

1.3.2 Ligand-Targeted Therapeutics: Types of Ligand; Advantages and Limitations

Researchers have widely embraced the concept of targeted delivery and a scientific database search on 'receptor targeted delivery' showed 862 studies (to 05/02/06), with 149 studies reported in 2005.

Ligand-targeted therapeutics refers to therapies directed against a receptor or epitope that is over-expressed in the diseased cell/tissue. It was the aim of this project to design a polyplex able to use RME to increase the internalisation of the polyplex and thereby increase the expression of a reporter gene. The targeting of cancer cells is complex as these cells are derived from normal cells but they are no longer growth regulated and can also evade attack by the immune system (Vander et al., 1994). Receptors that are being targeted in cancer therapy come from a diverse range having many different functions. Table 1.3 gives an overview of the receptors being targeted in non-viral gene therapy. Ideally a receptor or antigen is sought that is expressed only on tumour cells i.e. it is not present on cells elsewhere in the body. This is rarely, if ever, the case but cancer cells frequently over-express plasma membrane localised receptors due to their increased nutrient requirements for growth, invasion and metastasis (Prodi et al., 1998). Receptor over-expression can be used to differentiate tumour cells from normal cells and thus they provide a target for delivery systems. Although non-cancerous cells expressing the same receptors will take up targeted vector by receptor-mediated endocytosis, this uptake is expected to be less compared with uptake in cancerous cells over-expressing the receptor. The characteristics of each class of ligand are discussed below (Sections 1.3.2.1-1.3.2.4). The mechanisms involved with these receptors are often not truly characterised i.e. their endocytic pathway, rate of endocytosis. The mechanisms may also differ when the therapeutic is coupled to the ligand.

Ligands have been conjugated with the therapeutic e.g. fusion protein of urokinase and diphtheria toxin (Ramage et al., 2003), the carrier of the drug/imaging agent e.g. PK2 or form the therapeutic themselves e.g. Trastuzumab (Table 1.1). More discussion of uPAR as a target is developed in Section 1.4. The following parts of this section discuss the different targeting ligands being utilised and their advantages/disadvantages.

Table 1.3 – Multi-stage targeting methods

Mechanism of Targeting	Example	Reference
Temperature (increased in tumour and inflammation or applied locally)	Poly(N-isopropylacrylamide-coacrylamide), elastin like polypeptide	(Reviewed in Chilkoti et al., 2002) (Meyer et al., 2001)
Ultrasound (applied externally)	High intensity focused ultrasound increased β -galactosidase delivery	(Huber et al., 2003)
Magnetism (applied externally)	Superparamagnetic particles formed with PEI/DNA	(Reviewed in Plank et al., 2003)
*Antibody directed enzyme prodrug therapy (ADEPT)	fAb against CEA targets carboxypeptidase G2 which catalyses cleavage of a carbamate bond to activate a di-iodophenol mustard drug	(Francis et al., 2002, Monks et al., 2000)
Lectin-directed enzyme-activated prodrug therapy (LEAPT)	Galactose targeted α -L-rhamnosidase followed by rhamnose-doxorubicin activation	(Robinson et al., 2004)
Gene directed enzyme prodrug therapy (GDEPT)	Gene encoding for a non-native enzyme is delivered and expressed in the tumour, a prodrug is administered and activated in the tumour	(Reviewed in Denny, 2001)
Virus directed enzyme prodrug therapy (VDEPT)	A more specific case of GDEPT: nitroimidazole reductase gene delivered to tumour cells by adenoviral vector activates CB1954	(Chung-Faye et al., 2001)
Polymer directed enzyme prodrug therapy (PDEPT)	EPR accumulation of HPMa- β -lactamase followed by EPR accumulation of HPMa-copolymer-methacryloyl-glycine-glycine-cephalosporin-doxorubicin	(Satchi-Fainaro et al., 2003)
Protein targeted enzyme prodrug	VEGF fused to carboxypeptidase G2 to activate prodrug	(Spooner et al., 2003)

* - in phase I clinical trials, fAb – antibody fragment, CEA – carcinoembryonic antigen, CB1954 - 5-(aziridin-1-yl)-2,4-dinitrobenzamide