

Supporting Information on “Surface Property Induced Morphological Alterations of Human Erythrocyte”

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Contents

- Microscopic image of a dried blood droplet
- Detailed calculations regarding prediction of cluster sizes.
- FESEM images of protein coated samples
- Microscopic images affirming halting of deformations on BSA coated substrates

1. MICROSCOPIC IMAGE OF A DRIED BLOOD DROPLET

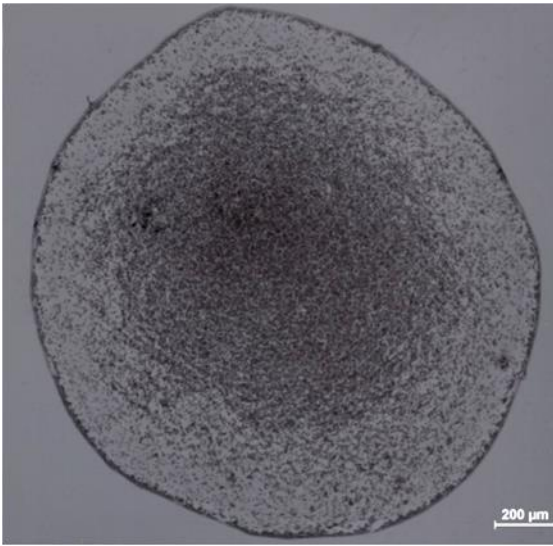


Figure S1: Dried blood droplet ($C = 0.01$) on substrate S1 (glass) showcasing the clusters in droplet center. **Scale bar:** 200 μm .

Dense RBC clusters are visible at the droplet center, whereas the distribution becomes relatively sparse as we move away from the center. However, near the peripheral rim, the concentration of RBC increase. This unique distribution is in complete contrast to the more popular distribution known as ‘*Coffee ring effect*’ and the details of this reversal in coffee ring effect will be dealt in the ensuing article.

2. DETAILED CALCULATIONS REGARDING PREDICTION OF CLUSTER SIZES.

From Smoluchowski equation, the coagulation rate of colloids for rapid coagulation is given by the following equation:

$$\frac{d\rho}{dt} = -16\pi D_p a \rho^2 \dots\dots\dots(1)$$

where,

ρ = Number of cells per unit volume

a = Radius of particle = 2.78×10^{-6} m

D_p = Diffusivity = 7.9×10^{-14} m²/s

Integration of equation (1) yields the following working form:

$$1 - \frac{\rho(t)}{\rho_0} = 1 - \frac{1}{1 + \frac{t}{\tau}} \dots\dots\dots(2)$$

where,

$\rho_{(t)}$ = number concentration of free cells left at time t

ρ_0 = number concentration of free cells left at time t=0

t = time required for drying

$$\tau = (16\pi D_p a \rho_0)^{-1} = \text{time constant for rapid coagulation} \dots \dots \dots (3)$$

So, total number of cells in the cluster is given by:

$$n_c(T) = \rho_{0V_L} \left(\frac{t}{1+\tau} \right) \dots \dots \dots (4)$$

where,

V_L = Initial drop volume = 1 μl = 10^{-9} m^3

The blood sample originally had **4×10^6 cells/ μl** .

For a concentration of 0.002% i.e. when rbc:buffer = 1:500,

The total number of cells after dilution comes down to **8×10^{12} cells/ m^3** ($=\rho_0$)

From equation (3), $\tau = 1.13 \times 10^4 \text{ s}^{-1}$

Upon substitution of t = (12 \times 60) s in equation (4), the total number of cells in the cluster ($n_c(T)$) comes out to be = 4.79×10^2

Now, mean projected area of a single RBC ($A_{cell}^{(1)}$) = $\pi \times a \times b$, where a = 7.0 μm and b = 3 μm

Hence, total surface area occupied by all the cells in the cluster = $3.16 \times 10^4 \mu\text{m}^2$

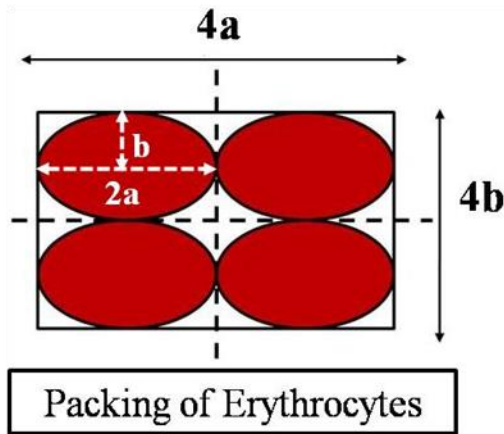


Figure S2 (a): Cells arranged in a square pitch

To take into account the void area, the cluster was assumed to elliptical cells arranged in square pitch.

Accordingly, the area fraction parameter (ϵ_A) = $\frac{\text{area of ellipse}}{\text{area of rectangular envelope}} = 0.78$

Hence, the occupied area by the cluster, which is same as that of the rectangular envelope becomes $4.05 \times 10^4 \mu\text{m}^2$

The calculated area is of the same order as obtained from the experiments.

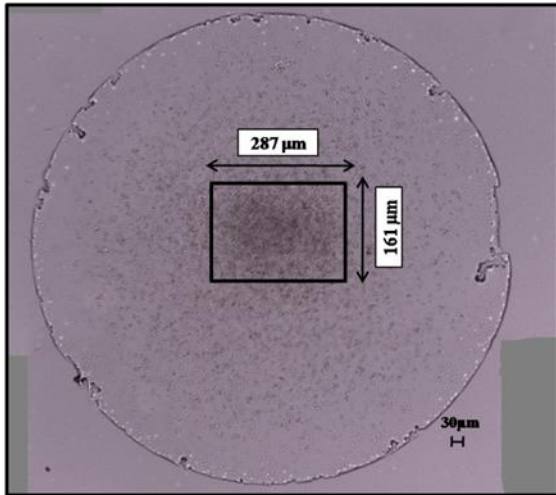


Figure S2 (b): A dried blood droplet of concentration 0.002

Area of the clustered region from experiments= $4.62 \times 10^4 \mu\text{m}^2$

The above area has been calculated by ImageJ software and the results obtained were comparable to the theoretically predicted values.

The same calculations were repeated for a different dilution of 0.02%. The results obtained both experimentally and theoretically were once again of the same order as shown below.

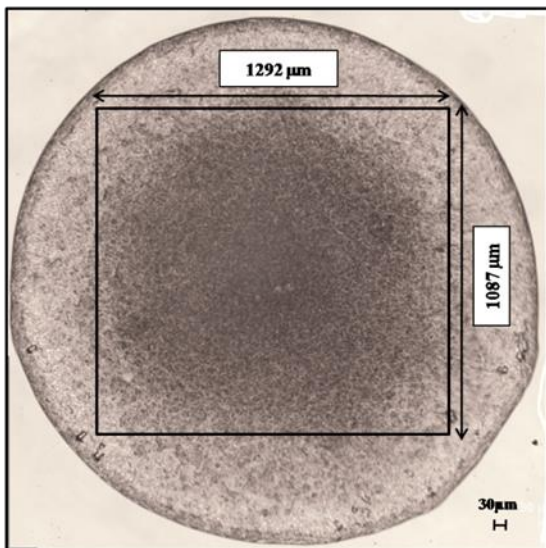


Figure S2 (c): A dried blood droplet of concentration 0.02

Theoretical area of the clustered region = $2.63 \times 10^6 \mu\text{m}^2$

Area obtained from the images (experimentally) = $1.40 \times 10^6 \mu\text{m}^2$

3. FESEM IMAGES OF PROTEIN COATED SAMPLES

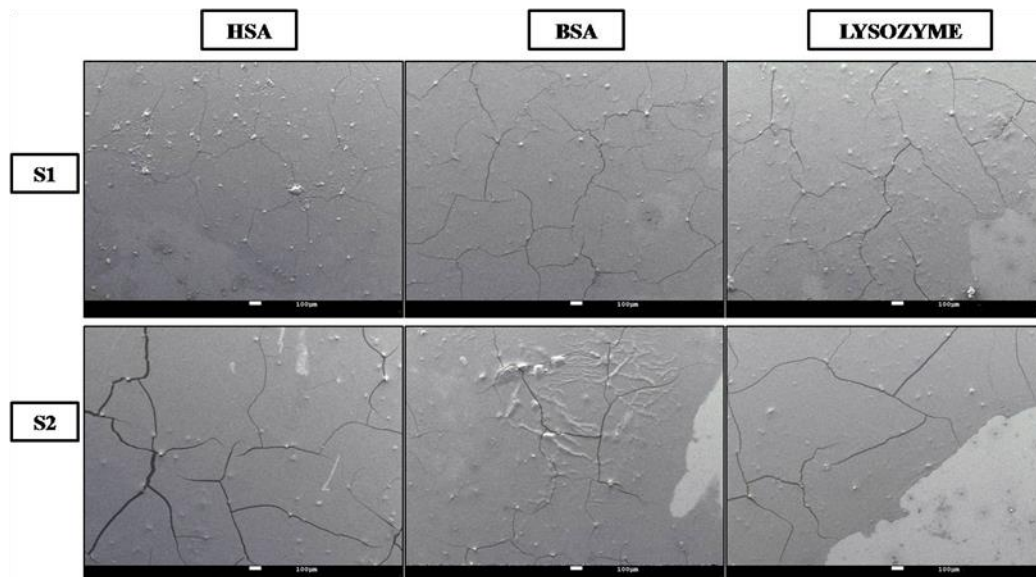


Figure S3: FESEM images of the protein covered S1 and S2 substrates . **Scale Bar:** 100 μm .

A section of the substrate S2 was covered with parafilm prior to dipping in the lysozyme solution. The part covered with parafilm showed no crack formation in the FESEM image (S2+ LYSOZYME), whereas the uncovered part showed distinct cracks. This ascertained the formation of the protein layer on the substrates. **Scale Bar:** 100 μm .

4. MICROSCOPIC IMAGES AFFIRMING HALTING OF DEFORMATIONS ON BSA COATED SUBSTRATES

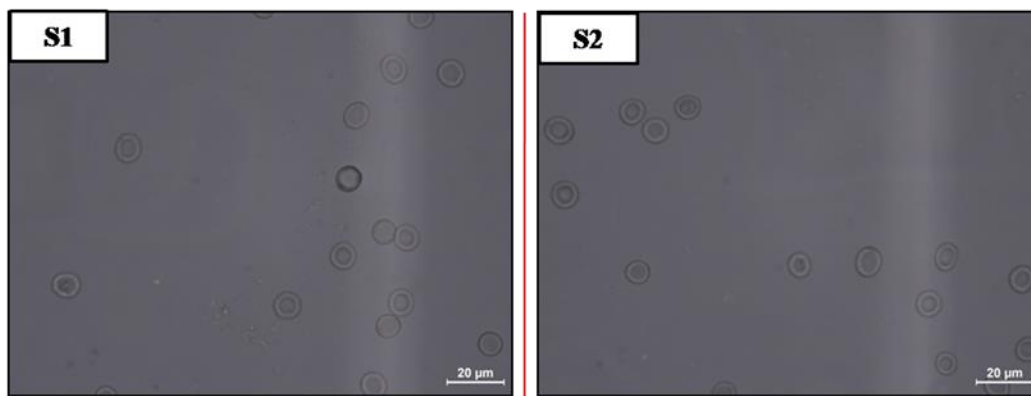


Figure S4: Microscopic images showcasing the effect of BSA on erythrocyte deformation on S1 and S2 substrates.

These microscopic images ascertain the fact that the erythrocytes retained their discocytic state when the substrates are coated with 1 mM BSA solution, similar to HSA and Lysozyme.