Cell-Penetrating Peptides, Electroporation, and Drug Delivery

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Short Abstract **— Certain short polycations, such as TAT and oligoarginine, rapidly pass through the plasma membranes of mammalian cells by a mechanism called transduction, as well as by endocytosis and macropinocytosis. These cell-penetrating peptides (CPPs) can carry with them cargos of 30 amino acids, more than the nominal limit of 500 Da and enough to be therapeutic. An analysis of the electrostatics of a charge outside the cell membrane and some recent experiments suggest that transduction may proceed by molecular electroporation. Ways to target diseased cells, rather than all cells, are discussed.**

Keywords **— Cell-penetrating paptides, transduction, molecular electroporation.**

We could cure cancer if we knew how to deliver a drug intact to the cytosol of every cancer cell, sparing healthy cells. The circulatory system can deliver a drug to every cell in the body, and certain chemical tricks protect drugs from peptidases and nucleases. But it's harder to cope with antibodies, spare healthy tissues, and get drugs past the plasma membrane, which blocks or endocytoses molecules in excess of 500 Da [1]. This work is about cell-penetrating peptides and other cations that can overcome the 500-Da restriction barrier and about tricks that may spare healthy cells.

In 1988, two groups [2, 3] working on HIV reported that the trans-activating transcriptional activator (TAT) of HIV-1 can cross cell membranes. The engine driving this 86-aa cell-penetrating peptide (CPP) is its residues 48–57 which carry a charge of $+8e$. Other CPPS were soon found. Antp is residues 43–58 of Antennapedia, a homeodomain of the fly; it carries a charge of $+7e$. R^n carries charge $+ne$. These and other polycations can penetrate the plasma membranes of live cells towing cargos that greatly exceed the 500 Da restriction barrier.

TAT carries cargos across cell membranes with high efficiency by at least two functionally distinct mechanisms according to whether the cargo is big or small [4]. Big cargos, such as proteins or quantum dots, enter via caveolae endocytosis and macropinocytosis [5], but relatively few escape the cytoplasmic vesicles in which they then are trapped [4]. Small cargos, such as peptides of 30 amino acids, enter both slowly by endocytosis and rapidly by an unknown mechanism, called transduction, that uses the membrane potential [4, 6]. Peptides fused to TAT enter cells within seconds [7].

I review therapeutic applications of CPPs and basic facts about plasma membranes and then describe a model [8, 9] of oligoarginine transduction as molecular electroporation. The model is supported by analytic work on the electrostatics of the bilayer, by Monte Carlo simulations of counterions, and by experiments [10]. I sketch a broader class of cell-penetrating molecules and suggest ways to target cancer cells.

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