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Review Article

Monoclonal antibodies in oncological malignancies: current status and future directions

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Abstract

Recently, extensive research has been done in the area of bio-oncology. Bio-oncology focuses on four targets in particular: angiogenesis, signal transduction, B-cell biology and apoptosis. Monoclonal antibodies (mAbs) are a novel technology that can offer a world of opportunity by interfering with these pathways. In the past decade, a number of antibody-based products have emerged for the treatment of patients with different types of cancer. To date, nine therapeutic mAbs have been approved by the FDA for the treatment of oncologic diseases. This review explores these mAbs and analyzes their mechanism of action, as well as side effects specific to each. It also focuses on the unique features and limitations of mAbs as a mode of therapy and looks at some of the possible future endeavors for this novel therapeutic class.

Introduction

Traditional chemotherapeutic agents usually exert their effects by inhibiting growth and stimulating cell death pathways in rapidly dividing cells. They often achieve this by interfering with DNA or RNA replication. Rapid proliferation is characteristic of tumor cells, but it is also characteristic of other cell types, such as epithelial and bone marrow cells; for this reason, typical adverse side effects of traditional chemotherapy include low blood counts, hair loss, mouth sores, skin changes and dysfunction of the gastrointestinal tract, resulting in vomiting, nausea, diarrhea, constipation, etc. (1).

The use of monoclonal antibody (mAb) therapy in the treatment of cancer represents a significant advance. MAbs are immunoglobulin G (IgG) antibodies whose Fab (antigen-binding fragment) portion contains variable domains designed to recognize antigens that are associated with various cancers. MAb therapy helps to mediate immune responses against tumor cells by facilitating the immunological recognition of receptors on the tumor cell surface that are usually rare or absent on the surface of healthy cells. These receptors are often involved in cellular signal transduction pathways associated with the inappropriate growth and proliferation of cancer cells (2).

MAbs can be classified by their mode of production. Murine mAbs are obtained by challenging mice with a desired antigen and then creating a hybridoma from an extracted B-cell, which can produce large amounts of mAbs specific to the antigen. Murine mAbs are highly immunogenic, resulting in a short serum half-life (3). Chimeric mAbs are made of approximately 65% human sequences and are produced by fusing murine variable regions onto human constant regions. Chimeric and humanized mAbs have less immunogenic content than murine mAbs and therefore have longer serum half-lives (4). Humanized mAbs on the other hand are approximately 95% human in their origin and are produced by placing murine hypervariable amino acid domains directly into human antibodies (5). Fully human mAbs can be produced by using either transgenic mice or phage display libraries (2, 6). Humanized mAbs have the least amount of immunogenic content of all the mAbs.

MAbs exert their anticancer effects through both immunological and nonimmunological mechanisms. The immunological mechanisms of action include stimulation of antibody-dependent cell-mediated cytotoxicity (ADCC) (7), complement activation through complement-dependent cytotoxicity (CDC) (8) and phagocytosis by macrophages after opsonization (9). Nonimmunological mechanisms of action include disruption of signaling pathways important for tumor cell growth or the stimulation of pathways that result in cell death.

Another attractive feature of mAbs is that they can be conjugated with other molecules to take on a secondary function. MAbs can be conjugated with radioactive isotopes, allowing for the delivery of high doses of radiation to cells targeted by mAbs. Murine mAbs are used preferentially in radioimmunotherapy because their high immunogenicity promotes rapid clearance, reducing unnecessary systemic radiation. MAbs can also be conjugated with drug-activating enzymes. Antibody-directed enzyme prodrug therapy (ADEPT) involves the systemic administration of an inactive chemotherapeutic agent, which can then be activated in select cells and locations according to the specificity of the linked mAb (10). MAbs conjugated with liposomes, carrying drugs or therapeutic nucleotides may also be a promising new tool in cancer therapy (11).

Adverse reactions

Adverse reactions to mAbs are fairly common and widely varied in their presentation. Some common side effects of mAbs include: hypersensitivity reactions, flu-like symptoms (chills, fatigue, fever, myalgia, etc.), nausea and vomiting, diarrhea, rash, hypo- and hypertension, and myelosuppression (resulting in neutropenia, thrombocytopenia, anemia, etc.). Some mAbs are associated with significant myelosuppression which can lead to serious complications, such as bleeding and infection. These reactions may be the result of cytokine release, a direct immune response to the drug, or may not involve the immune system directly (12). All of the mAbs discussed in this review are associated with the risk of infusion reactions. The exact mechanism by which mAbs elicit these allergy-like reactions has not been clearly defined. Severe infusion reactions to mAbs are rare, but mild and moderate infusion reactions are quite common, especially on the first exposure to the agent. Common mild to moderate infusion reactions typically present with fever, chills, rigors, urticaria or dyspnea. Severe reactions may present with airways obstruction, hypotension or anaphylaxis (13). Techniques designed to reduce the chances of infusion reactions include administering a test dose of the agent, extending the infusion time and, in some cases, pretreating the patient with other agents. The major adverse events characteristic of the mAbs discussed here are listed in Table I.

FDA-approved monoclonal antibodies

Anti-HER2

Trastuzumab (Herceptin®) is a humanized mAb with activity against the human epidermal growth factor receptor 2 (HER2, or erbB-2). Trastuzumab was approved by the FDA as monotherapy for patients with metastatic breast cancer who were previously on at least one chemotherapeutic regimen and tested positive for HER2 overexpression, as well as in combination with paclitaxel

Table I: FDA-approved monoclonal antibodies.

Monoclonal antibody	Target	Indications	Adverse reactions
Trastuzumab (Herceptin)	HER2	Early-stage and metastatic breast cancer	Cardiac toxicity (10%) Severe pulmonary complications (14%)
Gemtuzumab ozogamicin (Mylotarg)	CD33	AML	Severe hepatic VOD (3-15%) Myelosuppression (98%) Infection (30%)
Alemtuzumab (Campath)	CD52	B-CLL	Bone marrow hypoplasia, pancytopenia (6%) Infection (43%)
Rituximab (Rituxan)	CD20	NHL and other B-cell cancers	Tumor lysis syndrome Severe mucocutaneous reactions
Ibritumomab tiuxetan (Zevalin)	CD20	Follicular or transformed B-cell NHL	Severe cytopenia (< 5%) Melena (2%) Gastrointestinal hemorrhage (1%) Pruritus (9%) Urticaria (4%) Secondary malignancies (2%)
Tositumomab/iodine (I131) tositumomab (Bexxar)	CD20	NHL and other B-cell cancers	Severe cytopenia (71%) Secondary malignancies (6% at 5 years)
Cetuximab (Erbitux)	EGFR	Metastatic colorectal and head and neck cancer	Infusion reaction during first administration (90%) Severe infusion reactions (3%) Cardiopulmonary arrest (2%)
Panitumumab (Vectibix)	EGFR	Metastatic colorectal cancer	Severe dermatological toxicities (12%)
Bevacizumab (Avastin)	VEGF	Metastatic colorectal carcinoma, metastatic breast cancer and NSCLC	Gastrointestinal perforation (1.4-2%) Hemorrhage in patients with squamous histology (31%) Mucocutaneous hemorrhage (4%) Slower wound healing after major surgery (10-20%)

for metastatic breast cancer. More recently, trastuzumab was approved for the treatment of early-stage breast cancer after primary therapy (14). Trastuzumab has been used in combination with a number of other chemotherapeutic agents, including vinorelbine, gemcitabine, capecitabine, docetaxel and hormonal agents (15).

HER2 is a transmembrane tyrosine kinase that plays a role in the normal growth and differentiation of cells. Amplification of the *HER2* gene, which is characteristic of about 30% of metastatic breast cancers, results in HER2 overexpression (16). Trastuzumab binds to the extracellular domain of HER2 and induces apoptosis through ADCC (17).

In addition to the normal risks of mAb therapy, trastuzumab is associated with both pulmonary toxicity (14%) and cardiotoxicity (10%), which has led to the issuing of a black box warning by the manufacturer. The risk of cardiac failure while receiving trastuzumab was most elevated in patients who had received anthracycline-based chemotherapy or high-dose cyclophosphamide (17, 18).

Anti-CD33

Gemtuzumab ozogamicin (Mylotarg®), a humanized IgG_4 mAb linked to a cytotoxic calicheamicin derivative, targets CD33. CD33 is expressed on the surface of leukemic blast cells in more than 90% of patients with acute myeloid leukemia (AML). The FDA has approved gemtuzumab ozogamicin for the treatment of CD33-positive AML in first relapse in patients over 60 years of age and who are not considered candidates for cytotoxic therapy (15). Gemtuzumab ozogamicin may also be combined with other chemotherapeutic agents, such as daunorubicin, cytarabine, fludarabine or ciclosporin, for improved efficacy (3, 19).

When the gemtuzumab subunit binds to CD33, the mAb-receptor complex is internalized, creating an endosome that fuses with lysosomes. Once this fusion occurs, ozogamicin is released from the gemtuzumab subunit and localizes to the nucleus, where it makes double-stranded breaks in the DNA. These double-stranded breaks activate ATM, which phosphorylates downstream targets involved in cell cycle checkpoints, eventually resulting in apoptosis (20-22).

There are a number of serious adverse reactions associated with gemtuzumab ozogamicin, in addition to the normal risks of mAb treatment. Myelosuppression, for example, is observed in 98% of patients, resulting in a significant risk for the development of infection, as well as bleeding (21, 23). Hepatic veno-occlusive disease (VOD) occurs in 3-15% of patients. This serious and sometimes fatal adverse reaction is unpredictable with regard to both its incidence and timing (20, 24).

Anti-CD52

Alemtuzumab (Campath®) is a humanized mAb that is indicated for the treatment of B-cell chronic lymphocytic leukemia (B-CLL) and has also shown activity against T-cell prolymphocytic leukemia (25). The FDA has approved alemtuzumab for the treatment of B-CLL in patients who have been treated with alkylating agents and have failed fludarabine therapy (15). Alemtuzumab is most effective in the blood and bone marrow rather than in the spleen or lymph nodes, making it well suited for the treatment of B-CLL, which is characterized by malignant lymphocytosis in blood and infiltration of the bone marrow (26).

Alemtuzumab is directed at CD52, which is found on the surface of all B-cells and is highly expressed in B-CLL cells. *In vitro* studies implicate ADCC and CDC, as well as a nonclassical, caspase-independent pathway, as mediating the cell death induced by alemtuzumab in B-lymphoid cell lines and B-CLL cells (20, 27, 28).

Adverse reactions to alemtuzumab include pancytopenia/bone marrow hypoplasia in 6% of patients (18) and infectious complications (sepsis, pneumonia and opportunistic infections) in 43% of patients. These significant adverse reactions prompted the manufacturer to issue a black box warning (20). To protect against infections with pathogens such as *Pneumocystis carinii* pneumonia (PCP) and herpesviruses, prophylaxis with antibiotics is recommended (24). Additionally, cytomegalovirus (CMV) reactivation has been documented with alemtuzumab administration (29).

Anti-CD20

Currently there are three anti-CD20 therapies approved for cancer treatment: rituximab (Rituxan®), ibritumomab tiuxetan (Zevalin™) and tositumomab/iodine (I131) tositumomab (Bexxar®).

1. Rituximab

The FDA has approved rituximab, a chimeric mAb, for the treatment of patients with relapsed, refractory low-grade or follicular B-cell non-Hodgkin's lymphoma (NHL), as well as for first-line therapy in patients with NHL in combination with anthracycline-based or CHOP (cyclophosphamide, doxorubicin, vincristine, prednisone) chemotherapy (30). Recently, rituximab was approved for use in combination with CVP (cyclophosphamide, vincristine, prednisone) for the first-line treatment of naïve follicular NHL and the treatment of low-grade NHL (15). Rituximab is a beneficial addition to standard therapy for patients with NHL and other B-cell cancers because CD20 is expressed on about 95% of B-cell lymphoma cells, normal B-cells and stem cells. It is not expressed to a great extent, however, on B-CLL and plasma cells (31).

Numerous clinical trials have demonstrated benefits of using rituximab in combination with CHOP chemotherapeutic regimens, especially in elderly patients who may not be able to tolerate more aggressive treatments (31). In one of the largest clinical trials using rituximab plus CHOP therapy, a complete response was achieved in 76% of patients receiving rituximab plus CHOP compared to 63% receiving CHOP alone. The results from this trial

also showed that the 2-year survival rate was significantly greater in the rituximab plus CHOP group (70%) compared to CHOP alone (57%) (17). Studies are still needed to determine how the combination of rituximab and CHOP affects long-term survival rates.

Rituximab is thought to contribute to antitumor activity through ADCC and CDC, along with sensitizing cells to chemotherapy, possibly through the downregulation of antiapoptotic factors and IL-10 (a prosurvival cytokine), and has the ability to cross-link CD20 (32-34). The cross-linking of CD20 molecules can trigger intracellular signals that induce apoptosis (35). Cross-linked CD20 rapidly translocates to lipid rafts (36), where it is believed to form multimeric complexes that facilitate the influx of calcium from the extracellular space (37). The sustained influx of calcium into the cytosol can stimulate calcium-dependent signaling processes involved in cell cycle progression and apoptosis; for this reason, it has been hypothesized that the activity of rituximab may be due in part to its ability to modulate calcium levels in tumor cells (38, 39). There is also some evidence that lipid rafts facilitate complement activation or act as points where a cell is sensitive to complement penetration (40).

Although many mAbs appear to owe a portion of their antitumor activity to their ability to inhibit cell signaling of their target receptors, rituximab, on the other hand, may exert its antitumor effects in part through the stimulation of its target receptor. Additionally, there is some evidence that rituximab, like alemtuzumab, also mediates a non-classical, caspase-independent apoptotic pathway (28).

Serious side effects associated with rituximab include tumor lysis syndrome and severe mucocutaneous reactions. These adverse reactions have resulted in the issuing of a black box warning for rituximab (17). Tumor lysis syndrome occurs at a higher rate in patients with bulky disease and can lead to kidney failure. Pretreatment with fluids or allopurinol may decrease complications associated with tumor lysis syndrome. Mucocutaneous reactions can occur at any time within 13 weeks of initiation of rituximab treatment. They can manifest as Stevens-Johnson syndrome, lichenoid dermatitis, vesiculobullous dermatitis and toxic epidermal necrolysis.

Rituximab can also produce adverse hematological effects in patients, including severe lymphopenia (40%), severe leukopenia (4%), severe neutropenia (6%) and severe thrombocytopenia (2%). This myelosuppression leads to the development of infections in 31% of patients after treatment (30).

2. Ibritumomab tiuxetan

Ibritumomab tiuxetan is a radioimmunoconjugate consisting of a murine-human chimeric mAb linked to the chelator tiuxetan, to which a radioactive isotope can be added. The isotopes used are yttrium-90 (90Y) for treatment and indium-111 (111In) for imaging (41).This drug was the first radioimmunoconjugate to be approved by the FDA. It is indicated for the treatment of relapsed/refractory follicular/low-grade or transformed B-cell NHL and rituximab-refractory follicular NHL (15). Ibritumomab is also active in patients with bulky adenopathy, but the rate of response decreases as the tumor size increases (41). The ibritumomab tiuxetan therapeutic regimen involves a two-step infusion; a first infusion for imaging biodistribution of the drug and a second infusion with the determined therapeutic dose (42).

In addition to the normal adverse events associated with mAbs, ibritumomab tiuxetan has also been associated with severe cutaneous and mucocutaneous reactions, including: melena (2%; life-threatening in 1%), gastrointestinal hemorrhage (1%), pruritus (9%), rash (8%), urticaria (4%) and petechia (3%). There is also the risk (2%) that the patient will develop secondary malignancies, such as AML and myelodysplastic syndrome (MDS), as a result of the treatment (41).

The most common side effects seen with ibritumomab tiuxetan are hematological disturbances similar to those seen during rituximab treatment, including severe cytopenia in < 5% of patients (43). Since ibritumomab tiuxetan is administered in conjunction with rituximab, it is difficult to determine whether or not the adverse effects are due solely to rituximab, ibritumomab tiuxetan or a combination of the two (22).

3. Tositumomab/iodine (I131) tositumomab

Tositumomab is a murine γ mAb linked to I131 (41) and was originally approved by the FDA for the treatment of patients with CD20-positive follicular NHL, with or without transformation, whose disease is refractory to rituximab and has relapsed following chemotherapy. This approval was later expanded to include rituximab-naïve patients with relapsed or refractory low-grade follicular transformed CD20-positive NHL (15). The main mechanisms of action are ADCC and CDC. I131 not only kills the CD20-positive cells but also destroys surrounding tumor cells. Similar to ibritumomab tiuxetan, the therapeutic regimen involves two infusions.

Myelosuppression is the dose-limiting toxicity of this therapy. Severe cytopenia occurs in 71% of patients (44). Nonhematological adverse reactions include anorexia, arthralgia, rash, pruritus and increased thyroid-stimulating hormone (TSH) levels (41). There is also a risk of secondary malignancies (up to 6% 5 years after treatment) such as AML and treatment-related MDS.

Anti-EGFR

Currently there are two anti-epidermal growth factor receptor (anti-EGFR) mAbs that are approved for the treatment of cancer: cetuximab (Erbitux™) and panitumumab (Vecitibix®). It is known that EGFR is overexpressed in 25-80% of colorectal cancers and is generally associated with advanced disease. EGFR is a transmembrane receptor that has tyrosine kinase activity and is stimulated by growth factors such as transforming growth factor α (TGF-α) and EGF. EGFR is a good target for anticancer agents because it is a crucial factor for tumor cell proliferation, inhibition of apoptosis and other processes important for cancer progression (45).

1. Cetuximab

Cetuximab is a chimeric mAb that is highly selective for the EGFR and was approved by the FDA for the treatment of metastatic colorectal cancer that is irinotecan-refractory and for the treatment of naïve metastatic colorectal cancer. It has also been shown to be effective in squamous cell carcinoma of the head and neck and non-small cell lung cancer (NSCLC) and was approved for head and neck cancer in March 2006 (15, 45). It the first agent approved by the FDA for head and neck cancer that has shown a survival benefit in patients with unresectable disease (46). Cetuximab is generally used in combination with irinotecan, fluorouracil, folinic acid or topotecan because it enhances sensitivity to radiotherapy and chemotherapeutic agents (45). It has also proved effective when combined with tyrosine kinase inhibitors such as gefitinib for the treatment of NSCLC (47). Cetuximab is also being evaluated in phase III trials as a second-line therapy for colorectal cancer and in combination with radiation or chemotherapy for head and neck cancer. Currently it is also being studied in lung and pancreatic cancer (46).

Cetuximab has higher affinity for EGFR than EGF or TGF-α and competitively blocks the cellular action of these ligands by interfering with ATP binding (45, 46). Several mechanisms have been proposed for the antitumor activity of cetuximab, such as the inhibition of cell cycle progression in the G1 gap phase that occurs prior to DNA synthesis, leading to cancer cell apoptosis (45).

There is a particularly high incidence of infusion reactions associated with cetuximab; 90% of patients experience infusion reactions during their first administration and 3% of the infusion reactions caused by this drug can be classified as severe. There is also an infrequent occurrence of cardiopulmonary arrest (2%) documented with cetuximab (48).

2. Panitumumab

Panitumumab is different than cetuximab in that it is the first fully human mAb that targets EGFR for the treatment of solid tumors (15). It has been studied in lung, kidney and colorectal cancer and is currently approved for the treatment of metastatic colorectal cancer. As a fully human mAb, panitumumab is associated with a decreased risk of antibody formation, reducing the risk of hypersensitivity reactions (49).

The most common toxicity associated with panitumumab is rash, which can be mild to severe, although 12% of patients experience severe cases (49-51). Rash, which seems to be dose-dependent up to 2.5 mg/kg, usually appears during the first or second infusion and generally resolves within 4 weeks of cessation of treatment (49). It is still under investigation if the severity of the rash is correlated with clinical outcomes.

Overall, panitumumab is generally well tolerated when combined with other chemotherapeutic agents such as irinotecan/bolus 5-fluorouracil/leucovorin (49, 52). Like the other mAbs, there is generally a synergistic or additive effect seen when it is combined with chemotherapy or other agents (49).

Anti-VEGF

The vascular endothelial growth factor (VEGF) receptor is an excellent target for cancer treatment because there is an established relationship between vascular density, the expression of VEGF and the risk of tumor recurrence (53). VEGF is responsible for both angiogenesis and lymphangiogenesis, and may also stimulate the immune system through an increase in dendritic cell function. Besides affecting endothelial cells near tumor cells, targeting VEGF with mAbs may interfere with an autocrine survival pathway for cancer cells.

Bevacizumab (Avastin™) is a humanized murine-derived mAb that binds to VEGF and blocks its interaction with the VEGF receptor on endothelial cells, thereby preventing angiogenesis, as well as halting VEGF's ability to promote endothelial mitogenesis and vascular permeability (53-55). Bevacizumab has been approved for the first-line treatment of patients with metastatic colorectal carcinoma in combination with intravenous 5-fluorouracil-based chemotherapy (54). Bevacizumab was also approved as first-line treatment in patients with locally advanced, metastatic or recurrent NSCLC in combination with platinum-based therapy and for metastatic HER2-negative breast cancer in combination with paclitaxel (15).

In addition to the typical side effects of mAbs, bevacizumab has been associated with thrombotic events, nephrosis, proteinuria and increased bleeding. Bevacizumab has been known to cause gastrointestinal perforation (1.4-2%), mucocutaneous hemorrhaging (4%) (18), hemorrhaging in patients with squamous histology (31%) (56) and slower wound healing after major surgery (10-20%) (18). The manufacturer has issued a black box warning for gastrointestinal perforation, wound dehiscence and fatal hemoptysis (55). The FDA approved a change to the package insert that added a warning regarding dose administration for reversible posterior leukoencephalopathy syndrome and nasal septum perforation as a serious side effect.

Thrombotic events can be prevented by pretreating patients with anticoagulant agents such as warfarin. In the case of bevacizumab treatment, however, pretreatment with warfarin and other anticoagulants is not recommended because it may compound the risk of serious bleeding already associated with bevacizumab (55). Cardiac toxicity is also a risk associated with bevacizumab in patients who have undergone radiation therapy or chemotherapy with an anthracycline (18).

New directions

Investigational monoclonal antibodies

In response to the initial market success of mAbs, much research effort has been shifted to this area of therapeutics. Despite the introduction of nine monoclonal

oncologic medications to the market, a large number of potential molecular targets still exist. There are now over 400 clinical trials testing newly developed mAbs (57). Some of these new mAbs are listed in Table II.

Optimizing mAb treatment

There is much ongoing research devoted to improving the efficacy and optimizing treatment regimens of approved mAbs. Researchers are investigating the benefits of administering immunomodulatory agents in conjunction with mAbs, as well as designing mAbs that target important immune cell receptors. In addition, pharmacogenetic techniques may potentially allow clinicians to predict a patient's response to mAb therapy before treatment begins.

1. Immunomodulation

The most extensively studied immunomodulatory mAbs are those for CTLA-4, a T-cell-specific inhibitory receptor molecule. In human studies, blocking CTLA-4 was shown to stimulate tumor necrosis in some subjects (58). Other immunomodulatory mAbs that are currently being developed include those targeted at CD25 and 4-1BB. It is hoped that anti-CD25 mAbs will have the ability to deplete inhibitory T-cells and that 4-1BB-targeted mAbs will be able to stimulate T-cells and natural killer (NK) cells (59).

Two cytokines that have been investigated in combination with mAbs are interleukin-2 (IL-2) and interleukin-12 (IL-12). In phase I trials, the combination of IL-12 and trastuzumab, as well as IL-2 and trastuzumab, demonstrated enhanced NK-mediated ADCC activity against tumor cells (60). Another combination that is being actively investigated is IL-2 and rituximab. The combination has been used in phase I trials in patients with stage III-IV NHL. Adding IL-2 to the regimen correlated with increased levels of NK cells and enhanced rituximab-mediated ADCC (61-63).

2. Pharmacogenetics

A polymorphism has been identified in the gene for the Fc receptor FcγRIIIa that conveys enhanced receptor binding for IgG_1. This polymorphism is associated with improved responses to rituximab during the first-line treatment of follicular NHL (64). The genotyping of the FcγRIIIa of NHL patients may prove to be useful when deciding whether to use chemotherapy, rituximab or combination therapy.

Polyclonal recombinant antibodies

One promising area of new development is polyclonal recombinant antibodies. Our natural antibody response is polyclonal, with specificity for many different epitopes. New technologies are being developed that will allow the manufacture of a consistent mixture of recombinant antibodies with specificities for multiple epitopes (65). The development of polyclonal antibody libraries would allow for the rapid selection and expression of a multitude of mAbs with a high degree of specificity for multiple cancer cell epitopes (66). In theory, this type of antibody mixture would more closely mimic our body's natural immune response. Additionally, because a polyclonal antibody preparation would target many specific epitopes, a mutation of one of the epitopes would be less likely to confer resistance to the preparation as a whole. Also, having a more diverse arsenal of reactive antibodies may be able to increase the number and variety of effector functions a preparation can induce (67). At this time, many polyclonal products for the

Table II: Investigational monoclonal antibodies being developed for the treatment of cancer.

Monoclonal antibody	Target	Possible indications
Pertuzumab (Omnitarg)	HER dimerization inhibitor	Breast, prostate and ovarian cancer and NSCLC
Ipilimumab	CTLA-4	Follicular lymphoma, metastatic melanoma, renal cell carcinoma, prostate and breast cancers
Oregovomab (OvaRex)	CA125, HER1 receptors	Ovarian cancer
Lexatumumab	TRAIL receptor 2	Renal cell carcinoma and ovarian cancer
Matuzumab	EGFR	Platinum-resistant ovarian cancer (primary peritoneal malignancies), pancreatic cancer and NSCLC
Edrecolomab	EpCAM	Colorectal cancer
Catumaxomab	EpCAM	Head and neck cancer
Tocilizumab	IL-6	Renal cell carcinoma
Epratuzumab	CD22	NHL
Inotuzumab ozogamicin	CD22	NHL
Galiximab	CD80	NHL
Siplizumab	CD2	NHL
XmAb2513	CD30	NHL
SGN-40	CD40	NHL

treatment of cancer are reported to be in the pipeline, although none has received FDA approval as yet.

Nanobodies

Another promising area of antibody research that may prove beneficial in the treatment of cancer is nanobodies. Nanobodies are the smallest portion of an antibody that retains the ability to bind antigen. In simple terms, nanobodies consist of a heavy-chain variable region and lack a light chain (68). Heavy-chain-only antibodies occur naturally in several species of sharks and camels. Nanobodies have several distinct advantages over traditional antibodies, including their small size, superior solubility and decreased antigenicity, as well as ease of manufacture and purification. Cortez-Retamozo and colleagues demonstrated that nanobodies could be manufactured to selectively activate a cytotoxic prodrug at the site of a common cancer antigen, carcinoembryonic antigen (CEA) (69). Roovers *et al.* developed a nanobody specific for EGFR and demonstrated its ability to selectively prevent binding of EGF to EGFR without acting as an agonist itself (68). Obviously, more work in this area needs to be done before nanobody-based products can be used in humans, although initial positive studies indicate that this work is clearly warranted.

Conclusions

Despite some of the possible limitations of mAbs, this novel technology has demonstrated its clinical worth. The specificity of mAb therapy has given cancer patients effective new options that, generally speaking, have a more attractive toxicity profile than traditional chemotherapeutics. There is still a need for more research and development in this area of cancer treatment. Hopefully, new mAbs will be able to offer more curative treatments, greater specificity for a multitude of targets and a greater variety of effector functions. This area of research will continue to evolve and promises to be the future treatment of oncologic diseases.

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