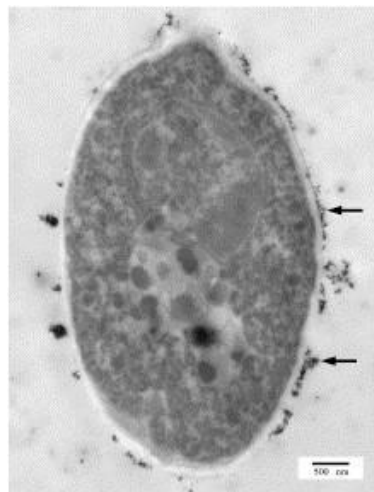


Fig. 1. TEM picture of *S. cerevisiae* incubated with 5 nm DMSA-MF. Observe MNP's attached to the cell wall (arrows). No particles were found inside the cell.

MR experiments have been successfully used to investigate biodistribution of MNPs introduced in living beings [4]. Fig. 2 shows the MR spectra of the two modified cells (open circles for the 5 nm and full circles for the 10 nm MNPs) plus the spectra of two 10 nm-size (dashed and solid lines), magnetite-based DMSA-MF samples containing 4.8×10^{16} (1:1) and 4.8×10^{13} particle/cm³ (1:1000). The huge dilution (from 1:1 to 1:1000) of the 10 nm-size MF sample causes a relative shift in the resonance field of about 73 G. However, the shift observed in the resonance field between the concentrated 10 nm-size MF sample (1:1) and the modified cells using the 10 nm-size, magnetite-based DMSA-MF sample, was about 250 G. This remarkable resonance field shift is the signature of a strong interaction among the MNP's

when attached to the *S. cerevisiae* cell wall. The MR spectrum of the modified cells using the 5 nm-core, magnetite-based DMSA-MF sample, is also shown in Fig. 2 (open circles).

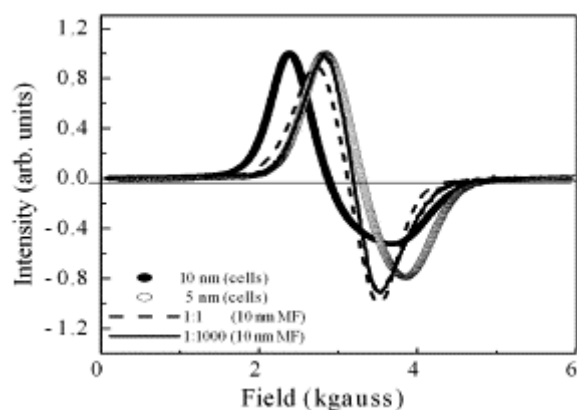


Fig. 2. Magnetic resonance spectra of the 5 (open circles) and 10 nm (full circles) nanoparticles in labeled cells. Solid and dashed lines represent the spectra of the 10 nm magnetite suspended as a stable magnetic fluid samples at different concentrations (see text).

In conclusion, 5 and 10 nm-sized magnetite particles, DMSA-coated, do attach to the *S. cerevisiae* cell wall after incubation of the yeast cells with the corresponding biocompatible magnetic fluid samples. Magnetic resonance data confirm that both nanoparticles are firmly attached to the cells as revealed by the TEM micrographs.

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References

- [1] I. Safarik, L. Ptackova, M. Safarikova, *Eur. Cells Mater.* 3 (S2) (2002) 52.
- [2] M.A.M. Martins, M.H. Cardoso, M.J. Queiroz, M.T. Ramalho, A.M.O. Campos, *Chemosphere* 38 (1999) 2455.
- [3] T. Goetze, C. Gansau, N. Buske, M. Roeder, P. Gornert, .M. Bahr, *J. Magn. Magn. Mater.* 252 (2002) 399.
- [4] L.M. Lacava, Z.G.M. Lacava, M.F. Da Silva, O. Silva, S.B. Chaves, R.B. Azevedo, F. Pelegrini, C. Gansau, N. Buske, D. Sabolovic, P.C. Morais, *Biophys. J.* 80 (2001) 2483.