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Preparation and evaluation of magnetite nanoparticles and novel magnetic dosage form for topical application

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ABSTRACT

The particles of magnetite were prepared using a chemical condensation method. The crystalline structure, morphology and the magnetic properties of the ferrite particles were studied by means of X-ray diffraction (XRD) and vibrating sample magnetometer (VSM). By an appropriate combination of the magnetic ointment bases prepared using different amounts of magnetic nanoparticles (MNPs) and different type of bases it was possible to obtain a system with a good texture and magnetic properties. The water absorption capacity of polyethylene glycol (PEG) formulations was related to the MNPs concentration. Magnetic measurements of a formulation PEG 400/1900 with MNPs 30% have demonstrated the increase of magnetization as a result of reduction of temperature from 300 K to 77 K and the transition from superparamagnetic to ferromagnetic behavior of a formulation (Hc = 100 Oe). These results suggest a potential of the new magnetic ointment bases for the development of modified dosage form for topical application.

1. Introduction

Magnetic nanotechnology is widely used in modern pharmaceutical science[1-5]. Magnetite nanoparticles (Fe_3O_4) are used as magnetic material of Magnetic Drug Delivery Systems[6-11].

Chronic tonsillitis is one of the most common otorhinolaryngological diseases. Treatments for tonsillitis focus on eliminating infection and inflammation associated with the condition. Chronic tonsillitis is corrected by tonsillectomy (surgical removal of the tonsils)[12].

The idea was to create the dosage form with the magnetite particles in the form of ointment. An ointment is a viscous semisolid preparation used topically on a variety of body surfaces. They compose of active ingredient and ointment base. Ointment bases are used as vehicles for medicated ointments. The magnetic ointment can be applied directly to the tonsils, then begin the influence of permanent magnetic field on the area of projection of the Palatine tonsils (anterolateral surface of the neck). Use of such a magnetic formulation with its applying and fixing via an external magnet on the tonsils will allow improving the efficacy of local treatment.

Topical therapy is desirable since, in addition to targeting the site of infection, it reduces the risk of systemic side effects thus the main aim of this study was to design novel magnetic topical dosage form for topical treatment of otorhinolaryngological infectious diseases such as tonsillitis. This investigation consisted of preparation and evaluation of magnetite nanoparticles (MNPs) and magnetic ointment bases using different amounts of MNPs and different type of bases to obtain a system with a good texture and magnetic properties. To our knowledge, no other reports exist of the preparation and evaluation of magnetic dosage form for topical treatment of otorhinolaryngological infectious diseases.

2. Materials and methods

2.1 Materials

Iron (III) chloride ($FeCl_3 \cdot 6H_2O$), iron (II) sulfate ($FeSO_4 \cdot 7H_2O$), $NH_3 \cdot H_2O$ (25% w/w aqueous solution) and polyethylene glycols were purchased from E. Merck Ltd. (India). Lugol iodine was purchased from Instamed Labchem (India); sea buckthorn oil from X.S. Biotech Co., Ltd. (China) and oleic acid from Beijing ChemWorks (China). All chemicals were of analytical grade.

2.2 Synthesis of magnetite nanoparticles

Ultrafine particles of $FeFe_2O_4$ were prepared by co-precipitating aqueous solutions of iron (II) salt ($FeSO_4 \cdot 7H_2O$) and iron (III) salt ($FeCl_3 \cdot 6H_2O$) in an alkaline medium (25% $NH_3 \cdot H_2O$). 13.89 g (0.05 mol) of $FeSO_4 \acute{} 7\ H_2O$ and 26.90 g (0.1 mol) of $FeCl_3 \acute{} 6H_2O$ were dissolved in 1 L of distilled water at heating (60-70°C). 25% aqueous ammonia solution was added drop-wise with continuous stirring until complete precipitation of the black ferrite was achieved (pH 9–11). After the system was cooled to room temperature, the precipitates were collected using magnetic separation and washed with distilled water until pH neutral, producing thus samples MNPs.

2.3 Preparation of ointment bases

Nine ointment bases of varying compositions (Table 1) were prepared by using different amounts of MNPs and different type of bases. The mixtures of PEG were prepared by melting together polyethylene glycol (PEG 400 and PEG 1500) on a hot plate/stirrer (at 70ºC). The PEG 400/1500 ratios weight were 9/1 (forms I-V) and 4/1 (forms VI-VII), respectively. MNPs were added to this molten base while stirring. The entire mixture was stirred while cooling. The prepared ointment bases were put in ointment jars and were stored at room temperature pending the evaluation. The ingredients of the different formulations (I to IX) are listed in **Table 1**.

2.4 Particle characterization

The X-ray diffraction (XRD) patterns of the samples were recorded on a Siemens D500 X-ray powder diffractometer using copper radiation. Slow scans

Table 1: Compositions used in the study to prepare ointment bases

Ingredients	Formulations								
	I	II	III	IV	V	VI	VII	VIII	IX
MNPs	20	25	30	35	40	25	30	10	50
PEG 400	72	67.5	63.0	58.5	54.0	60.0	56	-	-
PEG 1500	8	7.5	7.0	6.5	6.0	15.0	14	-	-
Lugol iodine	-	-	-	-	-	-	-	90	-
Sea buckthorn oil	-	-	-	-	-	-	-	-	47.5
Oleic acid	-	-	-	-	-	-	-	-	2.5

Table 2: Phase composition and characteristics of MNPs

Phase	Mass, %	Lattice parameters, (Å)	d, g/cm^3	Crystallite size and microstrain, nm/ %
	93.3(6)	a=8.3668(12)	5.28	16/0.14
γ-Fe_2O_3	6.2(2)	a=8.348(4)	4.89	9/0.13
α-Fe_2O_3	0.4(2)	a=5.0280	5.27	27/–
		c=13.651		

of the selected diffraction peaks were carried out in the step mode (step size 0.03°, measurement time 75 s. The crystallite size of the nanocrystalline samples was measured from the X-ray line broadening using the Debye-Scherrer formula after accounting for instrumental broadening. Magnetization measurements were performed in a vibrating sample magnetometer (VSM) at 300 K and 77K using a superconducting magnet to produce fields up 2 kOe.

2.5 Density measurement and rheological analysis of formulations

The density measurements of the formulations (I-IX) were carried out using a 50 ml pycnometer. For the hard samples, the pycnometer was filled with the melted sample. After 24 h storage at room temperature the mass was measured. Rheological analysis was performed on a rotary viscometer Rheotest RV2 (VEB MLW Medingen, Germany). The rheological behavior was studied by continuous shear investigations, which were performed in order to evaluate the shear stress (Pa) as a function of shear rate (s^{-1}). The study was started with a shear rate of 1 s^{-1} up to a maximum of 500 s^{-1} and back to 1 s^{-1}, and the resulting shear stress was measured at temperature 34°C (thermostatic water bath was used).

2.6 Water absorption capacity of ointment bases

The water absorption capacity of ointment bases (PEG 400/1500 mixture 9/1; PEG 400/1500 mixture 9/1 with magnetite 25% (PEG/MNPs25), 30% (PEG/MNPs30) and 35% (PEG/MNPs35) at 37 °C were measured *in vitro* through cellulose membrane inserted in the Franz diffusion cell. Distilled water was used as the medium. An electronic balance (AND, Model GF400, accuracy ± 0.001 g Japan)

Table 3: The parameters of the composition and physical characteristics of the ointment bases

Ingredients	Formulations								
	I	II	III	IV	V	VI	VII	VIII	IX
Mass part of MNPs, %	20	25	30	35	40	25	30	10	50
Volume part of MNPs, %	5.35	6.68	8.08	9.43	10.81	6.35	7.87	2.44	12.5
Density, g/cm^3	1.281	1.320	1.356	1.402	1.432	1.331	1.378	1.271	1.306
Saturation magnetization, emu/g	3.52	4.47	5.32	6.21	36.69	7.09	5.17	1.61	8.16

were used to measure weight of samples before and after immersion. In order to reduce error, all tests were performed in triplicate.

3. Results and discussion

3.1 Characterization of as-synthesized MNPs

The formation of Fe_3O_4 was confirmed by XRD, and is shown in Figure 1. The pattern has well defined peaks from reflection planes (220), (311), (400), (422), (511) and (440), which were indexed in the Fd3m(227) space group corresponding to a spinel cubic structure of magnetite (JCPDS PDF card No. 19–629). The presence of maghemite (γ-Fe_2O_3) and hematite (α-Fe_2O_3), beside to the major magnetite phase, have been confirmed by X-ray diffraction analysis (**Table 2**).

The magnetic measurement confirms that the synthesized particles exhibit superparamagnetic properties at room temperature. The magnetization curve for the MNPs exhibits immeasurable values of coercivity field and remnant magnetization. The particles have high saturation magnetization values (67 emu/g), which is consistent with the value reported in the literature for a magnetite sample with the same sizes [13].

3.2 Evaluation of the properties of magnetic ointment bases

To develop a successful magnetic dosage form formulation for topical treatment of otorhinolaryngological infectious diseases such as tonsillitis, it is important to study the rheology, texture, and magnetic properties. The results of comparing of the properties of the proposed compositions (**Table 3**) showed that the optimum formulations are II – IV because they have the best structure and one of the highest magnetic properties, which allowing its manipulation with an external magnetic field. Other formulations were not accepted because they have liquefied structure like formulations I, VIII-IX or too dense structures like formulations V-VII.

The magnetic interactions between the dispersed MNPs make them agglomerate and settle down. The highly dense PEG mixture provides steric hindrance preventing nanoparticle from aggregation. Therefore, the formulations II – IV with the PEG mixture were selected for further study.

Regarding flow behavior all the formulations show very similar characteristics (**Figure 2**). The flow curves of the tested formulations revealed a non-Newtonian shear-thinning behavior, with yield value. The formulations with higher yield values showed higher values of viscosity and shear stress

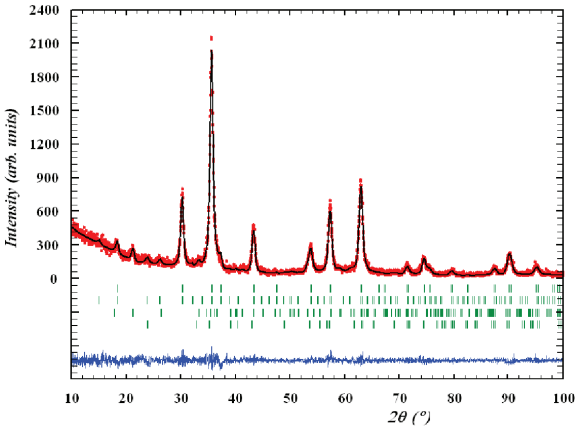

Figure 1. *X-ray diffraction pattern of MNPs*

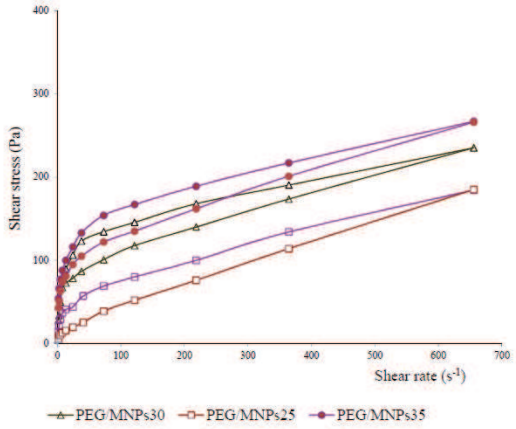

Figure 2. *Rheogram for the ointment bases PEG 400/1500 mixture 9/1 with magnetite 25% (PEG/MNPs25), 30% (PEG/MNPs30) and 35% (PEG/MNPs35)*

than the other formulations. Thixotropy, a reversible variation of viscosity with time, was observed in all formulations, demonstrated by the presence of hysteresis area between ascending and descending curves of the rheograms. Regarding the hysteresis area, this was slightly greater in the case of formulations PEG/MNPs30 and PEG/MNPs35.

PEGs are hydrophilic materials and are extensively used in pharmacy. Solid PEGs in the mixture with liquid PEGs will lead to a white, pasty ointment with good solubility in water, good dissolving properties and suitable for many active substances[14-16]. It is well known that, the type of the base used in formulating of a topical medication markedly affects the permeability of drug. Numerous actives can be dissolved in PEGs resulting in a good bioavailability[17-21]. Polyethylene glycol (PEG) and PEG derivatives are the most effective and widely used polymer for improving nanomaterial stability[22-26].

3.3 Water absorption capacity of ointment bases

All samples exhibit significant osmotic activity to distilled water (**Figure 3**). Incorporation of MNPs into the PEG mixture will decrease the water absorption activity of the formulations. As the content of active PEG decreases the activity of the samples naturally decreases.

The presence of a negative charge of Fe–O– groups

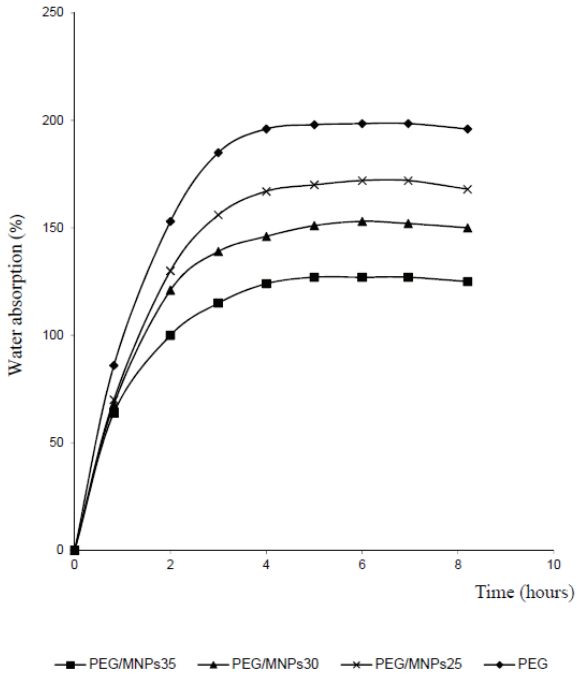

Figure 3. *The water absorption activity of PEG 400/1500 mixture 9/1(PEG); the ointment bases PEG 400/1500 mixture 9/1 with magnetite 25% (PEG/MNPs25), 30% (PEG/MNPs30) and 35% (PEG/MNPs35)*

on the surface of MNPs causes the disordering of intermolecular forces of colloidal solution. The effect of external magnetic field on absorption venues can

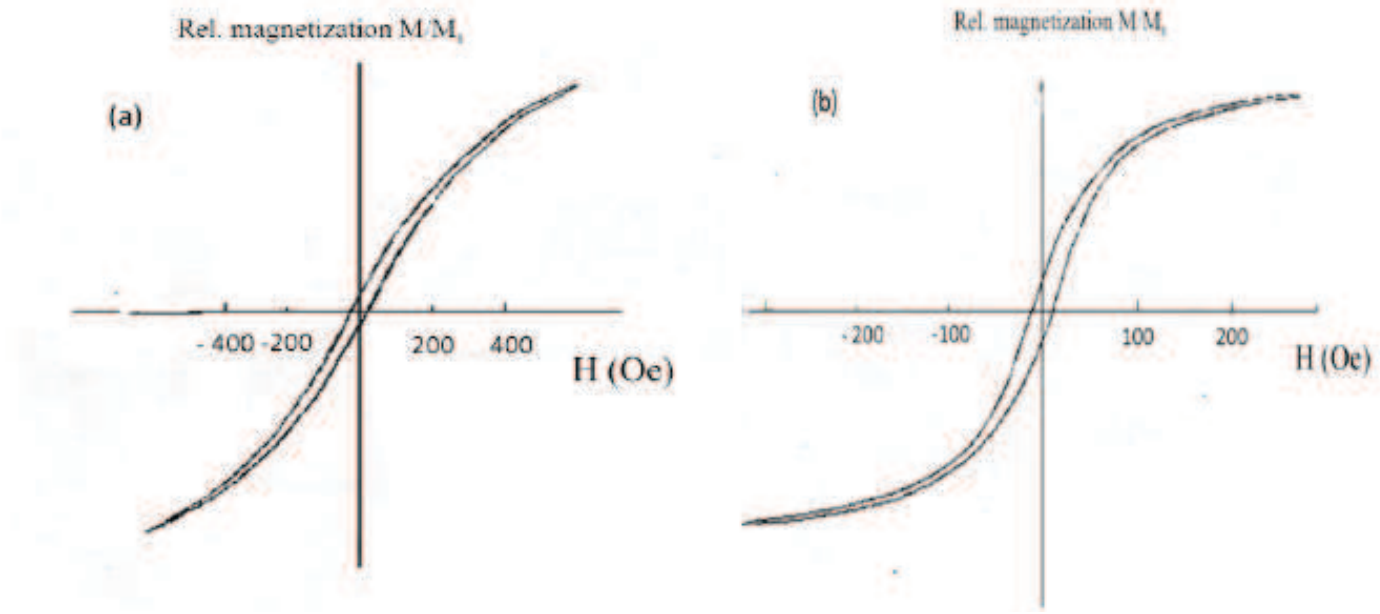

Figure 4. *Normalized magnetization loops at temperatures of 300K (a) and 77 K (b)*

be expected. However, no significant differences were observed on the water absorption activity of the formulations under the influence of the magnetic field (100 mT). Reducing of the absorption properties has a positive effect, because it is well known that the hydrophilic compounds (such as PEG) can damage cell membranes causing rashes, dry skin, contact dermatitis and surface damage to the skin[27].

3.4 Magnetic characterization of the formulation PEG/MNPs30

Magnetic behavior was studied for the formulation III with the optimal content of magnetite 30% (**Figure 4**).

Figure 4 shows the magnetization versus field plot at temperatures of 300 K (a) and 77 K (b). It can be seen that hysteresis disappeared with a little remanence and coercirvity (*Hc*), indicating the absence as a long range magnetic dipole-dipole interaction among the assemblies of superparamagnetic nanoparticles in the formulation at a room temperature. When the temperature was decreased to 77 K, the magnetization of the sample increased (from 5.32 emu/g till 6.24 emu/g) with a symmetric hysteresis loop, and showed a transition from superparamagnetic to ferromagnetic behavior of MNPs in the sample (*Hc* = 100 Oe). The difference in the intensity of the saturation between 300 K and 77 K is due to the magnetic ordering of superparamagnetic particles of the low-coercivity phase. Such behavior is typical for magnetite nanoparticles of single-domain size[28].

Based on the fact, that magnetite has a high value of thermal conductivity (9.7 $Wm^{-1}K^{-129}$), it is reasonable to use the prepared ointment base as a thermal conductivity medium for cryotherapy of the tonsils. Use of such a magnetic formulation with its applying and fixing via an external magnet on the area of pathologies will allow improving the efficacy of cryogenic treatment and destruction, will provide good adhesion and contact of the tonsils with metal applicators through which a cryogen passes. As it is realized, the final temperature level would be significantly lowered, the maximum freezing rate will be increased.

4. Conclusion

In this study a magnetic system has been developed,

PEG/MNPs formulation with a good texture and magnetic properties which might be a suitable ointment base for target treatment of otorhinolaryngological infectious diseases such as tonsillitis or can be used alone (e.g. as thermal conductivity medium for cryotherapy). □

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Conflict of Interests

The authors declare that they have no conflict of interests to disclose.

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