Journal of Magnetism and Magnetic Materials xx (xxxx) xxxx–xxxx

Contents lists available at ScienceDirect

Journal of Magnetism and Magnetic Materials

journal homepage: www.elsevier.com/locate/jmmm

Simple optical measurement of the magnetic moment of magnetically labeled objects

Alexandra Heidsieck[a,*], Sarah Rudigkeit[b], Christine Rümenapp[a], Bernhard Gleich[a]

[a] Zentralinstitut für Medizintechnik, Technische Universität München, Germany
[b] Physics Department, Technische Universität München, Germany

ARTICLE INFO

Keywords:
Magnetic moment
Magnetization
Magnetic nanoparticles
Velocity measurement
Magnetophoretic mobility
Characterization

ABSTRACT

The magnetic moment of magnetically labeled cells, microbubbles or microspheres is an important optimization parameter for many targeting, delivery or separation applications. The quantification of this property is often difficult, since it depends not only on the type of incorporated nanoparticle, but also on the intake capabilities, surface properties and internal distribution.

We describe a method to determine the magnetic moment of those carriers using a microscopic set-up and an image processing algorithm. In contrast to other works, we measure the diversion of superparamagnetic nanoparticles in a static fluid. The set-up is optimized to achieve a homogeneous movement of the magnetic carriers inside the magnetic field. The evaluation is automated with a customized algorithm, utilizing a set of basic algorithms, including blob recognition, feature-based shape recognition and a graph algorithm. We present example measurements for the characteristic properties of different types of carriers in combination with different types of nanoparticles. Those properties include velocity in the magnetic field as well as the magnetic moment. The investigated carriers are adherent and suspension cells, while the used nanoparticles have different sizes and coatings to obtain varying behavior of the carriers.

1. Introduction

Magnetically labeled cells, microbubbles or microspheres have become important tools for targeting, delivery or separation applications. Magnetic nanoparticles experience a force in an inhomogeneous magnetic field and are drawn towards the magnetic field source. Thereby, they can be diverted by an external magnetic field gradient. Magnetically labeled cells, microbubbles or microspheres incorporate up to several hundred magnetic nanoparticles and can thereby be manipulated more effectively by external magnetic fields.

However, the magnetic properties of those objects are often not quantified. In order to optimize the application and judge the efficiency, the magnetic moment of the objects must be known. This property can only be measured indirectly by observing the objects in a well-defined and known magnetic field. Most of the known methods to measure the magnetic moment of nanoparticles and magnetically labeled objects focus on the magnetization of the particles or the magnetic moment of a bulk of particles. Among the most commonly used methods are magnetization [1], force [2,3] and direct [4–6] or indirect [7,8] optical velocity measurements.

Since we are interested in the magnetic moment of the individual objects, we utilized optical velocity measurements. Complexes of nanoparticles, microspheres or cells, which are large enough to be seen under a microscope, can be tracked optically in a magnetic field. Häfeli et al. [4] as well as Zborowski and Chalmers et al. [5,6] used this method to track magnetic microspheres and magnetically labeled cells in a magnetic force field. This is also known as “Cell Tracking Velocimetry” or “Particle Tracking Velocimetry”. The measured quantity is often called “magnetophoretic mobility” and refers to the velocity of the objects normalized by the magnetic field. The magnetic set-ups vary from complex electromagnets to single large permanent magnets. Often, the cells are carried along by a laminar flow and diverted by a magnetic force perpendicular to their streamline. Based on the deviation from their original path, the magnetophoretic mobility and thereby the magnetic moment of the particles can be determined. While most available cell tracking velocimetry set-up can accomplish that, they are large and expensive. We aimed to design a simple set-up of a manageable size which can be used in any laboratory where a microscope with a digital camera is available.

Additionally, we aimed for a homogeneous force on the objects for the measurement of the magnetic moment to avoid acceleration. In the following, we present a method which is able to measure the magnetic

* Corresponding author.
E-mail address: aheidsieck@tum.de (A. Heidsieck).

http://dx.doi.org/10.1016/j.jmmm.2016.10.057
Received 24 June 2016; Received in revised form 28 September 2016; Accepted 11 October 2016
Available online xxxx
0304-8853/

moment of magnetic nanoparticle complexes, microbubbles and cells, based on the observation of their movement under well-defined conditions. Based on a Halbach cylinder, we developed a magnetic set-up in which the carriers move with a nearly constant velocity. Particle complexes, microbubbles or cells are added to a fluid in a high, but homogeneous magnetic flux density gradient field. Thereby, a homogeneous movement of the objects is assured. We observe the movement via optical microscopy and are able to draw conclusions about the magnetic moment of the object.

2. Theory

The magnetization $\vec{M}$ describes the strength of the magnetic dipole moment of the nanoparticle at a defined magnetic field strength and is defined as magnetic moment $\vec{\mu}$ per volume of the core material. Thereby, magnetic moment and magnetization are dependent on each other. Furthermore, the magnetization, as well as the magnetic moment depend on the strength of the external magnetic flux density field.

The magnetic force acting on a magnetically labeled object with the magnetic dipole moment $\vec{\mu}$ within an external inhomogeneous, static magnetic flux density field $\vec{B}$ is described by

$$\overrightarrow{F_{\text{mag}}} = (\overrightarrow{\mu}(B)\cdot\overrightarrow{\nabla})\overrightarrow{B} \tag{1}$$

and can – for an external, static magnetic field - be simplified to

$$\overrightarrow{F_{mag}} = \mu(B)\cdot\overrightarrow{\nabla}B \tag{2}$$

where the absolute value of the magnetic dipole moment $\mu = |\vec{\mu}|$ of a superparamagnetic nanoparticle depends on the absolute value of the local magnetic flux density $B = |\vec{B}|$ and can be described by

$$\mu(B) = \mu_{\text{sat}} L(B) \tag{3}$$

where L is the Langevin function [9]

$$L(B)=\coth\left(\frac{\mu_{sat}}{k_B T}B\right)-\frac{k_B T}{\mu_{sat}B} \tag{4}$$

and μ_{sat} is the saturation magnetization of the particles.

While the magnetization is usually constant for the same particle type at a certain flux density, the magnetic moment varies with the particle size. For larger objects which incorporate a larger amount of nanoparticles, we assume that the magnetic moment is proportional to the hydrodynamic surface area or volume of the object which are referred to as volume magnetization M_V and surface magnetization M_A.

The movement of the object with a velocity v causes friction between object and fluid which is described by the Stokes' drag force

$$\vec{F}_{\text{hydro}}=3\pi\eta d_h\vec{v}, \tag{5}$$

where η denotes the viscosity of the fluid and d_h is the hydrodynamic diameter of the corresponding object.

3. Materials and methods

3.1. Cell culture and media

MDA cells were maintained in complete culture medium composed of Dulbecco's Modified Eagle medium high glucose (DMEM, Biochrom) supplemented with 1 mM sodium pyruvate (Na-pyruvate (100 mM), Biochrom), 5% foetal bovine serum (FBS, Hyclone, perbio) and 0.1% Pen/Strep (Biochrom). Cells were passaged every 7 days at 2500 cells/cm^2 using Trypsin/EDTA-solution (0.05% /0.02%) in PBS w/o Ca^{2+}/Mg^{2+}(Biochrom) and 1 x PBS. Cell preparations were maintained at standard cell culture conditions 37 °C, 5% CO_2 and 95% humidity incubator (HERAEUS, Hanau, Germany).

3.2. Magnetic nanoparticles

The cells were loaded with three different types of magnetic nanoparticles:

- SO-Mag5 is a multi-core MNP made of maghemite cores closed up in a silica shell with a diameter of approximately 40 ± 14 nm. [10].
- MNP are bare magnetic nanoparticles made of maghemite with a distribution of the diameter of 5 ± 2 nm [11].
- MNP-APTES are MNP coated with (3-Aminopropyl)-triethoxysilane (APTES) (abcr GmbH) after the protocol of Ma et al. leaving free amino groups on their surface [12]. 10 ml of the bare particles were diluted in 10 ml of H_2O_{dd} and degased in an ultrasound bath for 15 min. Afterwards EtOH (absolute, Applichem GmbH) was added to the particles in a concentration of 0.02 M (according to the amount of MNP in mol) under nitrogen atmosphere. Two equivalents of APTES according to the amount of MNP were given to this suspension very carefully under vigorous stirring. The reaction was subsequently heated to 60 °C and incubated at 1000 rpm for 3 h. After cooling down to room temperature the particles were magnetically washed three times with 100 ml of EtOH and finally resuspended in 10 ml of H_2O_{dd}. The distribution of their diameter was 13.26 ± 4.32 nm as determined by dynamic light scattering (ZetaSizer Nano ZS, Malvern Instruments Ltd. Malvern, UK) with a number weighted distribution.

3.3. Cell preparation

MDA cells were transferred to a 24 Well plate 6 h before their loading with MNP. They were seeded with 80,000 cells per well, with 50,000 cells per ml and in quadruplets. The used MNP were added to the cells 100 pg Fe/cell for MNP, 80 pg Fe/cell for APTES-MNP and 20 pg Fe/cell for SO-Mag 5. After the incubation on a magnetic field for 24 h the cells were washed with PBS, resuspended in medium and transferred to Eppendorf tubes. Depending on their density each well was transferred to one tube, or two wells were combined into one.

3.4. Experimental set-up

For the observation of the cells, we designed a small set-up which can be placed under an inverted microscope. The layout of the set-up is shown in Fig. 1A. 47 small cuboidal rare-earth permanent magnets (height=50 mm, width=5 mm, NdFeB, N52; HKCM Engineering, Eckernförde, Germany) are arranged in two concentric Halbach cylinders (Inner array: 20 magnets on a circle with r =25.1 mm outer array: 27 magnets with r=33.1 mm). Thereby we achieved a quadrupolic magnetic vector potential which would ideally lead to a perfectly uniform magnetic flux density and a constant magnetic gradient. However, due to the limited number of magnets, we only approximate such a field as shown in Fig. 1. Inside the set-up a standard cell culture dish (inner diameter=34 mm, polystyrene (PS)) is placed. The whole set-up is placed under an inverted microscope (Zeiss Axiovert 200, Carl Zeiss AG, Oberkochen, Germany) equipped with a digital CMOS camera (Hamamatsu C11440, Hamamatsu Photonics, Hamamatsu-shi, Shizuoka, Japan).

The vertical position of the inner bottom of the cell culture dish is fixed due to the construction of the magnetic set-up. Due to the added volume of the cell suspension another 0.8 mm can be added to the medium position of the object.

The magnetic gradient in the radial direction is therefore only approximately linear. Fig. 1B shows the magnetic flux density and Fig. 1C the magnetic gradient; both are shown in the plane perpendicular to the z-axis indicated in Fig. 1A. We achieve a magnetic gradient of approximately 23.7 T/m at the vertical center of the set-up. The observation window is at a radial position of 6 mm, where there is a very homogeneous magnetic field gradient.