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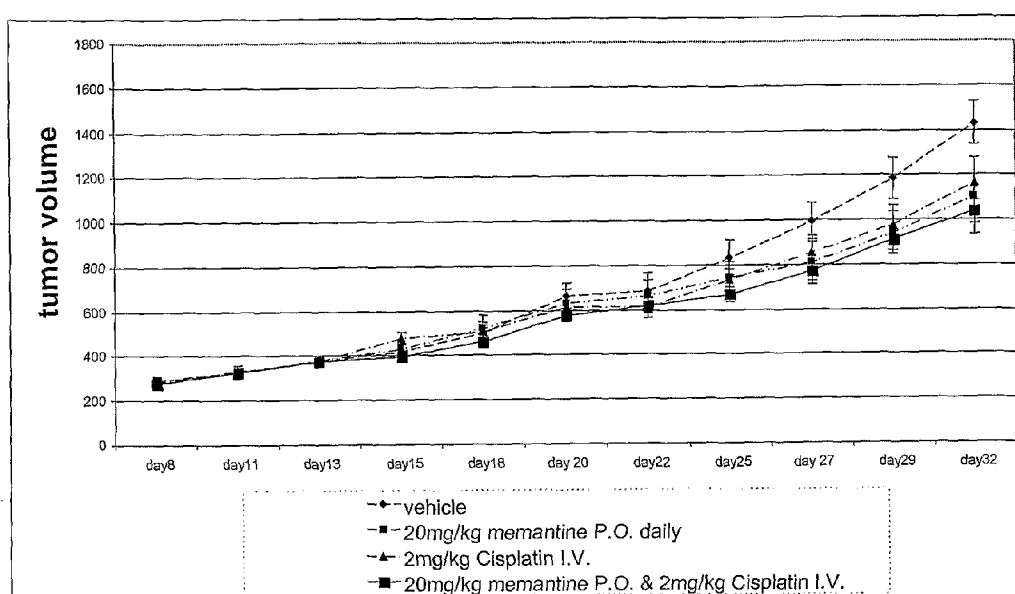
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[Continued on next page]

(54) Title: USE OF NMDA RECEPTOR ANTAGONISTS FOR TREATMENT OF UROLOGIC TUMORS

FIGURE 3



(57) Abstract: A method for treating a subject for urologic cancer or a urologic cancer related disorder comprises administering an NMDA receptor antagonist such as memantine to the subject.

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USE OF NMDA RECEPTOR ANTAGONISTS FOR TREATMENT OF UROLOGIC TUMORS

FIELD OF THE INVENTION

[0001] The present invention relates to the use of NMDA receptor antagonists such as memantine for treatment of urologic tumors, particularly bladder tumors.

BACKGROUND OF THE INVENTION

[0002] Bladder cancer is a common urologic cancer. The most common type of bladder cancer in the United States is urothelial carcinoma, formerly known as transitional cell carcinoma (TCC). The urothelium in the entire urinary tract may be involved, including the renal pelvis, ureter, bladder, and urethra.

[0003] Various treatments are known in the art for bladder cancer yet are not always effective, either because of the nature of the cancer or because of a delay in diagnosis of the disease, or a combination of both. Treatment may include surgical and pharmaceutical approaches, or a combination thereof. The description below centers on some of the available pharmaceutical treatments and their drawbacks.

[0004] For superficial (low grade) cancer, intravesical immunotherapy is available, such as Bacillus Calmette-Guérin (BCG) immunotherapy. However, treatment with BCG has drawbacks. Because BCG is a live attenuated organism, it can cause an acute disseminated tuberculosis-like illness if it enters the bloodstream (BCG sepsis), possibly resulting in death. Therefore, the use of BCG is contraindicated in patients with gross hematuria. More mild side effects may include granulomatous cystitis or prostatitis with bladder contraction. Other drawbacks include the fact that this therapy is less effective in reducing the 5-year recurrence rate for low-grade and low-stage bladder cancer than other available treatments.

[0005] Other forms of adjuvant intravesical chemotherapy for superficial bladder cancer include intravesical triethylenethiophosphoramide (thiotepa [Thioplex]), mitomycin-C, doxorubicin, and epirubicin. Valrubicin has recently been approved as intravesical chemotherapy for bladder cancer that is refractory to BCG. Unfortunately, valrubicin is presently not commercially available. Another treatment is interferon alpha or gamma, which

has been used in the treatment of superficial bladder cancer, either as a single agent therapy or in combination with BCG.

[0006] For more advanced bladder cancers, such as those cancers involved in muscle-invasive disease (clinical stage T2 and greater), other pharmaceutical therapies include adjuvant and neoadjuvant chemotherapy. Neoadjuvant chemotherapy prior to either radical cystectomy or external beam radiotherapy is controversial as it is not clear whether it provides additional benefits to patients.

[0007] The combination of gemcitabine and cisplatin (GC) is a newer regimen and has been shown^{*} to be as efficacious as MVAC (a combination therapy comprising cisplatin, methotrexate, doxorubicin and vinblastine), but with less toxicity. In addition, several novel compounds have shown activity against transitional cell bladder cancer and are now being tested in combination chemotherapy trials. Some of these promising agents are ifosfamide, paclitaxel, docetaxel, and carboplatin.

[0008] However, despite the number of different treatment options, many bladder cancers prove to be refractory to such treatments and/or may be diagnosed too late to benefit from these treatments, particularly since many treatments are most effective for earlier stage cancers.

[0009] Given the current difficulty of successfully treating bladder cancers, the likelihood of recurrence, and the fact that many patients do not respond to available treatments or alternatively respond initially but then stop responding, there is an unmet need for therapies which can successfully treat these cancers.

SUMMARY OF THE INVENTION

[0010] The present invention overcomes the disadvantages of previously available therapeutic options by providing a method for treating bladder cancer with an N-methyl-D-aspartate (NMDA) receptor antagonist. For example, the NMDA receptor antagonist may be memantine, topiramate, budipine, or combinations thereof. According to some embodiments of the invention, the NMDA receptor antagonist operates through a different, unique mechanism of action as compared to available treatments, and hence may be effective for many different types of bladder cancer patients, including patients with initial cancerous lesions, patients who do not respond to such available treatments, or who initially respond but then stop responding, and/or for recurrent bladder cancer.

[0011] In one aspect, the invention provides methods of treating urologic cancer in a patient in need thereof. In some embodiments, the methods comprise administering locally to the cancer; e.g. intravesically to the patient, a pharmaceutical composition comprising an NMDA receptor antagonist in a therapeutically effective amount to treat the urologic cancer. The urologic cancer can be, for example, bladder cancer or urothelial carcinoma. The NMDA receptor antagonist can be, for example, memantine, topiramate, budipine, and/or combinations thereof. In some embodiments, the urologic cancer cells overexpress NMDA receptor subunit NR1.

[0012] In some embodiments, the methods of treating urologic cancer comprise administering a secondary therapeutic agent. The secondary therapeutic agent can be, for example, cisplatin, carboplatin, BCG, valrubicin, thiotepa, mitomycin-C, doxorubicin, epirubicin, MVAC, taxol, taxotere, gemcitabine, GC, interleukin-2 (IL-2) and/or combinations thereof. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be cisplatin. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be carboplatin. The NMDA receptor antagonist and the secondary therapeutic agent can be administered sequentially or simultaneously.

[0013] In some embodiments, the methods of the present invention comprise the steps of administering the pharmaceutical composition into the patient's bladder; the composition being retained in the bladder for a composition retention period lasting at least about one hour; and the composition being expelled from the bladder after the composition retention period is complete. The pharmaceutical composition can be administered into the patient's bladder via a catheter such as a Foley catheter. In some embodiments, the methods comprise maintaining the patient supine for about 25% of the composition retention period, prone for about 25% of the composition retention period, on the patient's left side for about 25% of the composition retention period, and on the patient's right side for about 25% of the composition retention period. The therapeutic regimen can be performed, for example, about once weekly for at least about 6 weeks. These methods can also comprise administering a secondary therapeutic agent.

[0014] In another aspect, the invention provides pharmaceutical compositions comprising an NMDA receptor antagonist, wherein the pharmaceutical composition is formulated for local administration and the NMDA receptor antagonist is present in an amount that is therapeutically effective for treating urologic cancer when administered locally. The NMDA receptor antagonist

can be, for example, memantine, topiramate, budipine and/or combinations thereof. In some embodiments, the compositions can comprise a secondary therapeutic agent. The secondary therapeutic agent can be, for example, cisplatin, carboplatin, BCG, valrubicin, thiotepa, mitomycin-C, doxorubicin, epirubicin, MVAC, taxol, taxotere, gemcitabine, GC and combinations thereof. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be cisplatin. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be carboplatin.

[0015] The pharmaceutical compositions for local administration may comprise a combination therapy comprising an NMDA receptor antagonist and a secondary therapeutic agent, wherein the secondary therapeutic agent has a first effective dosage for treating urologic cancer when administered alone, and the dosage in the pharmaceutical composition is lower than the first effective dosage of the secondary therapeutic agent, the combination therapy achieving at least substantially the same efficacy in inhibiting urologic cancer cell growth as the secondary therapeutic agent achieves when administered alone in the first effective dosage.

[0016] In another aspect, the invention provides kits. In some embodiments, the kits comprise a pharmaceutical composition formulated for intravesical administration comprising a therapeutically effective amount of an NMDA receptor antagonist, a container, and a label associated with the container, wherein the label contains directions for intravesical administration of the pharmaceutical composition for treatment of urologic cancer. In some embodiments, the container is an IV saline bag or a syringe. The NMDA receptor antagonist can be, for example, memantine. In some embodiments, the pharmaceutical composition comprises a secondary therapeutic agent. In some embodiments, the label provides directions for administering the NMDA receptor antagonist and secondary therapeutic agent for treatment of urologic cancer.

[0017] In another aspect, the invention provides methods for predicting the effectiveness of NMDA receptor antagonist therapy for a urologic cancer of a patient. In some embodiments, the methods comprise measuring NMDA receptor activity in urologic tissue of the patient, and, for tissue having elevated NMDA receptor activity, identifying the patient as a candidate for treatment with an NMDA receptor antagonist. The method can further comprise administering an NMDA receptor antagonist to the patient. In some embodiments, the methods further comprise measuring NMDA receptor activity after administration of the NMDA receptor

antagonist, and if the tissue has a reduced post-administration NMDA receptor activity, identifying the cancer as responsive to treatment.

BRIEF DESCRIPTION OF THE DRAWINGS

[0018] **Figure 1** presents a comparison of NMDAR1 expression levels in human bladder cancer cell lines. NMDAR1 expression was determined in cell lysates by immunoblotting with an anti-NMDAR1 antibody (upper panel), and reprobing with anti-tubulin antibody (middle panel). NMDAR1 expression levels detected in the immunoblot were normalized to tubulin protein levels (lower panel).

[0019] **Figure 2** presents a series of graphs demonstrating the inhibition of cell proliferation in bladder cancer cells by the NMDAR1 antagonist memantine hydrochloride.

[0020] **Figure 2A** depicts the inhibition of cell proliferation in T24 cells by memantine. T24 cells were grown in medium containing 0.1% FBS and treated with the indicated concentrations of memantine hydrochloride. Cell proliferation was measured after 72 hrs using BrdU incorporation assay. Results are shown as relative growth compared to untreated cells.

[0021] **Figure 2B** depicts the inhibition of cell proliferation in HT1197 cells by memantine.

[0022] **Figure 2C** depicts the inhibition of cell proliferation in SCaBER cells by memantine.

[0023] **Figure 2D** depicts the dose response of T24 cell proliferation to memantine hydrochloride administration.

[0024] **Figure 2E** depicts the dose response of SCaBER cell proliferation to memantine hydrochloride administration.

[0025] **Figure 2F** depicts the effect of memantine hydrochloride in combination with carboplatin on proliferation of T24 cells.

[0026] **Figure 3** is a graph depicting tumor volume measurements, in mm³, of the T24 bladder cancer cell line xenographed in mice, when treated with memantine, cisplatin and their combination. Error bars represent SEM.

DETAILED DESCRIPTION

[0027] Embodiments of the invention are discussed in detail below. In describing embodiments, specific terminology is employed for the sake of clarity. However, the invention is not intended to be limited to the specific terminology so selected. A person skilled in the relevant art will recognize that other equivalent parts can be employed and other methods developed without parting from the spirit and scope of the invention. All references cited herein are incorporated by reference as if each had been individually incorporated.

[0028] “Treat” refers to preventing, curing, reversing, attenuating, alleviating, minimizing, suppressing or halting at least one of the symptoms or deleterious effects of the diseases, disorders or conditions described herein. Treatment refers to both therapeutic treatment and prophylactic or preventative measures. Those in need of treatment include those already with the disorder as well as those in which the disorder is to be prevented. Hence, the patient to be treated herein may have been diagnosed as having the disorder or may be predisposed or susceptible to the disorder. As used herein the term “treating” refers to preventing, curing, reversing, attenuating, alleviating, minimizing, suppressing or halting the deleterious effects of the above-described diseases, disorders or conditions.

[0029] A “disorder” is any condition that would benefit from treatment with the agent according to the present invention. This includes chronic and acute disorders or diseases including those pathological conditions which predispose the mammal to the disorder in question. These disorders include cancer, for example urologic cancers, and related disorders as described herein.

[0030] The “patient” according to the present invention is a mammal, such as a human, which is diagnosed with one of the diseases, disorders or conditions described herein, or alternatively is predisposed to at least one type of the diseases, disorders or conditions described herein. The compositions of the present invention can be administered to any mammal in need

of the composition that can experience the beneficial effects of the compounds of the invention. Any such mammal is considered a "patient." Such patients include humans and non-humans, such as humans, domestic and farm animals, and zoo, sports, or pet animals, such as dogs, horses, cats, cows, etc. The patient can be a man or a woman. The patient according to the present invention is a mammal, preferably a human which is diagnosed with one of the disease, disorder or conditions described hereinabove, or alternatively is predisposed to at least one type of the cluster or protein-related disease, disorder or conditions described hereinabove.

[0031] The term "therapeutically effective amount" refers to an amount of an NMDA receptor antagonist (and optionally one or more other agents as described herein) that is effective to treat urologic cancer and/or a related disease or disorder in a mammal. The therapeutically effective amount of the agent may reduce the number of cancer cells; reduce the tumor size; inhibit (i.e., slow to some extent and/or stop) cancer cell infiltration into peripheral organs; inhibit (i.e., slow to some extent and/or stop) tumor metastasis; inhibit, to some extent, tumor growth; and/or relieve to some extent one or more of the symptoms associated with the cancer. The therapeutically effective amount of the NMDA receptor antagonist antagonist may be different if a secondary therapeutic agent is included in the therapeutic regimen. The therapeutically effective amount of the NMDA receptor antagonist under these circumstances may readily be determined by a practitioner using animal and clinical trials and medical observations. To the extent that NMDA receptor antagonists (or another agent) may prevent growth and/or kill existing cancer cells, it may be cytostatic and/or cytotoxic. For cancer therapy, efficacy can, for example, be measured by assessing the time to disease progression (TTP) and/or determining the response rate (RR). For any composition used in the methods of the invention, the therapeutically effective amount or dose can be estimated initially from in vitro assays. For example, a dose can be formulated in animal models and such information can be used to more accurately determine useful doses in humans. Determination of a therapeutically effective amount is well within the capability of those skilled in the art.

[0032] A "disorder" is any condition that would benefit from treatment with the agent according to the present invention. This includes chronic and acute disorders or diseases including those pathological conditions which predispose the mammal to the disorder in question. These disorders include bladder cancer and bladder cancer-related disorders as described herein.

[0033] As used herein a "pharmaceutical composition" refers to a preparation of one or more of the active ingredients described herein with other chemical components such as physiologically suitable carriers and excipients. The purpose of a pharmaceutical composition is to facilitate administration of a compound to an organism.

Methods

[0034] In some embodiments, the invention provides methods for treating urologic cancer in a patient in need thereof comprising administering intravesically to the patient a pharmaceutical composition comprising an NMDA receptor antagonist in a therapeutically effective amount to treat the urologic cancer.

[0035] As used herein, "urologic cancer" includes, for example, adrenal cancer, bladder cancer, kidney cancer, penile cancer, prostate cancer, urethral cancer, and testicular cancer. "Bladder cancer" encompasses urothelial cancer, transitional cell cancer, transitional cell carcinoma, TCC, urothelial tumors, carcinoma in situ, CIS, squamous cell carcinoma and/or SCC when present in the urological system, and urothelial carcinoma. As used herein, "urothelial carcinoma" and "transitional cell carcinoma" are substantially equivalent terms. In bladder cancer, any part of the urothelium in the urinary tract may be involved, including any one or more of the renal pelvis, ureter, bladder, and urethra. Also encompassed within the term "bladder cancer" are metastatic cancers having their origin in any of the above cancers and/or locations.

[0036] Bladder cancer is a malignancy of the genitourinary system. Bladder cancer is often described as a polyclonal field change defect with frequent recurrences due to a heightened potential for malignant transformation. However, bladder cancer has also been described as a problem with implantation and migration from a previously affected site.

[0037] Bladder cancer is unique in several respects. First, bladder cancer exhibits polychronotropism, which is the tendency to recur over time and in new locations in the urothelial tract. It also exhibits multifocality – i.e., it is characterized by the formation of cancerous growths at several places in the bladder, rather than just a single tumor. Several theories have arisen to explain these characteristics. One such theory posits that these traits result from clonal expansion of transient cells (which is common to metastatic cancers); another theory attributes these traits to the existence and proliferation of cancer stem cells.. Bladder

cancer is often characterized by a mutation in one or more of the following genetic loci: KRAS, p53, EGF, p16, p21, BCL2.

[0038] The clinical course of bladder cancer can be associated with a broad spectrum of aggressiveness and risk. Low-grade, superficial bladder cancers have minimal risk of progression to death; however, high-grade muscle-invasive cancers are often lethal.

[0039] Almost all bladder cancers are epithelial in origin. The urothelium consists of a 3- to 7-cell mucosal layer within the muscular bladder. Of these urothelial tumors, more than 90% are transitional cell carcinomas. However, up to 5% of bladder cancers are squamous cell in origin, and 2% are adenocarcinomas. Non-urothelial primary bladder tumors are extremely rare and may include small cell carcinoma, carcinosarcoma, primary lymphoma, and sarcoma. Transitional cell sarcomas also found in kidney and urethra.

[0040] The World Health Organization classifies bladder cancers as low grade (grade 1 and 2) or high grade (grade 3). Tumors are also classified by growth patterns: papillary (70%), sessile or mixed (20%), and nodular (10%). Carcinoma in situ (CIS) is a flat, noninvasive, high-grade urothelial carcinoma. The most significant prognostic factors for bladder cancer are grade, depth of invasion, and the presence of CIS.

[0041] Upon presentation, 55-60% of patients have low-grade superficial disease, which is usually treated conservatively with transurethral resection and periodic cystoscopy. Forty to forty-five percent of patients have high-grade disease, of which 50% is muscle invasive and is typically treated with radical cystectomy.

[0042] Less than 5% of bladder cancers in the United States are squamous cell carcinomas (SCCs). However, worldwide, SCC is the most common form, accounting for 75% of bladder cancer in underdeveloped nations. In the United States, SCC is associated with persistent inflammation from long-term indwelling Foley catheters and bladder stones. In underdeveloped nations, SCC is associated with bladder infection by the parasite *Schistosoma haematobium*.

[0043] Adenocarcinomas account for less than 2% of primary bladder tumors. These tumors are observed most commonly in exstrophic bladders and respond poorly to radiation and chemotherapy. Radical cystectomy is the treatment of choice.

[0044] Small cell carcinomas are aggressive tumors associated with a poor prognosis and are thought to arise from neuroendocrine stem cells.

[0045] Carcinosarcomas are highly malignant tumors that contain both mesenchymal and epithelial elements.

[0046] For non-muscle invasive, superficial (low grade) forms of bladder cancer, intravesical treatment options are available. Examples of approved intravesical treatments include, for example, thiotepa, valrubicin and Bacillus Calmette-Guérin (BCG) immunotherapy.

[0047] BCG is a common and generally effective therapeutic option for non-muscle invasive bladder cancer. BCG immunotherapy may help to decrease the rate of recurrence and progression. It involves administering a live attenuated strain of *Mycobacterium bovis*, often mixed into a saline solution. Some early studies reported that an immune response against BCG surface antigens cross-reacted with putative bladder tumor antigens, and this was proposed as the mechanism for the therapeutic effect of BCG. However, multiple subsequent studies refute this claim and demonstrate that BCG induces a nonspecific, cytokine-mediated immune response to foreign protein.

[0048] Treatment with BCG also has drawbacks. Because BCG is a live attenuated organism, it can cause an acute disseminated tuberculosis-like illness if it enters the bloodstream (BCG sepsis), possibly resulting in death. Therefore, the use of BCG is contraindicated in patients with gross hematuria. More mild side effects may include granulomatous cystitis or prostatitis with bladder contraction. Other drawbacks include the fact that this therapy is less effective in reducing the 5-year recurrence rate for low-grade and low-stage bladder cancer.

[0049] Another treatment is interferon alpha or gamma. Interferon alpha or gamma has been used in the treatment of superficial bladder cancer, either as a single agent therapy or in combination with BCG. Its role has primarily been in post-BCG failure with early promising results. Although BCG with interferon has shown a 42% response with tolerable side effects after BCG failure, no evidence has indicated that re-treating with BCG with interferon is superior to re-treating with BCG alone.

[0050] Other forms of adjuvant intravesical chemotherapy for superficial bladder cancer include intravesical triethylenethiophosphoramide (i.e., thiotepa, which is marketed under the trade name Thioplex™). Thiotepa is used in breast, ovary and bladder cancers. Intravesical therapies for superficial bladder cancer also include mitomycin-C, doxorubicin, and epirubicin. Although these agents may increase the time to disease recurrence, no evidence indicates that

these therapies prevent disease progression. Also, no evidence suggests that these adjuvant therapies are as effective as BCG.

[0051] Valrubicin has been approved as intravesical chemotherapy for bladder cancer that is refractory to BCG. Valrubicin is an analog of doxorubicin. Doxorubicin is used to treat many types of cancer. In contrast, valrubicin is used specifically for bladder cancer because, being more toxic than doxorubicin, it generally can be tolerated only when administered locally (e.g., intravesically). In patients whose conditions do not respond to BCG, the overall response rate to valrubicin is approximately 20%, and in some patients it can delay time to cystectomy. Unfortunately, valrubicin is presently not commercially available.

[0052] Another treatment is interferon alpha or gamma. Interferon alpha or gamma has been used in the treatment of superficial bladder cancer, either as a single agent therapy or in combination with BCG. Its role has primarily been in post-BCG failure with early promising results. Although BCG with interferon has shown a 42% response with tolerable side effects after BCG failure, no evidence has indicated that re-treating with BCG with interferon is superior to re-treating with BCG alone.

[0053] For more advanced bladder cancers, such as those cancers involved in muscle-invasive disease (clinical stage T2 and greater), other pharmaceutical therapies include adjuvant and neoadjuvant chemotherapy. Neoadjuvant chemotherapy prior to either radical cystectomy or external beam radiotherapy is controversial as it is not clear whether it provides additional benefits to patients. Chemotherapeutic options for muscle invasive forms of bladder cancer include, for example, taxol.

[0054] In one small series, the T4 tumors of 45% of affected patients responded to chemotherapy, making potentially curative cystectomy possible. Although no definite evidence of benefit exists, patients with P3-P4 or N+ bladder cancer in the United States are typically advised to receive adjuvant chemotherapy. Chemotherapeutic agents for metastatic disease include MVAC, which is the standard treatment of metastatic bladder cancer. MVAC has an objective response rate of 57-70%, a complete response rate of 15-20%, and a 2-year survival rate of 15-20%. The combination of gemcitabine and cisplatin (GC) is a newer regimen and has been shown to be as efficacious as MVAC, but with less toxicity. GC is now considered a first-line treatment agent for bladder cancer.

[0055] In some embodiments, the methods of the present invention comprise administering an NMDA receptor antagonist to a patient. As used herein, "NMDA receptor antagonist" includes any agent that interferes with or inhibits the proper functioning of the NMDA receptor. For example, "NMDA receptor antagonist" includes memantine, topiramate and budipine. However, other NMDA receptor antagonists are contemplated, including competitive antagonists, noncompetitive antagonists (e.g., those that block the binding of glycine, another ligand that is necessary to proper NMDA receptor functioning); and uncompetitive antagonists (e.g., those that block the NMDA receptor channel).

[0056] "NMDA receptor antagonist" encompasses one or more of the following: topiramate (Johnson & Johnson); huperzine A (Georgetown University/Neuro Hi-Tech); Zenvia™ (AVANIR Pharmaceuticals); EVT-101 (Roche Holding AG); perzinfotel (Wyeth Research); dizocilpine (Merck & Co., Inc); CNS-5161 (chemical formula C₁₆ H₁₈ Cl N₃ S₂; lanicemine (chemical formula C₁₃ H₁₄ N₂); delucemine (NPS Pharmaceuticals); indantadol (Chiesi Farmaceutici SpA); intranasal ketamine (Javelin); AV-101 (University of Maryland/VistaGen); Neu-2000 (Neurotech Pharmaceuticals Inc); budipine (ALTANA Pharma AG); 5-(pentafluorobenzyl)aminosalicylic acid; methylphenylethynylpyridine (Novartis); neramexane (Merz & Co. GmbH); TAT-NR2B9c (NoNO and Arbor Vita); RGH-896 (Gedeon Richter and Forest Laboratories); ipenoxazone (Nippon Chemiphar Co. Ltd.); dexamabinol and related prodrugs (Hebrew University); acamprosate (Somaxon); gacyclidine (Ipsen); AD-529 (Sosei R&D Ltd (Sosei Group Corp); ED-1812 (Sosei R&D); memantine; CP-101616 (Pfizer, Inc.); NMDA glycine B antagonists (Grunenthal) (Merz & Co. GmbH); NMDA glycine B site antagonists (Merz); intravenous NMDA NR2B antagonists (RO-25-6981) (Evotec Neurosciences); NMDA/calcium channel antagonists (chemical formula C₂₀ H₂₃ N O₂) (Allelix); AZD-4282 (AstraZeneca plc); remacemide; bis-(7)-tacrine; kaitocephalin; CGX-1007, also referred to as conantokin-G (Cognetix and Medtronic); ifenprodil; aptiganel;; traxoprodil; BI-II-277-CL (Boehringer Ingelheim Corp.); glycine antagonists (GlaxoSmithKline) (CAS ID 476689-77-7); eliprodil; indole-2-carboxylates (Searle); ACEA-1021 (Acea Pharmaceuticals, Inc.); NMDA antagonists (Sumitomo) (CAS ID 295788-60-2); YT-1006 (Yaupon Therapeutics, Inc.); L-701324 (Merck & Co., Inc.); midafotel; SSNRAs (Purdue Neuroscience/Pfizer) (CAS ID 302799-86-6); NMDA antagonists (Vernalis) (CAS ID 22947-33-7); besonprodil; cinnamide-based NMDA antagonists (N-(2-(4-Hydroxyphenyl)ethyl)-4-chlorocinnamide) (CoCensys);

CNS-5788 (CeNeS Pharmaceuticals, Inc.); EAB-318 (chemical formula C₁₁ H₁₃ Cl N₃ O₅ P) (Wyeth Research); EMD-95885 (Merck KGaA); EN-3231 (MorphiDex, Endo Pharmaceuticals Holdings, Inc.); FR-115427 (Fujisawa Pharmaceutical Co., Ltd.); NPS-1407 (a diphenylpropylamine analog) (NPS Pharmaceuticals, Inc.); Ro-8-4304; selfotel; UK-315716 (Pfizer, Inc.); ZD-9379 (AstraZeneca plc); ACEA-1011, ACEA-1286, ACEA-1328, and ACEA-1416 (for all ACEA Purdue Neuroscience Corp); LY-235959 (Eli Lilly & Co.); L-689560 (Merck & Co., Inc.); HA-966 (CAS ID 1003-51-6); GPI-3000 (Scios Inc (Johnson & Johnson)); UPF-648 (chemical formula C₁₁ H₈ Cl₂ O₃) (Pharmacia & Upjohn SpA (Pfizer, Inc.)); NMDA antagonists (chemical formula C₂₂ H₂₄ N₄ O₂) (Tennessee University); ADCI (National Institutes of Health) ES-242-1 (chemical formula C₃₄ H₃₆ O₁₀); LY-233053 (Eli Lilly & Co.); LY-235723 (Eli Lilly & Co.); PD-158473 (Pfizer, Inc.); ACEA-1031 (ACEA Purdue Neuroscience Corp.); CP-283097 (Pfizer, Inc.); NPC-12626 (GlaxoSmithKline plc); himantane; CGP-39653 (Novartis AG); 5-propynylphosphorothioate; DD-B4 (DiverDrugs SL); FPL-15609 (Astra AB); Ro-24-6449 (Roche Holding AG); NC-1210 (Queens University at Kingston); MDL-100453 (Hoechst Marion Roussel, Inc. (Sanofi-Aventis)); L-703717 (Merck & Co., Inc.); L-708541 (Merck & Co., Inc.); kynureninase inhibitor (St. Andrews University) (CAS ID 440354-06-3); LY-274614 (Eli Lilly & Co.); NNC-07-9202 (Novo Nordisk A/S); WIN-67870-2 (Sterling Winthrop Products, Inc.); CNS-1044 (CeNeS Pharmaceuticals, Inc.); CNS-1118 (CeNeS Pharmaceuticals, Inc.); L-698544 (Merck & Co., Inc.); L-701252 (Merck & Co., Inc.); L-701273 (Merck & Co., Inc.); LY-235497 (Eli Lilly & Co.) ; PD-159913 (Pfizer, Inc.); Ro-24-6173 (Roche Holding AG); WIN-63480-2 (Sterling Winthrop Group Ltd.); RPR-118723 (Aventis Pharma AG (Sanofi-Aventis)); Conantokin-R; L-687414 (Merck & Co., Inc.); L-705022 (Merck & Co., Inc.); CGP-37849 (Novartis AG); MDL-100748 (Hoechst Marion Roussel, Inc.); MDL-29951 (Hoechst Marion Roussel, Inc.); EGIS-7444 (EGIS Gyogyszergyar RT); MK-801 analogs (Merck) (CAS ID 157874-71-0); MDL-105519 (Hoechst Marion Roussel, Inc.); ADD-17014 (University of Kentucky); CNS-5065 (CeNeS Pharmaceuticals Inc); CNS-1505 (CeNeS Pharmaceuticals, Inc.); CNS-1524 (CeNeS Pharmaceuticals, Inc.); CNS-1531 (CeNeS Pharmaceuticals, Inc.); NPS-1392 (chemical formula C₁₆ H₁₇ F₂ N); RPR-104632 (Rhone-Poulenc Rorer, Inc.); PD-138289 (Pfizer, Inc.); HO-473 (NPS Pharmaceuticals, Inc.); FPL-16283 (Fisons plc); L-703600 (Merck & Co., Inc.); LY-233536 (Eli Lilly & Co.); L-698532

(Merck & Co., Inc.); L-701315 (Merck & Co., Inc.); L-701376 (Merck & Co., Inc.); and/or CP-112116 (Pfizer, Inc.).

[0057] In some embodiments, the NMDA receptor antagonist is memantine. Memantine is the first in a novel class of Alzheimer's disease medications acting on the glutamatergic system. Memantine was developed by Merz and licensed to Forest for the U.S. and Lundbeck for selected European and international markets. Memantine is marketed under the brands Axura® and Akatinol® by Merz, Namenda® by Forest and Ebixa® by Lundbeck. As used herein, "memantine" encompasses, for example, memantine hydrochloride.

[0058] A dysfunction of glutamatergic neurotransmission, manifested as neuronal excitotoxicity, is involved in the etiology of Alzheimer's disease. Targeting the glutamatergic system, specifically NMDA receptors, offers a novel approach to treatment in view of the limited efficacy of existing drugs targeting the cholinergic system. Memantine is a moderate-affinity voltage-dependent uncompetitive antagonist at glutamatergic NMDA receptors.

[0059] By binding to the NMDA receptor with a higher affinity than magnesium ions, Memantine is able to inhibit the prolonged influx of calcium ions which forms the basis of neuronal excitotoxicity. The low affinity of memantine, however, preserves the physiological function of the receptor as it can still be activated by the relatively high concentrations of glutamate released following depolarization of the presynaptic neuron.

[0060] Surprisingly, according to the present invention, bladder cancer can be treated with an NMDA receptor antagonist, such as, for example, memantine. NMDA receptor antagonists, such as, for example, memantine, have a therapeutic effect for inhibiting cancer cell proliferation in such patients.

[0061] The therapeutically effective dosage for memantine can be determined by one of ordinary skill in the art. Such a dosage can be, for example, up to or at least about 1 mg/day, about 2 mg/day, about 3 mg/day, about 4 mg/day, about 5 mg/day, about 6 mg/day, about 7 mg/day, about 8 mg/day, about 9 mg/day, about 10 mg/day, about 11 mg/day, about 12 mg/day, about 13 mg/day, about 14 mg/day, about 15 mg/day, about 16 mg/day, about 17 mg/day, about 18 mg/day, about 19 mg/day, about 20 mg/day, about 21 mg/day, about 22 mg/day, about 23 mg/day, about 24 mg/day, about 25 mg/day, about 30 mg/day, about 35 mg/day, about 40 mg/day, about 55 mg/day, about 60 mg/day or more. When administered locally, other dosages may be used. Such a dosage can be, for example, up to or at least about 100 mg per

administration or about 200 mg, about 300 mg, about 400 mg, about 500 mg, about 600 mg, about 700 mg, about 800 mg, about 900 mg, about 1000 mg, about 1100 mg, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg per administration or more. These dosages can be given in a single unit dosage per day or administration, or in multiple unit dosages. Different unit dosages in a multiple unit dosage regimen may be equal or unequal in amount. The target dosage can be achieved by titrating up. For example, the starting dose of memantine can be 5 mg/day, then increased on a weekly basis by 5 mg/day increments until a target dosage of 20 mg/day is achieved. Other starting and target dosages are encompassed within the invention, in accordance with the therapeutically effective dosages described above and as would be understood by a person of skill in the art. As used herein, "about" may refer to a range from 10% below the referenced number to 10% above the referenced number. For example, "about 50" may mean from 45 to 55. Other meanings of "about" may be apparent from the context.

[0062] In some embodiments, the NMDA receptor antagonist is topiramate. Topiramate is an anticonvulsant drug that is used to treat epilepsy and migraines. It has also been used as a treatment for bipolar disorder, obesity and alcoholism. Topiramate enhances activation of gamma amino butyric acid (GABA) receptors and acts to inhibit the alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor, which, like NMDA receptors, is a type of glutamate receptor. Although topiramate may not be considered in the literature to be an NMDA receptor antagonist, it is included in the meaning of the term as used here. It is generally well-tolerated. The most commonly reported side effects are somnolence, fatigue, weight loss and nervousness.

[0063] The therapeutically effective dosage for topiramate can be determined by one of ordinary skill in the art. Such a dosage can be, for example, up to or at least about 10 mg/day, about 20 mg/day, about 30 mg/day, about 40 mg/day, about 50 mg/day, about 60 mg/day, about 70 mg/day, about 80 mg/day, about 90 mg/day, about 100 mg/day, about 120 mg/day, about 140 mg/day, about 160 mg/day, about 180 mg/day, about 200 mg/day, about 250 mg/day, about 300 mg/day, about 350 mg/day, about 400 mg/day, about 450 mg/day, about 500 mg/day, about 550 mg/day, about 600 mg/day, about 650 mg/day, about 700 mg/day, about 750 mg/day, about 800 mg/day or more. When administered locally, other dosages may be used. Such a dosage can be, for example, up to or at least about 1000 mg per administration, or about 1100 mg, about 1200

mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2200 mg, about 2400 mg, about 2600 mg, about 2800 mg, about 3000 mg, about 3500 mg, about 4000 mg, about 4500 mg, about 5000 mg, about 5500 mg, about 6000 mg, about 7000 mg, about 8000 mg, about 9000 mg, about 10,000 mg per administration or more. These dosages can be given in a single unit dosage per day or administration, or in multiple unit dosages. The target dosage can be achieved by titrating up. For example, a target dosage of 400 mg/day can be achieved by administering about 25 mg twice daily in week 1, about 50 mg twice daily in week 2, about 75 mg twice daily in week 3, about 100 mg twice daily in week 4, about 150 mg twice daily in week 5, and about 200 mg twice daily in week 6. Other starting and target dosages are encompassed within the invention, in accordance with the therapeutically effective dosages described above and as would be understood by a person of skill in the art. Different unit dosages in a multiple unit dosage regimen may be equal or unequal in amount.

[0064] In some embodiments, the NMDA receptor antagonist is budipine. Budipine acts as an uncompetitive inhibitor of the NMDA receptor. It has also been associated with several other mechanisms of action. For example, it has been shown to exert dopaminergic, antimuscarinic and anti-GABA effects. Eltze, M. J Neural Transm Suppl. 1999;56:83-105. Budipine is generally well-tolerated.

[0065] The therapeutically effective dosage for budipine can be determined by one of ordinary skill in the art. Such a dosage can be, for example, up to or at least about 1 mg/day, about 2 mg/day, about 3 mg/day, about 4 mg/day, about 5 mg/day, about 7.5 mg/day, about 10 mg/day, about 11 mg/day, about 12 mg/day, about 13 mg/day, about 14 mg/day, about 15 mg/day, about 16 mg/day, about 17 mg/day, about 18 mg/day, about 19 mg/day, about 20 mg/day, about 21 mg/day, about 22 mg/day, about 23 mg/day, about 24 mg/day, about 25 mg/day, about 26 mg/day, about 27 mg/day, about 28 mg/day, about 29 mg/day, about 30 mg/day, about 35 mg/day, about 40 mg/day, about 45 mg/day, about 50 mg/day, about 55 mg/day, about 60 mg/day or more. When administered locally, other dosages may be used. Such a dosage can be, for example, about 100 mg per administration or about 150 mg, about 200 mg, about 300 mg, about 400 mg, about 500 mg, about 600 mg, about 700 mg, about 800 mg, about 900 mg, about 1000 mg, about 1200 mg, about 1400 mg, about 1600 mg, about 1800 mg, about 2000 mg, about 2500 mg, about 3000 mg, about 3500 mg, about 4000 mg per

administration or more. These dosages can be given in a single unit dosage per day or administration, or in multiple unit dosages. The target dosage can be achieved by titrating up. Different unit dosages in a multiple unit dosage regimen may be equal or unequal in amount.

[0066] According to the method of the present invention, the NMDA receptor antagonist can be administered in any dosage that would be therapeutically effective in the route of administration chosen by the health care provider. For example, any of the NMDA receptor antagonists can be administered in an amount from about 1 mg/kg to about 250 mg/kg, or otherwise up to the solubility limit. For example, the NMDA receptor antagonist can be administered in an amount up to or at least about 1 mg/kg, about 5 mg/kg, about 10 mg/kg, about 15 mg/kg, about 16 mg/kg, about 17 mg/kg, about 18 mg/kg, about 19 mg/kg, about 20 mg/kg, about 21 mg/kg, about 22 mg/kg, about 23 mg/kg, about 24 mg/kg, about 25 mg/kg, about 30 mg/kg, about 35 mg/kg, about 40 mg/kg, about 45 mg/kg, about 50 mg/kg, about 60 mg/kg, about 70 mg/kg, about 80 mg/kg, about 90 mg/kg, about 100 mg/kg, about 120 mg/kg, about 140 mg/kg, about 160 mg/kg, about 180 mg/kg, about 200 mg/kg, about 225 mg/kg, about 250 mg/kg or more per administration.

[0067] The NMDA receptor is found in many tissues in the body. In neural tissue, the NMDA receptor contributes to excitatory synaptic transmission and is thought to play a role in learning and memory. It is unusual in that it is both a ligand-gated and voltage-gated ion channel. NMDA receptors bind glutamate, which regulates the proliferation, migration, and survival of neuronal progenitors and immature neurons during development (Guerrini L, Blasi F, Denis-Donini S. Synaptic activation of NF-kappa B by glutamate in cerebellar granule neurons in vitro. *Proc Natl Acad Sci U S A*. 1995 Sep 26;92(20):9077-81). Glutamate activates ionotropic and metabotropic receptors. The ionotropic glutamate NMDA receptors assemble from subsets of two subunits, NR1 and NR2. The NR1 subunit influences heteromeric NMDA channels ion permeability, whereas the NR2 subunits A, B, C, and D determine the electrophysiological properties of the channel (Takano T, Lin JH, Arcuino G, Gao Q, Yang J, Nedergaard M. Glutamate release promotes growth of malignant gliomas. *Nat Med*. 2001 Sep;7(9):1010-5). NMDA receptors are expressed both in neurons and in non-neuronal tissues. Activation of glutamate NMDA receptors in neurons is translated to the nucleus by the extracellular signal-regulated kinase (ERK1/2)-signaling cascade, leading to the phosphorylation of the cAMP-responsive element binding protein (CREB) and activation of genes promoting survival (Hansen

HH, Briem T, Dietko M, Siffringer M, Voss A, Rzeski W, Zdzisinska B, Thor F, Heumann R, Stepulak A, Bittigau P, Ikonomidou C. Mechanisms leading to disseminated apoptosis following NMDA receptor blockade in the developing rat brain. *Neurobiol Dis.* 2004 Jul;16(2):440-53). The NMDA ligand-gated ion-channel is permeable to the cations Na⁺, K⁺ and, importantly, Ca²⁺. Presence of the NMDA receptor (NMDAr) and its involvement in cellular proliferation is well-known in tumor cells derived from neuronal tissue, such as glioma and neuroblastoma and prostate (Yoshioka et al., 1997; Yoshioka A, Ikegaki N, Williams M, Pleasure D. Expression of N-methyl-D-aspartate (NMDA) and non-NMDA glutamate receptor genes in neuroblastoma, medulloblastoma, and other cells lines. *J Neurosci Res.* 1996 Oct 15;46(2):164-78; North et al., 1997; North WG, Fay MJ, Du J, Cleary M, Gallagher JD, McCann FV. Presence of functional NMDA receptors in a human neuroblastoma cell line. *Mol Chem Neuropathol.* 1997 Jan-Feb;30(1-2):77-94). However, not much is known about NMDAr expression and activity in other tumor types.

[0068] The NMDA receptor is a heterodimer of the NR1 and NR2 subunits. The NR1 subunit binds to the co-agonist glycine and the NR2 subunit binds glutamate. There are at least eight different isoforms of the NR1 subunit (designated NR1-1a and -1b; -2a and -2b; -3a and -3b; and -4a and -4b) and at least four isoforms of the NR2 subunit (designated NR2A, NR2B, NR2C, and NR2D). The NMDA receptors expressed in different tissues vary in the isoforms of each subunit expressed. For example, esophageal cancer cells express the NR2B isoform, but gastric cancer cell lines do not. Watanabe, K. et al. *Biochem. and Biophys. Res. Comm.* 367 (2008) 487-490. In addition, the effect of a particular subunit on proliferation varies across tissue types. For example, in esophageal cancer cells proliferation is inhibited by inactivation of the NR2B subunit. However, gastric cancer cells do not express the NR2B subunit. In gastric cancer cells, proliferation has been shown to be inhibited by inactivating the NR2A subunit instead. Watanabe, K. et al. *Biochem. and Biophys. Res. Comm.* 367 (2008) 487-490. According to the invention, NMDA receptor antagonists may exert their effects on the particular NMDA subunits present in urological cancers and responsible for cell proliferation there, as distinguished from different NMDA receptor compositions in other cell types, with different identity of the subunit that influences cell proliferation. Thus, the fact that particular NMDA receptor antagonists demonstrate efficacy in treating urological cancer is a significant and unexpected discovery.

[0069] In some embodiments, the urologic cancer cells overexpress NMDA receptor subunit NR1. As used herein, “overexpress” means that, for example, the receptor or NR1 subunit is present in an amount greater than that found in non-cancerous cells of the same tissue type. As used herein, “test amount” refers to the amount of a receptor or subunit in a patient and/or tissue with a urological cancer. As used herein, “control amount” refers to the amount of a receptor or subunit in a patient and/or tissue (taken from same patient or from a different patient) without the disease or condition. If the test amount of a receptor or subunit, such as NR1, exceeds the control amount for the receptor or subunit, the receptor or subunit is overexpressed. The test amount and control amount, as well as the difference between the two, can be conveyed either in terms of an absolute amount (e.g., microgram/ml) or a relative amount (e.g., relative intensity of signals).

[0070] In some embodiments, the methods of the present invention comprise administering a secondary therapeutic agent. As used herein, “secondary therapeutic agent” refers to any agent that can be administered simultaneously with, prior to or subsequently to the NMDA receptor antagonist in such a way as to be therapeutically effective according to the present invention. The secondary therapeutic agent may include any treatment protocol for bladder cancer, including but not limited to MVAC (cisplatin, methotrexate, doxorubicin and vinblastine), carboplatin, taxol, taxotere, gemcitabine, cisplatin and the combination of gemcitabine and cisplatin (i.e., GC), BCG, valrubicin, thiotepa, mitomycin-C, doxorubicin, epirubicin, and combinations thereof. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be cisplatin. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be carboplatin. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be gemcitabine. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be thiotepa. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be valrubicin. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be BCG. In each case, topiramate and/or budipine can replace memantine as the NMDA receptor antagonist. A combination of an NMDA receptor antagonist with one or more known treatments can result in a synergistic effect, and/or delay the occurrence of resistance. It will be appreciated that treatment of the above-described diseases according to the present invention may be

combined with other treatment methods known in the art (*i.e.*, combination therapy). Thus, treatment of malignancies using memantine may be combined with, for example, radiation therapy, antibody therapy and/or chemotherapy. As used herein, such therapies are encompassed within the term “secondary therapeutic agents.”

[0071] The NMDA receptor antagonist and the secondary therapeutic agent can be administered simultaneously or sequentially. For example, the NMDA receptor antagonist can be administered before or after the secondary therapeutic agent is administered. As used herein, “sequentially” encompasses administering the NMDA receptor antagonist before or after administering the secondary therapeutic agent. As used herein, “simultaneously” encompasses treatment regimens in which the start and end of both treatment with an NMDA receptor antagonist and treatment with a secondary therapeutic agent occur at the same time. It also encompasses situations in which the NMDA receptor antagonist and the secondary therapeutic agent treatment regimens overlap, without regard to whether they are started or completed at the same or different times. For example, if the NMDA receptor antagonist and the secondary therapeutic agent treatment regimens overlap in time at any point during the overall treatment regimen, they are considered to constitute simultaneous treatment as used herein. The invention contemplates administering the NMDA receptor antagonist early in therapy, and administering the secondary treatment later in therapy, optionally with an overlap, and optionally beginning both treatments simultaneously. The NMDA receptor antagonist and the secondary therapeutic agent can be administered via different routes of administration. For example, the NMDA receptor antagonist can be administered locally and the secondary therapeutic agent administered systemically; or the NMDA receptor antagonist can be administered systemically and the secondary therapeutic agent administered locally.

[0072] The methods described herein comprise administration of NMDA receptor antagonists for treatment of urologic cancer. These methods can be employed without regard to other uses for the NMDA receptor antagonists. For example, some NMDA receptor antagonists have been used in the treatment of various diseases or disorders for their neuroprotective effects. According to some embodiments of the present invention, NMDA receptor antagonists can be administered to a patient who does not require the administration of neuroprotective agents. In other words, NMDA receptor antagonists exhibit surprising efficacy when administered

intravesically to a patient suffering from bladder cancer, regardless of whether the patient is in need of any neuroprotective effects brought about by the NMDA receptor antagonist.

[0073] In some embodiments, the methods of the present invention comprise administering one or more NMDA receptor antagonists locally to a patient in need thereof, with or without concomitant administration of a secondary therapeutic agent. As used herein, "local" refers to administration of an active agent in such a way that a specific location or portion of the patient's body is exposed to the active agent. Examples of local administration include, for example, topical application of a composition comprising an active agent to an affected portion of a patient's skin. Another example of local administration is intravesical administration. As used herein, "intravesical" refers to administration into a patient's bladder. An example of intravesical administration involves treating a patient with BCG immunotherapy. However, as would be appreciated by a person of skill in the art, any clinically acceptable method of local administration of any active agent into a patient's bladder is contemplated within the term "intravesical," as used herein. As used herein, "systemic" refers to administration of an active agent in such a way that more than just a specific location in the patient's body is administered to the patient. Systemic administration can result in delivery to a portion of the patient's body or generally throughout the body. Examples of systemic administration routes include, without limitation, oral and intravenous administration.

[0074] According to certain embodiments of the invention, the NMDA receptor antagonist and/or the secondary therapeutic agent are delivered via whatever route of administration and in whatever dosage levels would, in the sound medical judgment of a qualified healthcare provider, deliver a therapeutically effective dose of the NMDA receptor antagonist and/or secondary therapeutic agent to the affected area of the patient. Local and/or systemic administration are encompassed within the invention. In addition, the active agent can be delivered to the affected area via, for example, the bloodstream, the lymphatic system, or through extracellular fluid. The invention contemplates delivery of metabolites of the NMDA receptor antagonist and/or the secondary therapeutic agent to the affected area in circumstances where the metabolites are effective to treat the cancer. The invention also contemplates metabolism of NMDA receptor antagonists, secondary therapeutic agents, and/or interactions between them and/or their metabolites that result in effective treatment of the cancer. In addition, the invention contemplates administering drugs locally via systemic administration in

circumstances where therapeutically effective doses can be delivered to the affected area without causing unacceptable side effects or toxicities. For example, oral administration of a composition of the present invention can result in delivery of the active agent to the bladder via normal excretory processes, where the agent or its metabolites remain effective to treat the cancer upon reaching the bladder, and treatment occurs while the active agent is resident in the bladder prior to excretion. Such a result can be achieved where, for example, the active agent passes through the kidneys or liver unaltered.

[0075] In some embodiments, the methods of the present invention relate to the intravesical administration of a pharmaceutical composition comprising one or more NMDA receptor antagonists. The intravesical administration can be carried out via any method known in the art for local administration of a pharmaceutically active agent. For example, the method can comprise the steps of administering the pharmaceutical composition into the patient's bladder; the composition being retained in the bladder for a composition retention period lasting at least about one hour; and the composition being expelled from the bladder after the composition retention period is complete. In some embodiments, the pharmaceutical composition can be administered into the patient's bladder via a catheter, such as a Foley catheter. The method can also comprise, without regard to order, maintaining the patient supine for about 25% of the composition retention period, prone for about 25% of the composition retention period, on the patient's left side for about 25% of the composition retention period, and on the patient's right side for about 25% of the composition retention period. The composition retention time can be, for example, at least about 1 hour. The composition retention time can also be about 2 hours, in which case the patient lies supine, prone, on the patient's left side, and on the patient's right side for about 15 minutes each, and the patient stands upright for about an additional 1 hour. This therapeutic regimen can be repeated on whatever schedule and for whatever period of time that a physician, acting within sound clinical judgment, deems appropriate. For example, the therapeutic regimen can be performed at least about once weekly for at least about 6 weeks. The pharmaceutical composition is generally formulated for intravesical delivery. Any of the aforementioned methods can also comprise administering a secondary therapeutic agent.

[0076] In some embodiments, the methods of the present invention comprise performing a secondary procedure. The term "secondary procedure" encompasses, without limitation,

radiation treatment, surgery or a combination thereof. "Secondary procedure" also encompasses any procedure that can accompany administration of an NMDA receptor antagonist for treatment of any of the disorders or conditions described herein. As used herein, "perform" encompasses completing the procedure to the point that the procedure is, or within reasonable medical judgment should, be effective to treat the cancer, whether alone or in combination with an NMDA receptor antagonist. As used herein, "secondary treatment" encompasses "secondary therapeutic agent" and "secondary procedure."

[0077] In some embodiments, the invention provides methods of treating bladder cancer or bladder cancer-related diseases, disorders or conditions comprising administering NMDA receptor antagonists to a patient in need thereof.

Compositions

[0078] In some embodiments, the invention provides pharmaceutical compositions comprising an NMDA receptor antagonist, wherein the pharmaceutical composition is formulated for local administration and the NMDA receptor antagonist is present in an amount that is therapeutically effective for treating urologic cancer when administered locally. The NMDA receptor antagonist can be, without limitation, memantine, topiramate, budipine and combinations thereof. In addition, the pharmaceutical compositions can also comprise a secondary therapeutic agent. Examples of such secondary therapeutic agents include, without limitation, cisplatin, carboplatin, BCG, valrubicin, thiotepa, mitomycin-C, doxorubicin, epirubicin, MVAC, taxol, taxotere, gemcitabine, GC and combinations thereof. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be cisplatin. For example, the NMDA receptor antagonist can be memantine and the secondary therapeutic agent can be carboplatin.

[0079] In addition to the advantages associated with administering an NMDA receptor antagonist for the treatment of urologic cancer, significant advantages arise when an NMDA receptor is administered in a combination therapy comprising a secondary therapeutic agent. For example, administering a combination therapy may permit the administration of a lower dosage of the secondary therapeutic agent than would be used if the secondary therapeutic agent were being administered alone, while still maintaining at least substantially the same therapeutic

effectiveness. For example, a combination therapy comprising an NMDA receptor antagonist and a secondary therapeutic agent according to the present invention may permit the health care provider to reduce the dosage of the secondary therapeutic agent to 1/2 (i.e., one-half) of its individual-use dosage, while still achieving at least substantially the same therapeutic effectiveness. In this way, the severity of side effects can be reduced, which can allow the patient to remain on the treatment regimen for longer periods. In addition, for patients who are unable to tolerate the individual dosage level of the secondary therapeutic agent, the reduction in dosage level achieved by a combination therapy may permit them to undergo treatment involving the secondary therapeutic agent when it may not have been possible otherwise. As used herein, "combination therapy" refers to a therapeutic regimen involving administration of an NMDA receptor antagonist as well as administration of one or more secondary therapeutic agents, whether simultaneously or sequentially. As used herein, "indicated dosage" refers to the dosage level appropriate to treatment of urologic cancer using the specified agent by itself, without a secondary therapeutic agent.

[0080] The pharmaceutical composition according to the present invention can be used for the treatment of bladder cancer or a bladder cancer-related disease, disorder or condition. The NMDA receptor antagonist can be provided to the subject per se, or as part of a pharmaceutical composition where they are mixed with a pharmaceutically acceptable carrier, which can also include one or more other agents.

[0081] The invention also provides uses for an NMDA receptor antagonist in the manufacture of a medicament for treatment of bladder cancer. The NMDA receptor antagonist can be, for example, memantine, topiramate, and/or budipine. The medicament so manufactured can also include one or more secondary therapeutic agents. The method of manufacture may also comprise manufacturing separate dosage forms for the NMDA receptor antagonist and the one or more secondary therapeutic agents.

[0082] Herein the term "active ingredient" refers to the component of the composition that is accountable for the biological effect. As used herein, "active ingredient" and "active agent" are substantially equivalent terms.

[0083] As used herein, the phrases "physiologically acceptable carrier" and "pharmaceutically acceptable carrier," which may be used interchangeably, refer to a carrier or a diluent that does not cause significant irritation to an organism and does not abrogate the

biological activity and properties of the active ingredient or ingredients. A pharmaceutically acceptable carrier can comprise more than one ingredient. A pharmaceutically acceptable carrier can comprise, for example, an adjuvant or an excipient. It can also comprise, for example, polyethylene glycol (PEG), a biocompatible polymer with a wide range of solubility in both organic and aqueous media (Mutter et al. (1979).

[0084] Herein the term "excipient" refers to an inert substance added to a pharmaceutical composition to further facilitate administration of an active ingredient. Examples of excipients include, for example, calcium carbonate, calcium phosphate, various sugars and types of starch, cellulose derivatives, gelatin, vegetable oils and polyethylene glycols.

[0085] Techniques for formulation and administration of drugs may be found in "Remington's Pharmaceutical Sciences," Mack Publishing Co., Easton, PA, latest edition, which is incorporated herein by reference.

[0086] Suitable routes of administration may, for example, include intravesical, oral, rectal, transmucosal, especially transnasal, intestinal or parenteral delivery, including intramuscular, subcutaneous and intramedullary injections as well as intrathecal, direct intraventricular, intravenous, intraperitoneal, intranasal, or intraocular injections. The pharmaceutical composition may be administered locally or systemically. For example, the composition can be administered locally via injection of the preparation directly into a specific region of a patient's body, such as for example through the urethra (for transurethral administration).

[0087] Pharmaceutical compositions of the present invention may be manufactured by processes well known in the art, e.g., by means of conventional mixing, dissolving, granulating, dragee-making, levigating, emulsifying, encapsulating, entrapping or lyophilizing processes.

[0088] Pharmaceutical compositions for use in accordance with the present invention may be formulated in conventional manner using one or more physiologically acceptable carriers comprising excipients and auxiliaries, which facilitate processing of the active ingredients into preparations which, can be used pharmaceutically. Proper formulation is dependent upon the route of administration chosen.

[0089] For injection, the active ingredients of the invention may be formulated in aqueous solutions, preferably in physiologically compatible buffers such as Hank's solution, Ringer's solution, or physiological salt buffer. For transmucosal administration, penetrants

appropriate to the barrier to be permeated are used in the formulation. Such penetrants are generally known in the art.

[0090] For oral administration, the compounds can be formulated readily by combining the active compounds with pharmaceutically acceptable carriers well known in the art. Such carriers enable the compounds of the invention to be formulated as tablets, pills, dragees, capsules, liquids, gels, syrups, slurries, suspensions, and the like, for oral ingestion by a patient. Pharmacological preparations for oral use can be made using a solid excipient, optionally grinding the resulting mixture, and processing the mixture of granules, after adding suitable auxiliaries if desired, to obtain tablets or dragee cores. Suitable excipients are, in particular, fillers such as sugars, including lactose, sucrose, mannitol, or sorbitol; cellulose preparations such as, for example, maize starch, wheat starch, rice starch, potato starch, gelatin, gum tragacanth, methyl cellulose, hydroxypropylmethyl-cellulose, sodium carboxymethylcellulose; and/or physiologically acceptable polymers such as polyvinylpyrrolidone (PVP). If desired, disintegrating agents may be added, such as cross-linked polyvinyl pyrrolidone, agar, or alginic acid or a salt thereof such as sodium alginate.

[0091] Dragee cores are provided with suitable coatings. For this purpose, concentrated sugar solutions may be used which may optionally contain gum arabic, talc, polyvinyl pyrrolidone, carbopol gel, polyethylene glycol, titanium dioxide, lacquer solutions and suitable organic solvents or solvent mixtures. Dyestuffs or pigments may be added to the tablets or dragee coatings for identification or to characterize different combinations of active compound doses.

[0092] The compositions may be formulated for intravesical administration. For such formulations, suitable vehicles can comprise saline, phosphate-buffered saline (i.e., PBS) and/or gelatin nanoparticles. Other suitable vehicles and methods of preparation will be apparent to a person of ordinary skill in the art.

[0093] Pharmaceutical compositions, which can be used orally, include push-fit capsules made of gelatin as well as soft, sealed capsules made of gelatin and a plasticizer, such as glycerol or sorbitol. The push-fit capsules may contain the active ingredients in admixture with filler such as lactose, binders such as starches, lubricants such as talc or magnesium stearate and, optionally, stabilizers. In soft capsules, the active ingredients may be dissolved or suspended in suitable liquids, such as fatty oils, liquid paraffin, or liquid polyethylene glycols. In addition,

stabilizers may be added. All formulations for oral administration should be in dosages suitable for the chosen route of administration.

[0094] For buccal administration, the compositions may take the form of tablets or lozenges formulated in conventional manner.

[0095] For administration by nasal inhalation, the active ingredients for use according to the present invention are conveniently delivered in the form of an aerosol spray presentation from a pressurized pack or a nebulizer with the use of a suitable propellant, e.g., dichlorodifluoromethane, trichlorofluoromethane, dichloro-tetrafluoroethane or carbon dioxide. In the case of a pressurized aerosol, the dosage unit may be determined by providing a valve to deliver a metered amount. Capsules and cartridges of, e.g., gelatin for use in a dispenser may be formulated containing a powder mix of the compound and a suitable powder base such as lactose or starch.

[0096] The preparations described herein may be formulated for parenteral administration, e.g., by bolus injection or continuous infusion. Formulations for injection may be presented in unit dosage form, e.g., in ampoules or in multidose containers with optionally, an added preservative. The compositions may be suspensions, solutions or emulsions in oily or aqueous vehicles, and may contain formulatory agents such as suspending, stabilizing and/or dispersing agents.

[0097] Pharmaceutical compositions for parenteral administration include aqueous solutions of the active preparation in water-soluble form. Additionally, suspensions of the active ingredients may be prepared as appropriate oily or water based injection suspensions. Suitable lipophilic solvents or vehicles include fatty oils such as sesame oil, or synthetic fatty acids esters such as ethyl oleate, triglycerides or liposomes. Aqueous injection suspensions may contain substances, which increase the viscosity of the suspension, such as sodium carboxymethyl cellulose, sorbitol or dextran. Optionally, the suspension may also contain suitable stabilizers or agents which increase the solubility of the active ingredients to allow for the preparation of highly concentrated solutions.

[0098] Alternatively, the active ingredient may be in powder form for constitution with a suitable vehicle, e.g., sterile, pyrogen-free water based solution, before use.

[0099] The preparation of the present invention may also be formulated in rectal compositions such as suppositories or retention enemas, using, e.g., conventional suppository bases such as cocoa butter or other glycerides.

[00100] Pharmaceutical compositions suitable for use in context of the present invention include compositions wherein the active ingredients are contained in an amount effective to achieve the intended purpose. More specifically, a therapeutically effective amount is an amount of one or more active ingredients effective to prevent, alleviate or ameliorate symptoms of disease or prolong the survival of the subject being treated.

[00101] Toxicity and therapeutic efficacy of the active ingredients described herein can be determined by standard pharmaceutical procedures in vitro, in cell cultures or experimental animals. The data obtained from these in vitro and cell culture assays and animal studies can be used in formulating a range of dosage for use in human. The dosage may vary depending upon the dosage form employed and the route of administration utilized. The exact formulation, route of administration and dosage can be chosen by the individual physician in view of the patient's condition. (See e.g., Fingl, et al., 1975, in "The Pharmacological Basis of Therapeutics", Ch. 1 p.1).

[00102] Depending on the severity and responsiveness of the condition to be treated, dosing can be of a single or a plurality of administrations, with course of treatment lasting from several days to several weeks or until cure is effected or diminution of the disease state is achieved.

[00103] The amount of a composition to be administered will, of course, be dependent on the subject being treated, the severity of the affliction, the manner of administration, the judgment of the prescribing physician, etc.

[00104] Compositions including the preparation of the present invention formulated in a compatible pharmaceutical carrier may also be prepared, placed in an appropriate container, and labeled for treatment of an indicated condition.

[00105] Pharmaceutical compositions of the present invention may, if desired, be presented in a pack or dispenser device, such as an FDA approved kit, which may contain one or more unit dosage forms containing the active ingredient. The pack may, for example, comprise metal or plastic foil, such as a blister pack. The pack or dispenser device may be accompanied by instructions for administration. The pack or dispenser may also be accommodated by a notice

associated with the container in a form prescribed by a governmental agency regulating the manufacture, use or sale of pharmaceuticals, which notice is reflective of approval by the agency of the form of the compositions or human or veterinary administration. Such notice, for example, may be of labeling approved by the U.S. Food and Drug Administration for prescription drugs or of an approved product insert.

Kits

[00106] In another aspect, the invention provides kits comprising a pharmaceutical composition formulated for intravesical administration comprising a therapeutically effective amount of an NMDA receptor antagonist, and a container. A label can be associated with the container, wherein the label contains directions for intravesical administration of the pharmaceutical composition for treatment of urologic cancer. In some embodiments, the NMDA receptor antagonist is memantine. The pharmaceutical composition can also comprise a secondary therapeutic agent.

[00107] The container can be any suitable container capable of holding and dispensing the dosage form and which will not significantly interact with the composition. The pack may, for example, comprise an IV saline bag, a syringe, a metal or plastic foil, such as a blister pack, or it can be any other suitable dispenser device. In some embodiments, the NMDA receptor is packaged in dry form and reconstituted by conventional methods prior to administration.

[00108] In some embodiments, a label is associated with the container. For example, the printed labeling may provide instructions for administering any of the compositions, including locally administering an NMDA receptor antagonist and a secondary therapeutic agent; using any of the kits; or performing any other method herein described. For example, the label can provide directions for locally administering the NMDA receptor antagonist and secondary therapeutic agent for treatment of urologic cancer. The container, pack or dispenser may also be accompanied by a notice associated with the container in a form prescribed by a governmental agency regulating the manufacture, use or sale of pharmaceuticals, which notice is reflective of approval by the agency of the form of the compositions or human or veterinary administration. Such notice, for example, may be of labeling approved by the U.S. Food and Drug Administration for prescription drugs or of an approved product insert. The labeling instructions

will be consistent with the methods of treatment described herein. The labeling may be associated with the container by any means that maintain a physical proximity of the two. By way of non-limiting example, they may both be contained in a packaging material such as a box or plastic shrink wrap or may be associated with the instructions being bonded to the container such as with glue that does not obscure the labeling instructions or other bonding or holding means.

Methods for Predicting Effectiveness

[00109] In some embodiments, the invention provides methods for predicting the effectiveness of NMDA receptor antagonist therapy for a urologic cancer of a patient. In some embodiments, the methods comprise measuring NMDA receptor activity in urologic tissue of the patient, and, for tissue having elevated NMDA receptor activity, identifying the patient as a candidate for treatment with an NMDA receptor antagonist. The method can further comprise administering an NMDA receptor antagonist to the patient. In some embodiments, the methods further comprise measuring NMDA receptor activity after administration of the NMDA receptor antagonist, and if the tissue has a reduced post-administration NMDA receptor activity, identifying the cancer as responsive to treatment.

[00110] Additional objects, advantages, and novel features of the present invention will become apparent to one ordinarily skilled in the art upon examination of the following examples, which are not intended to be limiting. Additionally, each of the various embodiments and aspects of the present invention as delineated hereinabove and as claimed in the claims section below finds experimental support in the following examples.

EXAMPLES

[00111] **EXAMPLE 1: Expression of the Glutamate Receptor Iontropic N-methyl D-aspartate 1 (or NMDAR1; NMDA receptor subunit 1) in Human Bladder Cancer Cell Lines**

[00112] Three human bladder cancer cell lines T24 (urinary bladder transitional cell carcinoma; ATCC, Cat.No. HTB-4), SCaBER (urinary bladder squamous cell carcinoma; ATCC, Cat.No. HTB-3), and HT1197 (urinary bladder carcinoma; ATCC, Cat.No. CRL-1473) were used to examine the expression of NMDAR1. The cells were lysed and the levels of NMDAR1 expression were determined by immunoblotting with an antibody against NMDAR1. Blots were reprobbed with an anti-Tubulin antibody, and the expression levels of NMDAR1 were normalized to Tubulin protein levels.

[00113] **Cell treatments and lysis:**

[00114] T24, SCaBER, and HT1197 cells were grown in T-75 flasks according to the ATCC guidelines. Medium was removed, cells were washed with ice-cold Dulbecco's PBS (Biological Industries, Israel, Cat. No. 02-023-1A), and 500 µl of lysis buffer (50 mM Tris pH 7.4, 1% NP-40, 2 mM EDTA, 100 mM NaCl) containing complete protease inhibitor cocktail (Roche, 1-873-580-001), were added to each flask. Flasks were incubated on ice for 30 minutes and tapped until cells dissociation occurred. Lysates were transferred to 1.5ml tube and centrifuged at 4°C for 10 minutes at 14,000 rpm, and the supernatant was transferred to new tubes.

[00115] **Immunoblot Analysis:**

[00116] Lysate samples were separated on 4-12% Bis-Tris gels (Invitrogen) in NuPAGE MOPS running (Invitrogen, NP0001). Proteins were transferred to nitrocellulose membranes using NuPAGE transfer buffer (Invitrogen, NP0006). After transfer, blots were stained with Ponceau S solution (Sigma, Cat. No. P-7170), and washed twice with TBS-T 0.1% (TBS with 0.1% Tween-20). Blocking was carried out at RT for 1hr with 5% nonfat dry milk (BD, Difco skim milk, 232100, Lot.4184250) in TBS-T 0.1%. Anti-NMDAR1, rabbit polyclonal Ab (Cell Signaling Technology, 4204, Lot.1) was added at 1:1000 in TBS-T 0.1% with 5% BSA, and incubated for overnight at 4°C. Blots were washed x3 in TBS-T 0.1%, and secondary Ab, peroxidase-conjugated goat anti-rabbit IgG (Jackson ImmunoResearch, 111-035-144) was added

in blocking solution at 1:25,000, for 2 hrs at RT. Blots were washed x3 in TBS-T 0.1% and SuperSignal West Pico Chemiluminiscent (Pierce, Cat. No. 34080) was used for detection of HRP. Equal volumes of each solution were mixed, the blot was immersed in the solution for 5min and exposed to film.

[00117] For reprobing with anti-Tubulin Ab, the blot was re-blocked with 5% nonfat dry milk for 1 hr, followed by incubation with 1:30,000 anti-Tubulin Ab (mouse monoclonal Ab) for overnight at 4°C. Blots were washed as above, and secondary Ab, goat-anti-mouse (Jackson ImmunoResearch, 115-035-146) was added at 1:25,000 in blocking solution, for 1hr at RT. Blots were washed again, and HRP detection was carried out with SuperSignal West Pico Chemiluminiscent as described above. Autoradiograms were scanned and levels of NMDAR1 protein were quantified by densitometry using ImageJ 1.36b software, and normalized to levels of Tubulin expression.

[00118] **Results:**

[00119] The results of immunoblotting described above are shown in Figure 1. As can be seen in Figure 1 all three bladder cancer cell lines express the NMDAR1 protein, though at different levels. T24 expresses the highest levels of NMDAR1, while HT1197 expresses the lowest levels of the three.

[00120] **EXAMPLE 2: Effect of Memantine on Cell Proliferation of Human Bladder Cancer Cells**

[00121] The effect of Memantine on cell proliferation of human bladder cancer cells was tested using three different bladder cancer cell lines, T24 (urinary bladder transitional cell carcinoma; ATCC, Cat.No. HTB-4), SCaBER (urinary bladder squamous cell carcinoma; ATCC, Cat.No. HTB-3), and HT1197 (urinary bladder carcinoma; ATCC, Cat.No. CRL-1473). Each cell line was treated with multiple doses of Memantine alone or with carboplatin. The effect was found to be synergistic with carboplatin.

[00122] Memantine was able to inhibit the growth of all three types of human bladder cancer cells. Without wishing to be limited by a single hypothesis, since all three cell lines were unexpectedly found to express the NMDAR1 protein (as shown in Example 1), which is the target of action of Memantine in the brain, the mechanism of action of Memantine for inhibition of bladder cancer cell proliferation may be occurring also through the NMDAR1 protein.

[00123] **Description of BrdU proliferation assay:**

[00124] T24, SCaBER, and HT1197 cells were grown according to the ATCC guidelines. Cells were seeded in 96-well microtiter plates at a concentration of 5,000 cells/well in a final volume of 200 μ l medium containing 10% FBS (Fetal bovine serum, Heat Inactivated, Biological Industries, Cat.No.04-121-1A). On the next day, cells were rinsed and supplemented with 200 μ l of medium containing 0.1% FBS for additional 24 hrs. After serum starvation, cells were treated with different concentrations of the NMDA receptor antagonist Memantine Hydrochloride (Sigma-Alrich, Israel, M9292, Lot.126k1103, dissolved in double distilled water) or with Carboplatin (Sigma-Alrich, Israel, C2538, Lot.066k1724, CAS number 41575-94-4, dissolved in double distilled water) as control, for 72 hrs. For the BrdU incorporation assay, BrdU was added at a final concentration of 10 μ M for 2 hrs. BrdU ELISA assay was performed according to the manufacturer instructions (Cell proliferation ELISA, Roche, Cat. No.11 647 229 001).

[00125] **Results:**

[00126] The results of the proliferation assays described above are shown in Figure 2. As can be seen in Figures 2A, 2B, and 2C, Memantine inhibits the proliferation of T24, HT1197, and SCaBER cells, respectively, as measured by BrdU incorporation at 100 μ M. The results are depicted as relative growth compared to untreated cells defined as 1.0. The histograms indicate a strong inhibition of proliferation by Memantine in all three cell lines, with correlation to expression levels.

[00127] Figures 2D and 2E show a dose response of T24 and SCaBER cells, respectively, to the concentration of Memantine used for treatment. T24 cells show higher sensitivity to the drug, in agreement with the higher levels of expression shown in Figure 1 (although again without wishing to be limited by a single hypothesis).

[00128] Figure 2F shows the affect of Carboplatin on proliferation of T24 cells with or without Memantine at different concentrations. Results indicate a synergistic effect for the two drugs. For example, the combination of 5 μ M of Carboplatin together with 60 μ M of Memantine shows a similar effect to that of 10 μ M Carboplatin.

[00129] **EXAMPLE 3: Effect of Memantine and Cisplatin on Induced Tumors in Mice**

[00130] **Material and methods:**

[00131] Test substance: The test substance was memantine (10mg tablets, Unipharm, Batch unknown) stored at room temperature. The test formulation was prepared by grinding a 10mg tablet and dissolving it in 2ml WFI to give a concentration of 5mg/ml (based on a mean weight of 25g). The resulting solution was centrifuged briefly in order to pellet insoluble component, and then 1.5 mls of soluble fraction was removed with a pipette and filter sterilized. Part of the resulting 5 mg/ml solution was used to dose mice in Group 1. A second part of the resulting solution was diluted to 2.5 mg/ml. This was used to dose mice in Group 2.

[00132] Animals: Female nude mice were obtained from commercial suppliers (e.g., Harlan Olac) for use in the study. The mice were 7-10 weeks old at the start of the study. Nude (nu/nu) athymic mice have been used extensively to generate human tumour xenografts using the T24 bladder cancer cell line, and remain the experimental method of choice for testing anti-tumour efficacy of new compounds prior to administration in man. The test substance was administered P.O. (i.e., oral administration) and intravenously.

[00133] The mice were divided into four groups of 10 mice each. The following table presents the treatment protocol each group received:

Group	Treatment Protocol
Group 1	Vehicle i.p. daily
Group 2	20mg/kg memantine P.O. daily
Group 3	2mg/kg Cisplatin i.v. on days 1, 4, 7 & 10
Group 4	20mg/kg memantine P.O. daily and 2mg/kg Cisplatin i.v. on days 1, 4, 7 & 10

[00134] Experimental procedure:

[00135] T24 cells were maintained *in vitro* in DMEM culture medium (Sigma, UK) containing 10% (v/v) heat inactivated foetal bovine serum (Sigma, Poole, UK) at 37°C in 5% CO₂ and humidified conditions. Cells from semi-confluent monolayers were harvested with 0.25% Trypsin-EDTA (Sigma), washed twice in the culture medium described above. Cells were then re-suspended, for *in vivo* administration at the concentration of 5x10⁶ cells in 200µl of Matrigel (BD Matrigel™ Basement Membrane Matrix, BD Biosciences), and were injected

subcutaneously into the left flank of 48 mice. 200µl of just Matrigel was injected subcutaneously into the left flank of 5 mice.

[00136] Tumour dimensions were measured using caliper measurement of length, width and tumour cross-sectional area, and volume was then calculated. Tumor dimensions were recorded from day 7 onwards, three times weekly. Measurements of tumor dimensions in the matrigel only group was taken in parallel. When the mean tumour volume across all mice was estimated to be approximately 150mm³ the mice were allocated to their treatment groups. Ten mice were allocated into each treatment group in such a way as to achieve a uniform mean tumour volume and weight across groups. Mice with estimated tumour size less than about 50mm³ were excluded from the study. Mice were evaluated daily by an experienced technician until termination.

[00137] Treatment: The treatment protocols were initiated when the mean tumour size across the study reached approximately 150mm³. Compounds were prepared as described above. The final concentration of the dosing suspension was determined according to the mean weight of the mice in each group at the start of the study and administered in a volume of 100µl. Treatment was administered for 2 weeks.

[00138] Termination: Each mouse remained in the study until terminated or until circumstances necessitated removal of that mouse from the study. Under the experimental protocol, animals were terminated during the study if the tumour size became excessive or any adverse effects were noted according to Home Office Project License PPL 40/2962. At the end of the study, 1 week after the final dose, the mice were anesthetized (Hypnorm/Hynovel). About 1ml of blood removed by cardiac puncture and processed for plasma in blood EDTA collection tubes, frozen and returned to the sponsor. The mice were then be terminated by an approved method. Tumours were excised and weighed and tumour volumes were determined.

[00139] Results:

[00140] Memantine administered orally (p.o.) at a dose of 20mg/kg reduced the tumour volume at day 32 by ~23%, which was statistically significant as compared to the vehicle group (p<0.01 AVOVA test). This reduction was also better than that achieved by cisplatin administered at 2mg/kg i.v. on days 1, 4, 7 and 10 (~19% reduction in tumour volume, p<0.01 AVOVA test compared to the vehicle group). The combination of the two drugs exhibited a synergistic effect in tumour volume reduction, achieving a 27% reduction (p<0.001 AVOVA test

compared to the vehicle group, and $p < 0.05$ AVOVA test compared to the cisplatin group). These results are presented in a Figure 3. The combination treatment also achieved a 20% reduction in tumour weight ($p = 0.007$ one-way ANOVA compared to vehicle).

[00141] Overall these results support the efficacy of memantine, either alone or in combination with other treatment agents such as cisplatin, in the treatment of bladder cancer. Local administration of effective doses of the NMDA receptor antagonist is not feasible in the model used here, but the results indicate the likelihood of more significant effects when administered locally, e.g. intravesically.

[00142] **EXAMPLE 4: Local Administration of Memantine Composition**

[00143] A pharmaceutical composition comprising a therapeutically effective amount of memantine is prepared by adding memantine to an IV bag. The composition is administered to the patient with bladder cancer intravesically from the IV bag using a urethral catheter. The patient maintains the composition in the bladder for about one hour. During the hour, the patient lies supine for about 15 minutes; prone for about 15 minutes; on the patient's left side for about 15 minutes; and on the patient's right side for about 15 minutes. The patient then expels the composition. This treatment regimen is repeated weekly for about 6 weeks. The patient's bladder cancer is effectively treated.

It is appreciated that certain features of the invention, which are, for clarity, described in the context of separate embodiments, may also be provided in combination in a single embodiment. Conversely, various features of the invention, which are, for brevity, described in the context of a single embodiment, may also be provided separately or in any suitable subcombination. Although the invention has been described in conjunction with specific embodiments thereof, it is evident that many alternatives, modifications and variations will be apparent to those skilled in the art. Accordingly, it is intended to embrace all such alternatives, modifications and variations that fall within the spirit and broad scope of the appended claims. All publications, patents and patent applications mentioned in this specification are incorporated herein in their entirety by reference into the specification, to the same extent as if each individual publication, patent or patent application was specifically and individually indicated to be incorporated herein by reference. In addition, citation or identification of any reference in this application shall not be construed as an admission that such reference is prior art to the present invention.

CLAIMS

1. A method for treating urologic cancer in a patient in need thereof comprising administering locally to cells of the cancer a pharmaceutical composition comprising an NMDA receptor antagonist in a therapeutically effective amount to treat the urologic cancer.
2. The method of claim 1, wherein the urologic cancer cells overexpress NMDA receptor subunit NR1.
3. The method of claim 1, wherein the urologic cancer is bladder cancer.
4. The method of claim 1, wherein the urologic cancer is urothelial carcinoma.
5. The method of claim 1, wherein the NMDA receptor antagonist is selected from the group consisting of memantine, topiramate, budipine and combinations thereof.
6. The method of claim 1, wherein the NMDA receptor antagonist is memantine.
7. The method of claim 1, wherein the NMDA receptor antagonist is topiramate.
8. The method of claim 1, wherein the NMDA receptor antagonist is budipine.
9. The method of claim 1, wherein the pharmaceutical composition is administered intravesically.
10. The method of claim 1, wherein the therapeutically effective amount is from about 1 mg/kg to about 250 mg/kg.
11. The method of claim 1, wherein the therapeutically effective amount is from about 1 mg/kg to about 50 mg/kg.

12. The method of claim 1, wherein the therapeutically effective amount is about 20 mg/kg.
13. The method of claim 1, further comprising administering a secondary therapeutic agent.
14. The method of claim 13, wherein the secondary therapeutic agent is selected from the group consisting of cisplatin, carboplatin, BCG, valrubicin, thiotepa, mitomycin-C, doxorubicin, epirubicin, MVAC, taxol, taxotere, gemcitabine, GC, interleukin-2 and combinations thereof.
15. The method of claim 13, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is cisplatin.
16. The method of claim 13, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is carboplatin.
17. The method of claim 13, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is gemcitabine.
18. The method of claim 13, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is thiotepa.
19. The method of claim 13, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is valrubicin.
20. The method of claim 13, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is BCG.
21. The method of claim 13, wherein the NMDA antagonist and the secondary therapeutic agent are administered sequentially.

22. The method of claim 13, wherein the NMDA antagonist and the secondary therapeutic agent are administered simultaneously.
23. The method of claim 13, wherein the NMDA receptor antagonist is administered locally and the secondary therapeutic agent is administered systemically.
24. The method of claim 1, wherein the patient does not require the administration of neuroprotective agents.
25. The method of claim 1, comprising performing a therapeutic regimen comprising the steps of:
- administering the pharmaceutical composition into the patient's bladder;
 - the composition being retained in the bladder for a composition retention period lasting at least about one hour; and
 - the composition being expelled from the bladder after the composition retention period is complete.
26. The method of claim 25, wherein administering the pharmaceutical composition into the patient's bladder is done via a urethral catheter.
27. The method of claim 25, further comprising performing the therapeutic regimen at least about once weekly for at least about 6 weeks.
28. The method of claim 25, further comprising administering a secondary therapeutic agent.
29. A pharmaceutical composition for local administration comprising a combination therapy comprising an NMDA receptor antagonist and a secondary therapeutic agent, wherein the secondary therapeutic agent has a first effective dosage for treating urologic cancer when administered alone, and the dosage in the pharmaceutical composition is lower than the first effective dosage of the secondary therapeutic agent, the combination therapy achieving at least substantially the same efficacy in inhibiting urologic cancer cell growth

as the secondary therapeutic agent achieves when administered alone in the first effective dosage.

30. A pharmaceutical composition comprising an NMDA receptor antagonist, wherein the pharmaceutical composition is formulated for local administration and the NMDA receptor antagonist is present in an amount that is therapeutically effective for treating urologic cancer when administered locally.
31. The pharmaceutical composition of claim 30, wherein the NMDA receptor antagonist is selected from the group consisting of memantine, topiramate, budipine and combinations thereof.
32. The pharmaceutical composition of claim 30, further comprising a secondary therapeutic agent.
33. The pharmaceutical composition of claim 32, wherein the secondary therapeutic agent is selected from the group consisting of cisplatin, carboplatin, BCG, valrubicin, thiotepa, mitomycin-C, doxorubicin, epirubicin, MVAC, taxol, taxotere, gemcitabine, GC and combinations thereof.
34. The pharmaceutical composition of claim 32, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is cisplatin.
35. The pharmaceutical composition of claim 32, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is carboplatin.
36. The pharmaceutical composition of claim 32, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is gemcitabine.
37. The pharmaceutical composition of claim 32, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is thiotepa.

38. The pharmaceutical composition of claim 32, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is valrubicin.
39. The pharmaceutical composition of claim 32, wherein the NMDA receptor antagonist is memantine and the secondary therapeutic agent is BCG.
40. A kit comprising a pharmaceutical composition formulated for intravesical administration comprising a therapeutically effective amount of an NMDA receptor antagonist, a catheter for intravesical administration, a container, and a label associated with the container, wherein the label contains directions for intravesical administration of the pharmaceutical composition for treatment of urologic cancer.
41. The kit of claim 40, wherein the container is an IV saline bag.
42. The kit of claim 40, wherein the container is a syringe.
43. The kit of claim 40, wherein the NMDA receptor antagonist is memantine.
44. The kit of claim 40, wherein the catheter is a Foley catheter.
45. The kit of claim 40, wherein the pharmaceutical composition comprises a secondary therapeutic agent.
46. The kit of claim 40, wherein the label provides directions for administering the NMDA receptor antagonist and secondary therapeutic agent for treatment of urologic cancer.
47. A method for predicting the effectiveness of NMDA receptor antagonist therapy for a urologic cancer of a patient, comprising:
measuring NMDA receptor activity in urologic tissue of the patient, and

for tissue having elevated NMDA receptor activity, identifying the patient as a candidate for treatment with an NMDA receptor antagonist.

48. The method of claim 47, further comprising administering an NMDA receptor antagonist to the patient.

49. The method of claim 48, further comprising measuring NMDA receptor activity after administration of the NMDA receptor antagonist, and if the tissue has a reduced post-administration NMDA receptor activity, identifying the cancer as responsive to treatment.

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FIGURE 1

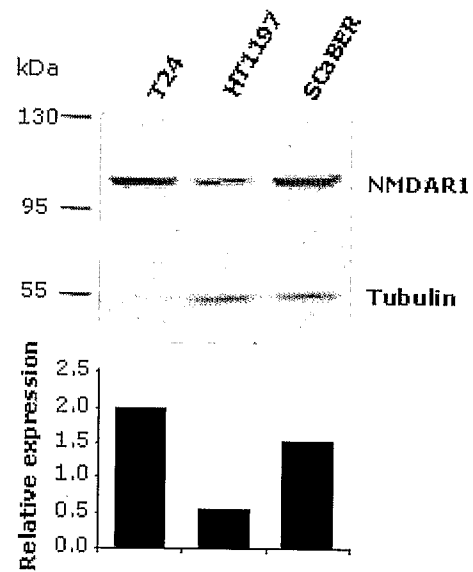
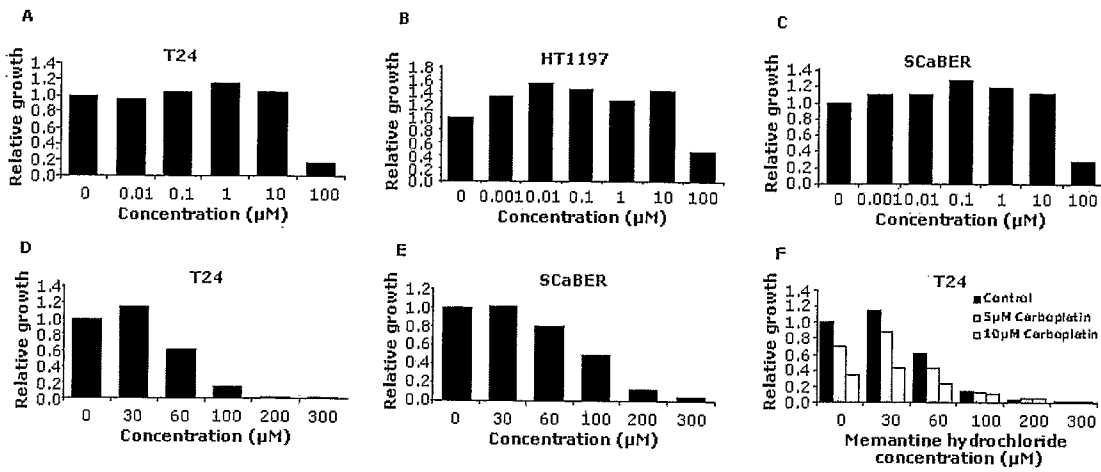


FIGURE 2



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FIGURE 3

