CLINICAL RESULTS WITH HUMANIZED AND HUMAN MONOCLONAL ANTIBODIES

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ABSTRACT. Monoclonal antibodies (MAbs) from animals may be humanized by the process of "CDR grafting". However, the CDRs from a murine MAb that are inserted into a human MAb framework may not retain their original shape. When this occurs, the humanized MAb will no longer bind the original target antigen. A computer-assisted molecular modeling procedure has been developed to overcome this inherent difficulty. More than a dozen murine MAbs have been humanized using this procedure which has permitted the humanized MAbs to retain their antigen binding ability. Two murine MAbs humanized by this procedure, SMART Anti-Tac (anti-CD25 / IL-2r) and SMART M195 (anti-CD33), have been entered into clinical trials. Relative to their murine counterparts, the humanized MAbs showed longer serum half-lives, improved effector cell function and, most importantly, were not immunogenic in humans. Nearly 40 patients have received single or multiple doses of these humanized MAbs and in some cases up to 12 doses have been administered over a period of 4 months with no immunogenic response. These clinical results to date, along with those with human (anti-viral) MAbs, indicate that humanized and human MAbs are capable of overcoming the disadvantages of short serum half-life and immunogenicity of murine MAbs.

1. Introduction

The therapeutic properties of immunoglobulins have long been recognized. Natural immunization was reportedly observed in the fifth century B.C. by Greek physicians and the practice of vaccination to induce protective levels of antibodies is used throughout the world today. In those cases where there is insufficient time for vaccination, the practice of passive immunization (administration of pre-formed immunoglobulins, or "anti-serums") has become an established medical procedure. The next major advance in immunotherapy came in 1975 when Köhler & Milstein[1] described a procedure to generate stable cell lines (hybridomas) that could produce large quantities of monoclonal antibodies (mAbs). These hybridomas provided the means to replace the heterogeneous and relatively low potency anti-serums with homogeneous preparations of relatively high potency monoclonal antibodies. Thus, for the first time, monoclonal antibodies (mAbs) could be produced in sufficient quantity for use as therapeutic pharmaceutical agents.

2. Monoclonal Antibodies

2.1 MURINE MONOCLONAL ANTIBODIES

Köhler & Milstein's pioneering work used mouse (or murine) cell lines. Thus, the first mAbs considered for therapeutic use were from mice because there were no comparable human myeloma cell lines with which to form human hybridomas. The targeting specificity of these murine mAbs seemed to embody Ehrlich's "magic bullet" and held great promise for the treatment of cancer and other diseases. However, clinical studies revealed that injection of these murine proteins into humans could lead to the formation of HAMA (Human Anti-

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Monoclonal Antibody) in which the patient produced antibodies against the murine antibody[2].

HAMA formation can neutralize a murine mAb causing its rapid clearance from the bloodstream and possibly causing further difficulties by the formation of immune complexes. Moreover, even in the absence of HAMA, murine mAbs tend to have relatively short serum half-lives of one to three days. This is much shorter than the serum half-life of human IgG antibodies which are generally on the order of three weeks. Thus, rather early on, it became apparent that murine mAbs would need to be replaced with less antigenic counterparts that still retain their extraordinary specificity of target antigen recognition.

2.2 HUMANIZED MONOCLONAL ANTIBODIES

Several approaches have been taken to generate mAbs with reduced antigenicity. One of the early solutions was to genetically engineer a "chimeric" mAb that was part murine, part human[3]. The murine portion contributed the variable regions for antigen recognition and the human portion provided the remaining constant regions common to human IgG immunoglobulins. The chimerization process reduced the antigenicity of the original murine mAb in some, but not all, instances and occasionally provided a longer serum half-life. Chimeric mAbs represented an improvement, but one which could be improved upon even further.

2.2.1 *CDR Grafting.* One of the ingenious solutions to the problem of mAb antigenicity was to further "humanize" a chimeric mAb by retaining additional portions of the human variable domain. A "humanized" mAb is essentially a human antibody to which only the hypervariable regions (also called the "complementarity regions" or CDRs) have been grafted from a murine antibody. There are a total of six CDRs in an IgG antibody that form the antigen binding site, three on the heavy chain and three on the light chain of the antibody. With the genetic engineering process, known as "CDR grafting", only those portions of the murine antibody directly involved in antigen recognition are grafted into the framework of a human antibody[4].

2.2.2 *The SMART™ Process for Humanization.* The initial demonstration of CDR grafting by Winter and colleagues produced an antibody that retained the ability to bind the target antigen[5]. This original description did not reveal, however, that the murine CDRs may not always fit correctly into a human framework. In fact, in most cases the grafted CDRs do not maintain the correct three-dimensional shape to bind the target antigen. In these cases, in order to preserve the required shape of the CDRs, certain "key contact" amino acids in the surrounding framework of the murine antibody must be transferred along with the CDRs to the humanized mAb[6]. These "key contact" amino acids are so named because they are in contact with, or otherwise influence the shape of, the CDRs so they are able to bind the target antigen. When both the CDRs and the "key contact" amino acids are grafted into the human framework regions, the humanized antibody retains the ability to bind the target antigen with an affinity similar to that of the original murine antibody.

These "key contact" amino acids can be identified by studying the three-dimensional structure of the antibody. Traditionally, this evaluation is carried out using X-ray crystallography of crystallized antibody fragments. However, crystallization of proteins is a difficult and time consuming task and the challenge has been how to identify these "key contact" amino acids quickly, and routinely, for any new murine antibody of interest. Protein Design Labs (PDL) has developed a SMART™ procedure to do this using computer-assisted molecular modeling. This proprietary molecular modeling process for antibody humanization involves three steps.

The first step is to sequence the murine antibody and, utilizing this information, select a known human framework whose amino acid sequence has the greatest homology to the framework regions in the original murine mAb. This begins the humanization process using a human framework most likely to accept the murine CDRs without distortion of their three-dimensional shape.

The second step involves identifying the "key contact" amino acids in the framework region of the murine antibody. This step uses a molecular modeling process based on unique

algorithms developed by Professor Michael Levitt of Stanford University. This proprietary software, which has been added to by the computational chemists at PDL, utilizes the similar three-dimensional shapes that have been determined for a number of antibodies by X-ray crystallography analysis. The three-dimensional model constructed with this modeling software allows the identification of the "key contact" amino acids which are then retained in the humanized antibody. Thus, with the appropriate software, computer-assisted modeling can rapidly predict the three-dimensional shape of an antibody and avoid time-consuming crystallization experiments and X-ray crystallography studies[7].

The third, and final, step in the humanization process is the selective replacement of any atypical amino acids in the framework region of the humanized mAb. Human immunoglobulins show a great deal of sequence homology within the framework regions. Occasionally, an atypical amino acid is found in one human antibody that is not normally present at that position in most other human framework regions. To further ensure the reduced antigenicity of the humanized antibody, each of these atypical amino acids is replaced with the consensus amino acid normally found at that position. Generally, there are a dozen or less of such atypical amino acids in any human framework region.

3. Clinical Results with Humanized Monoclonal Antibodies

3.1 SMART ANTI-TAC ANTIBODY

The first mAb humanized using the SMART process was the Anti-Tac antibody directed against the alpha chain (p55) of the human IL-2 receptor on activated T-lymphocytes ("Tac" refers to this T cell activation antigen). The anti-Tac murine antibody was generated by Dr. Thomas Waldmann and his colleagues at the National Institutes of Health of the United States Department of Health and Human Services[8]. The murine antibody was evaluated in a clinical trial where, among other observations, it was noted that the patients developed a HAMA response. The SMART humanization process was then used to generate a humanized version which had nearly the same antigen binding affinity as the original murine mAb[9]. Since this was the first antibody humanized by PDL, the process was repeated but this time using only CDR grafting and not using the optimization of the SMART process. This mAb was not able to bind the target antigen. This result confirmed that CDR grafting alone was not sufficient to generate a useful humanized antibody. Moreover, it was provided the first validation of the utility of the SMART process.

Worldwide clinical development of the SMART Anti-Tac antibody is being conducted by Hoffmann-La Roche for the treatment of immune-mediated diseases. In the initial Phase I trial directed by Dr. Claudio Anasetti, single doses of either 0.5, 1.0 or 1.5 mg/kg of the humanized Anti-Tac were administered to a total of twenty-four bone marrow transplant patients with steroid-resistant graft-versus-host disease (GvHD) at the Fred Hutchinson Cancer Research Center (Seattle, WA) and Vancouver General Hospital (Vancouver, BC). The interim results showed that the first thirteen patients tolerated the intravenous infusion of the Anti-Tac mAb with approximately 40% showing improvement in clinical markers of disease. Positive clinical responses were seen for the three key markers of GvHD: skin (6/11 patients); liver (2/8 patients); and gastrointestinal (5/8 patients). There were no appreciable side effects attributed to the humanized mAb with one patient each showing chills or diaphoresis. The mean serum half-lives for the three dose groups were 79, 88 and 114 hours. These values are less than observed for other humanized antibodies and may reflect binding of the free mAb by the target antigen on activated T-cells. Follow up assays showed that the Anti-Tac mAb was still bound to target antigen on circulating T-cells (IL-2 receptor alpha chain) twenty-eight days after treatment[10].

Most importantly, this Phase I trial with the SMART Anti-Tac mAb showed there was no HAMA development. This was the first "proof of principle" that the SMART humanization process could generate a non-antigenic mAb for use in humans. The positive results from this single-dose Phase I treatment trial led to a multi-national Phase II/III trial begun in April of 1993 in which multiple doses of the Anti-Tac mAb are being evaluated for the prevention

of GvHD. As of March 1994, approximately half of the anticipated number of patients have been enrolled in this trial which is expected to conclude patient enrollment by the end of 1994. In February, 1994, Roche began a multiple-dose Phase I trial administering the Anti-Tac mAb to patients receiving kidney transplants.

3.2 SMART M195 ANTIBODY

The second SMART antibody to enter clinical trials was HuM195. This humanized antibody was generated from the murine M195 mAb developed by Drs. Old, Scheinberg et. al. at the Memorial Sloan-Kettering Cancer Center in New York[11]. M195 is directed against the CD33 antigen which is present on leukemic cells in most patients with acute or chronic myeloid leukemia (AML or CML) but is not present on hematopoietic stem cells. Thus, the antigen specificity of M195 potentially provided a means to target leukemic cells for destruction while sparing the stem cells needed for hematopoiesis.

When administered as a ^{131}I radioimmunoconjugate, the mAb targeted leukemic cells and effectively reduced the number of peripheral blood and bone marrow blast cells in patients with relapsed or refractory AML. However, seven of nineteen patients developed HAMA. For two of these patients who initially showed positive responses to M195 and were then retreated with ^{131}I-M195, the HAMA response sharply reduced the serum half-life of the antibody and its ability to reduce the number of leukemic cells[12].

A humanized form of this mAb (HuM195), generated using the SMART process retained the original antigen specificity of the murine mAb and showed a slight increase in the binding affinity for CD33 antigen[13]. HuM195 was entered into a Phase I trial enrolling AML patients at the Memorial Sloan-Kettering Cancer Center under the direction of Dr. David A. Scheinberg[14]. Thirteen patients received six doses of the HuM195 at levels of 0.5, 1.0, 3.0 or 10.0 mg/m^2 over a period of eighteen days. Two of the patients received a second dosing regimen for a total of 12 doses (up to 216 mg cumulative dose of HuM195). The first dose given to each patient was trace labeled with ^{131}I to evaluate the targeting of HuM195 to leukemic cells and its pharmacokinetic parameters. The humanized HuM195 targeted sites of the disease as well as the murine mAb, was well tolerated and no HAMA was detected in any of the patients. This was a more critical test of antigenicity since nearly 40% of these same class of patients previously had shown HAMA development after administration of the murine form of the mAb.

3.3 OTHER SMART ANTIBODIES

The SMART process has been applied to over a dozen antibodies to date. These humanized mAbs are directed against potential targets for the treatment of cancer, inflammation, cardiovascular, autoimmune and infectious diseases as shown in the Table 1. In each case, the SMART process has produced a humanized antibody with nearly equal antigen binding capability of the original murine mAb.

Table 1. SMART (humanized) mAbs

Product in Research or Development	Potential Therapeutic Indications	Development Status (1)
Viral Infections		
SMART Anti-CMV Antibody	Cytomegalovirus (CMV) infections associated with AIDS, bone marrow and organ transplants	Preclinical
SMART Anti-Herpes Antibody	Genital and neonatal herpes	Preclinical
Autoimmune Disease and Inflammatory Disease		
SMART Anti-Tac Antibody (2)	Graft-versus-host disease	Phase II / III
	Organ transplant rejection and certain autoimmune diseases	Phase I
Recombinant Anti-Tac Immunotoxin (2)	Graft-versus-host disease, organ transplant rejection and certain autoimmune diseases	Phase I
SMART Anti-Gamma Interferon Antibody	Multiple sclerosis (MS), systemic lupus erythematosus (SLE)	Preclinical
Cancer		
SMART M195 Antibody	Myeloid leukemia (AML) and chronic myeloid leukemia CML)	Phase I / II
SMART Anti-Tac Antibody (3)	Chronic lymphocytic leukemia (CLL) and adult T cell leukemia (ATL) and certain lymphomas	Phase I / II
Recombinant Anti-Tac Immunotoxin (2)	Chronic lymphocytic leukemia (CLL) and adult T cell leukemia (ATL) and certain lymphomas	Preclinical
SMART ABL 364 Antibody	Certain epithelial cell cancers including lung, colorectal and breast tumors	Preclinical
Other Diseases		
SMART Anti-Platelet Antibody	Arterial thrombosis (cardiac infarction, stroke)	Preclinical
SMART Anti-CD18 Antibody	Reperfusion injury (myocardial infarction, stroke), adult respiratory distress syndrome (ARDS), trauma, shock, rheumatoid arthritis	Preclinical
SMART Anti-L-Selectin Antibody	Reperfusion injury (myocardial infarction, stroke), adult respiratory distress syndrome (ARDS), trauma, shock, rheumatoid arthritis	Preclinical

(1) "Preclinical Development" includes *in vitro* testing, efficacy and toxicology testing in animals, process development and manufacturing scale-up prior to possible submission of an IND.

(2) The SMART Anti-Tac Antibody, and the Recombinant Anti-Tac Immunotoxin, each may have possible applications to the treatment of both autoimmune conditions and cancer.

4. Clinical Results With Human Monoclonal Antibodies

Generation of human mAbs, using traditional methods for antibody production presents difficulties from both ethical and technical viewpoints. Ethical considerations do not allow for the immunization of human volunteers with antigens that may cause harm to the volunteers. In addition, many desired target antigens for human therapeutic mAbs are themselves human proteins such as, for example, the human IL-2 receptor. In such cases, immunization of a human volunteer with this "self" antigen probably would not lead to an antibody response, and if an antibody response were to occur, it could have potentially dangerous health effects.

One exception to this situation is the case of normal human pathogens such as viruses, bacteria, fungi or protozoa, to which individuals may have already been exposed and have developed a humoral immune response. Other techniques have been developed to generate human monoclonal antibodies to human antigens as possible therapeutic targets. These techniques have included fusion of human lymphocytes to form human hybridomas, transformation of human lymphocytes to continuous cell lines by Epstein-Barr virus infection, development of SCID mice with functional human lymphocytes, development of combinatorial libraries of human immunoglobulin genes, and long-term culture of human splenocytes capable of producing human antibodies following in vitro antigen stimulation. The latter two technologies are described elsewhere in this volume; what follows here is a description of the development of a particular type of human heterohybridoma termed a "trioma".

4.1 TRIOMAS

It is possible to generate a totally human mAb from the lymphocytes of patients who have been immunized against the target antigen (for example, hepatitis B virus) or where immunity may already exist from an earlier infection with, for example, any of the several herpes viruses. However, it has proven be difficult to produce a stable human hybridoma from the human lymphocytes. Years of experimental work with murine systems had provided several continuous myeloma cell lines, such as SP2/0, which functioned very well as fusion partners for murine lymphocytes to generate murine hybridomas. This has not been the case for the production of human hybridomas due to a lack of human myeloma cell lines capable of serving as fusion partners.

Fusion of human lymphocytes with murine myeloma cell lines invariably results in a genetically unstable hybridoma that gradually loses the human chromosomes encoding for human antibody. The most successful technique to overcome this difficulty has been to make use of the stable murine-human cell lines that do result from such fusions. Even those these cells do not produce antibody, some of them have adapted to accommodate human chromosomes. These stable murine-human hybridomas can then be used as a fusion partner for human lymphocytes. The result of this fusion (murine-human hybridoma x human lymphocyte) can yield a stable "heterohybridoma" or "trioma" capable of producing human monoclonal antibody.

This technique was developed independently by researchers at Stanford University[15] who termed their cell lines "heterohybridomas" and at the Sandoz Research Institute who termed the murine-human-human hybridomas as "triomas". The latter group, initially developed a murine-human fusion partner termed SPAZ-4 from fusion of the murine cell line SP2/0 with human peripheral blood lymphocytes[16]. The stable murine-human SPAZ-4 cell line has been used as the fusion partner with human lymphocytes to yield several triomas producing human antibodies, two of which are in preclinical development and two that are advancing in clinical trials as shown in Table 2.

Table 2. Human mAbs Produced by Triomas

Product in Research or Development	Potential Therapeutic Indications	Development Status (1)
Viral Infections		
Human Anti-CMV Antibody	Cytomegalovirus (CMV) infections associated with AIDS, bone marrow and organ transplants	Completed Phase I/II
Human Anti-Hepatitis B Antibody	Chronic active hepatitis B and end-stage liver disease from chronic active hepatitis B	Completed Phase I/II
Human Anti-Herpes Antibody	Genital and neonatal herpes	Preclinical
Human Anti-Varicella Zoster Antibody	Shingles (zoster) and severe neonatal infections.	Preclinical

4.2 HUMAN ANTI-CMV ANTIBODY MSL 109

The human anti-cytomegalovirus (anti-CMV) antibody designated MSL 109 was generated by Östberg et. al. using the trioma procedure which they originally developed. Human splenocytes were stimulated in vitro with lysates of CMV-infected fibroblasts and then fused with the SPAZ-4 (murine-human) fusion partner. The resulting MSL 109 trioma cell line was selected for its production of mAb with high reactivity to the neutralizing gH antigen of human CMV.

MSL 109 is an IgG1-kappa human mAb with an ED_{50} of less than 0.5 µg/mL for clinical isolates of human CMV. This human mAb acts additively with ganciclovir in vitro and in a rabbit CMV retinitis model developed by Dunkel et al[17]. Preclinical studies with non-human primates showed that MSL 109 is essentially free of toxicity after doses of up to 16 mg/kg and did not result in HAMA formation when administered to primates at doses of 0.5 mg/kg for up to one year[18].

4.2.1 *Bone Marrow Transplant Patients.* Two Phase I clinical trials with MSL 109 have been conducted with patients undergoing bone marrow transplantation. In the first study, eight patients received doses of 0.5 or 2.0 mg/kg every two weeks for a total of six doses. There were no signs or symptoms of toxicity attributed to this human mAb and no HAMA development. The trough serum levels of MSL 109 were far in excess of the ED_{50} values for CMV and ranged from 9 to 13 µg/mL for the low dose group and from 23 to 261 µg/mL for the high dose group[19]. The second Phase I trial administered doses of 0.05, 0.25 and 0.5 mg/kg approximately every three weeks for six months with minimal side effects, no evidence of dose-related adverse events, and no development of HAMA[20]. The average serum half-life of MSL 109 in patients was approximately six days in the first trial and approximately seventeen days in the second trial. The unexpectedly short half-life in the first trial may have been due to difficulties in defining a two-compartment model from the limited number of serum samples collected. Other humanized and human antibodies typically have exhibited serum half-lives on the order of two weeks.

4.2.2 *CMV Infections In AIDS Patients.* Two additional Phase I/II trials have been conducted with MSL 109 in AIDS patients. The first study enrolled twenty-three CMV seropositive patients who were shedding CMV into their urine[21]. Doses ranging from 0.125 mg/kg to 10 mg/kg were administered every two weeks for twenty-four weeks. All doses were well tolerated with only one adverse event reported (mild headache). Data analysis has not been completed for virus shedding and pharmacokinetics. The second trial enrolled AIDS patients receiving ganciclovir or foscarnet for CMV retinitis[22]. MSL 109 was given in doses from 1 mg/kg to 5 mg/kg, or fixed doses of 20 mg or 80 mg, every two weeks along with the standard ganciclovir or foscarnet maintenance therapy. As before, the MSL 109 was

well tolerated and there was preliminary evidence that progression of the retinitis was delayed (mean of 202 days) two-fold longer than historical controls with ganciclovir or foscarnet alone (100 days).

4.2.3 *Neonatal CMV Infections.* A Phase I study sponsored by the NIAID Collaborative Antiviral Study Group is underway in which neonates with congenital CMV infection without symptoms of CNS involvement will receive three doses of either 1, 5 or 25 mg/kg of MSL 109 at two week intervals. This study with MSL 109 was undertaken due to the severity of neonatal CMV infections, the need for more effective therapies, and the safety shown in preclinical studies in neonatal non-human primates and in clinical trials (above).

4.3 HUMAN ANTI-HBV ANTIBODY

Östberg et al also employed their trioma technology to generate an IgG1-lambda human mAb, termed OST 577, directed against the "a" determinant of the hepatitis B surface antigen (HBsAg) to which the OST 577 antibody binds tightly (Ka = 4 x 10_9 M-1). This human antibody was developed from peripheral blood lymphocytes from volunteers showing high titers of anti-HBV antibody after vaccination with a commercial HBV vaccine[23].

Preclinical safety studies showed that OST 577 was well tolerated in single dose (rodent) and multiple dose (non-human primates) studies. Pharmacokinetic studies conducted with rhesus monkeys (0.5 mg/kg) showed peak plasma levels of approximately 11 µg/mL and a plasma half-life of approximately two weeks and no evidence of immunogenicity after a one-month multiple dose regimen.

In a single-animal prophylaxis study, one dose of OST 577 (5 mg/kg) was able to delay the onset of hepatitis B infection in a chimpanzee to 41 weeks, approximately five-fold longer than the historic median value of eight weeks[24].

4.3.1 *Liver Transplant In Hepatitis B Patients.* The initial clinical trial with OST 577 enrolled patients undergoing liver transplant for end stage liver disease due to hepatitis B. The five patients in this trial received a total of 70 to 90 mg of OST 577 just before, during and after liver transplantation and maintenance doses of 10 mg at increasingly longer time intervals for up to 23 months. All five patients converted to seronegativity and none showed further symptoms of clinical hepatitis.

Four of the five patients have now been followed more than 3 years after treatment. One patient died of causes unrelated to hepatitis infection while the remaining four patients have now been followed more than three years[25]. Two of the four surviving patients have remained free of any evidence of HBV reinfection including the absence of HBeAg, HBsAg and negative findings for HBV DNA by PCR. The remaining two patients seroconverted to HBsAg+ approximately 250 days after transplantation but have not shown clinical signs of HBV infection. The HBV virus present in these patients exhibited alterations in the HBsAg gene resulting in reduced reactivity with the OST 577 mAb and a possible reduction in virulence that was confirmed in a chimpanzee challenge study[26]. Higher level maintenance doses (viz, > 10 mg per dose) may be able to prevent the emergence of these "escape" variants of HBV.

Overall, this trial showed that OST 577 could be given repeatedly over many months without adverse events, without the formation of HAMA and, even at low doses, protect against HBV re-infection of the transplanted liver. In a clinical situation where nearly all such transplanted patients' livers would be re-infected with HBV, these results indicate that prophylaxis with this human mAb could make such transplantation procedures possible.

4.3.2 *Chronic Active Hepatitis B (CAHB).* OST 577 has also been administered to twelve patients in a multiple-dose trial for the treatment of chronic active hepatitis B (CAHB)[27]. In this Phase I/II trial, patients received seven to nine doses of OST 577 at levels of 0.5, 1 or 2 mg/kg over a thirty-five to thirty-seven day period. The human mAb was well tolerated at doses of 0.5 and 1 mg/kg with one patient in the 1 mg/kg group showing hives that responded to treatment with an antihistamine. Two of the three patients receiving 2 mg/kg OST 577 experienced chills and one developed fever and hypotension, possibly from immune complex formation (these patients can exhibit very high serum titers of HBsAg). Several of the

surrogate markers for HBV infection were decreased by 50% or more with reductions in liver enzymes in 5 of 10 patients, reductions in HBsAg in 10 of 12 patients and in viral DNA for 5 of 9 patients. These early results suggest that continued treatment with the human OST 577 mAb beyond one month, as in the liver transplant trial, may provide even greater benefit for CAHB patients. Clinical trial protocols are being developed to test this hypothesis.

5. Conclusion

The ability to produce monoclonal antibodies on a large scale allowed this class of natural proteins to be considered for use as pharmaceutical agents. After more than a decade of clinical trials, it has become clear that many of the drawbacks to the clinical use of monoclonal antibodies is due to their murine origin. New methods have been developed to generate humanized and human antibodies which overcome the major difficulties of HAMA response and short serum half-life of murine antibodies. Results from on-going clinical trials with this new generation of antibodies indicate that these human and humanized antibodies will be successful therapeutic agents for the treatment of human disease.

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