DEVELOPING IMMUNOTHERAPIES FOR CHILDHOOD CANCER

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INTRODUCTION

The development of immune-based treatment (immunotherapy) for childhood cancer is a rapidly advancing field with impressive results already achieved in children with leukaemia.[1 2] For cancers resistant to conventional treatments, harnessing the power and specificity of the immune system to fight cancer is an attractive alternative approach. The immune system is essential for controlling cancer progression by continual surveillance and elimination of transformed cells. This protective process is hindered by the ability of cancer cells to develop mechanisms enabling them to 'hide' from immune destruction (including down-regulation of tumour-associated antigens and major histocompatibility complex (MHC) class I, and the creation of an immunosuppressive tumour microenvironment). The aims of cancer immunotherapy are to enhance existing anti-tumour immune responses (active immunotherapy), including cancer vaccines and immune check-point inhibitors, or to enable the immune system to specifically recognise and kill cancer cells (passive immunotherapy) (Table 1).

Passive immunotherapy	Active immunotherapy
Monoclonal antibodies	Cancer vaccines
Bispecific antibodies	Immune check-point inhibitors
CAR T-cells	
TCR-redirected T-cells	
Tumour infiltrating lymphocytes	
Viral reactive T-cells	
Donor lymphocytes	

Table 1. Classification of immune-based therapies for childhood cancer. Abbreviations: CAR; chimeric antigen receptor, TCR; T-cell receptor.

The identification of targetable tumour antigens is fundamental to the development of successful ‘passive’ immunotherapies. Ideally targets should be highly expressed on cancer cells with little or no expression on normal tissue in order to avoid the potential for ‘on-target, off-tumour’ toxicities. B-lymphocyte antigen CD19 and disialoganglioside GD2 have been selected as suitable antigens for paediatric leukaemia and neuroblastoma immunotherapy clinical trials, respectively.[1] [3] However, neither of these targets are 100% perfect as CD19-directed therapy causes depletion of healthy B-cells, and GD2 is expressed at low level on normal peripheral nerves.

This article gives a brief overview of the main types of immunotherapy currently under development (Table 2), and addresses some of the main caveats surrounding translation to clinical practice.

Immunotherapy	Description	Advantages	Limitations
Monoclonal antibodies (MAb)	Bind tumour antigen and augment antibody-dependent cell-mediated cytotoxicity (ADCC) e.g. anti-GD2 (*Ch14.18*)[3] and anti-CD20 (*rituximab*)[4]. MAb can also be linked to chemotherapeutics or radionuclides.	'Off the shelf' product Efficacious in Phase III clinical trials	Short half-life, requires repeated administration 'On target, off tumour' side effects Anaphylactoid reactions
Bispecific antibodies	Simultaneously bind tumour antigen and T-cell e.g. *blinatumomab*[5] binds CD19 and CD3	'Off the shelf' product	Short half-life, requires repeated administration Cytokine release syndrome Potential for 'antigen escape'
Checkpoint inhibitors	e.g. anti-PD-1 or anti-CTLA-4 (*Ipilimumab*) antibodies block inhibitory immune signals	'Off the shelf' product	Short half-life, requires repeated administration Cytokine release syndrome
Tumour vaccines	Most commonly *Ex vivo* production of autologous tumour antigen pulsed dendritic cells for injection	Generation of immunological memory	Patient specific therefore expensive to produce and requires gene therapy laboratory
Tumour infiltrating lymphocytes (TILs)	T-cells are extracted from the tumour itself and cultured *ex vivo* to large numbers for infusion	Tumour-specific Immunological memory	Patient specific Not reliably produced from all tumour samples Limited study in paediatric patients
Viral reactive T-cells	T-cells stimulated with viral antigen expressing antigen presenting cells	Immunological memory	Patient specific Small number of cells for infusion
TCR re-directed T cells	Autologous T-cells are genetically modified with tumour antigen specific T-cell receptors	Immunological memory Directly target tumour antigen Can also target intracellular antigens	Patient specific MHC-restricted Risk of mis-pairing with endogenous TCR Cytokine release syndrome
CAR re-directed T cells	Autologous T-cells are genetically modified with Chimeric antigen receptors	Immunological memory Can include 'safety switch' MHC-unrestricted Can be engineered with 'costimulatory' domains to enhance efficacy and persistence	Patient specific Can only target known cell surface antigens Cytokine release syndrome 'On-target, Off tumour' side effects Potential for 'antigen escape'

Table 2. Overview of cancer immunotherapy approaches.

Abbreviations: ALL; Acute Lymphoblastic Leukaemia, CNS; central nervous system, EBV; Epstein-Barr virus, CAR; chimeric antigen receptor, TCR; T-cell receptor, MHC; Major histocompatibility complex.

WHAT TYPES OF CANCER IMMUNOTHERAPY ARE BEING DEVELOPED?

Monoclonal antibodies

Over the last two decades, the development of monoclonal antibodies to treat cancer has yielded some of the greatest successes. Monoclonal antibodies directly targeting tumour antigens have now been incorporated into many standard paediatric treatment protocols[3]. Bispecific antibodies and bispecific T-cell engagers (BiTE®) bind two targets and can therefore simultaneously bind a tumour antigen and cytotoxic T-cell.[5]

Antibodies have also been engineered to block immune check-points. PD-1 and CTLA-4 are examples of inhibitory co-receptors that provide an 'immunological break' to uncontrolled T-cell activation. Monoclonal antibodies that target these checkpoints can augment existing inhibited immune responses to cancer. PD-1 blockade has shown great promise in clinical trials for metastatic melanoma[6] and other adult cancers, and its efficacy is now being tested in paediatric malignancies.

Adoptive cell therapy

Adoptive cell therapy (ACT) is an example of 'personalised medicine' where autologous tumour specific T-cells are manufactured in the laboratory before re-infusion back into the patient. Approaches in children include the culturing and genetic modification of T-cells to promote activation, proliferation and tumour specificity (Figure 1).

Tumour specificity of T-cells from peripheral blood can be achieved by genetic modification with antigen-specific T-cell receptors (TCRs) or chimeric antigen

receptors (CARs). CARs combine an extracellular antibody-derived antigen-binding domain with an intracellular T-cell activation domain (Figure 1). CARs have the additional advantage of being unrestricted by MHC, unlike TCRs.

Clinical trials using CD19-directed CAR T-cells for children with refractory leukaemia have achieved greater than 70% remission rates.[1] A research priority is now to translate expertise to solid tumours and a key challenge will be engineering CAR T-cells that effectively traffic to tumour sites, and form immunological memory.

Naturally occurring tumour-reactive T-cells can also be derived and propagated from tumour tissue itself under special culture conditions (known as tumour infiltrating lymphocytes, TILs), although there has been little clinical experience to date for childhood solid tumours.

Cancer vaccines

An example of active immunotherapy is through vaccination, however clinical trials aimed at inducing anti-tumour immune responses have so far been disappointingly ineffective in children with cancer.

TRANSLATING IMMUNOTHERAPY INTO CLINICAL PRACTICE

The development of novel immunotherapies must include rigorous pre-clinical testing to fully assess any potential harm to patients. Toxicities can be divided into two groups; those related to autoimmunity ('on-target, off tumour effects') and those relating to an increase in circulating cytokines (e.g. leading to cytokine-release syndrome).[7] Learning how to recognise and manage these toxicities will be key following translation to large-scale clinical trials.

The manufacture of a personalised immunotherapeutic is a highly complex and labour-intensive process that is currently restricted to just a few centres in the UK. Hence, currently treatment is limited to a small number of patients within a clinical trial setting. CD19 CAR T-cells however are now being commercialised for much wider application and one exciting development is 'off the shelf' rather than 'patient-

specific' therapies achievable through "genome editing" in which third party donor cells can be silenced for immune attack through deletion of genes such as MHC.

CONCLUSION

There has been a paradigm shift in adult oncology through the developments of cancer immunotherapy. For paediatrics, a major rate-limiting step has been the identification of optimal targetable tumour antigens. Combinational therapies with standard treatments or other immune-based treatments to overcome the immunoinhbitory microenvironment is a current research priority.

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FIGURE LEGENDS

Figure 1 – Production of autologous chimeric antigen receptor (CAR) engineered T-cells for patient infusion.