A Postulate Describing
Cellular Electromechanical Breakdown
by Electroporation

When blood cells, suspended in normal saline, are subjected to a single 60µS electric pulse, the after effect on the cells depends on electric field strength applied. Figure 1 – 5 provide five postulated illustrations (based on Oliver's experimental results) that describes the fate of the cell after it has received electrical treatment, ranging from less than 2 kV/cm to 3 kV/cm.

Although the experimental work was on red blood cells, the electrical breakdown processes occur for all cell types. But the combination of pulse duration and electric field strength that leads to critical membrane electroporation (CME), must be established for each individual person, treatment medication, cell type, cell condition and cell size.

Stages of Cellular Electromechanical Breakdown

1. **Less than 2 kV/cm:** An initial electromechanical breakdown of the membrane produces pore defects with water taken up. The membrane reseals the pore to produce an intact cell (***reversible electric breakdown***).

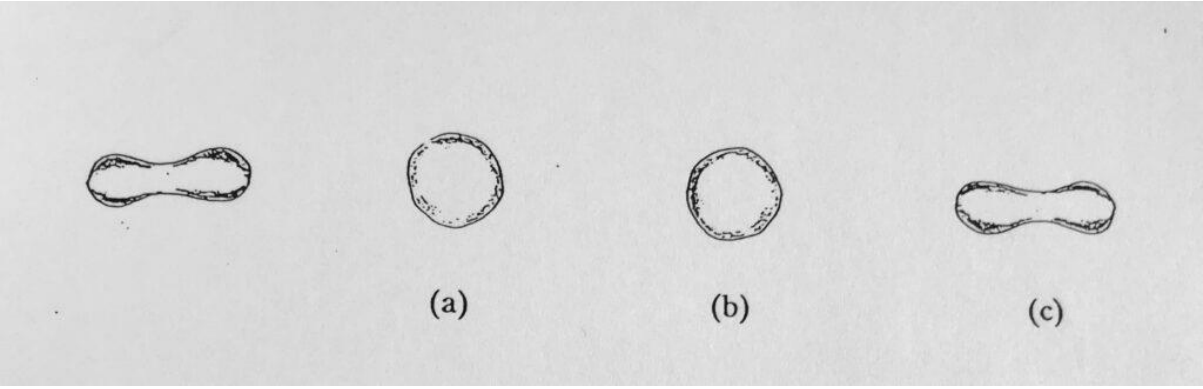

Figure 1. The initial electromechanical breakdown of the erythrocyte membrane at the membrane voltage (2 kV/cm) produces (a) pore defects, (b) water uptake and (c) a resealing of the membrane to form an intact cell.

2. When voltage is increased beyond this first voltage causing membrane breakdown, metabolite imbalance occurs. Water is expelled out of the cytosol via the pore defect and the cell shrinks (crenates)**.** Eventually, the cell re-seals the membrane, activates the $Na^+ K^+$ ATPase pump so that the cell eventually returns to a viable state. But the pump system seemingly over-compensates the ion and water balance before returning to its normal state again (***reversible electric breakdown***).

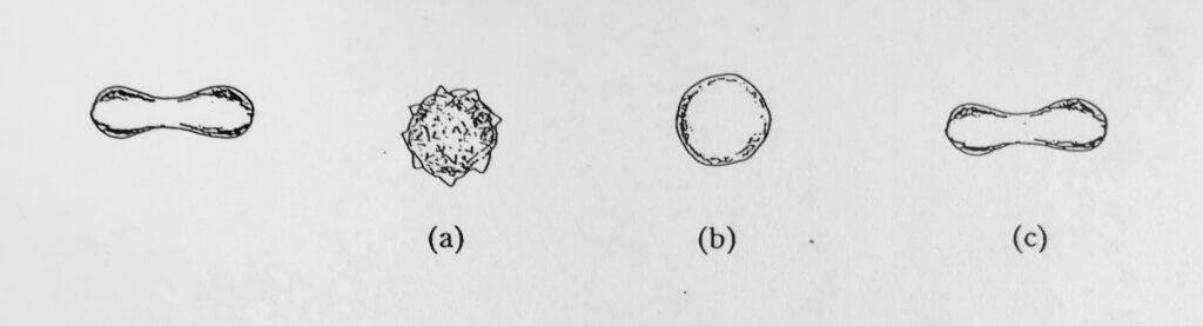

Figure 2. For a pulse with a voltage beyond the membrane breakdown level, (a) metabolite imbalance occurred with cell shrinkage, (b) the membrane eventually re-sealed and the uptake of water swelled the cell slightly more than normal size and (c) further water transfer across the membrane returns the cell to its quasi-normal state.

3. When the voltage of the pulse is sufficient to cause a defect in the membrane/cytoskeleton structure, the membrane fails to adequately repair and the uptake of water into the cell causes swelling and eventually haemolysis (haemoglobin leakage from the cell). Some stronger or young smaller cells could recover, others do not.

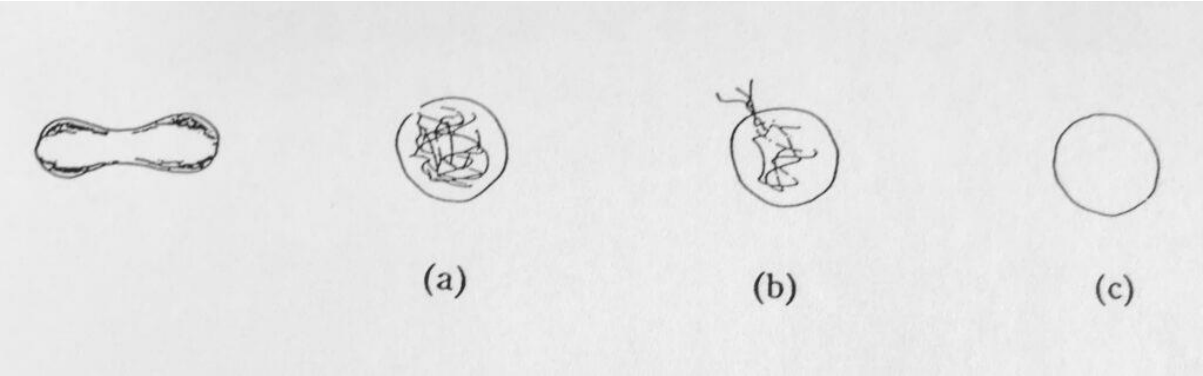

Figure 3. For a pulse voltage sufficient to cause a defect in the membrane/cytoskeleton structure, complete repair failed to occur in some of the cell population before (a) the uptake of water caused the cells to swell, (b) the haemoglobin was released from the cytosol and (c) some ghost cells were forced.

4. **Approximately 3 kV/cm:** For a voltage sufficient to cause major membrane rupture, immediate uptake of water follows. In a very large percentage of the cell population, the cells rapidly swell from increased internal water pressure until the membrane/cytoskeleton is stretched to the point of irreparable damage and lysis occurs. The electrical parameters for this irreversible electroporation (CME) process are described as ***critical cell lysis voltage*** **and** ***critical cell lysis pulse length*****.**

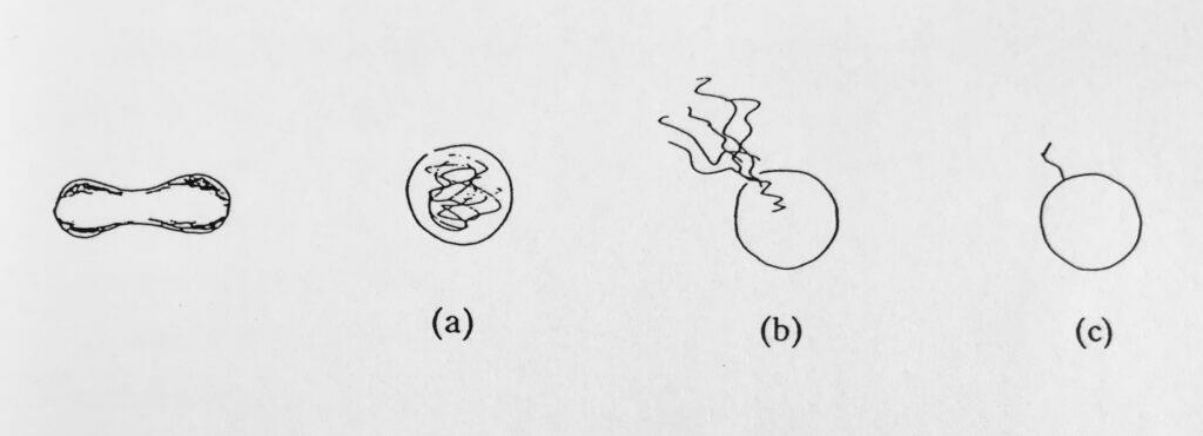

Figure 4. In the case when the pulse voltage was sufficient to cause a major rupture to a critical site within the membrane/cytoskeleton structure (approximately 3 kV/cm), (a) immediate uptake of water occurred in a large percentage of the cell population, (b) rapid lysis followed and (c) some cells showed fragmented 'tails' in the ghost cells.

5. **Greater than 3 kV/cm:** For further increased voltage exceeding critical lysis, critical pore defects are excessive in size and more violent osmotic ruptures involving the cytoskeleton, occur. The membrane violently ruptures. The most likely site is where the lipid/protein attaches to the cytoskeleton matrix;

A significant percentage of the cells become ghost cells or fragments with signs of denatured haemoglobin.

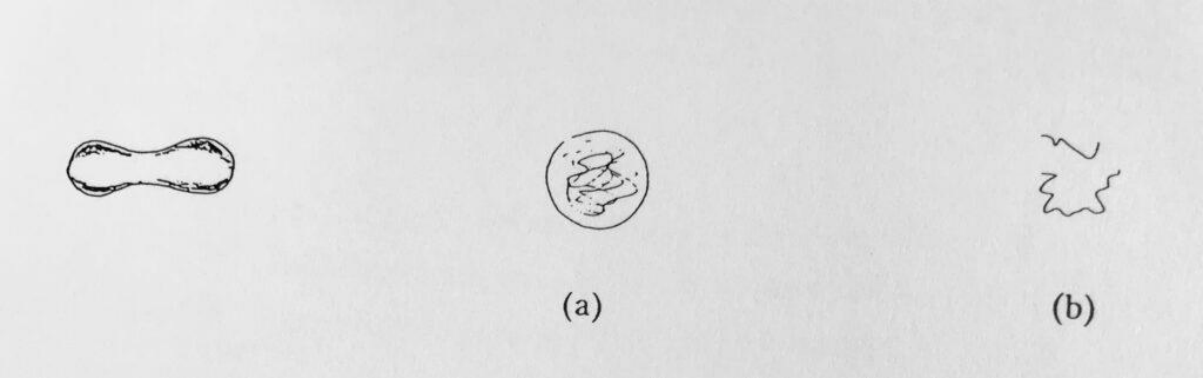

Figure 5. For a pulse voltage of excessive magnitude, (a) larger defects caused more violent osmotic rupture of the membrane/cytoskeleton and (b) cell fragmentation led to the loss of cells from intact cell and ghost cell population suspended in the media.