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One-step synthesis of iron oxide polypyrrole nanoparticles encapsulating ketoprofen as model of hydrophobic drug

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ABSTRACT

This study reports a novel one-step synthesis of hybrid iron oxide/polypyrrole multifunctional nanoparticles encapsulating hydrophobic drug and decorated with polyethylene glycol. The overall process is based on the *in situ* chemical oxidative polymerization of pyrrole along with the reduction of ferric chloride ($FeCl_3$) in the presence of ketoprofen as model drug and PEGylated surfactants. The final product is a nanocomposite composed of polypyrrole and a mixture of FeO/Fe_2O_3. Different concentrations of ketoprofen were encapsulated in the nanocomposite, and were characterized by Fourier transform infrared spectroscopy (FTIR) and differential scanning calorimetry (DSC). Encapsulation efficiency of the final product was measured by absorption, which can reach up to 98%. The release experiments confirmed complete drug release after about 3 h in PBS solution. Morphological characterization of the nanocomposites was performed by electron microscopy (scanning and transmission electron microscopy) which confirmed the spherical geometry and opaque nature of nanoparticles with average particle size well below 50 nm. The final product is multifunctional system, which could act both as a nanocarrier for drug molecules as well as a contrasting agent. Magnetic relaxometry studies confirmed their possible applications as potential contrast agent in the field of magnetic resonance imaging (MRI).

1. Introduction

Nanotechnology involves manipulating properties and structures at the nanoscale level offering many advantages and properties which otherwise are not possible at macroscopic level. One such area is the combination of nanotechnology with medical applications that has led to the emergence of a new field called nanomedicine. Important tools for the development of nanomedicine are the nano-carriers, which have received a special attention over the past few years. These systems have opened new ways to modify the biodistribution and pharmacokinetics of active principle ingredients (API) by improving their bioavailability and/or targeting specific sites at high concentrations while minimizing the side effects. Polymeric nanoparticles are one such organic strategy for nanomedicine which have high potential to revolutionize modern medicine (Nasongkla et al., 2006; Khemtong et al., 2009; Feng et al., 2010), liposomes (Ewesuedo and Ratain, 2003; Petros et al., 2008; Ferrara et al., 2009), dendrimers (Medina and El-Sayed, 2009) and inorganic NPs (Liong et al., 2008; Morgan et al., 2008; Kim et al., 2009) because of their numerous advantages like their possible biocompatibility, biodegradability, non-immunogenic properties, and their potential applications for controlled drug release into targeted cancer cells or tumor tissues (Haag and Kratz 2006). Some specific systems like polymer-drug

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conjugates can even increase the drug payloads, reduce the systemic toxicity, prolong drug circulation time, and improve drug solubility and targeting (Godwin et al., 2001; Ye et al., 2006). There are many synthesis protocols for the preparation of polymer based NPs, which can be broadly divided-up to (i) monomers polymerization in the form of nanoparticles, and (ii) nanoprecipitation of preformed polymers. Polymeric nanoparticles based on intrinsic conductive polymers (ICPs) such as polypyrrole, polyaniline, polythiophene are often prepared by polymerization of their monomers. Among these, polypyrrole possess many interesting properties like good conductivity, bio-degradability, biocompatibility and possess no threat to the environment which makes it a promising candidate for a wide range of applications. (Skotheim, 1986; Stupnisek-Lisac et al., 1992; Tarcha et al., 1992; Pope et al., 1996; Faverolle et al., 1998; Skotheim et al., 1998; Leclerc, 1999; Jager et al., 2001; Iroh and Su, 2002; Kros et al., 2002; An et al., 2004; Kros et al., 2004; Wang et al., 2004; White and Slade, 2004; Ramanathan et al., 2005; Reece et al., 2005; Sides and Martin, 2005; Wadhwa et al., 2006; Esrafilzadeh et al., 2013; Maity et al., 2014). Another advantage lies in their facile synthesis through various polymerization routes, *e.g.* from photosynthesis polymerization process (Yang and Lu, 2005; Hodko et al., 2009; Attia et al., 2013), electrochemical polymerization pathway (Ashraf et al., 1996; Li et al., 2005) to *in situ* chemical oxidative polymerization reaction (Fujii et al., 2007; Müllera et al., 2011; Wang et al., 2013; Ghadim et al., 2014) in the presence ferric chloride ($FeCl_3$), ferric nitrate ($Fe(NO_3)_2$), ammonium persulfate ($NH_4S_2O_8$), sodium persulfate ($Na_2S_2O_8$) as oxidizing agents. These reactions involve polymeric stabilizers or surfactants like polyvinylpyrolidone (PVP), polyvinylalchol (PVA), sodium dodecyl sulfate (SDS), cetyltriammonuim bromide (CTAB).

The properties of polypyrrole NPs can be further enhanced by making hybrid particles especially using the polymer for encapsulation of metal or inorganic particles *e.g.* gold, silver, copper, and iron inside the polymer core (Gangopadhyay and De, 2000; Martins et al., 2006). Such hybrid nanocomposites have found a broad range of applications as they often exhibit many new properties as well as improved physical and chemical properties over their single-component counterparts. Unfortunately, such hybrid materials are often met with some limitations like difficulty in process design, PPy mechanical performance, and poor water solubility, which limit their applications in biomedical fields and also potential industrial scaling-up. In order to overcome these limitations, many techniques have been developed based on the dispersion of ICPs in a matrix composed of an insulation polymer, which improves the process design and mechanical performance along with the electrical, magnetic and optical properties of ICPs (Wang and Fernandez, 1992; Vicentini et al., 2007).

In the present study we have proposed a novel nanocarrier system based on polypyrrole nanocomposite, capable of encapsulating ketoprofen as model drug. These nanoparticles were synthetized by a one-step *in situ* polymerization reaction, coated with a PEG chains, and are designed to show electromagnetic properties. The PEG layer not only controlled the *in vivo* behavior such as blood clearance (Soppimath et al., 2001) but also allow a precise control on the final size and monodispersion feature of the nanocarrier. This morphology of the nanoparticles as well as their ability to encapsulate a model drug, and their release profile have also been thoroughly investigated. Our main objectives in this study are focused on the design, characterization and physicochemical evaluation of a new nanoparticulate system. The main advantage is the simple one-pot synthesis protocol for the preparation of multifunctional polymeric nanoparticles. In addition, the materials herein used have already been used in biological application showing their biocompatibility (Alizadeh and Shamaeli 2014). The final material has been thoroughly characterized to confirm the size, change in morphology with composition, magnetic properties, as well as encapsulation and release profile of a model lipophilic drug (ketoprofen).

2. Experimental part

2.1. Reagents and materials

Prior to synthesis pyrrole (Aldrich) was distilled twice under reduced pressure, and was refrigerated in dark at low temperature in an inert environment. Iron (III) chloride hexahydrate ($FeCl_3{\cdot}6H_2O$, Aldrich) was used as an oxidant/dopant species and was employed without further purification. Ketoprofen as a hydrophobic drug model was purchased from Sigma–Aldrich. Kolliphor® HS 15 is a mixture of free PEG 660 and PEG 660 hydroxystearate (PEG 660 12-hydroxystearate, MW 870 Da) (BASF, Ludwigshafen, Germany). Distilled water and methanol were used as solvents, stock solution of phosphate buffer saline (PBS) pH 7.4 prepared by using double distillated water. Dialysis tubing (12,000–14,000 molecular weight cutoff) were obtained from Sigma Aldrich, USA. 0.45 μm syringe filters were purchased from Fisher (Germany).

2.2. Preparation of ketoprofen doped iron/PPy-PEG nanocomposites as drug delivery system

The drug-loaded nanocomposites (NCs) were synthesized according to the concept of *in situ* chemical oxidative polymerization, based on a modified reported method (Škodová et al., 2013). As illustrated in Fig. 1, $FeCl_3{\cdot}6H_2O$ was used as an oxidant to initiate the polymerization process, and the ferric ion(+III) was reduced during the chemical reaction (redox reaction) forming FeO and Fe_2O_3 in nanometric sizes that were embedded in the polymer matrix (Zong et al., 2015). Nonionic hydrophilic PEGylated surfactant (Kolliphor® HS 15) was introduced during the synthesis as emulsifier and stabilizer. In a typical procedure, an aqueous and organic phase have been prepared separately, (i) The surfactant (Kolliphor® HS 15) was added in the 55 mL aqueous phase (milliQ water) corresponding to 0, 5, 10, 20, 30, 40, 50 wt.% of the total amount of NCs under vigorous stirring. This was followed by addition of (2.8 mmoles, 1 equv.) of ferric chloride. (ii) The organic phase (methanol) was prepared by dissolving pyrrole monomer (11.2 mmoles, 4 equiv.), in 3 mL of methanol followed by addition of different amounts of ketoprofen (K) (5, 10, 20 wt.% of Ket/total NC ratio). After complete homogenization, the organic phase was added drop wise into the aqueous phase under vigorous stirring at 1000 rpm. The complete polymerization took about 12 h at room temperature, gradually getting converted into nanocomposites displaying a black aspect. The particles was then collected by centrifugation and the resulting nanoparticles were thoroughly washed three times with deionized water to remove any hydrophilic remnants, one time with ethanol, and finally with acetone to remove excess of free ketoprofen and free pyrrole monomer. The precipitate was finally dried under vacuum for 24 h.

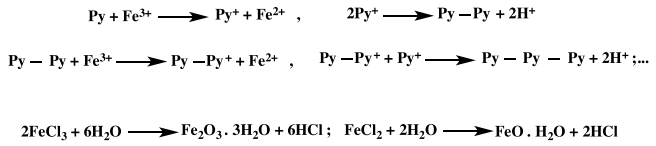

Fig. 1. Chemical equations for oxidation-reduction process during the polymerization of polypyrrole inducing iron oxide NPs imbedded into the NCs.