

sequently the mask layer and unwanted film stack are removed and cleaned off thoroughly. Then, the release layer is removed, lifting off nanoparticles which are the negative image of the mask layer. These particles are eventually imparted with surfactants and biomolecules. The ultra-smooth substrate can be reused after thorough cleaning and chemical mechanical polishing (CMP).

[0043] Alternatively, the nanoparticles can be fabricated with a subtractive fabrication method. In this case, the film stack is directly deposited on the release layer followed by a mask layer. The film stack is etched through the mask layer, and eventually released from the substrate. These nanoparticles result from a positive image of the mask layer as opposed to the case in the additive fabrication method.

[0044] The size of the magnetic nanoparticles suitable for use with the present invention is preferably comparable to the size of the target biomolecule to be worked with, such that the nanoparticles do not interfere with biological processes such as DNA hybridization. Consequently, the size of the magnetic nanoparticles is preferably from about 5 nm to about 250 nm (mean diameter), more preferably from about 5 nm to about 150 nm, and most preferably from about 5 nm to about 20 nm. For example, magnetic nanoparticles having a mean diameter of 5 nm, 6 nm, 7 nm, 8 nm, 9 nm, 10 nm, 11 nm, 12 nm, 13 nm, 14 nm, 15 nm, 16 nm, 17 nm, 18 nm, 19 nm, 20 nm, 25 nm, 30 nm, 35 nm, 40 nm, 45 nm, 50 nm, 55 nm, 60 nm, 70 nm, 80 nm, 90 nm, 100 nm, 110 nm, 120 nm, 130 nm, 140 nm, and 150 nm, as well as nanoparticles having mean diameters in ranges between any two of these values, are suitable for use with the present invention. Further, in addition to the more common spherical shape of magnetic nanoparticles, nanoparticles suitable for use with the present invention can be disks, rods, coils, or fibers.

[0045] Synthetic antiferromagnetic nanoparticles for use in this application may be considerably larger than ordinary ferromagnetic particles. This is because, to prevent clumping, the nanoparticle must have no net magnetic moment (or a very small magnetic moment) in zero applied field. Antiferromagnetic particles may have zero magnetic moment in zero field at all sizes, but for a ferromagnetic particle its size must be below the "superparamagnetic limit", which is typically 20 nm or less, usually less. To demonstrate the advantage of the synthetic antiferromagnetic particle we have made a calculation of the voltage produced in a spin valve detector for a synthetic antiferromagnetic particle 30 nm in diameter and 30 nm in height and compared it to the voltages produced by 16 nm diameter Fe_3O_4 and 11 nm diameter Co nanoparticles. We assume that 75% of the synthetic antiferromagnetic particle is ferromagnetic FeCo and that the spin valve detector is the same in all cases. Table 1 gives the results of these calculations and the caption of Table 1 gives the spin valve dimensions and operating conditions. Note that the spin valve signal from the synthetic antiferromagnet is nearly two orders of magnitude greater than for the ferromagnetic particles.

TABLE 1

Particle	Synthetic FeCo	Fe_3O_4	Co
Saturation magnetization	1950 emu/cc	480 emu/cc	1400 emu/cc
Size	30 nm diameter, 30 nm height	16 nm diameter	11 nm diameter
Net saturation moment	31 femtoemu @ 75 vol % magnetic	1.0 femtoemu @ 100 vol %	1.0 femtoemu @ 100 vol %
Vertical mode SV detector signal (bias field 100 Oe, tickling field 141 Oe)	193 μV	0.9 μV	0.8 μV

Spin valve signal voltage (peak-to-peak amplitude) versus magnetic tag. Only the data for the vertical detection mode (FIG. 2B) is listed here. The voltage is due to a single nanoparticle with its center 20 nm away from the midplane of the spin valve free layer. The sensor size is $3 \mu\text{m} \times 0.2 \mu\text{m}$ with an active length of $1 \mu\text{m}$. The sense current density is 10^8 A/cm^2 . The effect of the stray field from the spin valve sensor is included in the calculation. The synthetic FeCo is assumed to saturate at 30 Oe and the particle physically rotates with the applied field. Note that the room temperature magnetic moments of the superparamagnetic nanoparticles are reduced as described by the Langevin function, but that of the synthetic antiferromagnetically coupled nanoparticles is changed much less by superparamagnetism.

[0046] Note that the signal levels listed in Table 1 are for spin valve detectors. If replaced with an MTJ detector with a junction area of $0.2 \mu\text{m}$ by $0.2 \mu\text{m}$ and resistance-area product of $1 \text{ k}\Omega\text{-}\mu\text{m}^2$, operating with a magnetoresistance (MR) of 25% at a bias voltage of 250 mV, and $H_b=35 \text{ Oe}$, $H_t=100 \text{ Oe rms}$, the voltage signal from a single synthetic FeCo nanoparticle could reach greater than 1 mV. This signal level makes it detectable without a lock-in amplifier, greatly speeding up the entire MagArray™ detector readout process.

[0047] In addition to their advantageous signal level, the synthetic antiferromagnetically coupled nanoparticles can be saturated in different applied fields. This feature can be exploited for multiplex magnetic separation of cells. For the MagArray™ detector, different kinds of synthetic antiferromagnetically coupled nanoparticles with a series of saturation threshold fields can be used to label the biomolecules from different biological processes, thus achieving multiplex biological analysis such as "multi-color" gene expression analysis. For example, consider a "two-color" gene expression scheme with two types of magnetic particles, one saturating in 100 Oe and the other in 125 Oe. We can then interrogate the MagArray™ detector with a two-test sequence. The first test saturates the first type of magnetic particles and gives a voltage signal V_1 , then the second test saturates both types of magnetic particles and gives a voltage signal V_2 . Both types of particles contribute to the signals measured in the tests. If the numbers of the two types of particles at a given site are N_1 and N_2 , respectively, then the tested voltage signals should be:

$$V_1 = \alpha_{11} \times N_1 + \alpha_{12} \times N_2,$$

$$V_2 = \alpha_{21} \times N_1 + \alpha_{22} \times N_2,$$

[0048] where α_{ij} ($ij=11, 12, 21, \text{ or } 22$) are calibration constants. By solving the above equations, we can quantify both types of particles and the two types of genes (or other biomolecules) tagged to them.