Electronic Supporting Information

Removal of Cells from Body Fluids by Magnetic Separation in Batch and Continuous Mode: Influence of Bead Size, Concentration and Contact Time

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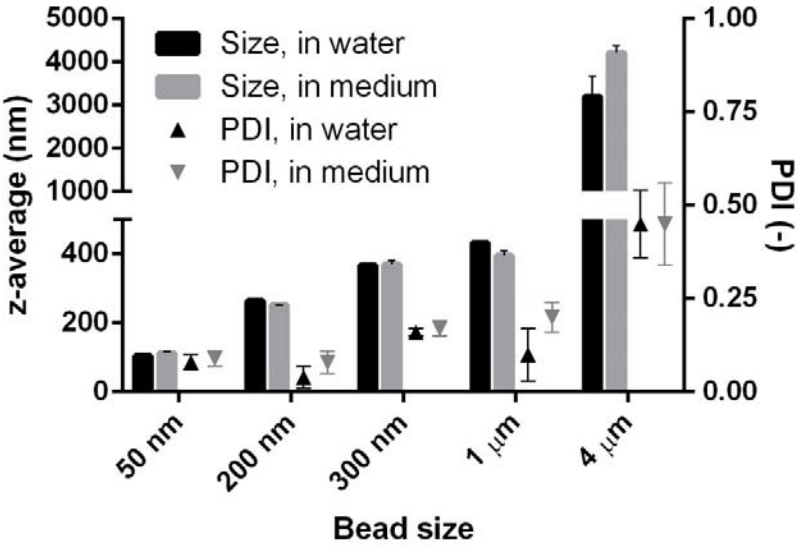

Figure S1: Particle size and polydispersity index (PDI). *Hydrodynamic size of magnetic beads in water and cell culture medium measured by Dynamic Light Scattering (DLS).*

Mathematical Model

Closed form solution of Equations (1-5)

In order to find the closed form solution of the equations, we can proceed as follows. The equations are first rendered dimensionless. The following dimensionless quantities are defined:

$$\begin{aligned}
&\tau = \frac{2k_bT}{3\eta}\left(R_C + R_{MP}\right)\left(\frac{1}{R_C} + \frac{1}{R_{MP}}\right)N_0 t \\
&\nu = \frac{N}{N_0} \\
&y_i = \frac{C_i}{N_0}
\end{aligned} \tag{1.1}$$

The equations can be written in dimensionless form:

$$\begin{aligned}
&\frac{d\nu}{d\tau} = -\nu \cdot \sum_{i=0}^{M-1}\left(1 - \frac{i}{M}\right)\cdot y_i \\
&\frac{dy_0}{d\tau} = -\nu \cdot y_0 \\
&\frac{dy_i}{d\tau} = \left(1 - \frac{i-1}{M}\right)\cdot \nu \cdot y_{i-1} - \left(1 - \frac{i}{M}\right)\cdot \nu \cdot y_i \\
&\frac{dy_M}{d\tau} = \frac{1}{M}\cdot \nu \cdot y_{M-1}
\end{aligned} \tag{1.2}$$

The initial conditions are:

$$\begin{aligned}
&y_0(0) = \frac{C_T}{N_0} = r \\
&y_i(0) = 0 \text{ for } 1 \le i \le M \\
&\nu(0) = 1
\end{aligned} \tag{1.3}$$

Using the conservation of particles and cells, we can rewrite the first equation as follows:

$$\frac{d\nu}{d\tau} = -\nu \cdot \left(r - \frac{1}{M}(1-\nu)\right) \tag{1.4}$$

This equation can be solved exactly, and the solution reads:

$$\nu = \frac{\left(r - \frac{1}{M}\right)\exp\left(\left(\frac{1}{M} - r\right)\tau\right)}{r - \frac{1}{M}\exp\left(\left(\frac{1}{M} - r\right)\tau\right)} \tag{1.5}$$

From this, the concentration of y_0 as a function of time can be obtained:

$$\frac{dy_0}{d\tau} = -\frac{\left(r-\frac{1}{M}\right)\exp\left(\left(\frac{1}{M}-r\right)\tau\right)}{r-\frac{1}{M}\exp\left(\left(\frac{1}{M}-r\right)\tau\right)}\cdot y_0 \Rightarrow$$

$$y_0 = r\cdot\left(\frac{r-\frac{1}{M}\exp\left(\left(\frac{1}{M}-r\right)\tau\right)}{r-\frac{1}{M}}\right)^{-M} \tag{1.6}$$

To integrate the other equations, we start taking the ratio of all cell balance equations with the one for y_0. In this manner, the concentration of particles disappears. We have:

$$\begin{aligned} \frac{dy_1}{dy_0} &= -1+\left(1-\frac{1}{M}\right)\cdot\frac{y_1}{y_0} \\ \frac{dy_i}{dy_0} &= -\left(1-\frac{i-1}{M}\right)\cdot\frac{y_{i-1}}{y_0}+\left(1-\frac{i}{M}\right)\cdot\frac{y_i}{y_0} \\ \frac{dy_M}{d\tau} &= -\frac{1}{M}\cdot\frac{y_{M-1}}{y_0} \end{aligned} \tag{1.7}$$

The solution can be written in the following general form:

$$y_i = \binom{M}{i} y_0^{1-\frac{i}{M}}\left(r^{\frac{1}{M}}-y_0^{\frac{1}{M}}\right)^i = \binom{M}{i} y_0\left(\left(\frac{r}{y_0}\right)^{\frac{1}{M}}-1\right)^i \tag{1.8}$$

One can easily verify that this solution satisfies the mass balance on all cells:

$$\sum_{i=0}^{M} y_i = \sum_{i=0}^{M}\binom{M}{i} y_0^{\frac{M-i}{M}}\left(r^{\frac{1}{M}}-y_0^{\frac{1}{M}}\right)^i = r \tag{1.9}$$

The average number of particles per cell can be obtained as follows:

$$\frac{d}{dr}\sum_{i=0}^{M} y_i = \sum_{i=0}^{M}\frac{i}{M}\binom{M}{i} y_0^{\frac{M-i}{M}}\left(r^{\frac{1}{M}}-y_0^{\frac{1}{M}}\right)^{i-1} r^{\frac{1}{M}-1} = 1 \Rightarrow$$

$$\sum_{i=0}^{M} i\cdot y_i = \sum_{i=0}^{M} i\binom{M}{i} y_0^{\frac{M-i}{M}}\left(r^{\frac{1}{M}}-y_0^{\frac{1}{M}}\right)^i = M\cdot r^{1-\frac{1}{M}}\left(r^{\frac{1}{M}}-y_0^{\frac{1}{M}}\right) \Rightarrow \tag{1.10}$$

$$\langle i\rangle = \frac{\sum_{i=0}^{M} i\cdot y_i}{\sum_{i=0}^{M} y_i} = M\cdot\left(1-\left(\frac{y_0}{r}\right)^{\frac{1}{M}}\right)$$

Finally, the final form of the solution is obtained by substituting Equation (1.6) into Equation (1.8)

$$y_i = \binom{M}{i} r \cdot \left(\frac{r - \frac{1}{M}\exp\left(\left(\frac{1}{M} - r\right)\tau\right)}{r - \frac{1}{M}} \right)^{-M} \left(\frac{r - \frac{1}{M}\exp\left(\left(\frac{1}{M} - r\right)\tau\right)}{r - \frac{1}{M}} - 1 \right)^{i} \tag{1.11}$$

Solution of Equations (7-9)

For the solution of Equations (7-9), we proceed in a similar manner. The following dimensionless quantities are defined:

$$\begin{aligned}
&\tau = \frac{2k_b T}{3\eta}\left(R_{1,C} + R_{MP}\right)\left(\frac{1}{R_{1,C}} + \frac{1}{R_{MP}}\right) N_0 t \\
&\nu = \frac{N}{N_0},\, y_i = \frac{C_{1,i}}{N_0},\, z_i = \frac{C_{2,i}}{N_0} \\
&\alpha = \frac{\left(R_{2,C} + R_{MP}\right)\left(\frac{1}{R_{2,C}} + \frac{1}{R_{MP}}\right)}{W\left(R_{1,C} + R_{MP}\right)\left(\frac{1}{R_{1,C}} + \frac{1}{R_{MP}}\right)}
\end{aligned} \tag{1.12}$$

The equations can be written in dimensionless form:

$$\begin{aligned}
&\frac{d\nu}{d\tau} = -\nu \cdot \left(\sum_{i=0}^{M_1-1}\left(1 - \frac{i}{M_1}\right)\cdot y_i + \alpha \sum_{i=0}^{M_2-1}\left(1 - \frac{i}{M_2}\right)\cdot z_i \right) \\
&\frac{dy_0}{d\tau} = -\nu \cdot y_0 \\
&\frac{dy_i}{d\tau} = \left(1 - \frac{i-1}{M_1}\right)\cdot \nu \cdot y_{i-1} - \left(1 - \frac{i}{M_1}\right)\cdot \nu \cdot y_i \\
&\frac{dy_{M_1}}{d\tau} = \frac{1}{M_1}\cdot \nu \cdot y_{M-1} \\
&\frac{dz_0}{d\tau} = -\alpha \cdot \nu \cdot z_0 \\
&\frac{dz_i}{d\tau} = \alpha \cdot \left(1 - \frac{i-1}{M_2}\right)\cdot \nu \cdot z_{i-1} - \alpha \cdot \left(1 - \frac{i}{M_2}\right)\cdot \nu \cdot z_i \\
&\frac{dz_{M_2}}{d\tau} = \frac{\alpha}{M_2}\cdot \nu \cdot z_{M-1}
\end{aligned} \tag{1.13}$$

The initial conditions are:

$$
\begin{aligned}
& y_0(0) = r_1 \\
& y_i(0) = 0 \text{ for } 1 \le i \le M_1 \\
& z_0(0) = r_2 \\
& z_i(0) = 0 \text{ for } 1 \le i \le M_2 \\
& \nu(0) = 1
\end{aligned}
\tag{1.14}
$$

The magnetic particles concentration conditions can be written as:

$$\sum_{i=1}^{M_1} i \cdot y_i + \sum_{i=1}^{M_2} i \cdot z_i + \nu = 1 \tag{1.15}$$

The solution of the cell populations equations in terms of y_0 and z_0, respectively, is:

$$
\begin{aligned}
y_i &= \binom{M_1}{i} y_0^{1-\frac{i}{M_1}} \left(r_1^{\frac{1}{M_1}} - y_0^{\frac{1}{M_1}} \right)^i \\
z_i &= \binom{M_2}{i} z_0^{1-\frac{i}{M_2}} \left(r_2^{\frac{1}{M_2}} - z_0^{\frac{1}{M_2}} \right)^i
\end{aligned}
\tag{1.16}
$$

The balance equation of particles can be reformulated as:

$$\frac{d\nu}{d\tau} = -\nu \cdot \left(r_1 + \alpha \cdot r_2 - r_1^{1-\frac{1}{M_1}} \left(r_1^{\frac{1}{M_1}} - y_0^{\frac{1}{M_1}} \right) - \alpha \cdot r_2^{1-\frac{1}{M_2}} \left(r_2^{\frac{1}{M_2}} - z_0^{\frac{1}{M_2}} \right) \right) \tag{1.17}$$

A relationship between y_0 and z_0 can be easily obtained:

$$\frac{dz_0}{dy_0} = \alpha \cdot \frac{z_0}{y_0} \Rightarrow \frac{z_0}{r_2} = \left(\frac{y_0}{r_1} \right)^{\alpha} \tag{1.18}$$

Then, the equation for the particle concentration will be integrated as a function of y_0:

$$
\begin{aligned}
& \frac{d\nu}{dy_0} = \left(\frac{r_1}{y_0} + \alpha \cdot \frac{r_2}{y_0} - \frac{r_1^{1-\frac{1}{M_1}}}{y_0} \left(r_1^{\frac{1}{M_1}} - y_0^{\frac{1}{M_1}} \right) - \frac{\alpha \cdot r_2^{1-\frac{1}{M_2}}}{y_0} \left(r_2^{\frac{1}{M_2}} - r_2^{\frac{1}{M_2}} \left(\frac{y_0}{r_1} \right)^{\frac{\alpha}{M_2}} \right) \right) \Rightarrow \\
& \nu = 1 + M_1 \cdot r_1 \left(\left(\frac{y_0}{r_1} \right)^{\frac{1}{M_1}} - 1 \right) + M_2 \cdot r_2 \left(\left(\frac{y_0}{r_1} \right)^{\frac{\alpha}{M_2}} - 1 \right)
\end{aligned}
\tag{1.19}
$$

The only equation to be solved (numerically) is the following one, providing the concentration of cells without bound particles, as a function of time:

$$\frac{dy_0}{d\tau} = -\nu \cdot y_0 \Rightarrow$$
$$\frac{dy_0}{d\tau} = -y_0 \cdot \left(1 + M_1 \cdot r_1 \left(\left(\frac{y_0}{r_1} \right)^{\frac{1}{M_1}} - 1 \right) + M_2 \cdot r_2 \left(\left(\frac{y_0}{r_1} \right)^{\frac{\alpha}{M_2}} - 1 \right) \right) \tag{1.20}$$

This last equation can be solved numerically.

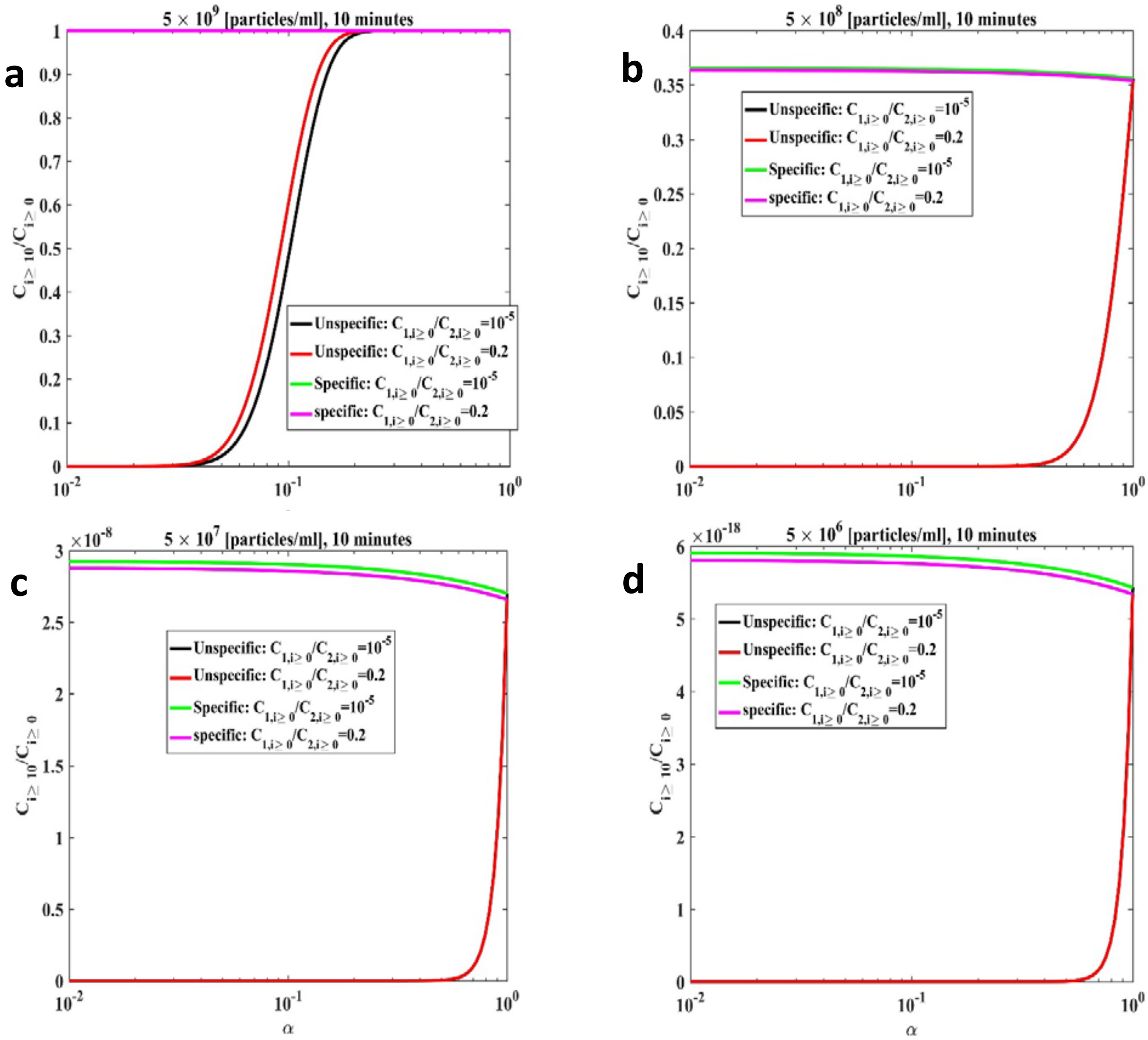

Figure S2: Mathematical modelling results for magnetic beads with a size of 300 nm. *Unspecific binding was included through the parameter α. Two different ratios of specific versus unspecific cells were investigated: 0.2 and 10^{-5}. Total contact time: 10 minutes. Change in the fraction of cells with at least 10 magnetic particles as a function of α, for both specific and unspecific cells. Four particles number concentrations: (a) 5 x 10^9 beads per mL. (b) 5 x 10^8 beads per mL. (c) 5 x 10^7 beads per mL. (d) 5 x 10^6 beads per mL.*

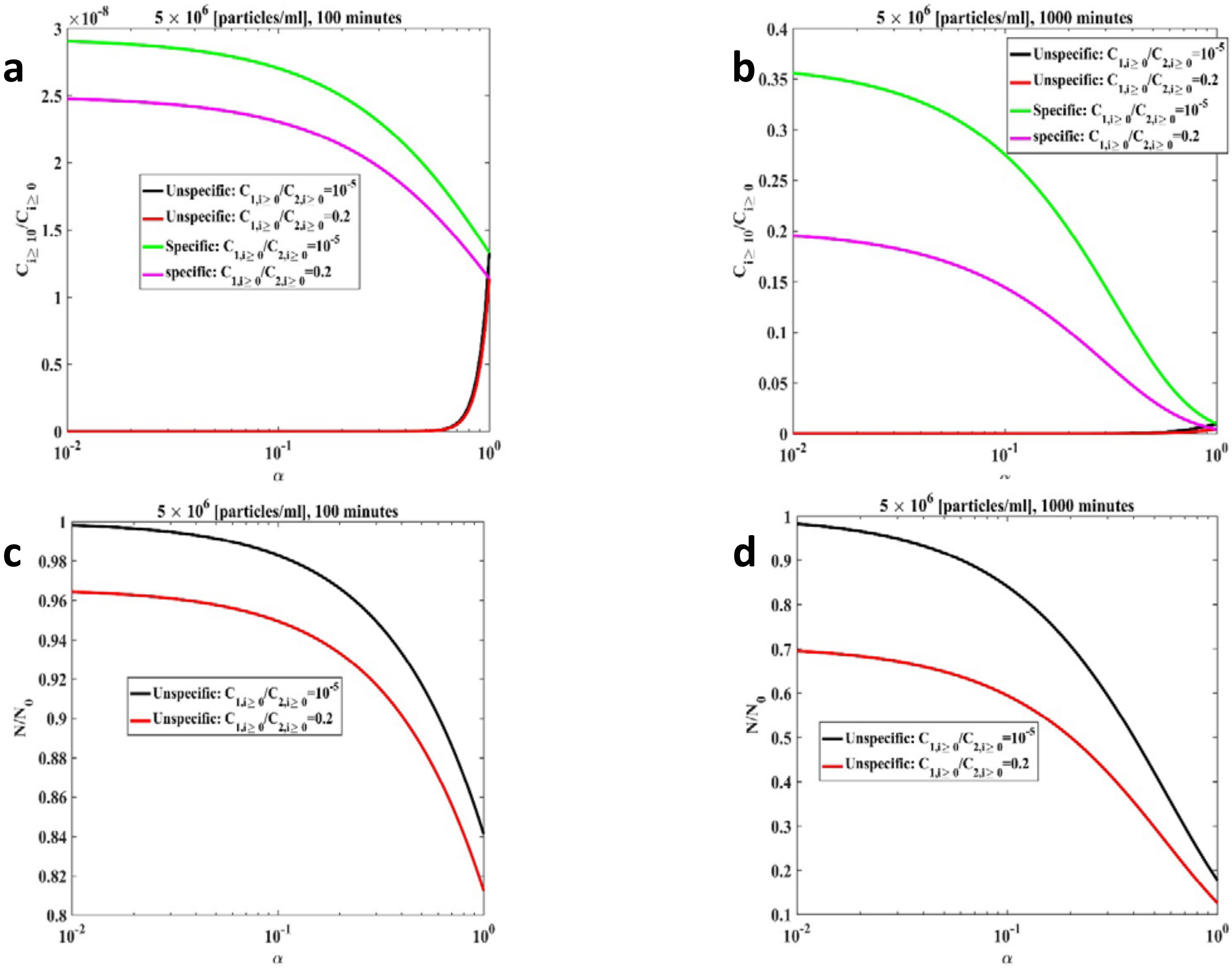

Figure S3: Mathematical modelling results for magnetic beads with a size of 300 nm. *Unspecific binding was included through the parameter α. Two different ratios of specific versus unspecific cells were investigated: 0.2 and 10^{-5}. Particles number concentration 5 x 10^{6} beads per mL. (a) Change in the fraction of cells with at least 10 magnetic particles as a function of α, for both specific and unspecific cells, contact time 100 minutes. (b) Change in the fraction of cells with at least 10 magnetic particles as a function of α, for both specific and unspecific cells, contact time 1000 minutes. (c) Change in the concentration of magnetic particles as a function of α, for both specific and unspecific cells, contact time 100 minutes. (d) Change in the concentration of magnetic particles as a function of α, for both specific and unspecific cells, contact time 1000 minutes.*