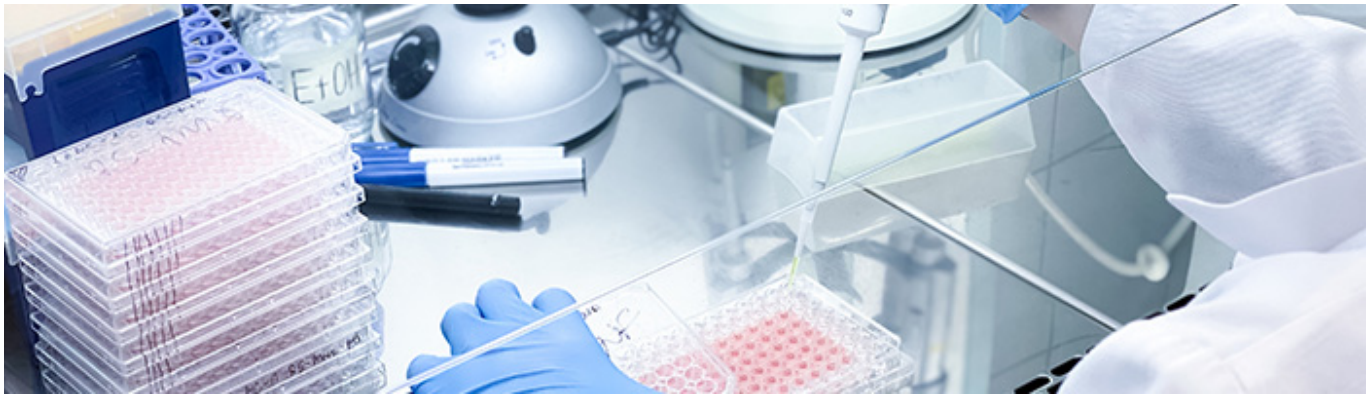
In June 1986, muromonab-CD3 became the first monoclonal antibody approved by the U.S. Food and Drug Administration (FDA) for treating human patients. Since then, the market for antibody therapeutics has grown exponentially, and there are now more than 100 approved products, comprising multiple formats and being used to treat a broad range of disease states. With demand for antibody therapeutics continuing to rise, high-quality research reagents are essential to support their development. Read on for a brief look at the journey of therapeutic antibodies.



**Muromonab-CD3 was a gamechanger**

Muromonab-CD3 (marketed as Orthoclone OKT®3) is a monoclonal mouse IgG2a antibody that targets T cell co-receptor CD3, effectively blocking all cytotoxic T cell function. It was originally approved to help prevent acute rejection after renal transplantation, and its indications were later extended to include other solid organ transplants. However, the frequency of severe adverse reactions, including cytokine storm syndrome, led to muromonab-CD3 being withdrawn from the market in 2010, when these challenges had largely been addressed by advances in antibody engineering[1,2].

**Chimeric monoclonal antibodies address adverse effects**

The main reason for the adverse effects seen following treatment with muromonab-CD3 is that it is a mouse antibody,  meaning that the protein is identified as foreign by the patient's immune system. To try and circumvent this issue, researchers developed chimeric monoclonal antibodies, which are formed by replacing the constant region of a parent murine monoclonal antibody with a human equivalent. Early examples of approved chimeric monoclonal antibodies include abciximab (ReoPro®), a glycoprotein IIb/IIIa receptor antagonist which was approved in 1993 to lessen the risk of heart attack by preventing platelet aggregation, and rituximab (Rituxan®), a CD20-directed cytolytic antibody approved in 1997 for treating relapsed or refractory non-Hodgkin's lymphoma[3,4].

**Humanization limits the number of foreign epitopes**

Despite successes in the clinic, many chimeric monoclonal antibodies failed to reach the market due to the associated production of anti-drug antibodies (ADAs) and unwanted side effects. This prompted the development of humanized monoclonal antibodies, in which all but the complementarity-determining regions (CDRs) are replaced with human counterparts. Daclizumab (Zenapax®) is a humanized monoclonal antibody that binds the alpha subunit (CD25) of the IL-2 receptor to inhibit various T-cell

functions. It was approved in 1997 to prevent transplant organ rejection, although its use was suspended in 2018 following reports of serious inflammatory brain disorders[5]. A more recent example, otelixizumab, is currently in phase III clinical trials for treating type I diabetes[6].

**Introducing fully human monoclonal antibodies**

As antibody engineering technologies have evolved, it has become possible to generate fully human monoclonal antibodies. These are typically produced in one of two ways: via phage display or by engineering mice such that they carry human antibody genes. Adalimumab (Humira®) is an example of the former, which was first approved in 2002 for treating rheumatoid arthritis. As adalimumab's various patents have expired, several biosimilars have been developed[7]. Panitumumab (Vectibix®) is a fully human monoclonal antibody produced in transgenic mice, which was approved in 2006 for treating patients with epidermal growth factor receptor (EGFR) positive metastatic colorectal carcinoma[8].

**Targeted delivery with antibody-drug conjugates (ADCs)**

As well as being engineered to limit adverse effects, monoclonal antibodies have been modified to exhibit additional functions. Antibody-drug conjugates are produced using chemical ligands to couple effector molecules to thiols, amines, or carbohydrates, and are mainly used for targeting cancer cells. Examples include gemtuzumab ozogamicin (Mylotarg®), which was approved in 2000 and delivers a calicheamicin to CD33-positive cells to treat acute myeloid leukemia, and tisotumab vedotin (Tivdak®), which was granted accelerated approval in 2021 and targets monomethyl auristatin E to the tissue factor expressing tumors presented by patients with recurrent or metastatic cervical cancer[9,10].

**Bispecific antibodies**

Bispecifics are therapeutic antibodies with two different binding sites directed at two distinct antigens or two epitopes on the same antigen. Currently, there are three bispecific antibodies on the market. Two of these harness the cells of the immune system to treat cancer– catumaxomab (Removab®), which binds EpCAM and CD3 and was approved in 2009 for treating malignant ascites caused by metastasizing tumors, and blinatumomab (Blincyto®), which binds CD3 and CD19 and was approved in 2014 for treating relapsed or refractory precursor B-cell acute lymphoblastic leukemia. The third approved bispecific, emicizumab (Hemlibra®, approved in 2017), restores the function of missing activated factor VIII (FVIII) by bridging FIXa and FX to treat bleeding due to hemophilia A[11]. More recently, trispecific antibodies have been investigated as an alternative approach to anticancer immunotherapy. For example, earlier this year, an antibody capable of binding HER2, CD3, and CD28 was shown to stimulate the regression of breast cancers in a humanized mouse model by inhibiting tumor cell cycle progression[12].

**Future perspectives**

The 2022 'Antibodies to Watch' article offers an easily digestible overview of current trends within the field of antibody therapeutics[13]. It covers the primary indications in the United States and European Union, of which oncology, immune-mediated disorders, and infectious diseases rank highest, and includes an

overview of common therapeutic targets. Notably, the authors suggest that 2022 may be the year in which the record number of US and EU approvals (13) is exceeded.

Jackson ImmunoResearch specializes in producing secondary antibodies for life science applications. Our portfolio encompasses a wide selection of products that are validated for common research applications, including many techniques used during therapeutic antibody development.

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