



(51) International Patent Classification:

A61K 39/395 (2006.01) A61P 13/12 (2006.01)
C07K 16/28 (2006.01)

(21) International Application Number:

PCT/US2016/066033

(22) International Filing Date:

9 December 2016 (09.12.2016)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

62/265,322 9 December 2015 (09.12.2015) US

(71) Applicant: CEDARS-SINAI MEDICAL CENTER
[US/US]; 8700 Beverly Blvd., Los Angeles, California
90048 (US).

(72) Inventors: JORDAN, Stanley; 2104 Pine Avenue, Man-
hattan Beach, California 90266 (US). VO, Ashley; 18857
Beechtree Lane, Porter Ranch, California 91326 (US).
CHOI, Jua; 11662 Ricasoli Way, Porter Ranch, California
91326 (US).

(74) Agents: VAKHARIA-RAO, Hema et al.; Nixon Peabody
LLP, 300 South Grand Avenue, Suite 4100, Los Angeles,
California 90071-3151 (US).

(81) Designated States (unless otherwise indicated, for every
kind of national protection available): AE, AG, AL, AM,
AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY,
BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM,
DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT,
HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KH, KN,
KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA,
MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG,
NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS,
RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY,
TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN,
ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every
kind of regional protection available): ARIPO (BW, GH,
GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ,
TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU,
TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE,
DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU,
LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK,
SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ,
GW, KM, ML, MR, NE, SN, TD, TG).

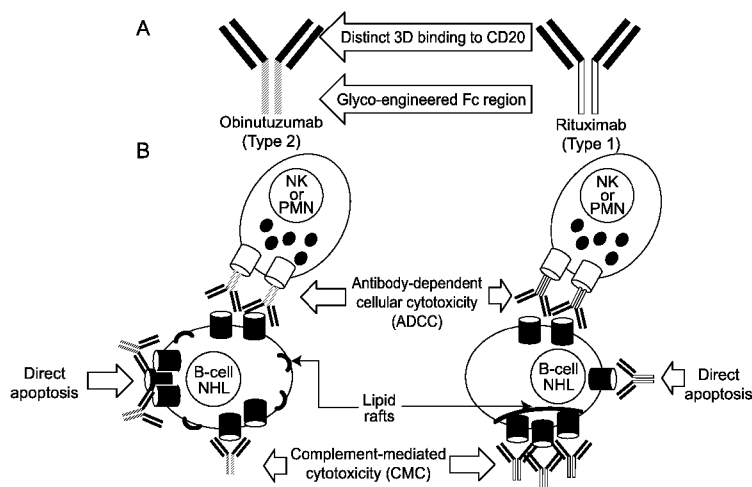
Published:

— with international search report (Art. 21(3))

(54) Title: METHODS FOR TREATING NEPHROTIC SYNDROME

FIG. 1

Obinutuzumab v. Rituximab: Distinct Mechanism of Action of Depleting B-cells



(57) Abstract: Described herein are methods for treating nephrotic syndrome using an anti-CD20 antibody. In one embodiment, that anti-CD20 antibody is Obinutuzumab.

METHODS FOR TREATING NEPHROTIC SYNDROME

CROSS-REFERENCE TO RELATED APPLICATION

[0001] This application claims the benefit of U.S. Provisional Application No. 62/265,322, filed December 9, 2015, which is hereby incorporated by reference herein in its entirety

[0002] TECHNICAL FIELD

[0003] The invention relates to treatment for nephrotic syndrome, including Rituximab-resistant nephrotic syndrome.

BACKGROUND

[0004] All publications herein are incorporated by reference to the same extent as if each individual publication or patent application was specifically and individually indicated to be incorporated by reference. The following description includes information that may be useful in understanding the present invention. It is not an admission that any of the information provided herein is prior art or relevant to the presently claimed invention, or that any publication specifically or implicitly referenced is prior art.

[0005] Nephrotic syndrome (NS) is a group of symptoms that include protein in the urine, low blood protein levels, high cholesterol levels, high triglyceride levels and swelling. NS is caused by different disorders that damage the kidneys and results in release of too much protein in the urine. Exemplary conditions that result in NS include cancer, genetic disorders, immune disorders, diabetes, systemic lupus, multiple myeloma, infections and so on.

[0006] Obinutuzumab (Obi) is a novel FDA approved glycol-rich monoclonal antibody which binds CD20 at a different epitope than Rituximab, another anti-CD20 antibody also approved by the FDA. Thus, Obinutuzumab has been termed a type 2 anti-CD20 and also has a different mechanism of action than Rituximab. Obi depletes B-cells primarily through homotropic aggregation and antibody-dependent cellular cytotoxicity. These features give it many distinctions from Rituximab. The differences in mechanism of action between Obi and Rituximab are shown in **Fig. 1**. Obi was thought to be more effective in depleting B-cells in remote areas of the body such as the spleen and lymph nodes which could still act as mediators of disease. Indeed, this was found to be true in the pivotal trials of Obi in chronic lymphocytic leukemia and non-Hodgkin's Lymphoma (NHL) where Obi was found superior to Rituximab in inducing long-term remissions of these diseases. Analysis of lymphoid and

splenic tissues from treated patients showed a marked difference in removal of CD20+ tumor cells in Obi patients compared to Rituximab.

[0007] Due to either significant side effects or relative inefficacies of many existing therapeutic agents for nephrotic syndrome, there is an unmet clinical need for novel therapies for treatment of nephrotic syndrome that are more effective and less toxic. Most patients with NS have an immunologic cause for the disease and current therapies are directed to modification of the immune system. There are no FDA approved therapies and current therapies have to be maintained on a daily basis, often for life which is often toxic with limited efficacy. Accordingly, provided herein are methods for treating nephrotic syndrome using Obinutuzumab.

SUMMARY

[0008] The following embodiments and aspects thereof are described and illustrated in conjunction with compositions and methods which are meant to be exemplary and illustrative, not limiting in scope.

[0009] Provided herein are methods for treating, inhibiting or reducing severity of nephrotic syndrome in a subject in need thereof comprising, consisting of or consisting essentially of providing an anti-CD20 antibody and administering an effective amount of the antibody to the subject, so as to treat, inhibit or reduce the severity of nephrotic syndrome in the subject.

[0010] Also provided herein are methods for treating, inhibiting or reducing severity of nephrotic syndrome in a subject in need thereof comprising, consisting of or consisting essentially of administering an effective amount of an anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of nephrotic syndrome in the subject.

[0011] Further provided herein are methods are treating, inhibiting or reducing severity of glomerulonephritis (GN) in a subject in need thereof comprising, consisting of or consisting essentially of providing an anti-CD20 antibody; and administering an effective amount of the antibody to the subject, so as to treat, inhibit or reduce the severity of glomerulonephritis in the subject.

[0012] Also provided herein are methods are treating, inhibiting or reducing severity of glomerulonephritis (GN) in a subject in need thereof comprising, consisting of or consisting

essentially of administering an effective amount of an anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of glomerulonephritis in the subject.

[0013] In some embodiments, glomerulonephritis is membranous glomerulonephritis, focal segmental glomerulosclerosis (FSGS), minimal change nephrotic syndrome, cryoglobulinemic glomerulonephritis, IgA nephropathy.

[0014] In some embodiments, the anti-CD20 antibody is Obinutuzumab, Rituximab or a combination thereof. In one embodiment, the anti-CD20 antibody is Obinutuzumab.

[0015] In one embodiment, the subject has drug-resistant nephrotic syndrome. In an embodiment, the drug-resistant nephrotic syndrome is Rituximab-resistant nephrotic syndrome.

[0016] In some embodiments, nephrotic syndrome in the subject is caused by any one or more of minimal change disease (MCD), focal segmental glomerulosclerosis (FSGS), membranous glomerulonephritis (MGN), IgA nephropathy and cryoglobulinemic glomerulonephritis.

[0017] In some embodiments, the subject has been administered standard-of-care treatment prior to administration of anti-CD20 antibody. In an embodiment, the subject does not respond to standard-of-care treatment prior to administration of anti-CD20 antibody.

[0018] In some embodiments, the subject has not been administered standard-of-care treatment prior to administration of anti-CD20 antibody. In some embodiments, the anti-CD20 antibody is administered simultaneously or sequentially with standard-of-care treatment.

[0019] In some embodiments, the standard-of-care treatments are any one or more of high dose prednisone (steroids), mycophenolic acid (cellcept) and optionally calcineurin inhibitors. In some embodiments, the calcineurin inhibitors are cyclosporine, tacrolimus, or a combination thereof.

[0020] In some embodiments, the effective amount of the anti-CD20 antibody (for example, is Obinutuzumab) is about 0.01-0.05 mg/kg/day, 0.05-0.1 mg/kg/day, 0.1 to 0.5mg/kg/day, 0.5 to 1mg/kg/day, 1 to 2mg/kg/day, 2 to 3mg/kg/day, 3 to 4mg/kg/day, 0.5 to 5 mg/kg/day, 1 to 5mg/kg/day, 3 to 5mg/kg/day, 5 to 10 mg/kg/day, 10 to 20 mg/kg/day, 20 to 50 mg/kg/day, 50 to 100 mg/kg/day, 100 to 200 mg/kg/day, 200 to 300 mg/kg/day, 300 to 400 mg/kg/day,

400 to 500 mg/kg/day, 500 to 600 mg/kg/day, 600 to 700mg/kg/day, 700 to 800mg/kg/day, 800 to 900mg/kg/day or 900 to 1000 mg/kg/day. In some embodiments, the anti-CD20 antibody (for example, is Obinutuzumab) is administered to the subject before, during, or after the subject develops nephrotic syndrome. In some embodiments, the anti-CD20 antibody (for example, is Obinutuzumab) is administered to the subject 1-3 times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody (for example, is Obinutuzumab) is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, or 1-5 years.

[0021] In various embodiments, the anti-CD20 antibody or fragment thereof is administered to the subject before, during, or after the subject develops the nephrotic syndrome. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject 1-3 times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, or 1-5 years.

[0022] In some embodiments, the subject has been administered standard-of-care treatment for nephrotic syndrome prior to administration of the anti-CD20 antibody (for example, prior to administration of Obinutuzumab). In some embodiments, the subject does not respond to or poorly responds to standard-of-care treatment for nephrotic syndrome prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the subject has not been administered standard-of-care treatment for nephrotic syndrome prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the standard-of-care treatment for nephrotic syndrome is administered simultaneously or sequentially with the anti-CD20 antibody (for example, Obinutuzumab).

[0023] In exemplary embodiments, the standard-of-care treatment for nephrotic syndrome is steroid, cyclophosphamide, cyclosporine, tacrolimus and Rituximab. Nephrotic syndrome treatments include high dose prednisone (steroids), mycophenolic acid (cellcept) and occasionally calcineurin inhibitors (cyclosporine and tacrolimus). All are used in various combinations. In addition, high dose Cytoxan is used in severe cases. This has a high side effect profile and is not used routinely. Because of this, and the relative inefficacy of many of the drugs, there is a large unmet need for new therapies that are less toxic in patients with nephrotic syndrome

[0024] Also provided herein are methods for treating, inhibiting or reducing severity of nephrotic syndrome in a subject in need thereof comprising, consisting of or consisting essentially of providing an anti-CD20 antibody and administering an effective amount of an anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of nephrotic syndrome in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years, every 10 year, for lifetime of the subject or combinations thereof. In an embodiment, the subject is human.

[0025] Also provided herein are methods for treating, inhibiting or reducing severity of nephrotic syndrome in a subject in need thereof comprising, consisting of or consisting essentially of administering an effective amount of an anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of nephrotic syndrome in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years, every 10 year, for lifetime of the subject or combinations thereof. In an embodiment, the subject is human.

[0026] Further provided herein are methods are treating, inhibiting or reducing severity of glomerulonephritis (GN) in a subject in need thereof comprising, consisting of or consisting essentially of providing an anti-CD20 antibody and administering an effective amount of the antibody to the subject, so as to treat, inhibit or reduce the severity of glomerulonephritis in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram

Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years, every 10 year, for lifetime of the subject or combinations thereof. In an embodiment, the subject is human.

[0027] Also provided herein are methods are treating, inhibiting or reducing severity of glomerulonephritis (GN) in a subject in need thereof comprising, consisting of or consisting essentially of administering an effective amount of an anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of glomerulonephritis in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years, every 10 year, for lifetime of the subject or combinations thereof. In an embodiment, the subject is human.

[0028] Also provided herein are methods for desensitizing a subject to antibody mediated transplant rejection (ABMR) in the subject in need thereof. The methods include providing an anti-CD20 antibody (for example, Obinutuzumab) and administering an effective amount of the antibody to the subject so as to desensitize the subject to ABMR. In some embodiments, the subjects are desensitized prior to transplant. In some embodiments, the subjects are desensitized during transplant. In some embodiments, the subjects are desensitized prior to, during and after transplant. In various embodiments, desensitizing the subject includes treating the subject with the anti-CD20 antibody (for example, Obinutuzumab) and Intravenous immunoglobulin (IVIG), with or without plasmapheresis. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are

administered sequentially. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are administered simultaneously. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2.

[0029] Also provided herein are methods for desensitizing a subject to antibody mediated transplant rejection (ABMR) in the subject in need thereof. The methods include administering an effective amount of an anti-CD20 antibody (for example, Obinutuzumab) to the subject so as to desensitize the subject to ABMR. In some embodiments, the subjects are desensitized prior to transplant. In some embodiments, the subjects are desensitized during transplant. In some embodiments, the subjects are desensitized prior to, during and after transplant. In various embodiments, desensitizing the subject includes treating the subject with the anti-CD20 antibody (for example, Obinutuzumab) and Intravenous immunoglobulin (IVIG), with or without plasmapheresis. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are administered sequentially. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are administered simultaneously. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2.

BRIEF DESCRIPTION OF THE DRAWINGS

[0030] Exemplary embodiments are illustrated in referenced figures. It is intended that the embodiments and figures disclosed herein are to be considered illustrative rather than restrictive.

[0031] **Fig. 1** depicts in accordance with various embodiments of the invention, the mechanism of action of Rituximab (Type I) versus Obinutuzumab (Type II).

[0032] **Fig. 2A-Fig. 2C** depict in accordance with various embodiments of the invention, (**Fig. 2A**) renal biopsy revealing membranous lupus glomerulonephritis; (**Fig. 2B**) the effect of Rituximab on anti-nuclear antibody (ANA) titers at the time of diagnosis and over 3 years; (**Fig. 2C**) anti-dsDNA Antibodies at Systemic Lupus Erythematosus (SLE) relapse and up to 8 weeks post-treatment

[0033] **Fig. 3** depicts in accordance with various embodiments of the invention, the effect of Obi in a patient with recurrence of focal segmental glomerulosclerosis (FSGS) and with antibody mediated rejection (ABMR).

[0034] **Fig. 4** depicts in accordance with various embodiments of the invention, the effect of Obi in a patient with drug- and Rituximab-resistant minimal change disease nephrotic syndrome (NS).

[0035] **Fig. 5** depicts in accordance with various embodiments of the invention, pre- and post-Obi urine total protein/creatinine ratios (UTP/Cr) through 12M with linear trends of the 12 nephrotic syndrome patients.

[0036] **Fig. 6** depicts in accordance with various embodiments of the invention, UTP/Cr values over time. Significant reductions were seen in these values after Obi therapy in the nephrotic syndrome patients.

[0037] **Fig. 7.** depicts in accordance with various embodiments of the invention, the effects of Obi on anti-PLA2R in a patient with primary MGN and anti-dsDNA antibodies in a patient with membranous SLE.

[0038] **Fig. 8** depicts in accordance with various embodiments of the invention, mean serum creatinine (mg/dl) values post treatment in the nephrotic syndrome patients. Significance was seen at 1M post treatment but overall creatinine remained stable in this patient population.

DETAILED DESCRIPTION

[0039] All references cited herein are incorporated by reference in their entirety as though fully set forth. Unless defined otherwise, technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Allen *et al.*, *Remington: The Science and Practice of Pharmacy* 22nd ed., Pharmaceutical Press (September 15, 2012); Hornyak *et al.*, *Introduction to Nanoscience and Nanotechnology*, CRC Press (2008); Singleton and Sainsbury, *Dictionary of Microbiology and Molecular Biology* 3rd ed., revised ed., J. Wiley & Sons (New York, NY 2006); Smith,

March's Advanced Organic Chemistry Reactions, Mechanisms and Structure 7th ed., J. Wiley & Sons (New York, NY 2013); Singleton, *Dictionary of DNA and Genome Technology 3rd ed.*, Wiley-Blackwell (November 28, 2012); and Green and Sambrook, *Molecular Cloning: A Laboratory Manual 4th ed.*, Cold Spring Harbor Laboratory Press (Cold Spring Harbor, NY 2012), provide one skilled in the art with a general guide to many of the terms used in the present application. For references on how to prepare antibodies, see Greenfield, *Antibodies A Laboratory Manual 2nd ed.*, Cold Spring Harbor Press (Cold Spring Harbor NY, 2013); Köhler and Milstein, *Derivation of specific antibody-producing tissue culture and tumor lines by cell fusion*, Eur. J. Immunol. 1976 Jul, 6(7):511-9; Queen and Selick, *Humanized immunoglobulins*, U. S. Patent No. 5,585,089 (1996 Dec); and Riechmann *et al.*, *Reshaping human antibodies for therapy*, Nature 1988 Mar 24, 332(6162):323-7.

[0040] For references on pediatrics, see Schwartz *et al.*, *The 5-Minute Pediatric Consult 4th ed.*, Lippincott Williams & Wilkins, (June 16, 2005); Robertson *et al.*, *The Harriet Lane Handbook: A Manual for Pediatric House Officers 17th ed.*, Mosby (June 24, 2005); and Hay *et al.*, *Current Diagnosis and Treatment in Pediatrics (Current Pediatrics Diagnosis & Treatment) 18th ed.*, McGraw-Hill Medical (September 25, 2006).

[0041] One skilled in the art will recognize many methods and materials similar or equivalent to those described herein, which could be used in the practice of the present invention. Other features and advantages of the invention will become apparent from the following detailed description, taken in conjunction with the accompanying drawings, which illustrate, by way of example, various features of embodiments of the invention. Indeed, the present invention is in no way limited to the methods and materials described. For convenience, certain terms employed herein, in the specification, examples and appended claims are collected here.

[0042] Unless stated otherwise, or implicit from context, the following terms and phrases include the meanings provided below. Unless explicitly stated otherwise, or apparent from context, the terms and phrases below do not exclude the meaning that the term or phrase has acquired in the art to which it pertains. The definitions are provided to aid in describing particular embodiments, and are not intended to limit the claimed invention, because the scope of the invention is limited only by the claims. Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs.

[0043] As used herein the term “comprising” or “comprises” is used in reference to compositions, methods, and respective component(s) thereof, that are useful to an embodiment, yet open to the inclusion of unspecified elements, whether useful or not. It will be understood by those within the art that, in general, terms used herein are generally intended as “open” terms (e.g., the term “including” should be interpreted as “including but not limited to,” the term “having” should be interpreted as “having at least,” the term “includes” should be interpreted as “includes but is not limited to,” etc.). Although the open-ended term “comprising,” as a synonym of terms such as including, containing, or having, is used herein to describe and claim the invention, the present invention, or embodiments thereof, may alternatively be described using alternative terms such as “consisting of” or “consisting essentially of.”

[0044] Unless stated otherwise, the terms “a” and “an” and “the” and similar references used in the context of describing a particular embodiment of the application (especially in the context of claims) can be construed to cover both the singular and the plural. The recitation of ranges of values herein is merely intended to serve as a shorthand method of referring individually to each separate value falling within the range. Unless otherwise indicated herein, each individual value is incorporated into the specification as if it were individually recited herein. All methods described herein can be performed in any suitable order unless otherwise indicated herein or otherwise clearly contradicted by context. The use of any and all examples, or exemplary language (for example, “such as”) provided with respect to certain embodiments herein is intended merely to better illuminate the application and does not pose a limitation on the scope of the application otherwise claimed. The abbreviation, “e.g.” is derived from the Latin *exempli gratia*, and is used herein to indicate a non-limiting example. Thus, the abbreviation “e.g.” is synonymous with the term “for example.” No language in the specification should be construed as indicating any non-claimed element essential to the practice of the application.

[0045] As used herein, the terms “treat,” “treatment,” “treating,” or “amelioration” when used in reference to a disease, disorder or medical condition, refer to both therapeutic treatment and prophylactic or preventative measures, wherein the object is to prevent, reverse, alleviate, ameliorate, inhibit, lessen, slow down or stop the progression or severity of a symptom or condition. The term “treating” includes reducing or alleviating at least one adverse effect or symptom of a condition. Treatment is generally “effective” if one or more symptoms or clinical markers are reduced. Alternatively, treatment is “effective” if the

progression of a disease, disorder or medical condition is reduced or halted. That is, “treatment” includes not just the improvement of symptoms or markers, but also a cessation or at least slowing of progress or worsening of symptoms that would be expected in the absence of treatment. Also, “treatment” may mean to pursue or obtain beneficial results, or lower the chances of the individual developing the condition even if the treatment is ultimately unsuccessful. Those in need of treatment include those already with the condition as well as those prone to have the condition or those in whom the condition is to be prevented.

[0046] “Beneficial results” or “desired results” may include, but are in no way limited to, lessening or alleviating the severity of the disease condition, preventing the disease condition from worsening, curing the disease condition, preventing the disease condition from developing, lowering the chances of a patient developing the disease condition, decreasing morbidity and mortality, and prolonging a patient’s life or life expectancy. As non-limiting examples, “beneficial results” or “desired results” may be alleviation of one or more symptom(s), diminishment of extent of the deficit, stabilized (i.e., not worsening) state of pancreatic cancer, delay or slowing of nephrotic syndrome, and amelioration or palliation of symptoms associated with nephrotic syndrome.

[0047] “Diseases”, “conditions” and “disease conditions,” as used herein may include, but are in no way limited to any form of nephrotic syndrome.

[0048] As used herein, the term “administering,” refers to the placement of an agent or a composition as disclosed herein into a subject by a method or route which results in at least partial localization of the agents or composition at a desired site. “Route of administration” may refer to any administration pathway known in the art, including but not limited to oral, topical, aerosol, nasal, via inhalation, anal, intra-anal, peri-anal, transmucosal, transdermal, parenteral, enteral, or local. “Parenteral” refers to a route of administration that is generally associated with injection, including intratumoral, intracranial, intraventricular, intrathecal, epidural, intradural, intraorbital, infusion, intracapsular, intracardiac, intradermal, intramuscular, intraperitoneal, intrapulmonary, intraspinal, intrasternal, intrathecal, intrauterine, intravascular, intravenous, intraarterial, subarachnoid, subcapsular, subcutaneous, transmucosal, or transtracheal. Via the parenteral route, the agent or composition may be in the form of solutions or suspensions for infusion or for injection, or as lyophilized powders. Via the enteral route, the agent or composition can be in the form of

capsules, gel capsules, tablets, sugar-coated tablets, syrups, suspensions, solutions, powders, granules, emulsions, microspheres or nanospheres or lipid vesicles or polymer vesicles allowing controlled release. Via the topical route, the agent or composition can be in the form of aerosol, lotion, cream, gel, ointment, suspensions, solutions or emulsions. In an embodiment, agent or composition may be provided in a powder form and mixed with a liquid, such as water, to form a beverage. In accordance with the present invention, “administering” can be self-administering. For example, it is considered as “administering” that a subject consumes a composition as disclosed herein.

[0049] As used herein, a “subject” means a human or animal. Usually the animal is a vertebrate such as a primate, rodent, domestic animal or game animal. Primates include chimpanzees, cynomolgous monkeys, spider monkeys, and macaques, e.g., Rhesus. Rodents include mice, rats, woodchucks, ferrets, rabbits and hamsters. Domestic and game animals include cows, horses, pigs, deer, bison, buffalo, feline species, e.g., domestic cat, and canine species, e.g., dog, fox, wolf. The terms, “patient”, “individual” and “subject” are used interchangeably herein. In an embodiment, the subject is mammal. The mammal can be a human, non-human primate, mouse, rat, dog, cat, horse, or cow, but are not limited to these examples. In an embodiment, the subject is human. In addition, the methods described herein can be used to treat domesticated animals and/or pets.

[0050] “Mammal” as used herein refers to any member of the class Mammalia, including, without limitation, humans and nonhuman primates such as chimpanzees and other apes and monkey species; farm animals such as cattle, sheep, pigs, goats and horses; domestic mammals such as dogs and cats; laboratory animals including rodents such as mice, rats and guinea pigs, and the like. The term does not denote a particular age or sex. Thus, adult and newborn subjects, as well as fetuses, whether male or female, are intended to be included within the scope of this term.

[0051] A subject can be one who has been previously diagnosed with or identified as suffering from or having a condition in need of treatment (e.g., nephrotic syndrome) or one or more complications related to the condition, and optionally, have already undergone treatment for the condition or the one or more complications related to the condition. Alternatively, a subject can also be one who has not been previously diagnosed as having a condition or one or more complications related to the condition. For example, a subject can be one who exhibits one or more risk factors for a condition or one or more complications

related to the condition or a subject who does not exhibit risk factors. For example, a subject can be one who exhibits one or more symptoms for a condition or one or more complications related to the condition or a subject who does not exhibit symptoms. A “subject in need” of diagnosis or treatment for a particular condition can be a subject suspected of having that condition, diagnosed as having that condition, already treated or being treated for that condition, not treated for that condition, or at risk of developing that condition.

[0052] A therapeutically or prophylactically significant reduction in a symptom is, e.g. at least about 10%, at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, at least about 100%, at least about 125%, at least about 150% or more in a measured parameter as compared to a control or non-treated subject or the state of the subject prior to administering the anti-CD20 antibody (for example, Obinutuzumab). Measured or measurable parameters include clinically detectable markers of disease, for example, elevated or depressed levels of a biological marker, as well as parameters related to a clinically accepted scale of symptoms or markers for fibrosis and/or inflammation. It will be understood, however, that the total daily usage of the compositions and formulations as disclosed herein will be decided by the attending physician within the scope of sound medical judgment. The exact amount required will vary depending on factors such as the type of disease being treated, gender, age, and weight of the subject.

[0053] Provided herein are methods for treating, inhibiting, reducing the severity of and/or delaying progression of nephrotic syndrome in a subject in need thereof. The methods comprise, consist of or consist essentially of providing an anti-CD20 antibody and administering an effective amount of the anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of nephrotic syndrome in the subject. In some embodiments, the methods comprise, consist of or consist essentially of administering an effective amount of an anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of nephrotic syndrome in the subject. In one embodiment, nephrotic syndrome is Rituximab-resistant nephrotic syndrome. In one embodiment, the subject is human. In some embodiments, the anti-CD20 antibody is Obinutuzumab, Rituximab or a combination thereof. In one embodiment, the anti-CD20 antibody is Obinutuzumab. In some embodiments, nephrotic syndrome in the subject is caused by any one or more of minimal change disease, focal sclerosis syndrome, membranous glomerulonephritis, IgA nephropathy and cryoglobulinemic glomerulonephritis.

In various embodiments, the anti-CD20 antibody or a fragment thereof described herein is administered to the subject before, during, or after the subject develops the nephrotic syndrome. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject 1-3 times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, 1-5 years, 5-10 years or for lifetime of the subject. In some embodiments, the subject has been administered standard-of-care treatment for nephrotic syndrome prior to administration of the anti-CD20 antibody (for example, prior to administration of Obinutuzumab). In some embodiments, the subject does not respond to or poorly responds to standard-of-care treatment for nephrotic syndrome prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the subject has not been administered standard-of-care treatment for nephrotic syndrome prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the standard-of-care treatment for nephrotic syndrome is administered simultaneously or sequentially with the anti-CD20 antibody (for example, Obinutuzumab). In exemplary embodiments, the standard-of-care treatment for nephrotic syndrome is steroid, cellcept, cyclophosphamide, cyclosporine, tacrolimus and Rituximab. Nephrotic syndrome treatments include high dose prednisone (steroids), mycophenolic acid (cellcept) and occasionally calcineurin inhibitors (cyclosporine and tacrolimus). All are used in various combinations. In addition, high doses of Cytoxan are used in severe cases. Cytoxan has a high side effect profile and is not used routinely.

[0054] Also provided herein are methods for treating, inhibiting, reducing the severity of and/or delaying progression of glomerulonephritis (GN) in a subject in need thereof. The methods comprise, consist of or consist essentially of providing an anti-CD20 antibody and administering an effective amount of the anti-CD20 antibody to the subject so as to treat, inhibit, reduce the severity of and/or delay progression of glomerulonephritis in the subject. In some embodiments, the methods comprise, consist of or consist essentially of administering an effective amount of an anti-CD20 antibody to the subject so as to treat, inhibit, reduce the severity of and/or delay progression of glomerulonephritis in the subject. In some embodiments, glomerulonephritis is membranous glomerulonephritis, cryoglobulinemic glomerulonephritis, IgA nephropathy, minimal change disease, focal segmental glomerulosclerosis, recurrent nephritis, nephrotic syndrome after kidney transplantation and transplant glomerulopathy with proteinuria and nephrotic syndrome in

kidney transplant patients. In one embodiment, the subject is human. In some embodiments, the anti-CD20 antibody is Obinutuzumab, Rituximab or a combination thereof. In one embodiment, the anti-CD20 antibody is Obinutuzumab. In various embodiments, the anti-CD20 antibody or a fragment thereof described herein is administered to the subject before, during, or after the subject develops glomerulonephritis. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject 1-3 times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, 1-5 years, 5-10 years or for lifetime of the subject. In some embodiments, the subject has been administered standard-of-care treatment for glomerulonephritis prior to administration of the anti-CD20 antibody (for example, prior to administration of Obinutuzumab). In some embodiments, the subject does not respond to or poorly responds to standard-of-care treatment for glomerulonephritis prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the subject has not been administered standard-of-care treatment for glomerulonephritis prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the standard-of-care treatment for glomerulonephritis is administered simultaneously or sequentially with the anti-CD20 antibody (for example, Obinutuzumab). In exemplary embodiments, the standard-of-care treatment for glomerulonephritis includes high dose prednisone (steroids), mycophenolic acid (cellcept), plasmapheresis and/or Rituximab. All are used in various combinations.

[0055] Further provided herein is a method for treating, inhibiting, reducing the severity of and/or delaying progression of Rituximab-resistant nephrotic syndrome. The methods comprise, consist of or consist essentially of providing an anti-CD20 antibody and administering an effective amount of the anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of Rituximab-resistant nephrotic syndrome in the subject. In some embodiments, the methods comprise, consist of or consist essentially of administering an effective amount of an anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of Rituximab-resistant nephrotic syndrome in the subject. In one embodiment, the subject is human. In one embodiment, the anti-CD20 antibody is Obinutuzumab. In various embodiments, the anti-CD20 antibody or a fragment thereof described herein is administered to the subject before, during, or after the subject develops Rituximab-resistant nephrotic syndrome. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject 1-3

times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, 1-5 years, 5-10 years or for lifetime of the subject. In some embodiments, the subject has been administered Rituximab and does not respond to or responds poorly to Rituximab.

[0056] Also provided herein is a method for treating, inhibiting, reducing the severity of and/or delaying progression of membranous glomerulonephritis. The methods comprise, consist of or consist essentially of providing an anti-CD20 antibody and administering an effective amount of the anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of membranous glomerulonephritis in the subject. In some embodiments, the methods comprise, consist of or consist essentially of administering an effective amount of an anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of membranous glomerulonephritis in the subject. In one embodiment, the subject is human. In some embodiments, the anti-CD20 antibody is Obinutuzumab, Rituximab or a combination thereof. In one embodiment, the anti-CD20 antibody is Obinutuzumab. In various embodiments, the anti-CD20 antibody or a fragment thereof described herein is administered to the subject before, during, or after the subject develops membranous glomerulonephritis. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject 1-3 times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, 1-5 years, 5-10 years or for lifetime of the subject. In some embodiments, the subject has been administered standard-of-care treatment for membranous glomerulonephritis prior to administration of the anti-CD20 antibody (for example, prior to administration of Obinutuzumab). In some embodiments, the subject does not respond to or poorly responds to standard-of-care treatment for membranous glomerulonephritis prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the subject has not been administered standard-of-care treatment for membranous glomerulonephritis prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the standard-of-care treatment for membranous glomerulonephritis is administered simultaneously or sequentially with the anti-CD20 antibody (for example, Obinutuzumab). In exemplary embodiments, the standard-of-care treatment for membranous glomerulonephritis is high dose prednisone (steroids), mycophenolic acid (cellcept), plasmapheresis and/or Rituximab. All are used in various combinations.

[0057] Further provided herein is a method for treating, inhibiting, reducing the severity of and/or delaying progression of cryoglobulinemic glomerulonephritis (CGN). The methods comprise, consist of or consist essentially of providing an anti-CD20 antibody and administering an effective amount of the anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of cryoglobulinemic glomerulonephritis in the subject. In some embodiments, the methods comprise, consist of or consist essentially of administering an effective amount of an anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of cryoglobulinemic glomerulonephritis in the subject. In one embodiment, the subject is human. In some embodiments, the anti-CD20 antibody is Obinutuzumab, Rituximab or a combination thereof. In one embodiment, the anti-CD20 antibody is Obinutuzumab. In various embodiments, the anti-CD20 antibody or a fragment thereof described herein is administered to the subject before, during, or after the subject develops cryoglobulinemic glomerulonephritis. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject 1-3 times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, 1-5 years, 5-10 years or for lifetime of the subject. In some embodiments, the subject has been administered standard-of-care treatment for cryoglobulinemic glomerulonephritis prior to administration of the anti-CD20 antibody (for example, prior to administration of Obinutuzumab). In some embodiments, the subject does not respond to or poorly responds to standard-of-care treatment for cryoglobulinemic glomerulonephritis prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the subject has not been administered standard-of-care treatment for cryoglobulinemic glomerulonephritis prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the standard-of-care treatment for cryoglobulinemic glomerulonephritis is administered simultaneously or sequentially with the anti-CD20 antibody (for example, Obinutuzumab). In exemplary embodiments, the standard-of-care treatment for cryoglobulinemic glomerulonephritis includes high dose prednisone (steroids), mycophenolic acid (cellcept), plasmapheresis and/or Rituximab. All are used in various combinations. Many patients with CGN also have hepatitis C or B and may be at risk for progression of liver failure if excessive immunosuppression is used. Newer anti-hepatitis C therapies have not altered the course of CGN. Because of this, and the relative inefficacy of many of the drugs, there is a large unmet need for new therapies that are less toxic in patients with CGN.

[0058] Also provided herein is a method for treating, inhibiting, reducing the severity of and/or delaying progression of IgA nephropathy (IgAN). The methods comprise, consist of or consist essentially of providing an anti-CD20 antibody and administering an effective amount of the anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of IgA nephropathy in the subject. In some embodiments, the methods comprise, consist of or consist essentially of administering an effective amount an anti-CD20 antibody to the subject so are to treat, inhibit, reduce the severity of and/or delay progression of IgA nephropathy in the subject. In one embodiment, the subject is human. In some embodiments, the anti-CD20 antibody is Obinutuzumab, Rituximab or a combination thereof. In one embodiment, the anti-CD20 antibody is Obinutuzumab. In various embodiments, the anti-CD20 antibody or a fragment thereof described herein is administered to the subject before, during, or after the subject develops IgA nephropathy. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject 1-3 times per day or 1-7 times per week. In some embodiments, the anti-CD20 antibody or a fragment thereof is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, 1-5 years, 5-10 years or for lifetime of the subject. In some embodiments, the subject has been administered standard-of-care treatment for IgA nephropathy prior to administration of the anti-CD20 antibody (for example, prior to administration of Obinutuzumab). In some embodiments, the subject does not respond to or poorly responds to standard-of-care treatment for IgA nephropathy prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the subject has not been administered standard-of-care treatment for IgA nephropathy prior to administration of anti-CD20 antibody (for example, Obinutuzumab). In some embodiments, the standard-of-care treatment for IgA nephropathy is administered simultaneously or sequentially with the anti-CD20 antibody (for example, Obinutuzumab). In exemplary embodiments, the standard-of-care treatment for IgA nephropathy is high dose steroid, mycophenolate mofetil, angiotensin-converting enzyme inhibitor (ACEI) or angiotensin receptor blocker (ARB), fish oil, azathioprine, and cyclophosphamide. Most patients with IGAN have mild to moderate disease, and can be controlled with ACEI/ARB therapy without immunosuppression. However, a sizeable number do have more aggressive disease and progress to renal failure. Currently there are no approved drugs for treating IgAN. Inventors' work has shown resolution of clinical and laboratory features of IgAN with Rituximab and cellcept, supporting the use of Obi therapy to treat IgAN especially since IgAN is an autoimmune disease.

[0059] Also provided herein are methods for treating, inhibiting or reducing severity of nephrotic syndrome in a subject in need thereof comprising providing an anti-CD20 antibody and administering an effective amount of the anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of nephrotic syndrome in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years or combinations thereof. In an embodiment, the subject is human.

[0060] Also provided herein are methods for treating, inhibiting or reducing severity of nephrotic syndrome in a subject in need thereof comprising administering an effective amount of an anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of nephrotic syndrome in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years or combinations thereof. In an embodiment, the subject is human.

[0061] Further provided herein are methods are treating, inhibiting or reducing severity of glomerulonephritis (GN) in a subject in need thereof comprising providing an anti-CD20 antibody and administering an effective amount of the antibody to the subject, so as to treat, inhibit or reduce the severity of glomerulonephritis in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some

embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years or combinations thereof. In an embodiment, the subject is human.

[0062] Also provided herein are methods are treating, inhibiting or reducing severity of glomerulonephritis (GN) in a subject in need thereof comprising administering an effective amount of an anti-CD20 antibody to the subject, so as to treat, inhibit or reduce the severity of glomerulonephritis in the subject. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. In some embodiments, effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years or combinations thereof. In an embodiment, the subject is human.

[0063] Also provided herein are methods for desensitizing a subject to antibody mediated transplant rejection (ABMR) in the subject in need thereof. The methods include providing an anti-CD20 antibody (for example, Obinutuzumab) and administering an effective amount of the antibody to the subject so as to desensitize the subject to ABMR. In some embodiments, the subjects are desensitized prior to transplant. In some embodiments, the subjects are desensitized during transplant. In some embodiments, the subjects are desensitized prior to, during and after transplant. In various embodiments, desensitizing the subject includes treating the subject with the anti-CD20 antibody (for example, Obinutuzumab) and Intravenous Gammaglobulin (IVIG), with or without plasmapheresis. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are administered sequentially. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are administered simultaneously. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two

doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. The effective amount of IVIG will be apparent to a person of skill in the art.

[0064] Also provided herein are methods for desensitizing a subject to antibody mediated transplant rejection (ABMR) in the subject in need thereof. The methods include administering an effective amount of an anti-CD20 antibody (for example, Obinutuzumab) to the subject so as to desensitize the subject to ABMR. In some embodiments, the subjects are desensitized prior to transplant. In some embodiments, the subjects are desensitized during transplant. In some embodiments, the subjects are desensitized prior to, during and after transplant. In various embodiments, desensitizing the subject includes treating the subject with the anti-CD20 antibody (for example, Obinutuzumab) and Intravenous Gammaglobulin (IVIG), with or without plasmapheresis. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are administered sequentially. In various embodiments, the anti-CD20 antibody (for example, Obinutuzumab) and IVIG are administered simultaneously. In one embodiment, the effective amount of the antibody is two doses of 1gram Obinutuzumab per dose. In some embodiments, Obinutuzumab is administered intravenously. In some embodiment, the two doses of 1gram per dose of Obinutuzumab are administered about 15 days apart. In some embodiments, a first dose is administered on the same day or split over a period of two days. In an embodiment, the first dose is split over two days wherein 100mg of Obinutuzumab is administered on day 1 and 900mg of Obinutuzumab is administered on day 2. The effective amount of IVIG will be apparent to a person of skill in the art.

Dosages

[0065] In some embodiments of the invention, the effective amounts of the anti-CD20 antibody (for example, Obinutuzumab) can be in the range of about 10-50mg/day, 50-100mg/day, 100-150mg/day, 150-200mg/day, 100-200mg/day, 200-300mg/day, 300-400mg/day, 400-500mg/day, 500-600mg/day, 600-700mg/day, 700-800mg/day, 800-900mg/day, 900-1000mg/day, 1000-1100mg/day, 1100-1200mg/day, 1200-1300mg/day, 1300-1400mg/day, 1400-1500mg/day, 1500-1600mg/day, 1600-1700mg/day, 1700-1800mg/day, 1800-1900mg/day, 1900-2000mg/day, 2000-2100mg/day, 2100-2200mg/day,

2200-2300mg/day, 2300-2400mg/day, 2400-2500mg/day, 2500-2600mg/day, 2600-2700mg/day, 2700-2800mg/day, 2800-2900mg/day or 2900-3000mg/day. In one embodiment the anti-CD20 antibody is Obinutuzumab.

[0066] In further embodiments of the invention, the effective amount of the anti-CD20 antibody (including Obinutuzumab) for use with the claimed methods may be in the range of 1-5mg/kg, 5-10mg/kg, 10-50mg/kg, 50-100mg/kg, 100-150mg/kg, 150-200mg/kg, 100-200mg/kg, 200-300mg/kg, 300-400mg/kg, 400-500mg/kg, 500-600mg/kg, 600-700mg/kg, 700-800mg/kg, 800-900mg/kg or 900-1000mg/kg. In one embodiment the anti-CD20 antibody is Obinutuzumab. In one embodiment, the effective amount of Obinutuzumab is 2 doses of 1gram Obinutuzumab (1g x 2). The second dose may be administered, for example, 1-3, 1-5, 1-10, 5-10, 10-15, 11-15, 12-15, 13-15, 14-15 or 15-20 days after the first dose. The first dose may be infused over a period of 2 days; on the first day 100mg may be infused at 25mg/hr x 4hours and on the second day, 900mg may be infused at 50mg/hr and increased every 30 minutes to a maximum rate of 400mg/hr. The second dose (for example, administered day 15) of 1000mg may start at 100mg/hr and increased 100mg/hr increments every 30 minutes to a maximum of 400mg/hr.

[0067] Typical dosages of an effective amount of anti-CD20 antibody, such as Obinutuzumab, can be in the ranges recommended by the manufacturer where known therapeutic compounds are used, and also as indicated to the skilled artisan by the in vitro responses or responses in animal models. For example, Obinutuzumab is currently recommended for the treatment of chronic lymphocytic anemia as administering 100 mg on day 1, 900mg on day 2 and 1000mg on day 8, and day 15. The same or similar dosing can be used in accordance with various embodiments of the present invention, or an alternate dosage may be used in connection with alternate embodiments of the invention. The actual dosage can depend upon the judgment of the physician, the condition of the patient, and the effectiveness of the therapeutic method based, for example, on the in vitro responsiveness of relevant cultured cells or histocultured tissue sample, or the responses observed in the appropriate animal models

[0068] For treatment of nephrotic syndrome, Obinutuzumab (for example, 1g) may be given every 13-15 days for a total of 2 doses. The first dose may be split over 2 days: 100mg on day 1 and 900mg on day 2, however the full 1g may be infused, for example, once the package insert for Obinutuzumab is updated. The 2nd dose may be infused at a rate of 100mg/hr and

increased by 100mg/hr increments every 30 minutes to a maximum of 400mg/hr. Package labeling updates may be published over time to also accommodate faster infusion rates. Repeat doses may be administered per clinical response after a dose response period of at least 2 weeks.

Pharmaceutical Composition

[0069] In various embodiments, the present invention provides a pharmaceutical composition. The pharmaceutical composition includes an anti-CD20 antibody (for example, Obinutuzumab). The pharmaceutical compositions according to the invention can contain any pharmaceutically acceptable excipient. "Pharmaceutically acceptable excipient" means an excipient that is useful in preparing a pharmaceutical composition that is generally safe, non-toxic, and desirable, and includes excipients that are acceptable for veterinary use as well as for human pharmaceutical use. Such excipients may be solid, liquid, semisolid, or, in the case of an aerosol composition, gaseous. Examples of excipients include but are not limited to starches, sugars, microcrystalline cellulose, diluents, granulating agents, lubricants, binders, disintegrating agents, wetting agents, emulsifiers, coloring agents, release agents, coating agents, sweetening agents, flavoring agents, perfuming agents, preservatives, antioxidants, plasticizers, gelling agents, thickeners, hardeners, setting agents, suspending agents, surfactants, humectants, carriers, stabilizers, and combinations thereof.

[0070] In various embodiments, the pharmaceutical compositions according to the invention may be formulated for delivery via any route of administration. "Route of administration" may refer to any administration pathway known in the art, including but not limited to aerosol, nasal, oral, transmucosal, transdermal, parenteral or enteral. "Parenteral" refers to a route of administration that is generally associated with injection, including intraorbital, infusion, intraarterial, intracapsular, intracardiac, intradermal, intramuscular, intraperitoneal, intrapulmonary, intraspinal, intrasternal, intrathecal, intrauterine, intravenous, subarachnoid, subcapsular, subcutaneous, transmucosal, or transtracheal. Via the parenteral route, the compositions may be in the form of solutions or suspensions for infusion or for injection, or as lyophilized powders. Via the parenteral route, the compositions may be in the form of solutions or suspensions for infusion or for injection. Via the enteral route, the pharmaceutical compositions can be in the form of tablets, gel capsules, sugar-coated tablets, syrups, suspensions, solutions, powders, granules, emulsions, microspheres or nanospheres or lipid vesicles or polymer vesicles allowing controlled release. Typically, the compositions

are administered by injection. Methods for these administrations are known to one skilled in the art.

[0071] The pharmaceutical compositions according to the invention can contain any pharmaceutically acceptable carrier. “Pharmaceutically acceptable carrier” as used herein refers to a pharmaceutically acceptable material, composition, or vehicle that is involved in carrying or transporting a compound of interest from one tissue, organ, or portion of the body to another tissue, organ, or portion of the body. For example, the carrier may be a liquid or solid filler, diluent, excipient, solvent, or encapsulating material, or a combination thereof. Each component of the carrier must be “pharmaceutically acceptable” in that it must be compatible with the other ingredients of the formulation. It must also be suitable for use in contact with any tissues or organs with which it may come in contact, meaning that it must not carry a risk of toxicity, irritation, allergic response, immunogenicity, or any other complication that excessively outweighs its therapeutic benefits.

[0072] The pharmaceutical compositions according to the invention can also be encapsulated, tableted or prepared in an emulsion or syrup for oral administration. Pharmaceutically acceptable solid or liquid carriers may be added to enhance or stabilize the composition, or to facilitate preparation of the composition. Liquid carriers include syrup, peanut oil, olive oil, glycerin, saline, alcohols and water. Solid carriers include starch, lactose, calcium sulfate, dihydrate, terra alba, magnesium stearate or stearic acid, talc, pectin, acacia, agar or gelatin. The carrier may also include a sustained release material such as glyceryl monostearate or glyceryl distearate, alone or with a wax.

[0073] The pharmaceutical preparations are made following the conventional techniques of pharmacy involving milling, mixing, granulation, and compressing, when necessary, for tablet forms; or milling, mixing and filling for hard gelatin capsule forms. When a liquid carrier is used, the preparation will be in the form of a syrup, elixir, emulsion or an aqueous or non-aqueous suspension. Such a liquid formulation may be administered directly p.o. or filled into a soft gelatin capsule.

[0074] The pharmaceutical compositions according to the invention may be delivered in a therapeutically effective amount. The precise therapeutically effective amount is that amount of the composition that will yield the most effective results in terms of efficacy of treatment in a given subject. This amount will vary depending upon a variety of factors, including but not limited to the characteristics of the therapeutic compound (including activity,

pharmacokinetics, pharmacodynamics, and bioavailability), the physiological condition of the subject (including age, sex, disease type and stage, general physical condition, responsiveness to a given dosage, and type of medication), the nature of the pharmaceutically acceptable carrier or carriers in the formulation, and the route of administration. One skilled in the clinical and pharmacological arts will be able to determine a therapeutically effective amount through routine experimentation, for instance, by monitoring a subject's response to administration of a compound and adjusting the dosage accordingly. For additional guidance, see Remington: The Science and Practice of Pharmacy (Gennaro ed. 20th edition, Williams & Wilkins PA, USA) (2000).

[0075] Before administration to patients, formulants may be added to the rAAV vector, the cell transfected with the rAAV vector, or the supernatant conditioned with the transfected cell. A liquid formulation may be preferred. For example, these formulants may include oils, polymers, vitamins, carbohydrates, amino acids, salts, buffers, albumin, surfactants, bulking agents or combinations thereof.

[0076] Carbohydrate formulants include sugar or sugar alcohols such as monosaccharides, disaccharides, or polysaccharides, or water soluble glucans. The saccharides or glucans can include fructose, dextrose, lactose, glucose, mannose, sorbose, xylose, maltose, sucrose, dextran, pullulan, dextrin, alpha and beta cyclodextrin, soluble starch, hydroxethyl starch and carboxymethylcellulose, or mixtures thereof. "Sugar alcohol" is defined as a C4 to C8 hydrocarbon having an -OH group and includes galactitol, inositol, mannitol, xylitol, sorbitol, glycerol, and arabitol. These sugars or sugar alcohols mentioned above may be used individually or in combination. There is no fixed limit to amount used as long as the sugar or sugar alcohol is soluble in the aqueous preparation. In one embodiment, the sugar or sugar alcohol concentration is between 1.0 w/v % and 7.0 w/v %, more preferable between 2.0 and 6.0 w/v %.

[0077] Amino acids formulants include levorotary (L) forms of carnitine, arginine, and betaine; however, other amino acids may be added.

[0078] In some embodiments, polymers as formulants include polyvinylpyrrolidone (PVP) with an average molecular weight between 2,000 and 3,000, or polyethylene glycol (PEG) with an average molecular weight between 3,000 and 5,000.

[0079] It is also preferred to use a buffer in the composition to minimize pH changes in the solution before lyophilization or after reconstitution. Most any physiological buffer may be used including but not limited to citrate, phosphate, succinate, and glutamate buffers or mixtures thereof. In some embodiments, the concentration is from 0.01 to 0.3 molar. Surfactants that can be added to the formulation are shown in EP Nos. 270,799 and 268,110.

[0080] Another drug delivery system for increasing circulatory half-life is the liposome. Methods of preparing liposome delivery systems are discussed in Gabizon et al., *Cancer Research* (1982) 42:4734; Cafiso, *Biochem Biophys Acta* (1981) 649:129; and Szoka, *Ann Rev Biophys Eng* (1980) 9:467. Other drug delivery systems are known in the art and are described in, e.g., Poznansky et al., *DRUG DELIVERY SYSTEMS* (R. L. Juliano, ed., Oxford, N.Y. 1980), pp. 253-315; M. L. Poznansky, *Pharm Revs* (1984) 36:277.

[0081] After the liquid pharmaceutical composition is prepared, it may be lyophilized to prevent degradation and to preserve sterility. Methods for lyophilizing liquid compositions are known to those of ordinary skill in the art. Just prior to use, the composition may be reconstituted with a sterile diluent (Ringer's solution, distilled water, or sterile saline, for example) which may include additional ingredients. Upon reconstitution, the composition is administered to subjects using those methods that are known to those skilled in the art.

Kits

[0082] In various embodiments, the present invention provides a kit for treating or inhibiting nephrotic syndrome. The kit is an assemblage of materials or components, including an anti-CD20 antibody. Thus in some embodiments, the kit contains Obinutuzumab.

[0083] The exact nature of the components configured in the inventive kit depends on its intended purpose. In one embodiment, the kit is configured particularly for human subjects. In further embodiments, the kit is configured for veterinary applications, treating subjects such as, but not limited to, farm animals, domestic animals, and laboratory animals.

[0084] Instructions for use may be included in the kit. "Instructions for use" typically include a tangible expression describing the technique to be employed in using the components of the kit to effect a desired outcome, such as to treat or inhibit nephrotic syndrome in a subject. Optionally, the kit also contains other useful components, such as, measuring tools, diluents, buffers, pharmaceutically acceptable carriers, syringes or other useful paraphernalia as will be readily recognized by those of skill in the art.

[0085] The materials or components assembled in the kit can be provided to the practitioner stored in any convenient and suitable ways that preserve their operability and utility. For example the components can be in dissolved, dehydrated, or lyophilized form; they can be provided at room, refrigerated or frozen temperatures. The components are typically contained in suitable packaging material(s). As employed herein, the phrase “packaging material” refers to one or more physical structures used to house the contents of the kit, such as inventive compositions and the like. The packaging material is constructed by well-known methods, preferably to provide a sterile, contaminant-free environment. As used herein, the term “package” refers to a suitable solid matrix or material such as glass, plastic, paper, foil, and the like, capable of holding the individual kit components. Thus, for example, a package can be a bottle used to contain suitable quantities of an inventive composition containing anti-CD20 antibody, such as Obinutuzumab. The packaging material generally has an external label which indicates the contents and/or purpose of the kit and/or its components.

EXAMPLES

[0086] The following examples are not intended to limit the scope of the claims to the invention, but are rather intended to be exemplary of certain embodiments. Any variations in the exemplified methods which occur to the skilled artisan are intended to fall within the scope of the present invention.

Example 1

[0087] **Fig. 1** describes the differences in mechanism of action between Rituximab and Obinutuzumab (Obi). This figure shows the differences between Type I and Type II anti-CD20 molecules. Type 1 molecules bind to CD20 localized in lipid rafts on B-cells. This can result in incomplete B-cell depletion due to endocytosis of the CD20/anti-CD20 complex. Type 1 anti-CD20 depletes primarily through complement dependent cytotoxicity with low antibody-dependent cell-mediated cytotoxicity (ADCC). However, the glyco-engineered Obinutuzumab has much stronger affinity for the low affinity immunoglobulin gamma Fc region receptor III-A (FcγRIIIa) and results in enhanced depletion of B-cells through ADCC. Obinutuzumab has approximately 5-50-fold greater potency for depletion in ADCC compared to Rituximab. Obinutuzumab also kills B-cells directly through a mechanism known as homotypic aggregation. Overall, clinical studies in B-cell malignancies have shown superior B-cell depletion in peripheral blood, spleen and lymph nodes of patients treated with Obinutuzumab versus Rituximab.

[0088] For treatment of nephrotic syndrome, 1gram(g) of Obinutuzumab may be administered every 15 days for a total of 2 doses. The first dose may be split over 2 days: 100mg on day 1 and 900mg on day 2. However the full 1g of Obinutuzumab may be infused (for example, once the package insert is updated). The 2nd dose may be infused at a rate of 100mg/hr and increased by 100mg/hr increments every 30 minutes to a maximum of 400mg/hr.

[0089] As shown in **Fig. 2A** and **Fig. 2B**: Case Presentation - A 31year old African American male with systemic lupus erythematosus (SLE) presented with nephrotic syndrome, acute renal failure (ARF) and arthralgia with right costovertebral angle (CVA) tenderness. The patient's Albumin level was 0.6 g/dl (normal range is 3.5-5g/dl) on admission with 10g proteinuria (normal is <1g). The patient's auto-antibody levels were: positive for Anti-Nuclear antibody (ANA) with the antibody titer of 1:640 (normal is <1:40 antibody titer); anti-ds DNA was initially negative (normal <1:10 titer) and then increased to 1:640; complement C3 was 40mg/dl (normal is 79-152mg/dl), and complement C4 was 11mg/dl (normal is 16-38mg/dl). A computerized tomography (CT) scan of chest revealed R pulmonary embolism. Biopsy showed membranous lupus nephritis (see **Fig. 2A**). Treatment: the patient received high-dose steroids with taper. The patient received two doses of 1g Rituximab and 750mg bid (twice a day) of mycophenolate mofetil (cellcept). Results: At 2 weeks post-Rituximab patient developed complete remission for 3 year. ANA nadir was 1:320. The patient had an initial remission, but relapsed 3 years later. The ANA titers were not well controlled with Rituximab therapy. (see **Fig. 2B**).

[0090] As shown in **Fig. 2C**, Case Follow Up: The above patient again presented with similar symptoms and weight loss and right CVA tenderness were again noted. The patient also complained of hemoptysis. Right pulmonary embolism was diagnosed. The patient also had ANA 1:640 (normal titer is <1:40) and now anti-dsDNA was 1:640 (normal titer is <1:10). The patient was treated in a similar manner; however, the patient received Obinutuzumab instead of Rituximab and excellent clinical response was seen. Specifically, the patient received 2 doses of 1 gram Obi intravenously (IV) and each dose was separated by 15 days. The first dose was split over 2 days. One week after the first dose of 1 gram Obinutuzumab, the anti-ds-DNA titer was decreased to 1:40 and two months after the second dose of 1 gram Obinutuzumab, the anti-ds-DNA titer decreased to 1:20. This is a powerful demonstration of the efficacy of Obinutuzumab in promoting a rapid remission of SLE.

[0091] The patient shown in **Fig. 3** is a 39 year old Asian American female who received kidney transplant from her brother in 2011. The patient had recurrence of focal segmental glomerulosclerosis (FSGS) in her allograft and was not responsive to Rituximab therapy. She also had antibody mediated rejection (ABMR). The patient received 2 doses of 1g Obinutuzumab intravenously and each dose was separated by 15 days. The first dose was split over 2 days. One month after the 2 doses of Obinutuzumab, the patient's proteinuria (decreased urine total protein/creatinine ratio, UTP/Cr) and renal dysfunction had completely resolved, further supporting the use of Obinutuzumab in treatment of nephrotic syndrome.

[0092] **Fig. 4** shows the course of a 65 year old patient with Rituximab-resistance (also known as drug resistant NS or steroid resistant NS) nephrotic syndrome due to minimal change disease after administration of steroids or Rituximab. The patient was uncontrolled for several years. The patient received two doses of 1g Obinutuzumab per dose intravenously; the first dose was split over two days and the second dose was administered 15 days later. After the above treatment, the patient lost 30lbs of mainly water weight which presented as edema and showed resolution of proteinuria marked by decrease in UTP/Cr ratio.

Example 2

[0093] There is extensive data with Rituximab (RTX, Type I anti-CD20) for desensitization (DES) and treatment of ABMR. There is also a comparable data in treatment of glomerulonephritis (GN) and nephrotic syndrome (NS). However, Rituximab may not always be effective. The inventors report on the use of Obinutuzumab in GN and human leukocyte antigen (HLA) sensitized patients and show that Obinutuzumab is more effective in depletion of B-cells than Rituximab.

[0094] **Methods:** We identified 11 patients with primary GN and NS, patients with recurrent GN and NS post-transplant and patients with RTX-resistant DES and ABMR who were subsequently treated with Obinutuzumab. Obinutuzumab was administered on day 1 (100mg), day 2 (900mg) and day 15 (1000mg) as per chronic lymphocytic leukemia (CLL) dosing guidelines. Pre-medication with acetaminophen 650mg by mouth (p.o.), diphenhydramine 25mg p.o. and methylprednisolone 40 mg intravenously (i.v) were given. Patients were monitored for relevant clinical and laboratory parameters specific to their condition. These included: donor specific antibody (DSAs) (pre and 1month (M) post-treatment), auto-

antibodies (anti-dsDNA and anti-AT1R) pre and 1 month post-treatment, serum creatinine (SCr) and degrees of proteinuria.

[0095] Results: From a safety standpoint, Obinutuzumab was very well tolerated in this non-B-cell malignancy population. No infusion adverse event/significant adverse event (AE/SAE) was noted. One patient developed *Clostridium difficile* (*C. diff*) diarrhea 1 month post-Obinutuzumab, which resolved. The details of responses to OBI treatment are shown in Table 1.

[0096] **Table 1:** Obinutuzumab use in drug resistant GN and ABMR. Relative intensity scale (RIS) [0 point = No DSA; 2 points = <5000 mean fluoresce intensity {weak}; 5 points = 5000-10000 MFI {moderate}; 10 points = > 10,000 {strong}].

N=11	SCr (mg/dl)		UTP/Cr		Antibodies	
	Pre	Post	Pre	Post	Pre	Post
Primary Glomerular Disease (n=2)						
SLE relapse	1.1	0.8	8.7	0.83	640	20
Minimal change disease (MCD-1)	1.1	0.9	8.9	5.58	-	-
MCD-2	1.5	0.9	18.2	5.2	-	-
Recurrent GN post kidney transplant (n=5)						
Membranous GN (MGN)	1.7	1.92	4.68	0.95	-	-
FSGS-1	2.1	2	2.4	2	-	-
FSGS-2	1.6	1.3	3.94	3.41	-	-
FSGS	0.9	0.9	1.3	0	-	-
Desensitization (n=1)						
AT1R antibody strong	5.1(Pre-Tx)	0.9	-	-	AT1Rab 40	AT1Rab 11
ABMR post kidney transplant (n=3)						
Bx Proven ABMR and FSGS recurrence	0.9	0.9	84.1	<6	DR53 MFI 3750-5000	DR53 2500-3750
BX proven ABMR, s/p PLEX+I+ R, Anti-IL6R antibody	1.8	1.95	-	-	DQ8 RIS 5	n/a
BX proven Acute ABMR s/p PLEX + IVIG	1.1	1.2	-	-	Multiple DSA RIS: 47	Multiple DSA RIS: 27
Bx Proven ABMR and Strong DSA s/p IVIG	1.7	1.1	-	-	DQ8 RIS 10	DQ8 RIS 10

[0097] Briefly, 3 patients with primary GN and NS showed rapid and significant response to Obinutuzumab. One patient with recurrent SLE showed rapid resolution of symptoms and

decline in anti-dsDNA antibodies (from 1:640 to 1:20) in 2 weeks. Patients with Rituximab-resistant MCD showed rapid improvements in proteinuria and resolution of NS in 2 cases. In 3 patients with recurrent GN, reduction in proteinuria and stabilization of SCr was seen. 2 out of 3 patients with Rituximab-resistant ABMR showed reductions in DSAs and had stable SCr.

[0098] These data with Obinutuzumab in renal transplant and GN patients suggests that Obinutuzumab appears safe and has efficacy beyond that seen with Rituximab. This is likely due to Obinutuzumab's novel mechanism of action (MOA) that results in superior B-cell depletion compared to Rituximab.

Example 3

[0099] Here we examine the safety and limited efficacy of Obinutuzumab in patients with severe nephrotic syndrome, autoimmune diseases and antibody mediated rejection (ABMR) who had failed multiple treatments including Rituximab. A total of 25 patients were treated with Obinutuzumab, 12 patients with steroid-resistant and Rituximab-resistant nephrotic syndrome, membranous SLE, membraneoproliferative GN (MPGN) and primary membranous glomerulonephritis and 13 post kidney transplant patients with biopsy proven ABMR with presence of donor specific antibody (DSA) who failed to respond to Rituximab. Patients with ABMR were treated with Obinutuzumab day 1 (100mg), day 2 (900mg). Patients with nephrotic syndrome were treated with Obinutuzumab day 1 (100mg), day 2 (900mg), and day 15 (1000mg) as per CLL dosing guidelines. Premedication of acetaminophen 650mg oral, diphenhydramine 25-50mg oral, and solumedrol 40-80mg intravenous was administered 30 minutes prior to each infusion. Patients were monitored for relevant clinical and laboratory parameters specific to their condition which included: DSAs (pre and 1M post-treatment), SCr, and degrees of proteinuria. Patients with ABMR had before and 1 month post treatment renal function and DSAs assessed. There were 4 of 13 patients who did not have post treatment DSAs drawn. All infusions were monitored for infusion-related AEs and long-term assessments of AEs/SAEs were also carried out.

[00100] Table 2: Demographic Table of Rituximab Resistant Nephrotic Syndrome Patients (N-12). Most Recent Serum Creatinine and DSA RIS were 1-2M post Obinutuzumab (Gazyva) treatment.

	N = 12	P
M/F	5/7	
Mean Age (y)	49.8±21.8 (19-83)	
Nephrotic Syndrome		
FSGS	4	
MGN	3	
SLE	1	
MCD	2	
IgG-4 Related Disease	1	
MPGN type 1	1	
Mean Serum Cr (mg/dl)		
Pre Gazyva	1.8±0.9	
1M Post Gazyva	1.5±0.9	0.015
3M Post	1.7±1.1	0.164
6M Post	1.6±1.4	0.961
12M Post	1.6±1	
Mean Urine Total Protein/Cr		
Pre Gazyva	13.3±11.2	
1M Post Gazyva	5.1±3.7	0.016
2M Post	3.7±4	0.012
4M Post	1.7±1.8	0.055
6M Post	0.4±0.4	0.16
12M Post	0.7	

[00101] Table 3: Demographic Table of Post Kidney Transplant Patients with Rituximab Resistant ABMR. *Relative Intensity Scale (RIS) [0 points = No DSA; 2 points = <5000MFI {weak}; 5 points = 5000-10,000 MFI {moderate}; 10 points = >10,000MFI {strong}]. Most Recent Serum Creatinine and DSA RIS were 1-2M post Obi treatment.

	N=13	P
M/F	7/6	
Mean Age (y)	44.1 ± 15.1 (17-67)	
LD/DD	5/8	
Mean Serum Cr (mg/dl)		0.896
Pre Gazyva	1.92±0.67	
Most Recent Post Gazyva	1.87±0.93	
Mean DSA RIS*		0.187
Pre Gazyva	11.77±11.65	
Most Recent Post Gazyva	9.67±8.93	
Mean Time from Transplant to Treatment	7.22±5.44	

[00102] From this preliminary experience with Obinutuzumab for the treatment of Rituximab and steroid resistant nephrotic syndrome and GN (see **Table 2**), we saw significant benefits in reducing proteinuria up to one year post treatment as well as improvements in serum creatinine (mg/dl). No major AE/SAEs including infusion related side effects were seen in this patient population. For post- kidney transplant patients with Rituximab resistant ABMR (see **Table 3**), no significant differences for DSAs in aggregate were seen however specific patients showed reductions within one to three months post treatment, with improvement in serum creatinine. One post-transplant Obinutuzumab treated patient developed CMV viremia and another developed both *C. diff* colitis and urinary tract infection (UTI) which resolved with treatment.

[00103] **Fig. 5** shows pre-Obinutuzumab urine total protein/creatinine ratios (UTP/Cr) through 12M post-Obi treatment with linear trends of the 12 nephrotic syndrome patients. Significant improvements of UTP/Cr were seen over the 12 month period post treatment.

[00104] **Fig. 6** shows pre-Obinutuzumab UTP/Cr through 12M post-Obinutuzumab treatment. Significant reductions were seen at 1 month post treatment ($p=0.016$), 2 month post treatment ($p=0.0012$) and 4 month post treatment ($p=0.05$) compared to baseline.

[00105] **Fig. 7** shows linear trends of autoantibody levels at pre-Obinutuzumab up to 6M post Obinutuzumab. Trends show non-significant decrease of autoantibody levels from baseline.

[00106] **Fig. 8** shows mean serum creatinine (Cr, mg/dl) from pre-Obinutuzumab up to 12 months post Obinutuzumab. Significant decrease was seen at one month post Obinutuzumab, $p=0.014$, and overall Cr remained stable from baseline up to 12 month post treatment.

[00107] Based on the effects of Obinutuzumab treatment of patients with glomerular diseases and ABMR, we can conclude that the drug appears safe when administered to patients with these difficult conditions who have failed multiple other therapies. The most impressive findings were the reversal of NS and elimination of pathogenic autoantibodies. Obinutuzumab stabilized renal function but did not significantly decrease DSAs over this short observation period in patients with ABMR.

[00108] The various methods and techniques described above provide a number of ways to carry out the application. Of course, it is to be understood that not necessarily all

objectives or advantages described can be achieved in accordance with any particular embodiment described herein. Thus, for example, those skilled in the art will recognize that the methods can be performed in a manner that achieves or optimizes one advantage or group of advantages as taught herein without necessarily achieving other objectives or advantages as taught or suggested herein. A variety of alternatives are mentioned herein. It is to be understood that some preferred embodiments specifically include one, another, or several features, while others specifically exclude one, another, or several features, while still others mitigate a particular feature by inclusion of one, another, or several advantageous features.

[00109] Furthermore, the skilled artisan will recognize the applicability of various features from different embodiments. Similarly, the various elements, features and steps discussed above, as well as other known equivalents for each such element, feature or step, can be employed in various combinations by one of ordinary skill in this art to perform methods in accordance with the principles described herein. Among the various elements, features, and steps some will be specifically included and others specifically excluded in diverse embodiments.

[00110] Although the application has been disclosed in the context of certain embodiments and examples, it will be understood by those skilled in the art that the embodiments of the application extend beyond the specifically disclosed embodiments to other alternative embodiments and/or uses and modifications and equivalents thereof.

[00111] Preferred embodiments of this application are described herein, including the best mode known to the inventors for carrying out the application. Variations on those preferred embodiments will become apparent to those of ordinary skill in the art upon reading the foregoing description. It is contemplated that skilled artisans can employ such variations as appropriate, and the application can be practiced otherwise than specifically described herein. Accordingly, many embodiments of this application include all modifications and equivalents of the subject matter recited in the claims appended hereto as permitted by applicable law. Moreover, any combination of the above-described elements in all possible variations thereof is encompassed by the application unless otherwise indicated herein or otherwise clearly contradicted by context.

[00112] All patents, patent applications, publications of patent applications, and other material, such as articles, books, specifications, publications, documents, things, and/or the like, referenced herein are hereby incorporated herein by this reference in their entirety for all

purposes, excepting any prosecution file history associated with same, any of same that is inconsistent with or in conflict with the present document, or any of same that may have a limiting affect as to the broadest scope of the claims now or later associated with the present document. By way of example, should there be any inconsistency or conflict between the description, definition, and/or the use of a term associated with any of the incorporated material and that associated with the present document, the description, definition, and/or the use of the term in the present document shall prevail.

[00113] It is to be understood that the embodiments of the application disclosed herein are illustrative of the principles of the embodiments of the application. Other modifications that can be employed can be within the scope of the application. Thus, by way of example, but not of limitation, alternative configurations of the embodiments of the application can be utilized in accordance with the teachings herein. Accordingly, embodiments of the present application are not limited to that precisely as shown and described.

[00114] Various embodiments of the invention are described above in the Detailed Description. While these descriptions directly describe the above embodiments, it is understood that those skilled in the art may conceive modifications and/or variations to the specific embodiments shown and described herein. Any such modifications or variations that fall within the purview of this description are intended to be included therein as well. Unless specifically noted, it is the intention of the inventors that the words and phrases in the specification and claims be given the ordinary and accustomed meanings to those of ordinary skill in the applicable art(s).

[00115] The foregoing description of various embodiments of the invention known to the applicant at this time of filing the application has been presented and is intended for the purposes of illustration and description. The present description is not intended to be exhaustive nor limit the invention to the precise form disclosed and many modifications and variations are possible in the light of the above teachings. The embodiments described serve to explain the principles of the invention and its practical application and to enable others skilled in the art to utilize the invention in various embodiments and with various modifications as are suited to the particular use contemplated. Therefore, it is intended that the invention not be limited to the particular embodiments disclosed for carrying out the invention.

[00116] While particular embodiments of the present invention have been shown and described, it will be obvious to those skilled in the art that, based upon the teachings herein, changes and modifications may be made without departing from this invention and its broader aspects and, therefore, the appended claims are to encompass within their scope all such changes and modifications as are within the true spirit and scope of this invention.

CLAIMS

1. A method of treating, inhibiting or reducing severity of nephrotic syndrome in a subject in need thereof comprising:
 - (i) providing an anti-CD20 antibody; and
 - (ii) administering an effective amount of the antibody to the subject,so as to treat, inhibit or reduce the severity of nephrotic syndrome in the subject.
2. A method of treating, inhibiting or reducing severity of glomerulonephritis (GN) in a subject in need thereof comprising:
 - (i) providing an anti-CD20 antibody; and
 - (ii) administering an effective amount of the antibody to the subject,so as to treat, inhibit or reduce the severity of glomerulonephritis in the subject.
3. The method of claim 2, wherein glomerulonephritis is membranous glomerulonephritis, focal segmental glomerulosclerosis (FSGS), minimal change nephrotic syndrome, cryoglobulinemic glomerulonephritis, IgA nephropathy.
4. The method of claim 1 or 2, wherein the anti-CD20 antibody is Obinutuzumab, Rituximab or a combination there.
5. The method of claim 1 or 2, wherein the anti-CD20 antibody is Obinutuzumab.
6. The method of claim 1, wherein the subject has drug-resistant nephrotic syndrome
7. The method of claim 1, wherein the subject has Rituximab-resistant nephrotic syndrome.
8. The method of claim 1, wherein nephrotic syndrome in the subject is caused by any one or more of minimal change disease, focal segmental glomerulosclerosis, membranous glomerulonephritis, IgA nephropathy and cryoglobulinemic glomerulonephritis.
9. The method of claim 1 or 2, wherein the subject has been administered standard-of-care treatment prior to administration of anti-CD20 antibody.
10. The method of claim 9, wherein the subject does not respond to standard-of-care treatment prior to administration of anti-CD20 antibody.

11. The method of claim 1 or 2, wherein the subject has not been administered standard-of-care treatment prior to administration of anti-CD20 antibody.
12. The method of claim 1 or 2, wherein the anti-CD20 antibody is administered simultaneously or sequentially with standard-of-care treatment.
13. The method of claim 9, wherein standard-of-care treatment are any one or more of high dose prednisone (steroids), mycophenolic acid (cellcept) and optionally calcineurin inhibitors.
14. The method of claim 13, wherein the calcineurin inhibitors are cyclosporine, tacrolimus, or a combination thereof.
15. The method of claim 1 or 2, wherein the effective amount of the anti-CD20 antibody is about 0.1 to 0.5mg/kg/day, 0.5 to 5 mg/kg/day, 5 to 10 mg/kg/day, 10 to 20 mg/kg/day, 20 to 50 mg/kg/day, 50 to 100 mg/kg/day, 100 to 200 mg/kg/day, 200 to 300 mg/kg/day, 300 to 400 mg/kg/day, 400 to 500 mg/kg/day, 500 to 600 mg/kg/day, 600 to 700mg/kg/day, 700 to 800mg/kg/day, 800 to 900mg/kg/day or 900 to 1000 mg/kg/day.
16. The method of claim 1 or 2, wherein the composition is administered to the subject before, during, or after the subject develops nephrotic syndrome.
17. The method of claim 1 or 2, wherein the anti-CD20 antibody is administered to the subject 1-3 times per day or 1-7 times per week.
18. The method of claim 1 or 2, wherein the anti-CD20 antibody is administered to the subject for 1-5 days, 1-5 weeks, 1-5 months, or 1-5 years.
19. The method of claim 1 or 2, wherein the effective amount is two doses of 1g Obinutuzumab per dose.
20. The method of claim 19, wherein Obinutuzumab is administered intravenously.
21. The method of claim 19, wherein the two doses are administered 15 days apart.
22. The method of claim 19, wherein a first dose is administered on the same day or over two days.
23. The method of claim 22, wherein the first dose is administered in the amount of 100mg on day 1 and 900mg on day 2.

24. The method of claim 19, wherein the effective amount of the Obinutuzumab is administered to the subject every 6 months, every 1 year, every 18 months, every 2 years, every 3 years, every 4 years, every 5 years or combinations thereof.
25. The method of claim 1 or 2, wherein the subject is human.

FIG. 1

Obinutuzumab v. Rituximab: Distinct Mechanism of Action of Depleting B-cells

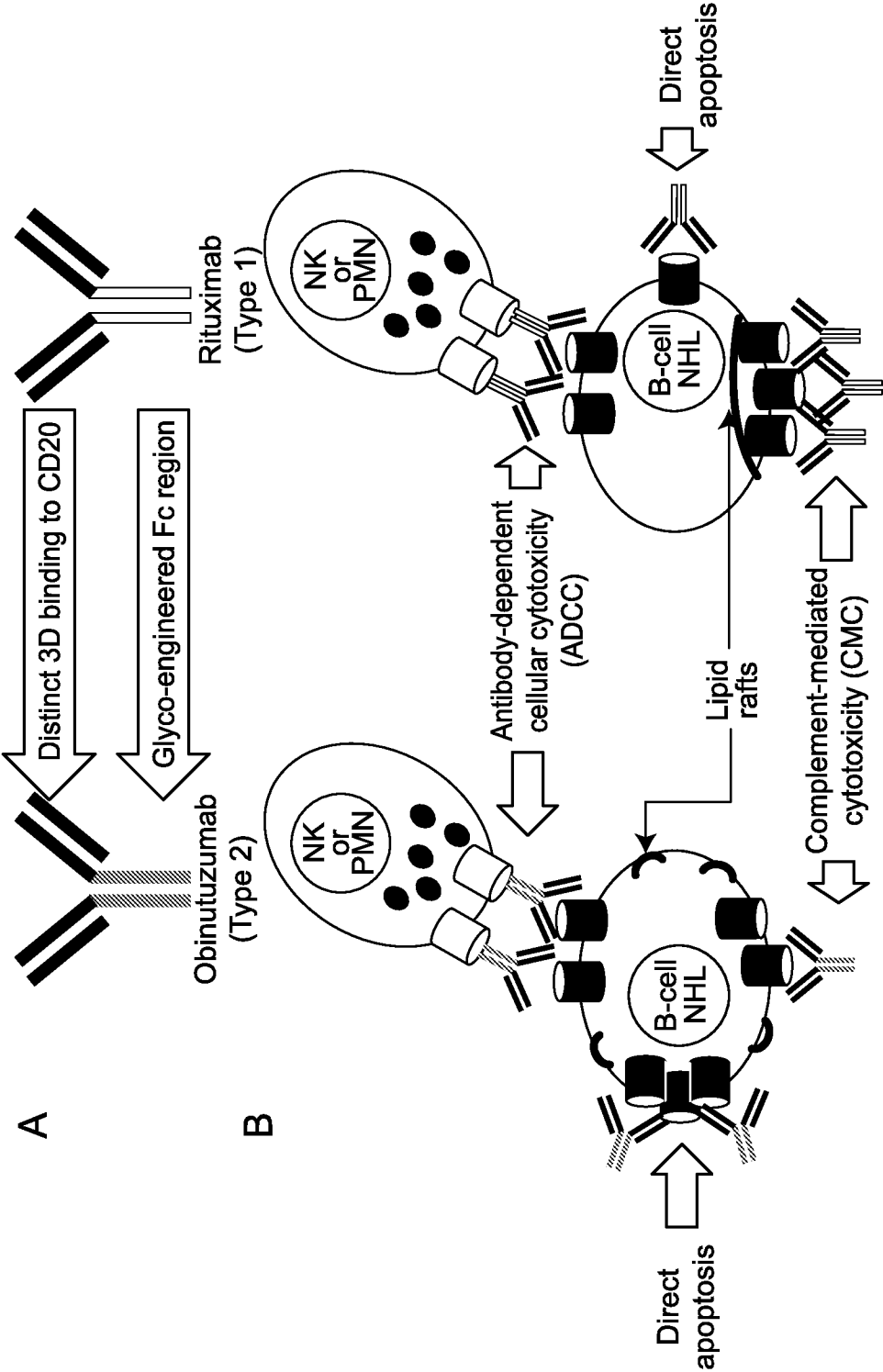


FIG. 2A

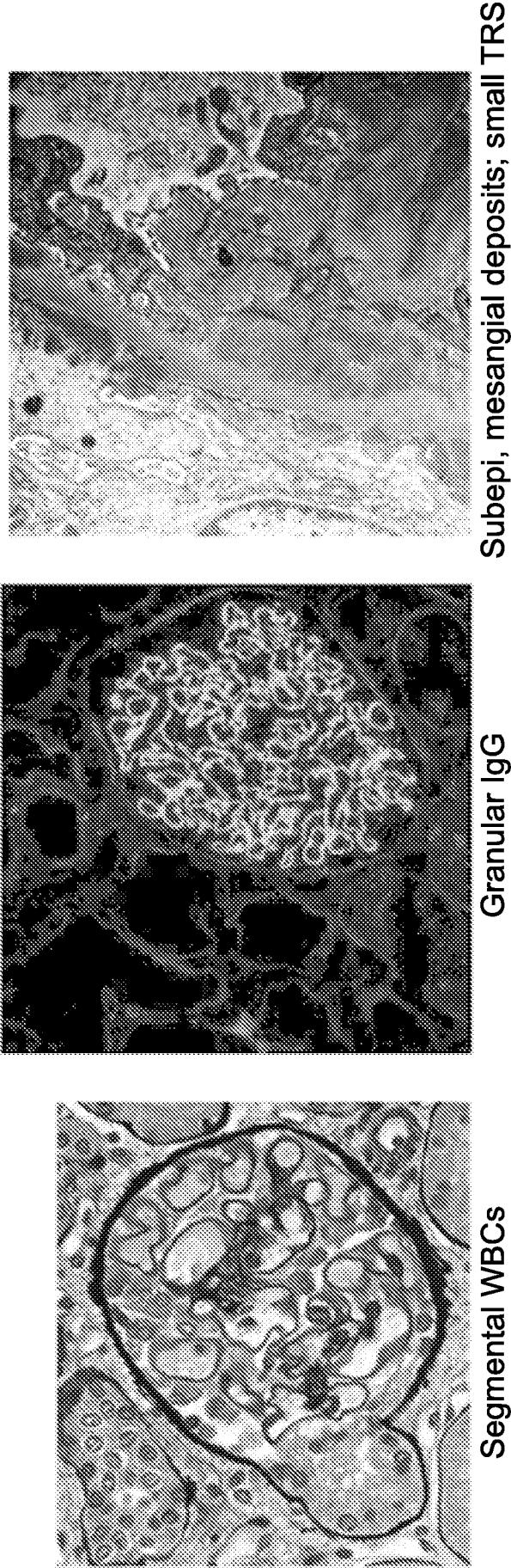


Fig. 2B

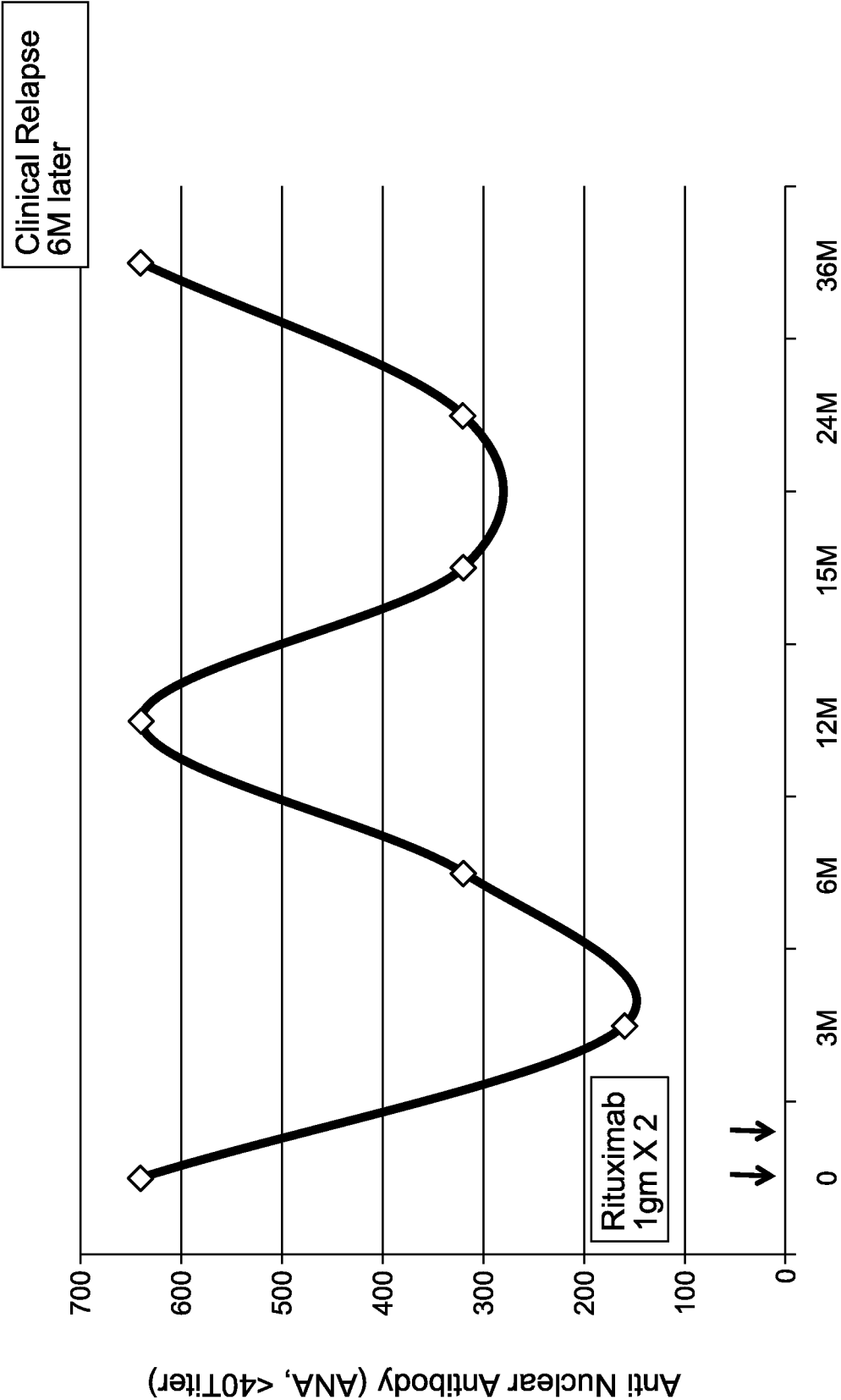


FIG. 2C
Anti-dsDNA Antibodies at Systemic Lupus Erythematosus (SLE)
Relapse and Up to 8 Weeks Post-Treatment

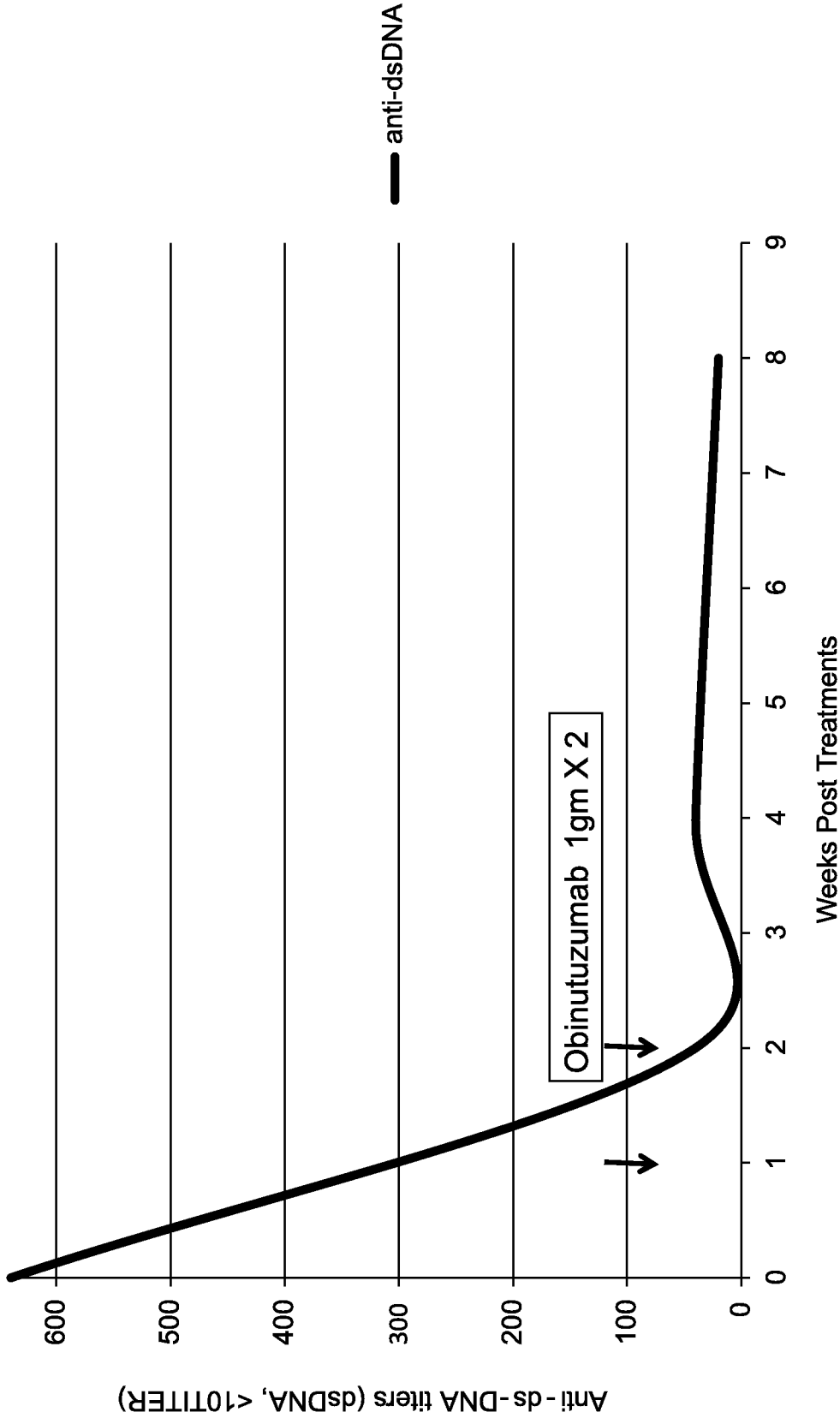


FIG. 3

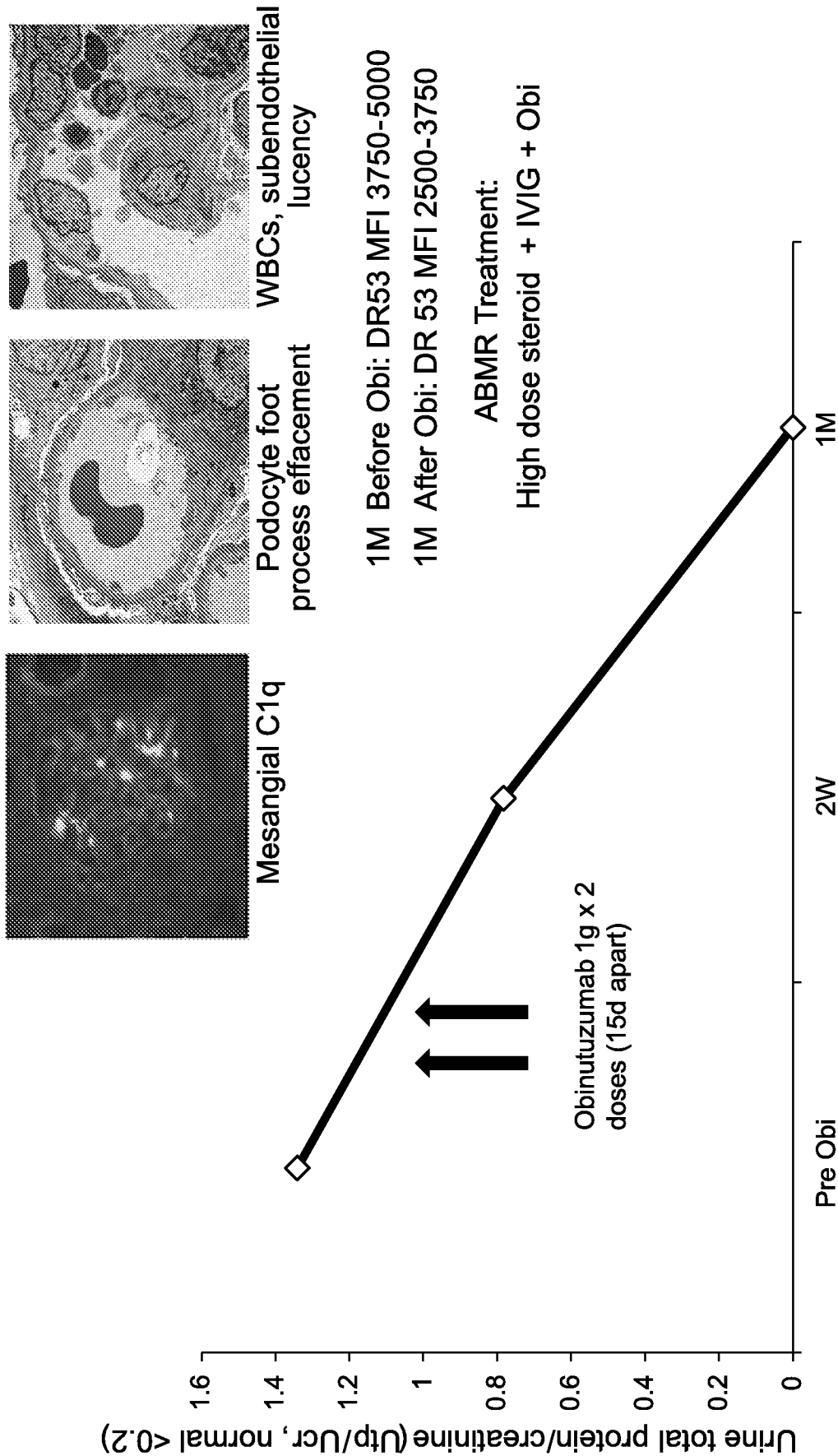


FIG. 4

Obinutuzumab in Minimal Change NS Resistant Other Therapies

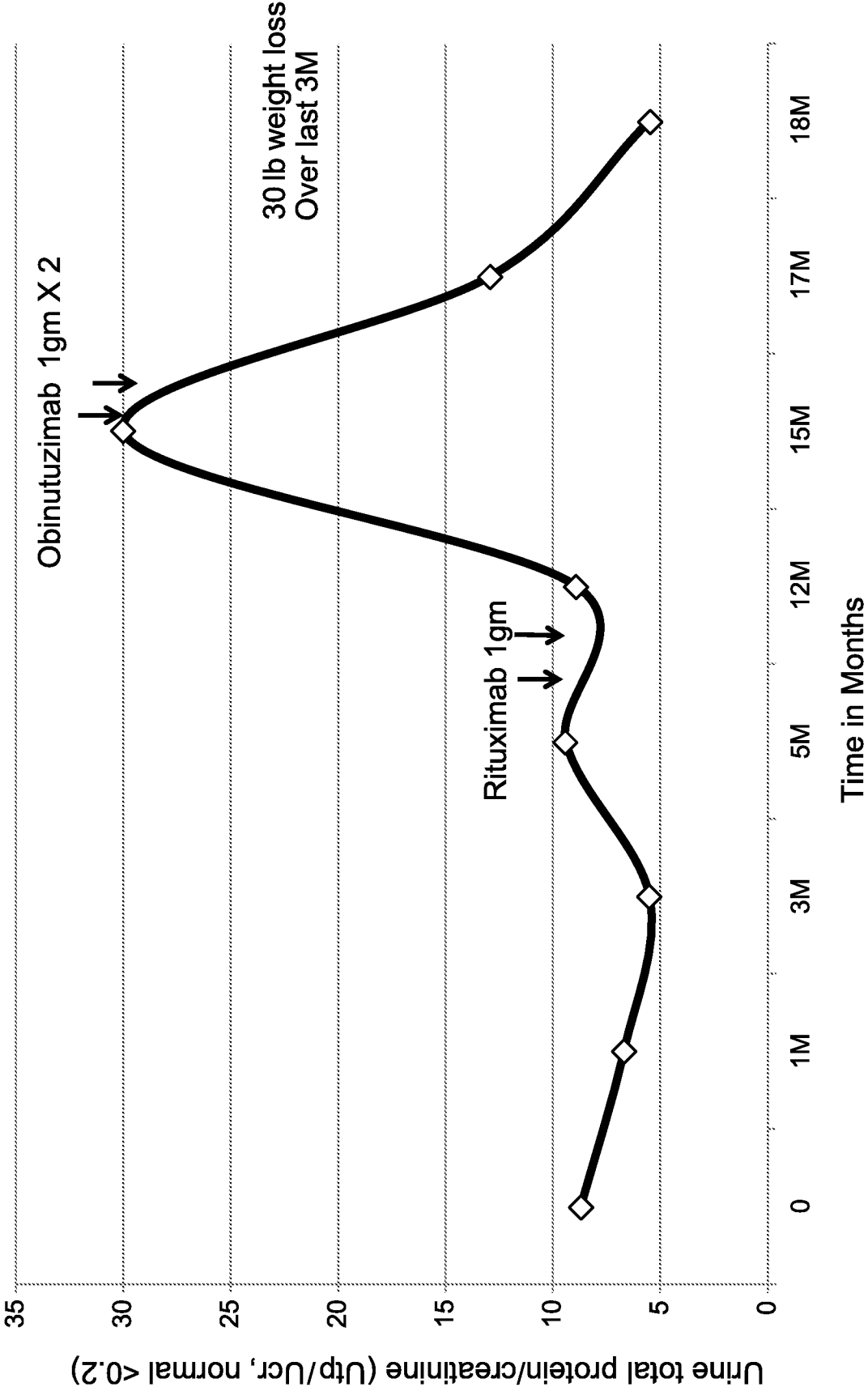


FIG. 5

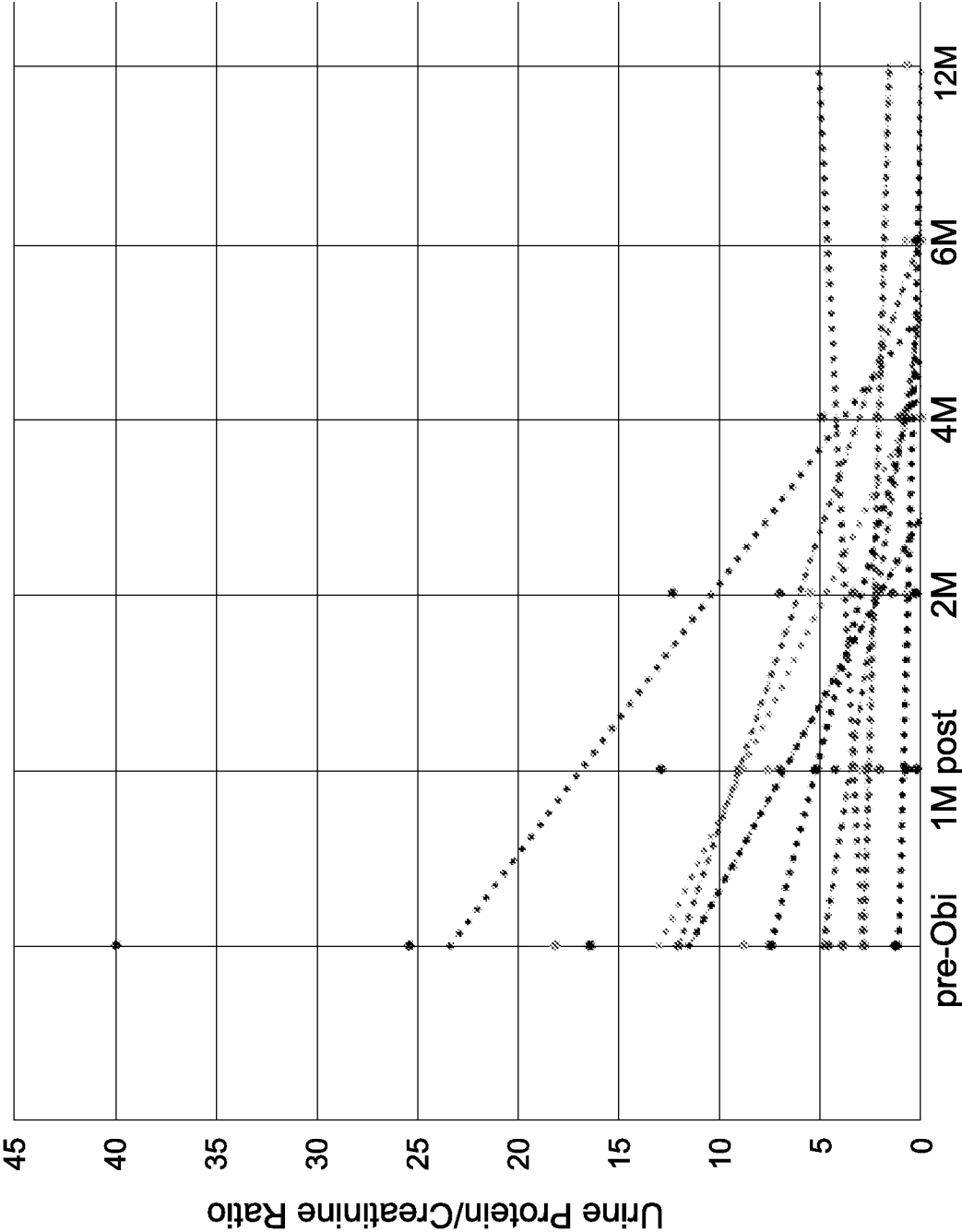


FIG. 6

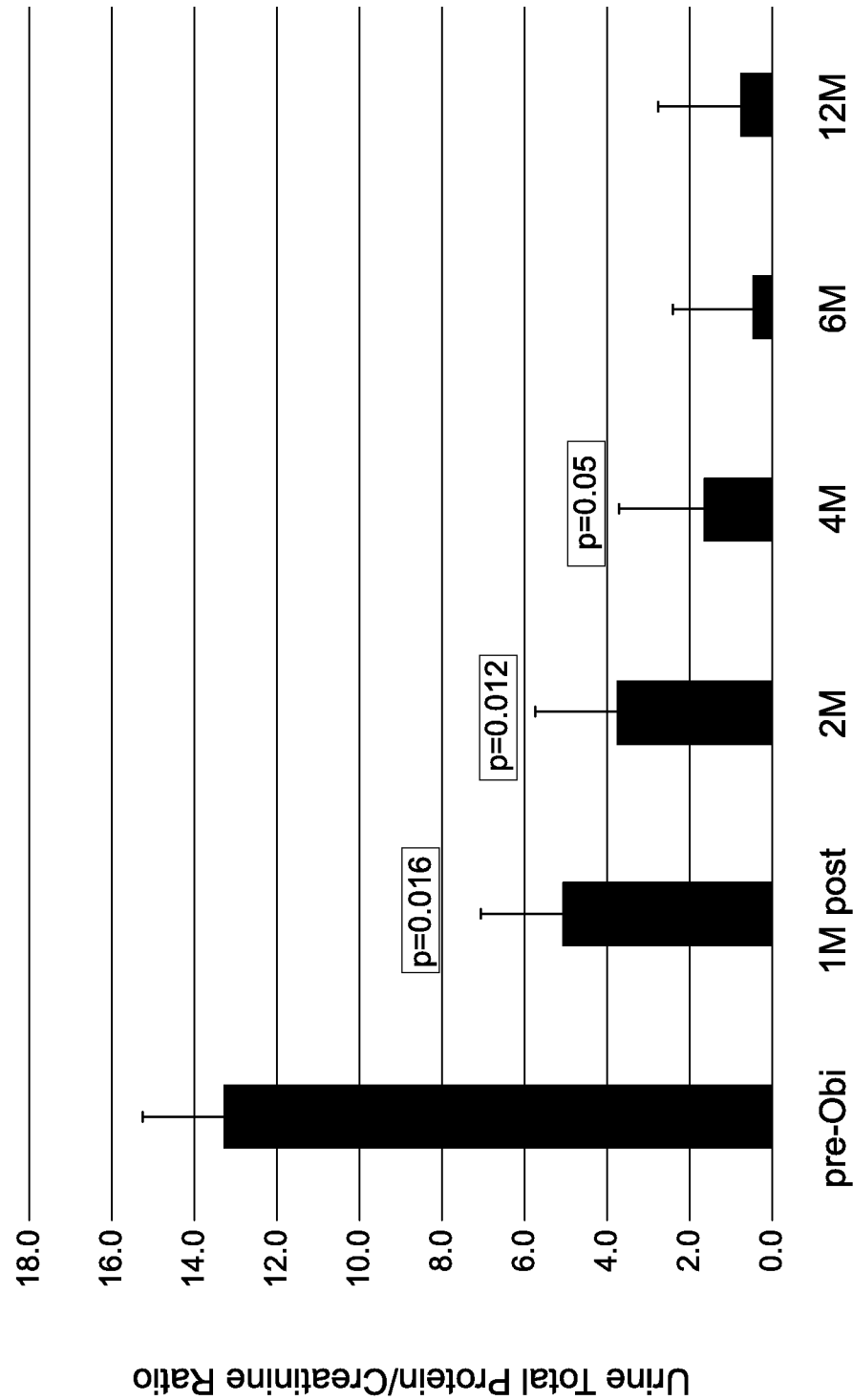


FIG. 7

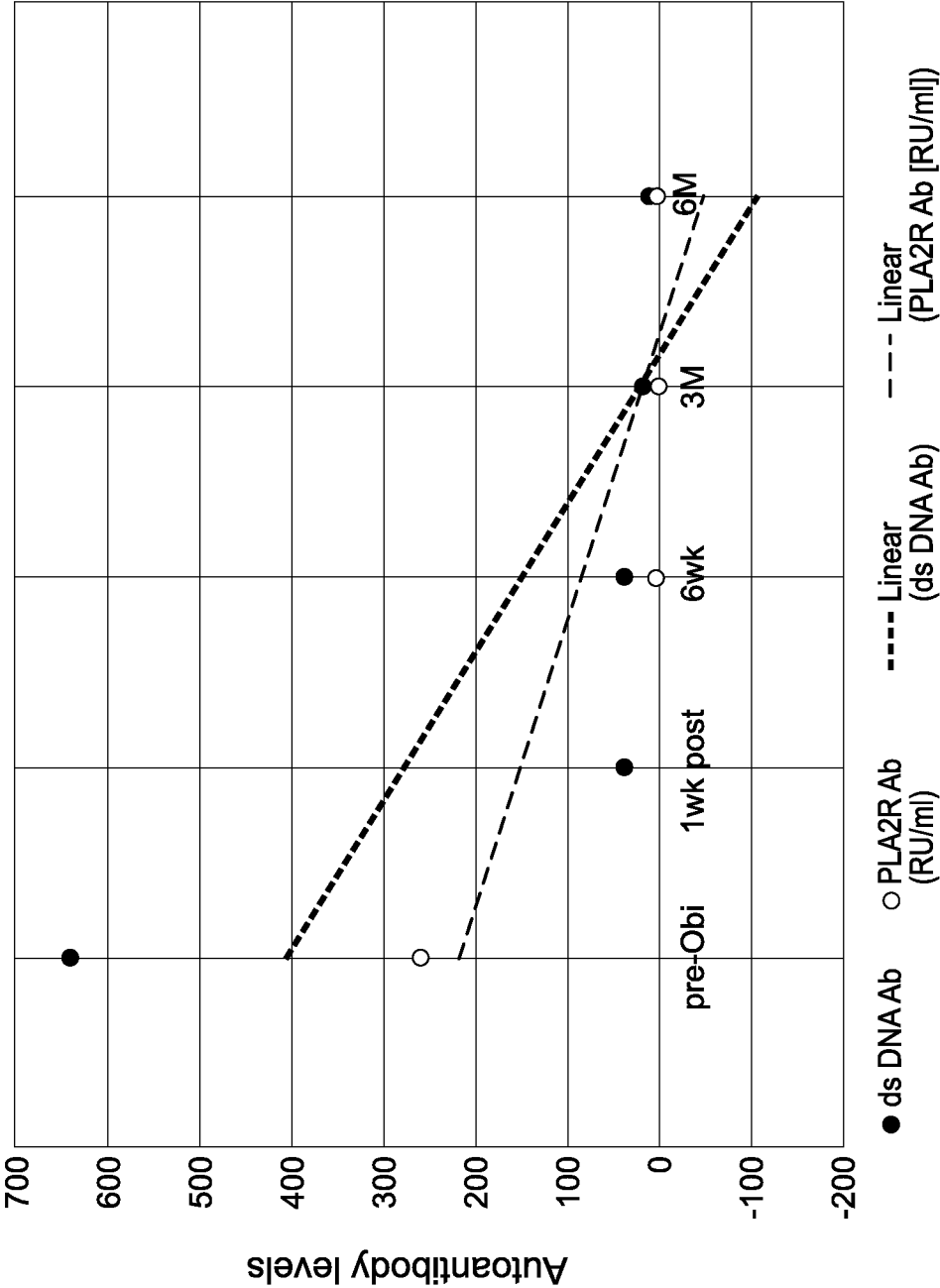
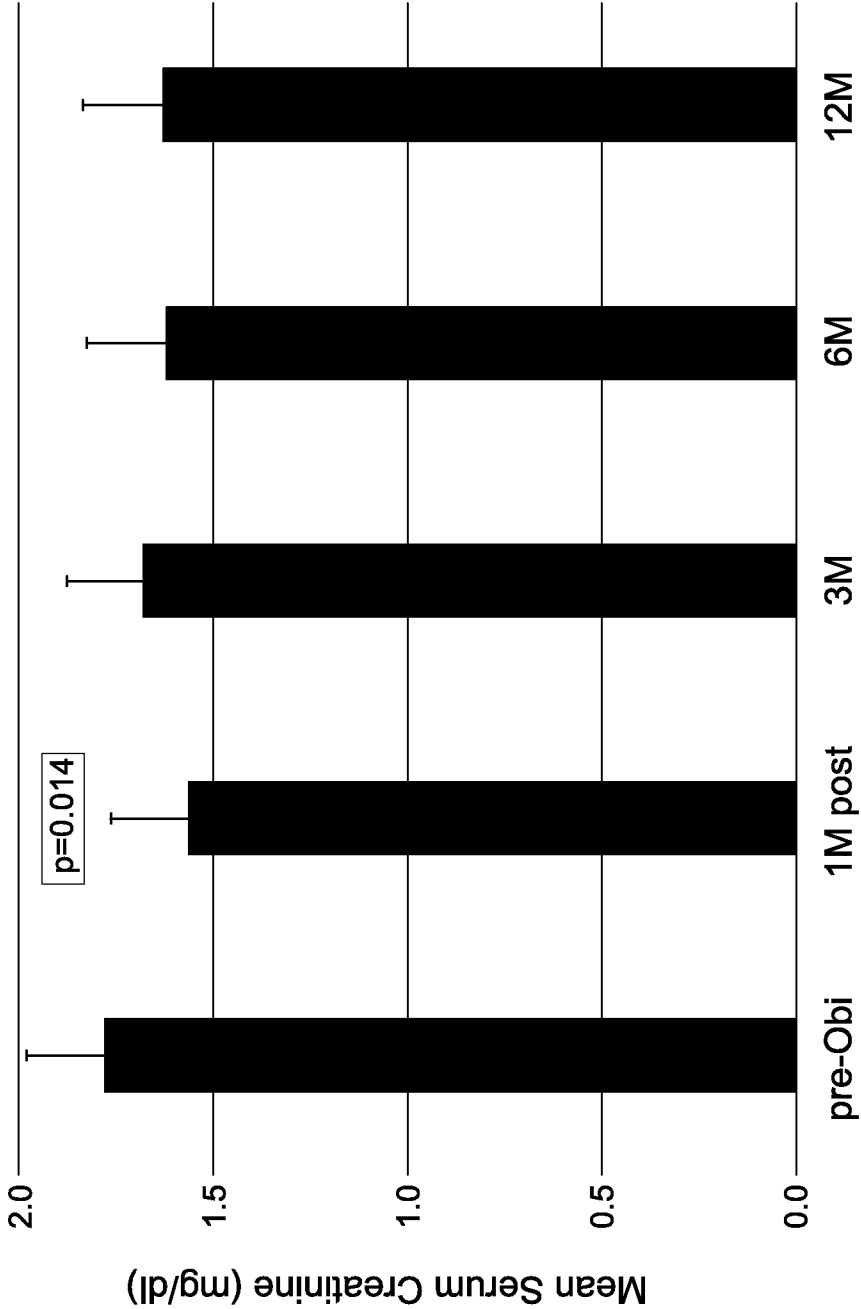


FIG. 8



INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 16/66033

A. CLASSIFICATION OF SUBJECT MATTER

IPC(8) - A61K 39/395, C07K 16/28, A61P 13/12 (2017.01)

CPC - A61K 39/3955, C07K 16/2887, A61K 2039/505

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC(8) - A61K 39/395, C07K 16/28, A61P 13/12 (2017.01)

CPC - A61K 39/3955, C07K 16/2887, A61K 2039/505

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

CPC - A61K 39/395

(KEYWORD LIMITED; TERMS BELOW)

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

PatBase, Google Patents, Google Scholar

Search terms: CD20 or anti-CD20 antibody, Obinutuzumab, Rituximab, nephrotic syndrome, glomerulonephritis, drug-resistant nephrotic syndrome, Rituximab-resistant nephrotic syndrome, pharmacokinetics

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2014/0314748 A1 (F. HOFFMANN- LA ROCHE AG et al.) 23 October 2014 (23.10.2014), para [0038]; para [0096-0098]; para [0270-0272]; Claim 1; Claim 44; Claim 45; Claim 84	1-5, 15-17, 19, 20, 25
Y		6-14, 18, 21-24
Y	BASU, Ofatumumab for Rituximab-Resistant Nephrotic Syndrome. N. Engl J Med, 27 March 2014, Vol 370, No. 13, pp 1200-1270. Especially pg 1270, col 1, para 2, pg 1200, col 2, last para; pg 1269, col 1, para 1	6-10, 13, 14
Y	US 2009/0311255 A1 (BRUNETTA et al.,) 17 December 2009 (17.12.2009), para[0095] ; para [0100]; para [0103]	11-14, 21
Y	SEHN et al., A phase 1 study of obinutuzumab induction followed by 2 years of maintenance in patients with relapsed CD20-positive B-cell malignancies. Blood. 31 May 2012, Vol 119, No. 22: pp 5118-5125. Especially Abstract	18, 24
Y	GOEDE et al., Obinutuzumab plus Chlorambucil in Patients with CLL and Coexisting Conditions N. Engl J Med, 20 March 2014, Vol 370, No. 12: pp 1101-1110. Especially pg 1103, col 1, para 2	22, 23
Y	BROWN et al., Obinutuzumab plus fludarabine/cyclophosphamide or bendamustine in the initial therapy of CLL patients: the phase 1b GALTON trial. Blood. 30 April 2015, Vol 125, No. 18: pp 2779-2785. Especially pg 2780, col 1, last para	23

☐ Further documents are listed in the continuation of Box C.


* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

25 January 2017

Date of mailing of the international search report

03 MAR 2017

Name and mailing address of the ISA/US

Mail Stop PCT, Attn: ISA/US, Commissioner for Patents
P.O. Box 1450, Alexandria, Virginia 22313-1450

Facsimile No. 571-273-8300

Authorized officer:

Lee W. Young

PCT Helpdesk: 571-272-4300

PCT OSP: 571-272-7774