Magnetically Responsive (Nano) Biocomposites

Ivo Šafařík, Kristýna Pospíšková, Kateřina Horská, Zdeňka Maděrová and Mirka Šafaříková

Abstract Many biological materials have been successfully used in various areas of bioscience, biotechnology and environmental technology applications. Biological materials are mainly diamagnetic, which means they do not interact significantly with external magnetic field. Using various postmagnetization procedures, biological materials can be converted into magnetically responsive composite materials which can be efficiently separated using simple magnets or magnetic separation systems. The prepared magnetic biocomposites can be used for many applications, such as immobilization of target compounds, as parts of biosensors, as whole cell biocatalysts or for magnetic removal and separation of xenobiotics and biologically active compounds.

Keywords Biological materials · Postmagnetization · Magnetic composite materials · Magnetic separations

Abbreviations

DBT	Dibenzothiophene
EPEC	Enteropathogenic *Escherichia coli*
PS	Phosphatidylserine
MRI	Magnetic resonance imaging
MSCs	Mesenchymal stem cells
VTEC	Verocytotoxigenic *Escherichia coli*

I. Šafařík (✉) · K. Horská · Z. Maděrová · M. Šafaříková
Department of Nanobiotechnology, Institute of Nanobiology and Structural Biology of GCRC, Na Sádkách 7, 370 05 České Budějovice, Czech Republic
e-mail: ivosaf@yahoo.com
URL: http://www.nh.cas.cz/people/safarik/

I. Šafařík · K. Pospíšková
Regional Centre of Advanced Technologies and Materials, Palacký University, Šlechtitelů 11, 783 71 Olomouc, Czech Republic

1 Introduction

Magnetically responsive nano- and microparticles and related structures have found many important applications in various areas of biosciences, medicine, biotechnology and environmental technology (Safarik and Safarikova 2012; Safarik et al. 2012d). Different types of responses of such materials to external magnetic field enable their various applications, namely their selective separation, targeting and localization using an external magnetic field (e.g. using an appropriate magnetic separator, permanent magnet, or electromagnet), heat generation (which is caused by magnetic particles subjected to high frequency alternating magnetic field), increase of a negative T2 contrast by magnetic iron oxides nanoparticles during magnetic resonance imaging or great increase of apparent viscosity of magnetorheological fluids when subjected to a magnetic field. In addition, magnetite nanoparticles can exhibit peroxidase-like activity (Gao et al. 2007; Safarik et al. 2012d; Safarik and Safarikova 2012). Currently large amount of various magnetic nano- and micromaterials can be obtained commercially. Alternatively such materials are produced in research laboratories, using many different basic principles (Laurent et al. 2008; Safarik et al. 2011a; Li et al. 2013). Many research groups are involved in the fine tuning of procedures leading to the production of homogeneous magnetic particles with defined size, structure, composition, coating etc. (Safarik et al. 2012d).

Different types of non-magnetic particulate materials including adsorbents, catalysts, chromatography materials, carriers, microbial cells, waste biological materials etc. are available. In many cases the application potential of these materials could be improved by their modification leading to the formation of magnetically responsive materials. Such a modification can substantially simplify separation of magnetic materials from complex systems. So, in addition to the main currently used strategies focused on specific preparation of target magnetic structures, alternative strategies leading to the formation of interesting magnetically responsive materials have been developed. These procedures are based on postmagnetization of already existing diamagnetic ("non-magnetic") or paramagnetic ("weakly" magnetic) particulate materials found in nature or prepared in the laboratory or industry (Safarik et al. 2012d). The term "postmagnetization" and the whole process of magnetic modification of non-magnetic materials was invented by Mosbach (Mosbach and Andersson 1977; Griffin et al. 1981) at the end of 70s and beginning of 80s of the 20th century, when gel particles for column affinity chromatography were magnetically modified using appropriate ferrofluid (magnetic fluid); such a modification led to the preparation of magnetic derivatives with unaltered biospecificity when applied to general ligand affinity chromatography studies (Safarik et al. 2012d).

Postmagnetization usually leads to the formation of strongly magnetic composite materials, where the "original" diamagnetic or paramagnetic structure is responsible for biological, catalytic, carrier or adsorption function of the formed composite, while magnetic label (most often in the form of magnetic iron oxides

nano- and microparticles, which are usually deposited on the surface or within the pores of treated materials) is responsible for the strong magnetic behavior of the formed composite materials (Safarik and Safarikova 2009; Safarik et al. 2012d). Alternatively non-magnetic materials can be modified by erbium ions which preserve their exceptionally high atomic magnetic dipole moment in various chemical structures (Zborowski et al. 1993) or by co-entrapment of nonmagnetic and magnetic particles in a gel material (Al-Dujaili et al. 1979; Safarik et al. 2012a). Postmagnetization can be applied to a broad variety of inorganic materials [e.g., clays (Bartonkova et al. 2007; Mockovciakova et al. 2010; Safarik et al. 2012b)], activated charcoal (Safarik et al. 1997, 2012a; Nakahira et al. 2007; Schwickardi et al. 2006), synthetic polymer particles (Cumbal and SenGupta 2005), biopolymer particles (Griffin et al. 1981; Mosbach and Andersson 1977; Torchilin et al. 1985), waste plant materials (Safarik et al. 2007a, 2011b, 2012c; Safarik and Safarikova 2010; Tian et al. 2011; Pospiskova and Safarik 2013), microbial cell walls (Patzak et al. 1997), whole microbial and algae cells (Safarik et al. 2007b; Pospiskova et al. 2013; Safarikova et al. 2008, 2009; Prochazkova et al. 2013) and many others.

Postmagnetization procedures can substantially increase the amount of useful magnetically responsive materials and subsequently the amount of their important applications in various areas of biosciences, biotechnology, food technology (bio)analytical chemistry, environmental chemistry and technology, removal of radionuclides, etc. (Safarik et al. 2012d).

As already mentioned, many different types of "non-magnetic" materials can be converted into the magnetic form. Table 1 shows examples of described postmagnetization procedures for the modification of inorganic materials, polymers and carbon materials. In this chapter, the attention will be mainly paid to postmagnetization of materials taken from living nature, namely plant derived materials and both prokaryotic and eukaryotic cells. Magnetic derivatives of these materials have been already successfully used e.g. as adsorbents for the removal of both organic and inorganic xenobiotics, carriers for the immobilization of biologically active compounds, whole cell biocatalysts and biosensor elements. However, the potential of magnetically responsive biocomposites is substantially greater.

2 Postmagnetization of Biomaterials

The ideal postmagnetization procedure should be cheap, easy-to-perform, scalable and tunable, leading to a stable magnetic product, both in dry state and water suspension. Preparation of magnetic derivatives of originally non-magnetic biomaterials can follow selected procedures used already for the postmagnetization of non-biological materials, as exemplified in Table 1. However, only selected procedures can be used for postmagnetization of biomaterials due to their specific characters. One of the general approaches is based on suspending the treated

Table 1 Examples of described postmagnetization procedures for the modification of inorganic materials, polymers and carbon materials

Non-magnetic material	Way of postmagnetization	Reference
Activated carbon	Impregnation of activated carbon with $Fe(NO_3)_3$ solution followed by drying at 90 °C and heating to a temperature of 700 °C under argon and benzene vapor	Schwickardi et al. (2006)
Bentonite	A suspension of bentonite in water was mixed with $FeSO_4$, then KNO_3 and KOH were added, then mixture was heated up to 90 °C	Bartonkova et al. (2007)
Bentonite	Precipitation of iron oxides from $FeSO_4$ and $FeCl_3$ by NH_4OH in the presence of bentonite in nitrogen atmosphere, followed by drying at 70 °C	Mockovciakova et al. (2010)
Carbon nanotubes	Filling of carbon nanotubes with ferrofluid followed by drying to leave deposited magnetic nanoparticles	Korneva et al. (2005)
Carbon nanotubes	Mixing of $(NH_4)_2Fe(SO_4)_2$ with hydrazine followed by the addition of carbon nanotubes, pH increase and heating	Li et al. (2010)
Charcoal	Co-entrapment of charcoal and magnetite particles in polyacrylamide gel	Al-Dujaili et al. (1979)
Charcoal	Precipitation of magnetite from $FeSO_4$ and $Fe_2(SO_4)_3$ by NaOH in the presence of charcoal, followed by aging for 24 h and heating at 473 K	Nakahira et al. (2007)
Charcoal	Precipitation of iron oxides from $FeSO_4$ and $FeCl_3$ by NaOH in the presence of charcoal, followed by drying at 100 °C for 3 h	Oliveira et al. (2002)
Charcoal	Precipitation of hydrated iron oxides from $FeSO_4$ by NaOH in the presence of charcoal, followed by heating to 100 °C for 1 h	Safarik et al. (1997)
Chromatography gels	Circulation of water based ferrofluid through a bed of chromatography gel	Mosbach and Andersson (1977), Griffin et al. (1981)
Montmorillonite	One g of powder was thoroughly mixed in a small beaker with 1 mL of water based ferrofluid stabilized with perchloric acid. This mixture was allowed to dry completely at temperatures not exceeding 50 °C	Safarik et al. (2012b)

materials in the solution of iron ions and after increase of pH and heating magnetic iron oxides particles are formed, thus modifying the treated material (Safarik et al. 1997). More gentle modification procedures are usually based on the deposition of pre-prepared magnetic iron oxides nano- or microparticles on the surface or within the pores of the treated biomaterials (Safarik et al. 2007a; Safarik and Safarikova 2014). In addition, several other alternative procedures can also be employed, such as covalent binding of magnetic particles to the treated materials (or vice versa,

depending on the size), cross-linking of cells, isolated cell walls or other biomaterials with a bifunctional reagent in the presence of magnetic particles, by the biologically driven precipitation of paramagnetic compounds on the cell surface or by biospecific binding of immunomagnetic particles to the target antibody containing materials (especially cells). Also entrapment of the modified materials into synthetic polymer, biopolymer or inorganic gels containing magnetic particles can be employed, as well as labeling with erbium cation (Safarik and Safarikova 2007; Safarik et al. 2011c, 2012d).

Recently, new and efficient postmagnetization procedures have been developed. One of them employed water based magnetic fluid stabilized with perchloric acid, which was directly mixed with material to be modified and dried completely. This procedure is extremely simple and various biological, inorganic and polymer materials have been successfully transferred into their magnetic derivatives (Safarik et al. 2012b).

Another postmagnetization process is based on microwave irradiation of suspensions containing the material to be modified and iron hydroxides (formed from ferrous sulfate after increasing the pH to 10–12 by the addition of sodium hydroxide); during the microwave treatment magnetic iron oxides nano- and microparticles are formed and subsequently bound on the surface of the treated material. Using this procedure a large amount of materials has been postmagnetized (Safarik et al. 2013).

A very efficient modification of the microwave assisted postmagnetization procedure enables to modify also heat- and pH- sensitive nonmagnetic materials. At first magnetic iron oxides nano- and microparticles have been synthesized from ferrous sulfate at high pH in a microwave oven. After their washing with water the suspension of magnetic particles was mixed thoroughly with nonmagnetic material to be modified. After complete drying stable magnetically responsive materials have been formed. This procedure is extremely inexpensive, very simple, scalable and tunable. Also rather sensitive biological materials including starch grains and insoluble proteins particles have been successfully magnetized (Safarik and Safarikova 2014).

2.1 Magnetic Modification of Non-living Biomaterials

A very simple procedure leading to the formation of postmagnetized materials has been developed, using water based magnetic fluids (ferrofluids) as the modifying agent. The most frequently used magnetic fluid (stabilized with perchloric acid) can be prepared using the "classical" procedure developed by Massart (Massart 1981). This ferrofluid was usually composed of maghemite nanoparticles with the diameter bellow 15 nm (Mosiniewicz-Szablewska et al. 2007, 2010) (Fig. 1). The material to be modified was suspended in methanol and appropriate amount of ferrofluid was then added. Magnetic iron oxide nanoparticles were deposited on the surface of the treated materials after short mixing, both in the form of

Fig. 1 TEM image of nanoparticles present in the ferrofluid stabilized with perchloric acid used for postmagnetization of biological materials (bar = 200 nm). Reproduced, with permission from (Safarik et al. 2012d)
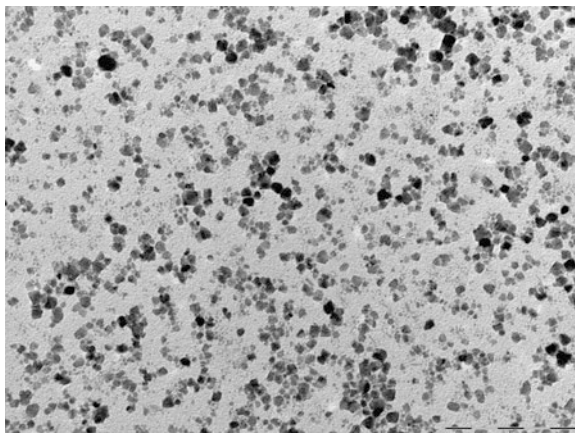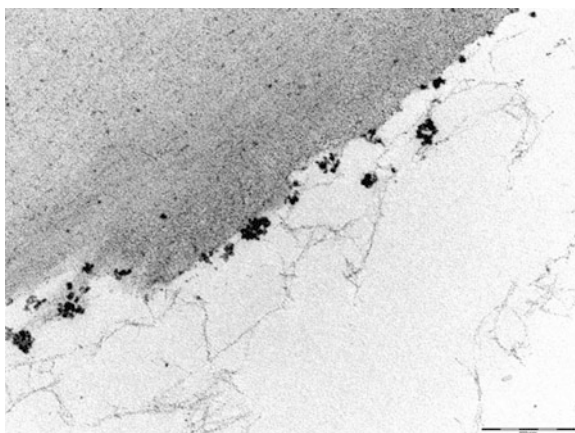

Fig. 2 Ultrathin section of magnetic sawdust particle observed in transmission electron microscopy (bar = 200 nm). Reproduced, with permission, from (Safarik et al. 2007a)

individual nanoparticles, or their aggregates (Mosiniewicz-Szablewska et al. 2007, 2010) (Fig. 2). Methanol-based procedure is especially useful for the treatment of non-living or dead biological materials, like different types of lignocellulose materials [e.g., sawdust (Safarik et al. 2007a), spent barley grains (Safarik et al. 2011b; Pospiskova and Safarik 2013), peanut husks (Safarik and Safarikova 2010) or spent coffee grounds (Safarik et al. 2012c)]. However, other materials including e.g. non-living microbial biomass or clays can be modified in this way, too. Alternatively, acetate buffer was used as a suspending medium for magnetic modification of both dead yeast cells (Safarik et al. 2007b; Safarikova et al. 2005) (Fig. 3) and algae cells (Safarikova et al. 2008). The amount of magnetic iron oxides nanoparticles deposited on the treated material surface can be influenced by the amount of ferrofluid used. Detailed magnetic characteristics of ferrofluid modified materials can be found elsewhere (Mosiniewicz-Szablewska et al. 2007, 2010). Direct magnetic modification with magnetic fluid (without the use of

Fig. 3 TEM picture of magnetically modified dried fodder yeast (*Kluyveromyces fragilis*) cells (bar = 200 nm). Reproduced, with permission, from (Safarik et al. 2007b)
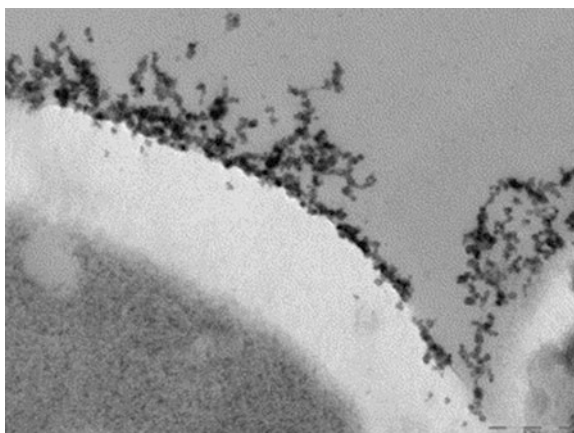
methanol or a buffer) was used for magnetization of sawdust, spent tea leaves, spent coffee grounds, spent barley grains, cellulose and starch (Safarik et al. 2012b).

As already described, microwave assisted modification was used for magnetization of large amount of biomaterials, such as microcrystalline cellulose, sawdust, spent tea leaves, spent coffee grounds, spent barley grains, powdered peanut husk or dried marine algae, in addition to other inorganic and synthetic materials (Safarik et al. 2013). The newest postmagnetization procedure employing microwave synthesized magnetic iron oxides nano- and microparticles can be used for magnetization of broad variety of materials, including sensitive biological materials (Safarik and Safarikova 2014).

2.2 Magnetic Modification of Living Biomaterials

Water-based ferrofluids have been successfully used to modify living microbial cells in order to prepare magnetically responsive whole cell biocatalysts (Safarikova et al. 2009), as shown in Fig. 4. Appropriate buffers had to be used to maintain the viability of the treated cells. Stable magnetically responsive yeast cell aggregates (Fig. 5) were formed when *S. cerevisiae* cells were mixed with magnetic iron oxides nano- and microparticles prepared by microwave assisted synthesis from ferrous sulfate (Pospiskova et al. 2013). Alternative approaches were based on the polyelectrolyte mediated deposition of magnetic iron oxides nanoparticles on the cell surface. *S. cerevisiae* yeast cells were coated with polyelectrolytes using alternating deposition of poly(allylamine hydrochloride) and poly(sodium polystyrene sulfonate) layers on the cells surface followed by deposition of non-coated magnetic nanoparticles (Fakhrullin et al. 2010a). In other experiments magnetic iron oxides nanoparticles stabilized with poly(allylamine
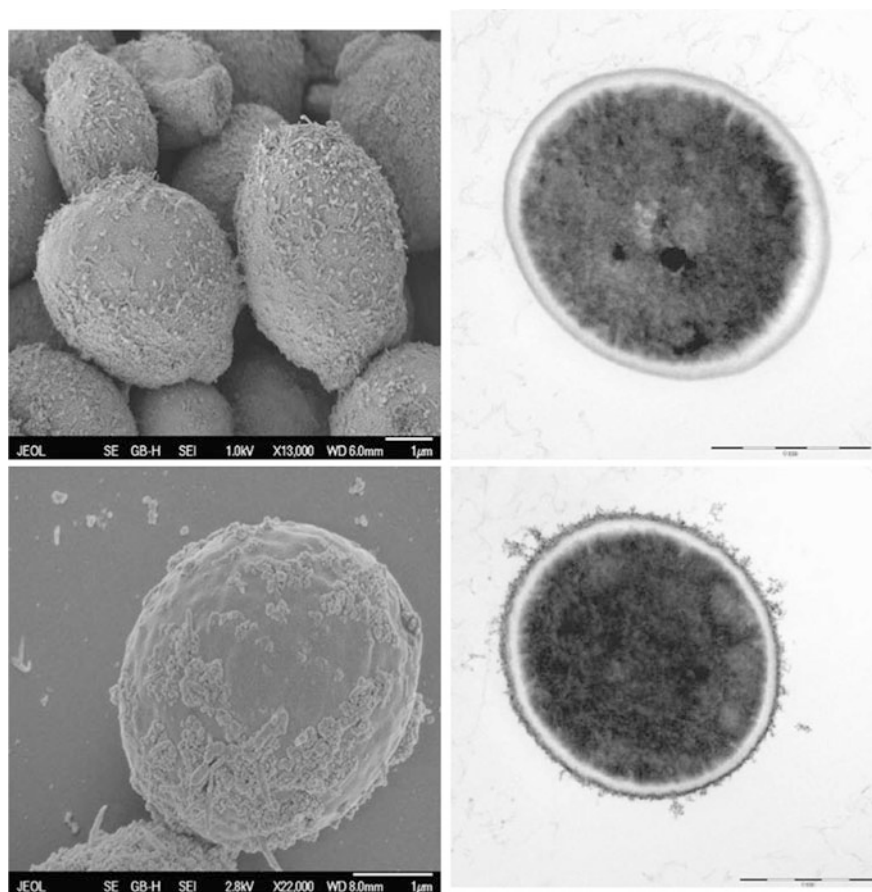
Fig. 4 *Left* SEM micrographs of ferrofluid modified *Saccharomyces cerevisiae* cells showing attached magnetic nanoparticles and their aggregates on the cell surface (bars = 1 μm). *Right* TEM micrographs of *Saccharomyces cerevisiae* cells (bars = 1 μm). *Top* Native cell. *Bottom* Ferrofluid modified cell with attached magnetic iron oxide nanoparticles on the cell wall. Reproduced, with permission, from (Safarikova et al. 2009)

hydrochloride) were used to modify *Chlorella pyrenoidosa* cells (Fakhrullin et al. 2010b) and living human cells (Dzamukova et al. 2011). The same magnetic nanoparticles were even used for magnetic modification of multicellular organisms, namely soil nematode *Caenorhabditis elegans* (Minullina et al. 2011).

Specific target cells are usually magnetically modified using immunomagnetic nano- or microparticles (Safarik and Safarikova 1999, 2012) (Fig. 6). This approach enables to detect and magnetically label cells bearing target epitopes on their surface and subsequently separate the labeled cells using flow-through or batch magnetic separation. Many immunomagnetic particles are commercially available, such as those used for the detection of microbial pathogenic bacteria
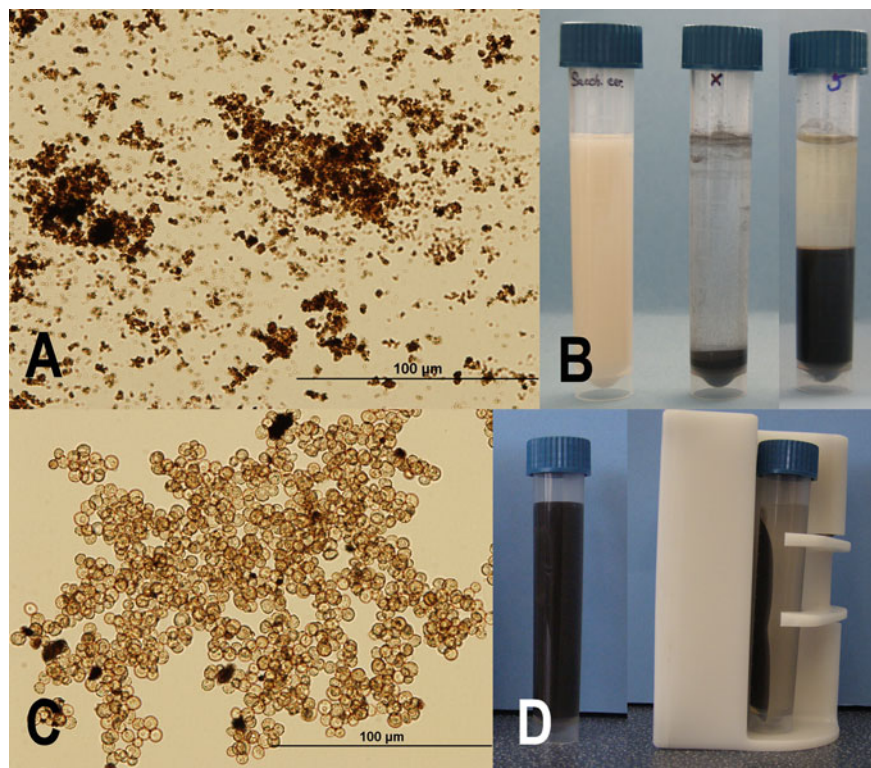
Fig. 5 Optical microscopy of magnetic iron oxides microparticles prepared by microwave assisted synthesis (**a**); process of magnetic modification of yeast cells (*left tube*—*S. cerevisiae* cells suspension; *middle tube*—sedimented iron oxides microparticles for magnetic modification; *right tube*—sedimented magnetically modified yeast cells) (**b**); optical microscopy of *S. cerevisiae* cells modified by iron oxides microparticles (**c**); magnetic separation of magnetically modified yeast cells (**d**). Reproduced, with permission, from (Pospiskova et al. 2013)

from Invitrogen, namely Dynabeads anti-*Salmonella*, Dynabeads anti-*Escherichia coli* O157, Dynabeads EPEC/VTEC O26, Dynabeads EPEC/VTEC O103, Dynabeads EPEC/VTEC O111, Dynabeads EPEC/VTEC O145, Dynabeads anti-*Legionella* and Dynabeads anti-*Listeria*. The same company also produces immunomagnetic particles for the separation of parasitic protozoa, e.g. *Cryptosporidium* oocysts (Dynabeads anti-*Cryptosporidium*) and simultaneous separation of *Cryptosporidium* oocysts and *Giardia* cysts (Dynabeads GC-Combo).

Different types of lectins, such as those produced from *Triticum vulgaris* and *Agaricus bisporus*, or concanavalin A, were immobilized on magnetic microspheres and used to magnetically label specific bacterial pathogens, such as *E. coli*. Recovered cell populations were free from environmental impurities and a high percentage of the culturable cells was extracted (Payne et al. 1993; Porter and Pickup 1998; Porter et al. 1998).
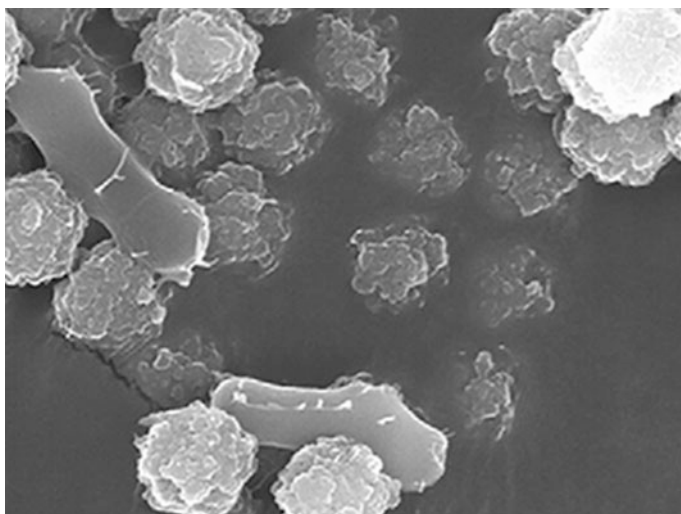
Fig. 6 Electron microscopy of *Legionella pneumophila* bound to immunomagnetic beads (Dynabeads My One Streptavidin (Invitrogen) with bound biotinylated polyclonal anti-*Legionella* antibody). Reproduced, with permission, from (Reidt et al. 2011)

Magnetic (nano)particles with immobilized annexin V have been employed for simple and efficient magnetic modification and subsequent separation of apoptotic cells from normal culture. This procedure is based on the fact that annexin V is a Ca^{2+}-dependent phospholipids-binding protein with high affinity for negatively charged phosphatidylserine (PS), which is redistributed from the inner to the outer plasma membrane leaflet in apoptotic or dead cells. Once on the cell surface, PS becomes available for binding to annexin V and any of its magnetic conjugates (Makker et al. 2008; Dirican et al. 2008; Safarik and Safarikova 2012).

Mesenchymal stem cells (MSCs), which can differentiate into multiple mesodermal tissues, were magnetically labeled using cationic magnetoliposomes (leading to the concentration of 20 pg of magnetite per cell) in order to enrich them magnetically from bone marrow. The magnetoliposomes exhibited no toxicity against MSCs in proliferation and differentiation to osteoblasts and adipocytes. During subsequent culture, substantially higher density of cells was obtained, compared to culture prepared without magnetoliposome treatment (Ito et al. 2004).

Superparamagnetic iron-oxide nanoparticles, such as MRI contrast agent Endorem or dextran-based magnetic nanoparticles MicroBeads (Miltenyi Biotec) have been used to label the stem cells. Nanoparticles can often be internalized by cells during cultivation by endocytosis (Sykova and Jendelova 2005).

The alternative magnetization procedures are based on the entrapment of nonmagnetic materials into appropriate (bio)polymer gel matrix, together with magnetic nano- or microparticles. Such a procedure can be very mild, enabling magnetic modification of living cells and subsequent employing their biological

Fig. 7 Magnetically responsive alginate microbeads containing entrapped *Saccharomyces cerevisiae* cells and magnetite microparticles. The scale bar corresponds to 50 μm. Reproduced, with permission, from (Safarik et al. 2008)

activities. In a typical example magnetically responsive alginate beads containing entrapped *S. cerevisiae* cells and magnetite microparticles were prepared (Safarik et al. 2008). Larger beads (2–3 mm in diameter) were prepared by dropping the mixture into calcium chloride solution, while microbeads (the diameter of majority of particles ranged between 50 and 100 μm) were prepared using the water-in-oil emulsification process (Fig. 7).

An exceptional magnetization procedure employed erbium chloride as a magnetic label of a variety of cells. Erbium ions have a high affinity for the external cell surface and preserve their exceptionally high atomic magnetic dipole moment (9.3 Bohr magnetons) in various chemical structures. Both Gram-positive and -negative bacteria can be magnetically modified (Zborowski et al. 1993; Safarik and Safarikova 2007).

3 Application of Magnetically Modified Biological Materials

3.1 Magnetic Plant-Derived Materials

Plant-derived materials, such as sawdust, spent grains, spent coffee grounds, straw etc. are typical representatives of low cost (sometimes even waste) materials originating from agricultural and food industries. Plant materials have been frequently used as low-cost biosorbents for the removal of important organic and inorganic xenobiotics (Srinivasan and Viraraghavan 2010; Volesky and Holan 1995).
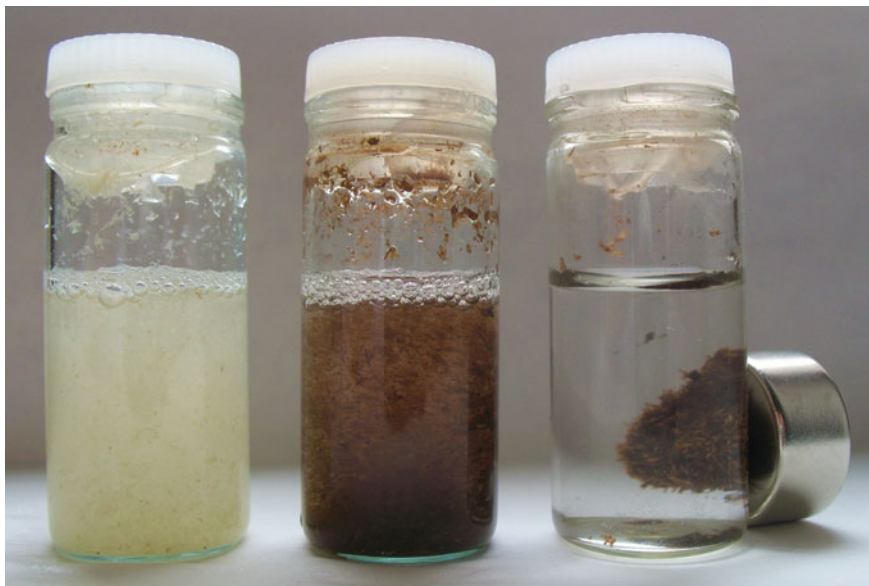
Fig. 8 Appearance of original lignocellulose suspension (*left*), suspension of lignocellulose after magnetic modification (*middle*) and demonstration of magnetic separation of magnetically modified lignocellulose (*right*)

Postmagnetization of such materials enables their simple magnetic manipulation (Fig. 8) and subsequently the possible development of magnetic separation-based technologies for xenobiotics removal (Safarik et al. 2011c, 2012d). Currently there are only a few examples of magnetically responsive plant-based materials applicable for xenobiotics removal. Especially, the adsorption of water soluble dyes was studied. Plant materials, modified with water-based magnetic fluid stabilized with perchloric acid or magnetized using a microwave assisted modification, was applied in laboratory adsorption experiments. In most cases analytes adsorption followed the Langmuir isotherm. This model allows calculating maximum adsorption capacities which are very important parameters for evaluation of adsorbents properties. The maximum adsorption capacities of the developed materials often exceeded values 100 mg of adsorbed dyes per one gram of magnetic biosorbent which is fully comparable with other described non-magnetic biosorbents (Srinivasan and Viraraghavan 2010; Safarik et al. 2011c, 2013).

3.2 Magnetically Modified Cells

Microbial cells, either in free or immobilized form, can be used for the preconcentration or removal of metal ions, organic and inorganic xenobiotics or biologically active compounds. Especially low cost biomaterials such as baker's yeast

(*Saccharomyces cerevisiae*), fodder yeast (*Kluyveromyces fragilis*) and algae *Chlorella vulgaris* are of special interest. The maximum adsorption capacities of magnetically modified yeast and algae cells for dyes removal can reach very high values, up to 430 mg of adsorbed dye per one gram of magnetic adsorbent (Safarik et al. 2011c).

In addition to organic xenobiotics, magnetically modified microbial cells can be efficiently used also for the removal of heavy metal ions, such as mercury (Yavuz et al. 2006) and copper (Uzun et al. 2011) ions. Magnetic fodder yeast represents a promising adsorbent which can be used to concentrate and separate Sr^{2+} ions from liquid high level waste originating during spent fuels reprocessing in nuclear power plants (Ji et al. 2010).

Magnetic modification of living microbial cells can lead to the formation of magnetically responsive whole cell biocatalysts. Both direct modification of cell walls with appropriate water-based magnetic fluids (Fig. 4), formation of cell aggregates caused by magnetic iron oxides nano- and microparticles synthesized from ferrous sulphate during microwave irradiation (Fig. 5) or target cells entrapment into biocompatible (bio)polymer gels in the presence of magnetic particles (Fig. 7) can be performed in a simple way (Safarikova et al. 2009; Fakhrullin et al. 2010a; Safarik et al. 2008; Pospiskova et al. 2013). The intracellular enzyme activities were not decreased substantially after the modification, as shown by hydrogen peroxide degradation and sucrose hydrolysis by intracellular enzymes catalase and invertase present in magnetically modified *S. cerevisiae* cells (Safarikova et al. 2009; Safarik et al. 2008; Pospiskova et al. 2013).

Rhodococcus erythropolis IGST8 cells decorated with magnetic Fe_3O_4 nanoparticles (45–50 nm in diameter) were used for the biodesulfurization of dibenzothiophene (DBT) and for the post-reaction separation of the bacteria from the reaction mixture. It was found that the decorated cells had 56 % higher DBT desulfurization compared to the undecorated cells. Based on the fact that the nanoparticles enhanced membrane permeability of black lipid membranes, the authors proposed that magnetic nanoparticles increased the permeability of the bacterial membrane, thus facilitating the mass transport of the reactant and product (Ansari et al. 2009).

Magnetically modified microbial and algae cells can be used as a part of biosensor systems, both in microfluidics configuration (Fakhrullin et al. 2010a) and as a part of screen printed electrodes (Zamaleeva et al. 2011); the cells can be used for genotoxicity and cytotoxicity measurements and also for the determination of herbicides such as atrazine and propazine.

Immunomagnetic separation is one of the most frequently used techniques for magnetic modification and subsequent separation of target cells. This procedure is very important e.g. for the isolation of stem or cancer cells. In microbiology and parasitology area, detection of important microbial pathogens (e.g., *Salmonella*, *Legionella*, *Listeria monocytogenes*, verocytotoxin-producing *E. coli*) and parasites

(e.g., *Cryptosporidium* and *Giardia*) is of great importance. Detailed information can be found in several review articles (Safarik and Safarikova 1999, 2012; Olsvik et al. 1994; Safarik et al. 1995).

The cells magnetically labeled with appropriate magnetic biocompatible nanoparticles enabled either their in vitro detection by staining for iron to produce ferric ferrocyanide (Prussian blue), or in vivo detection using MRI visualization, due to the selective shortening of T2-relaxation time, leading to a hypointense (dark) signal. MRI can be used to evaluate the cells engraftment, the time course of cell migration and their survival in the targeted tissue (Sykova and Jendelova 2005).

Magnetically labeled cells were successfully used for magnetic force-based tissue engineering to develop functional substitutes for damaged tissues. Labeled cells can be manipulated by using a magnet which enabled to seed labeled cells onto a low-adhesive culture surface by using magnetic force to form a tissue construct. Complex cell patterns (curved, parallel, or crossing motifs) can be successfully fabricated from several cell types. Magnetically labeled keratinocytes were accumulated using a magnet, and stratification was promoted by a magnetic force to form a sheet-like 3D construct (Ino et al. 2007). An excellent review describing various aspects of magnetic tissue engineering was published recently (Ito and Kamihira 2011).

4 Future Trends

Described examples of postmagnetization of non-magnetic biological materials represent just selected collection of many diverse magnetically responsive materials which can be prepared from enormous amount of "non-magnetic" precursors, both from biology and non-biology area. Several new, efficient procedures for postmagnetization have been described recently (Safarik et al. 2012b, 2013; Safarik and Safarikova 2014) which will enable to use really inexpensive materials and precursors, as well as simple technologies to magnetically modify broad range of materials.

Besides the already described utilizations, many other applications can be found in the future. Some magnetically modified plant materials could become very interesting carriers for immobilization of broad range of enzymes. Recently, technically important enzymes were immobilized on magnetic spent grain (Pospiskova and Safarik 2013) and other magnetic lignocellulose materials (Safarik et al. 2013). At least some of these materials can find interesting applications in food industry. Magnetic spent grain, which originates as a by-product of beer industry, is an excellent example of highly food-technology compatible magnetic carrier prepared by simple modification of the raw waste material (Safarik et al. 2012d).

5 Conclusions

Different types of biological materials, including by-products from food and agricultural industries, living and dead prokaryotic and eukaryotic cells and even multicellular organisms, can be successfully magnetized and subsequently utilized for many interesting applications, such as biosorbents for xenobiotics and biologically active compounds separation and removal, carriers for target compounds immobilization, whole cell biocatalysts and parts of sensing systems. Conversion of these originally "non-magnetic" biological materials into "smart materials" exhibiting response to external magnetic field may be one of the possible ways how to improve applicability of these materials, enabling their selective magnetic separation from difficult-to-handle systems. Of course, the same basic principles can be used for postmagnetization of many other materials, including inorganic and organic adsorbents and carriers, and other materials with interesting properties (Safarik et al. 2012d).

Despite the fact, that currently magnetically responsive biocomposites are tested mainly in laboratory experiments, there is a great potential for their large-scale applications in the near future. Postmagnetization of appropriate "non-magnetic" materials and formation of smart magnetically responsive materials will become a very useful tool for both laboratory and large scale applications. The list of available magnetic materials can increase dramatically in the near future which can lead to broader application of magnetic techniques.

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