Potentially New And Innovative Treatments For Superficial, Muscle-Invasive, And Metastatic Transitional Cell Carcinoma (TCC) Of The Bladder

I Secasan, D Pop, C Secasan

Citation

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Abstract

Purpose: To present a novel,innovative treatment for superficial, muscle-invasive and metastatic transitional cell carcinoma (TCC) of the bladder, which is based on a combination of:

a currently used anticancer drug (or a combination of anticancer drugs) - (DRUG-x)
a specific,autologous anti-cancer vaccine , VAX-x, made of in-vitro-heat/radiation - killed (IVHRK) cancer cells (or subunits of such cancer cells), prelevated from the patient, and bearing on their genomic/biochemical structure the mutations that would be induced into these cancer cells by the following, to come anticancer drug, or anticancer drugs combination(DRUG-x).

The subunits of the in-vitro-heat/radiation - killed (IVHRK) cancer cells may be certain proteins, enzymes (e.g. telomerase), DNA, or other cancer-cell antigens, bearing on their genomic/biochemical structure the mutations that would be induced into these cancer-cell- parts/subunits by the following, subsequent, to come anticancer-drug, or anticancer-drugs combination (DRUG-x), and may form the VAX-x component of the (VAX - x, DRUG - x)- couple). We define the combination of a specific anticancer drug (DRUG - x) and its
pre-(DRUG - x)- administrated, corresponding , autologous cancer vaccine (VAX-x), made of in-vitro-heat/radiation - killed (IVHRK) cancer cells from the patient, and bearing on their genome the mutations that would be induced into these cancer cells by the specific drug (DRUG - x), as a (VAX - x, DRUG - x)- couple. The order of the 2 components within the (VAX - x, DRUG - x)- couple clearly indicates that the autologous , (therapeutic) , anti- cancer vaccine, (VAX - x) , is pre-administrated to its corresponding (DRUG - x) couple-partner. The role of VAX-x , within a (VAX - x, DRUG - x)-couple, is to prevent the emergence of DRUG-x -resistant cancer cells (since it contains/encodes/encompassesthe drug-resistance-mutations that would be induced into cancer-cells by couple-partner DRUG-x) , wheras the role of DRUG-x is to kill DRUG-x-sensitive cancer cells, in order to cure bladder cancer in particular, and any other type of cancer in general. The existance of (VAX - x, DRUG - x)-couples is made possible by the process of mutagenesis, and is based on the need and capacity of cancer cells (and living pathogens) to mutate under drug-pressure.

Materials and Methods: The Internet was used in order to locate and collect some of the most relevant scientific articles necessary to review the current status of bladder-cancer treatment, and we have used our own creativity and innovative imagination in order to develop a successful and powerful strategy and practical solution for curing bladder cancer in particular, and all other cancer types in general.

Results: The term (vaccine-drugs)-couples, (VAX - x, DRUG - x)- couples, has been invented , defined, and coined, and thus a new strategy and concept to cure bladder cancer has been developed. (VAX - x, DRUG - x)- couples may prove to be far more efficient and powerful in curing superficial, muscle-invasive, and especially advanced-metastatic bladder cancer. (VAX - x, DRUG - x)- couples may proove to be a far better option for treating superficial bladder cancer than BCG-immunotherapy plus chemotherapy, since VAX- x is defined as a specific, autologous vaccine, compared to BCG which is a non-specific vaccine, and since the sinergy between VAX - x and DRUG - x is practically perfect, while sinergy between BCG and chemotherapy is not always sinchronized or sinergetic. Also (VAX - x, DRUG - x)- couples may proove to be a much better choice for treating

muscle-invasive and metastatic bladder cancer than currently used standard chemotherapy (MVAC), since (VAX - x, DRUG - x)- couples, at limit, may be written as (VAX - x, MVAC- x)- couples, and the sinergy between VAX - x, MVAC- x and the immune system would represent an impressive plus in comparison with MVAC alone. The adverse local and systemic effects, side effects, and toxicity of (VAX - x, DRUG - x)- couples may be significantly lower than those associated with BCG immunotherapy + chemotherapy and also considerably lower than those seen with current standard chemotherapy (MVAC) for muscle-invasive and metastatic bladder cancer. (VAX - x, DRUG - x)- couples and especially (VAX - x, MVAC - x)- couples may be able to cure superficial, muscle-invasive and even metastatic bladder cancers.

Conclusions: (VAX - x, DRUG - x)- couples may prove to be able to cure superficial, muscle-invasive,and even metastatic transitional cell carcinomas(TCC-s)of the bladder,and especially those (TCC-s) which are refractory to current standard treatments and to all other existing alternative treatments. In addition, (VAX - x, DRUG - x)- couples may prove to be far more efficient and effective in curing superficial, muscle-invasive, and metastatic bladder cancers, as well as all other types of cancers, and to be less toxic than comparable current conventional therapies, having the potential to become the standard treatment of care and cure for all bladder cancers, for all other types of cancer, and for absolutely all infectious diseases for which at least one drug exists. thus representing a revolution in modern medicine. The existance of (VAX - x, DRUG - x)- couples is made possible by the process of mutagenesis, and is based on the need and capacity of cancer cells (and living pathogens) to mutate under drug-pressure. The (Telomerase, MVAC - x)- couple might become one of the most powerful anticancer therapies, by possibly being able to cure high-stage and metastatic bladder cancer, as well as all other types of cancer.

INTRODUCTION

The successful treatment of bladder cancer remains a challenge for urologists and oncologists. There have been substantial changes in the therapeutic options for the management of superficial, muscle-invasive, and metastatic bladder cancer in the first 4 years of the new millennium.

For superficial bladder cancer, intravesical instillation of chemotherapeutic agents after transurethral resection is the standard of care. Novel therapeutic approaches under investigation include anti-cancer vaccines([[[1a]]]), antisense oligodeoxynucleotides ([[[1b]]]), magnetically targeted carriers ([[[1c]]]) and bio-adhesive microspheres ([[[1d]]]).

For muscle-invasive bladder cancer, systemic perioperative chemotherapy is being used with increasing frequency and the latest preclinical research efforts are focused on the inhibition of angiogenesis and other processes predisposing to metastatic disease.

So far, treatment goals for bladder cancer of any stage have been complete removal of the initial tumor, prevention of recurrence and progression to advanced disease with the ultimate aim of reducing mortality. The myriad of novel therapeutic modalities currently being explored suggest that bladder cancer will soon become a long-term manageable disease.

CURRENT TREATMENTS OF SUPERFICIAL BLADDER CANCER (IE, CARCINOMA IN SITU, TA, AND T1)

Superficial bladder cancer can be resected with minimal morbidity, but the patients remain at high risk for tumor recurrence and progression . Tumors can be divided into low-, intermediate-, and high-risk categories based on tumor grade, stage, and pattern of recurrence. Low-risk tumors are best treated with a single instillation of chemotherapy such as thiotepa, doxorubicin, or mitomycin.

Intermediate-risk tumors can be treated with chemotherapy, but, similar to high-risk tumors, will often require immunotherapy. High-risk tumors are best treated with intravesical bacille Calmette-Guerin (BCG) using a 3-week maintenance schedule. Side effects of BCG immunotherapy can be reduced by logarithmic reductions in the dosage of BCG. Patients who fail BCG may be rescued with BCG plus interferon-alfa or radical cystectomy.

Many patients with superficial bladder tumors treated with endoscopic surgery alone have recurrence or tumor progression at some point in their follow-up, and, in these patients, the need for adjuvant treatment becomes a major concern.

One of the most potent immunotherapies presently used is the application of Bacillus Calmette Guerin (BCG) to prevent recurrences of superficial bladder cancer. Despite its successful use, nonresponders and certain side effects remain

a major obstacle.

In 2002 Sylvester et al ([[[1e]]]) carried out a meta-analysis of randomised trials that showed that intravesical BCG (following transurethral resection) reduced the risk of progression in papillary tumours and carcinoma in situ (CIS) when maintenance BCG was used. However, the adverse local and systemic effects, side effects, and toxicity of BCG should not be neglected, and in low risk patients intravesical chemotherapy is considered to be the optimal treatment.

Current studies aim at developing recombinant BCG (rBCG) strains to further improve the effectiveness of the therapy. In BCG-treated patients a strong local induction of Th1-like cytokines was observed. For this reason rBCG-strains secreting Th1-like cytokines might be potentially useful agents to improve this type of immunotherapy. Recently, Arnold et al ($_{2}$) concluded that recombinant BCG has an enhanced immunostimulatory potential when compared to wild-type control BCG, and might offer new opportunities in the treatment of bladder cancer. With a low-dose treatment regimen for murine orthotopic bladder cancer, rBCG-IFNgamma significantly prolonged survival, whereas the therapeutic effect of wild-type control BCG did not reach statistical significance. Cervenakov et al ($_{3}$) have have succesfully used Alpha 2-b interferon and farmarubicin in the prophylaxis of recurrence of superficial transitional cell carcinoma of the urinary bladder. Mitsumori et al ($_{4}$) found that patients who received a high-dose epirubicin instillation had a significantly lower recurrence rate but the benefit of early instillation was not confirmed, as the study group was too small. In a study on 622 patients Kuroda et al. ($_{5}$) concluded that the greatest effect of intravesical instillation of epirubicin after TUR-BT was shown by the regimen using the highest concentration (40mg/40ml) of the drug solution which was administered during a short period of time (4 months, 9doses, 360mg total dose).

Milonas et al ($_{6}$) compared the efficacy of transurethral resection alone or transurethral resection followed by bladder instillations of Doxorubicin for 1 year in patients with superficial bladder carcinoma, and followed them long term for the incidence of recurrence and progression to muscle invasion. In regard to time of first recurrence and disease free survival this study indicated that adjuvant chemotherapy with Doxorubicin is superior to transurethral resection alone. However, progression in stage or recurrence rate was not influenced by the treatment regimen.

The combination of epirubicin and meglumine gamma-linolenic acid have been found to be a logical choice of combination therapy for patients with superficial bladder carcinoma by Harris et al. ($_{7}$) . The efficacy of epirubicin was enhanced significantly when it was used in combination with most concentrations of MeGLA (< 300 microg/mL), and the two agents acted synergistically. There was a corresponding increase in epirubicin uptake by cells under these conditions. At high MeGLA concentrations, however, anthracycline solubility was compromised, and drug synergy was lost ($_{7}$).

In order to prevent the relapse of bladder neoplasms, a 34 patients study ($_{8}$) was designed to explore the effect of intravesical instillation of pirarubicin (THP) together with polyvinylpyrrolidone (PVP) on patients with superficial bladder cancer who had undergone surgical operation. Intravesical instillation of THP/PVP is effective for prevention of postoperative recurrence of superficial bladder cancer with fewer side effects. However, further study is needed for wide use in such way($_{8}$).In the treatment of superficial bladder cancer, valrubicin, pirarubicin and gemcitabine are novelties. In a randomized trial conducted by Huang et al ($_{9}$) the efficacy and side effects of intravesical mitoxantrone instillation with those of doxorubicin in superficial bladder cancer following transurethral resection, were compared. The instilled doses of doxorubicin and mitoxantrone were 30 and 14 mg, respectively. Thirty-three patients received mitoxantrone, whereas 30 patients used doxorubicin. The recurrence rate in the doxorubicin group was 30% (95% CI: 19.8%-38.8%), while it was 27.3% (95% CI: 17.5%-36.8%) in the mitoxantrone group. The median recurrence-free survival in the mitoxantrone group and in the doxorubicin group was 22 and 20 months, respectively (p=0.580). The results revealed that the efficacy and side effects of mitoxantrone were similar to those of doxorubicin. Especially for patients with pulmonary tuberculosis or aged patients with primary bladder tumors, mitoxantrone and doxorubicin may be the tolerable and effective intravesical agents($_{9}$).endovesical thermochemotherapy appears to be more effective than standard endovesical chemotherapy as an adjuvant treatment for superficial bladder tumors at 24-month follow-up, despite an increased but acceptable local toxicity.The intravesical administration of mitomycin C could be safely performed in the form of both thermo-chemotherapy ($_{10}$) and electromotive drug approach with an increased ablative success rate on small superficial tumor involving only minimal local side effects. In a multicentric

study comparing intravesical chemotherapy alone and with local microwave hyperthermia for prophylaxis of recurrence of superficial transitional cell carcinoma ([11]) endovesical thermochemotherapy appeared to be more effective than standard endovesical chemotherapy as an adjuvant treatment for superficial bladder tumors at 24-month follow-up, despite an increased but acceptable local toxicity.

CURRENT TREATMENTS OF MUSCLE-INVASIVE AND METASTATIC TRANSITIONAL CELL CARCINOMA (TCC)

In the 1990s randomized controlled trials (RCT) of muscle-invasive and metastatic TCC focused on determining the efficacy of chemotherapy regimens combining cisplatine, vinblastine and methotrexate (CMV) or these drugs plus doxorubicin (MVAC). ([12],[13],[14]) Improvements in overall response rate, progression-free survival and survival were seen, but at the expense of increased toxicity, side-effects and toxic deaths.

Randomized controlled trials (RTI-s) in muscle-invasive and metastatic TCC reported this decade have focused on improving the effectiveness of MVAC by comparing it with combinations of gemcitabine-cisplatin, high dose intensity MVAC plus granulocyte-colony stimulating factor (G-CSF) and cisplatin plus

5-fluorouracil and alfa-interferon([15],[16],[17]). Malmstrom et al. ([18]) found that neoadjuvant chemotherapy improves long-term survival after cystectomy in patients with stages T3 to T4a bladder carcinoma, while no survival benefit was found for stages T1 to T2 disease.Grossman et al. ([19]) concluded that as compared with radical cystectomy alone, the use of neoadjuvant methotrexate, vinblastine, doxorubicin, and cisplatin followed by radical cystectomy increases the likelihood of eliminating residual cancer in the cystectomy specimen and is associated with improved survival among patients with locally advanced bladder cancer. Millikan et al ([20]) found that there is an improved cure fraction by the combination of multiagent chemotherapy and surgery, although they found no preferred sequence. Importantly, it is possible to select appropriate patients for such therapy on the basis of clinical staging information. Freiha et al. ([21]) concluded that treatment with CMV (cisplatin, vinblastine and methotrexate) chemotherapy after radical cystectomy is an acceptable approach in patients with stages p3b and p4N0 or N1 transitional cell carcinoma of the bladder. Further studies must be performed to determine whether these results can be extrapolated to patients with more limited disease (stages p2 and p3a) who are currently treated with radical cystectomy or definitive irradiation ([21]).

Three cycles of neoadjuvant chemotherapy before cystectomy or radiotherapy did not give the 10% improvement in 3-year survival that was judged to be necessary for introduction into routine use. The chemotherapy regimen was associated with a higher pathological complete-response rate in primary tumours, but there was no clear evidence that it would increase survival ([22]). Neoadjuvant chemotherapy with cisplatin and methotrexate did not significantly improve disease-free or overall survival in 153 randomized patients with invasive bladder cancer([23]). Two cycles of neoadjuvant methotrexate, cisplatin, and vinblastine (MCV) chemotherapy were not shown to increase the rate of clinical complete response over that achieved with a standard induction therapy or to increase freedom from metastatic disease([24]). M-VECA was found to be a safe and effective regimen for the treatment of patients with metastatic urothelial tumors ([25]). The results achieved in the 60 patients included in a study comparing the combination of carboplatin, methotrexate and vinblastine (M-CAVI) and M-VAC indicated that M-CAVI is better tolerated than M-VAC, although both treatment regimens had similar overall response rates, pathological response rates and survival in patients with locally advanced and locoregional bladder cancer([26]). The combination of methotrexate, vinblastine, doxorubicin, and cisplatin (MVAC) has dominated chemotherapy for advanced bladder cancer for over 15 years. Randomized studies have shown (MVAC) superiority over cisplatin alone or in combination with cyclophosphamide and doxorubicin. However, (MVAC) exhibits a significant toxicity profile and achieves only a slight impact on overall survival. The combination of gemcitabine and cisplatin represents a new standard alternative of treatment for bladder cancer, based on a similar efficacy to and lower toxicity than the classic MVAC regimen. Future drug development will focus on the clinical usefulness of three-drug regimens (including gemcitabine, paclitaxel or docetaxel, and a platinum salt), and nonplatinum-based combinations. The regimen of gemcitabine and cisplatin has been found to be equally efficacious with less toxicity than methotrexate, vinblastine, doxorubicin, and cisplatin(MVAC). It has been adopted as the standard arm in a phase III trial for advanced bladder cancer, comparing it with the triplet of gemcitabine, paclitaxel, and cisplatin. Other active agents in bladder cancer include ifosfamide, carboplatin, docetaxel, and

vinorelbine, and various doublets of these agents are being tested in phase II trials, with promising results([27]). Triple combinations like gemcitabine/paclitaxel/cisplatin and gemcitabine/paclitaxel/carboplatin have high levels of activity with overall and complete response rates of 76% and 26%, respectively, for the former and 68% and 32%, respectively, for the latter combination. The role of gemcitabine-based multiagent combinations compared with standard therapy awaits evaluation in prospectively randomized trials([28]). Combination paclitaxel, carboplatin, and gemcitabine is active; an encouraging number of patients with advanced urothelial carcinoma treated with this regimen experienced complete remission ([29]).The overall response rates for two-drug regimens of cisplatin-paclitaxel, carboplatin-paclitaxel and cisplatin-gemcitabine range from 63 to 72%, 14 to 65% and 42 to 66%, respectively. The overall response rates for platinum-paclitaxel-gemcitabine three-drug regimens range from 58 to 80%. The potential clinical benefit of these new three-drug combinations in the treatment of TCC needs to be tested in future phase III studies([30]). Paclitaxel, carboplatin, and methotrexate were well tolerated and active in advanced TCC. The high response rate to this regimen despite frequent p53 mutation is consistent with the p53-independent mechanism of paclitaxel.

Whether this regimen is superior to ethotrexate/vinblastine/doxorubicin/cisplatin, other paclitaxel-based regimens, or to paclitaxel alone will require comparative trials([31]). The combination of paclitaxel, carboplatin, and methotrexate holds promise to be well tolerated and active in advanced TCC([32]).The MCNO (methotrexate, carboplatin, mitoxantrone (Novantrone) and vincristine (Oncovin))([33]) regimen appears to have a lower efficacy than that obtained with cisplatin-based regimens for the treatment of metastatic disease and rather similar efficacy for the treatment of locally advanced urothelial-cell cancer. Therapy with this regimen, though less toxic, may not be a reliable alternative in elderly patients with visceral metastases ([33]). Patients treated with CP(carboplatin and paclitaxel) ([34]) had a median survival of 13.8 months compared with 15.4 months for patients treated with M-VAC. Patients treated with CP appeared in general to better tolerate their treatment; however, there were no significant differences noted with regard to measured quality of life parameters ([34]).

A gemcitabine/cisplatin regimen has been shown to lead to comparable survival in a phase III comparison to methotrexate/vinblastine/doxorubicin/cisplatin in the metastatic setting with less toxicity ([35]). A number of additional doublet combinations have thus been investigated. Substitution of carboplatin for cisplatin is feasible but leads to an apparent lower complete response rate. A combination of doxorubicin and gemcitabine has been reported to lead to a 36% complete response rate([35]). In addition to its antiproliferative and antiangiogenic effects, IFN-alpha has been shown to limit tumor invasion by restoring the normal balance between MMP-9 and E-cadherin and to enhance the activity of systemic chemotherapy ([36]).

Radiotherapy and concomitant 5-fluorouracil has been compared with radiotherapy alone in a prospective randomized study by Edland et al. ([37]). A prospective randomized phase III study based on neoadjuvant cisplatin chemotherapy before radical cystectomy in invasive transitional cell carcinoma of the bladder has been carried out by Martinez-Pineiro et al ([38]). The survival rates were 48.6% for patients with pN0 disease, 37.5% for pN1 and 5% for pN2-4. Toxicity of cisplatin was minimal and there were no differences in perioperative morbidity between the arms ([38]). Some urinary bladder cancer tumors may respond favorable to Herceptin therapy ([39]). Based on the success seen with anti-HER2 monoclonal antibodies (Herceptin) and the promising results with epidermal growth factor receptor (EGFR) targeted agents (IMC-C225 Cetuximab, ZD1389 Iressa, OSI-774 Tarceva, GW 57016) in other tumor types, and based on the results obtained in preclinical models, there is a great interest in assessing these agents in patients with bladder cancer([40]).Evidence from breast cancer suggests that only tumours with HER2/neu gene amplification respond to the anti-HER2/neu therapy trastuzumab (Herceptin; Genentech, Inc., South San Francisco, CA). If this were true for bladder cancer, only 4/75 (5%) of G3 pT2 TCCs would be suitable for treatment([41]). The role of trastuzumab, a humanized antibody to HER-2, in these tumours remains untested at present. However, specific treatments targeted toward oncogenes expressed in cancer cells are currently under development. Patients with urothelial carcinomas showing HER-2/ neu (human epidermal growth factor receptor 2) overexpression are candidates for such a specific treatment (trastuzumab)([42]). Telomere ends are known to be shortened at every division of the cells. Telomerase is a ribonucleoprotein which compensate for the telomere ends and indispensable for the immortalization of the cells. Responses of transitional cell carcinoma of the bladder

(TCC) to commonly used chemotherapy agents such as mitomycin C (MMC), cisplatin and gemcitabine are often disappointing. The expression of human telomerase reverse transcriptase (hTERT) is associated with cellular aging and tumorigenesis. It was found in nearly all cancer types but not in most normal, somatic cells. Since human telomerase reverse transcriptase (hTERT) is tumor specifically expressed and contributes to the immortality and malignancy of the majority of tumors, it is regarded as a suitable antitumor target. It is reported that the enzyme is activated in a variety of cancer cells. Enhancement of cytotoxic drug effects on the growth of (TCC) cells by hTERT antisense AS- oligodeoxynucleotides ODNs allows a dose decrease in chemotherapy and confirms the suitability of hTERT as a target in a specific therapy approach([43]).Specific hTERT inhibition causes remarkable short- and long-term effects on the growth of bladder cancer cells and represents a promising new treatment option of solid tumors. Kraemer et al. ([44]) consider that this alternative treatment could be applied in terms of an instillation therapy. In solid tumor oncology, decisions regarding treatment are governed by histologic diagnosis. Despite this reliance on histology and the assumption that histology defines the disease, underlying molecular heterogeneity likely differentiates among patients' outcomes ([45]). Betenski et al ([45]) concludes that molecular heterogeneity, if it confers different risks to patients and is unaccounted for in the design of a randomized study, can result in a clinical trial that is underpowered and fails to detect a truly effective new therapy for cancer. These authors feel that there is a need for individual approaches in the treatment of patients with bladder cancer, and we strongly agree with them. The study of Yang et al ([46]) supports intratumoral vaccination as a strategy for immunotherapy of established tumors. Keyhole limpet hemocyanin (KLH) is a high-molecular-weight copper-containing respiratory protein antigen collected from the haemolymph of the sea mollusk Megathura crenulata, a non-specific immunomodulator, that induces both a cell-mediated and a humoral response KLH has demonstrated efficacy and has induced long- term remissions against carcinoma in situ (CIS) in a limited number of cases ([47]) . However, most patients with CIS progressed over time whatever the substance instilled, whether KLH or BCG ([47]).

Bropirimine is an orally-active immunostimulant that has an antitumor effect on superficial transitional cell carcinoma of the bladder([48]). Bacteria such as Salmonella and Listeria can be attenuated by genetically-defined mutations and provide effective vehicles for DNA vaccines encoding tumor-associated antigens. Salmonella and nonpathogenic strains of Clostridium can selectively accumulate in tumors in vivo, providing attractive delivery systems to target immunomodulatory molecules and therapeutic agents to the tumor site([49]).

RESULTS

(VAX - x, DRUG - x)- Couples

We define the combination of a specific cancer drug (DRUG - x) and its pre-administrated , corresponding , autologous cancer vaccine (VAX-x), made of dead/killed cancer cells from the patient, and bearing on their genome the mutations that would be induced into these cancer cells by the specific drug (DRUG - x), as a (VAX - x, DRUG - x)- couple, clearly indicating that the autologous , (therapeutic) , cancer vaccine, VAX - x , is pre-administrated to its corresponding DRUG - x couple-partner.

(VAX - x, DRUG - x)- Couples are formed by :

1. a currently used anticancer drug (or a combination of anticancer drugs) - termed (DRUG-x),
2. a specific, anti-cancer, autologous vaccine , VAX-x, made of dead/killed cancer cells (or cancer cells parts/subunits), prelevated from the patient and bearing on their genome/biochemical structure the mutations that would be induced into these cancer cells by the following, to come anticancer drug, or anticancer drugs combination.

The role of a VAX-x , within a (VAX - x, DRUG - x)-couple, would be to prevent the emergence of DRUG-x - resistant cancer cells, wheras the role of DRUG-x would be to kill DRUG-x-sensitive cancer cells in order to cure bladder cancer in particular, and any other type of cancer in general.

Our innovative idea to introduce (VAX - x, DRUG - x) - Couples as a completely new treatment approach for superficial, muscle-invasive, and metastatic transitional cell carcinoma (TCC) of the bladder,was inspired and originates from our earlier theoretical research studies in the field of HIV-1([50,51]).

The Immune Response Corporation (Nasdaq: IMNR), a biopharmaceutical company ([52]) dedicated to becoming a leading immune-based therapy company in HIV, has based

its activity on its patented whole-killed virus technology, co-invented by Company founder Dr. Jonas Salk, to stimulate HIV immune responses. REMUNE® ([[[52a]]]), currently in Phase II clinical trials, is being developed as a first-line treatment for people with early-stage HIV and might be used as a non-specific anti-cancer vaccine in the treatment of superficial bladder cancer. Other interesting anti HIV-1 vaccines / therapeutic vaccines have been produced by Therion Biologics Corporation ([[[52b]]]) - based on the HIV-1 env/gag/pol genes (TBC-3B) and MN r-gp120/HIV-1 in alum., Apollon, Inc. - an HIV-1 gag-pol DNA Vaccine (APL-400-047) and ADVENTRX Pharmaceuticals ([[[52c]]]) who developed EradicAide , an HIV therapeutic vaccine, composed of six synthetic peptides, which stimulate a killer T-cell response to clear HIV-infected cells. A unique feature of this treatment is that it is designed to not elicit an antibody response. It is antibody-negative.

All these anti-HIV-1 vaccines / therapeutic-vaccines may be used as non-specific anti-cancer vaccines in the treatment of superficial bladder cancer, in a way similar to BCG.

Affymetrix ($_{53}$) , a leading US company in DNA-chip technology has developed GeneChip oligonucleotide probe arrays that are manufactured using a high resolution photolitographic fabrication process adapted from the semiconductor industry, for HIV-1 mutations determinations. This high-tech company has probably also the technical and scientific capacity to determine the specific mutations which cancer drugs ($_{55}$) induce into cancer cells, or on certain cancer - cell parts/subunits which can be found only in cancer cells and cannot be found in normal human cells. In this way, the VAX-x component of (VAX - x, DRUG - x)- couples may be standardized and produced in large scale by the bio-pharmaceutical industry.

The review of Coulie ($_{54}$) has been focused on the identification of several tumour antigens, their molecular nature, and how they can be used to develop anti-cancer vaccines.

Anti-cancer vaccines are normally used only as a treatment, after the cancer has been found in a patient. Anti - cancer vaccines (VAX-x) can be made from the person's own cancer cells (i.e. autologous vaccine - meaning that the antigens are derived from the same individual they are used to treat.). The cancer cells are treated with heat or radiation, so that they cannot multiply and grow, and to make sure that they cannot cause harm. While the cells are dead, the antigens are still recognized by the the immune system, which responds by attacking the dead cells. The immune system will also attack the live cancer cells carrying the antigen that was displayed on the dead cells. Autologous anti-cancer vaccines in development include those utilizing whole cells, tumor lysates , RNA or heat shock proteins . Early attempts at autologous cancer vaccines involved whole cell preparations or lysates of a patient's tumor, which were shown to be unsuccessful, as unpurified cellular contents send both stimulatory and suppressive immune signals.

Let us now suppose that the person's cancer cells are exposed, in vitro, to all currently existing and currently approved cancer drugs ($_{55}$) in order to assess the most appropriate treatment for the respective person. Some anti-cancer drugs will surely prove to be more efficient and effective than others in killing cancer cells, but in vitro results do not necessarily correlate with in vivo results. For most, if not for all cancer drugs ($_{55}$) , drug -resistant cancer cells may select naturally or may be grown by adaptive techniques in which the cancer cells are exposed to increasing doses of a certain cytostatic (Drug-x). These one-drug (naturally) resistant cancer cells can now be killed with heat or radiation, so that they preserve the exact mutations induced into their genomic/biochemic structure by the respective one-drug (DRUG-x), and can be used as the VAX-x component of a (VAX - x, DRUG - x)- Couple. VAX - x should be administrated before DRUG-x in order to allow the immune system to develop an immune response against DRUG-x resistant cancer cells before they even get the chance to arise. As soon as a good immune response to these in - vitro heat - or -radiation - killed (IVHRK) DRUG-x resistant cancer cells is recorded, the treatment with DRUG-x can commence. The sinergetic effect of VAX-x followed by DRUG-x acts like a scissor on that person's cancer cells and may eradicate all of them. Successive series of different (VAX - x, DRUG - x)- couples may ultimately lead to eradication of high stage and even metastatic cancer. Another advantage of (VAX - x, DRUG - x)- couples over conventional chemotherapy (e.g. MVAC) is the fact that successive one drug couples can be designed , which are less toxic : e.g. (VAX - 1, methotrexate), -----> (VAX - 2, vinblastine), ------> (VAX - 3, doxorubicin)------> (VAX - 4, cisplatin)

This series may also be followed by other monodrug-couples, by 2- and 3- drug combination based couples, and

even by a 4 drugs based couple (VAX - x, MVAC)

Alternatively, certain proteins may be taken from the cancer cells and used to make a cancer vaccine. These include antigens (the proteins on the cell surface which can stimulate an immune response). Unique antigens are antigens that occur only in cancer cells and do not exist in normal tissues. They arise from mutations that occur as the tumor cells grow. The vast majority of unique antigens are not only specific to cancer cells but also specific to each individual patient. Antigen vaccines use tumor-specific antigens -- proteins displayed on a tumor cell -- to stimulate the immune system. By injecting these antigens into the cancerous area of the patient, the immune system will produce an increased amount of antibodies or cytotoxic T lymphocytes, also known as killer T cells, to attack cancer cells that carry that specific antigen.

Heat shock proteins (HSPs) are believed to play a role in the presentation of antigens on the cell surface to help the immune system recognize diseased cells and therefore they can be conveniently used as potential anti-cancer vaccines. Also the entire repertoire of unique antigens from an individual patient's tumor can be used to stimulate a strong tumor-specific immune response. And again the entire repertoire of antigens unique to a person's cancer cells can be exposed to each FDA-approved cancer drug ($_{55}$) and drug induced mutations on the biochemical structure may be recorded ($_{53}$). DRUG - x modified antigens from an individual patient's tumor can then be used as the VAX-x component of a (VAX - x, DRUG - x)- Couple.

Dendritic cells break the antigens on the cancer cell surfaces into smaller pieces. The dendritic cells then display those antigen pieces to the killer T cells. To make a dendritic cell vaccine, some of the patient's dendritic cells are extracted and immune cell stimulants are used to reproduce large amounts of dendritic cells in the lab. These dendritic cells are then exposed to antigens from the patient's cancer cells. This combination of dendritic cells and antigens is then injected into the patient, and works like an anti-cancer vaccine. By analogy dendritic cells and DRUG - x exposed antigens can be injected into a patient as a VAX - x component of a (VAX - x, DRUG - x)- Couple.

Telomerase, a protein found in all major human cancers has proven to stimulate immune cells which are then able to kill multiple cancer cells and slow tumor growth. Almost all cancers express the telomerase protein. Thus telomerase might form the basis of a possible universal cancer vaccine. The TERT vaccine, made of a certain part of the telomerase protein, was able to stimulate an immune response that killed a wide variety of cancer cells and it showed that it could slow the growth of melanoma, bladder and breast cancers implanted into genetically unrelated mice. No other vaccine has been able to induce such a broad immune response. However, an effective anti-cancer vaccine has to display the antigens exactly as the cancer cells do. The effects of all FDA-approved anti-cancer drugs ($_{55}$) on the biochemical structure of telomerase should be studied. Since telomerase may constitute the main component of an universal anti-cancer vaccine, it should be administrated as the main and sole component of VAX - x in a DRUG-x-modified formula within (VAX - x, DRUG - x)- couples, and in its pristine, original, unmodified biochemical formula at the end of all successive (VAX - x, DRUG - x)- couples treatments, as a final vaccine sealing the victory of combined vaccine and chemotherapy science over cancer.

Farmarubicin-exposed telomerase (TERT-farmarubicin) should be used as the VAX-x component within the (TERT-farmarubicin, FARMARUBICIN) - couple in order to treat superficial bladder cancer. Other (VAX - x, DRUG - x)-couples suitable to treat superficial bladder cancer would be e.g. (TERT-thiotepa, THIOTEPA) , (TERT-doxorubicin, DOXORUBICIN), (TERT-mitomycin, MITOMYCIN). Accordingly, a successive series of 4 couples may be employed to replace MVAC chemotherapy in the treatment of muscle-invasive and metastatic bladder cancer: (TERT-methotrexate, METHOTREXATE), ------>(TERT-vinblastine, VINBLASTINE), ----->(TERT-doxorubicin, DOXORUBICIN), ------> (TERT-cisplatin, CISPLATIN).

At a limit , a powerful (TERT-MVAC, MVAC)-couple should be ultimately considered for the eradication of high stage and/or metastatic bladder cancers.

CONCLUSIONS

(VAX - x, DRUG - x)- Couples may be used in the treatment of all bladder cancers in particular, as well as in the treatment of all other types of cancer. This new strategy, theory and practical solution to treat cancer patients speaks for the individualization of anti-cancer treatments, rather than for large randomized clinical trials (RCT-s), since the patient's own cancer cells are used as a therapeutic vaccine.The main advantages of (VAX - x, DRUG - x)-Couples, when compared to conventional standard chemotherapy (e.g. MVAC) consist in the following:

1. higher efficiency in eradicating cancer in any patient, due to the sinergy between VAX - x, the immune system and DRUG - x ;

2. much lower toxicity and fewer side effects, since anti-cancer drugs can be administrated in monotherapy , doublets and triplets, compared to the 4-drug MVAC-therapy. The administration of successive (VAX - x, DRUG - x)-Couples, each based on a one-drug DRUG-x component, being also possible.

3. the use of (VAX - x, DRUG - x)- Couples creates an anti-cancer sinergy between the VAX - x vaccine, the patient's immune system, and the employed anticancer drugs, and this sinergetical effect is decissive in the eradication of cancer.

(VAX - x, DRUG - x)- couples may prove to be able to cure superficial, muscle-invasive,and even metastatic transitional cell carcinomas(TCC-s)of the bladder,especially those(TCC-s) which are refractory to current standard treatments and to all other existing alternative treatments.In addition, (VAX - x, DRUG - x)- couples may also prove to be more efficient in curing advanced and even metastatic cancers and to be less toxic than current conventional therapies,having the potential to become the standard treatment of care and cure for all bladder cancers, for all other types of cancer, and for absolutely all infectious diseases.

References

1. Lenarczyk A, Le TT, Drane D, Malliaros J, Pearse M, Hamilton R, Cox J, Luft T, Gardner J, Suhrbier A ISCOM based vaccines for cancer immunotherapy. Vaccine. 2004 Feb 25;22(8):963-74.
2. Fuessel S, Kueppers B, Ning S, Kotzsch M, Kraemer K, Schmidt U, Meye A, Wirth MP. Systematic in vitro evaluation of survivin directed antisense oligodeoxynucleotides in bladder cancer cells. J Urol. 2004 Jun;171(6 Pt 1):2471-6. (susanne.fuessel@mailbox.tu-dresden.de)
3. Leakakos T, Ji C, Lawson G, Peterson C, Goodwin S. Intravesical administration of doxorubicin to swine bladder using magnetically targeted carriers.Cancer Chemother Pharmacol. 2003 Jun;51(6):445-50. Epub 2003 Mar 22. (tleakakos@ferx.com)
4. Le Visage C, Rioux-Leclercq N, Haller M, Breton P, Malavaud B, Leong K. Efficacy of paclitaxel released from bio-adhesive polymer microspheres on model superficial bladder cancer.J Urol. 2004 Mar;171(3):1324-9.
5. Sylvester R. , Van der Meijden A.P.M. , Lamm D.L. , Intravesicle Bacillus Calmette-Guerin reduces the risk of progression in patients with superficial bladder cancer. A meta-analysis of the published results of randomised trials , J. Urol. 2002 : 168 : 1964 ;
6. Arnold J, de Boer EC, O'Donnell MA, Bohle A, Brandau S. Immunotherapy of experimental bladder cancer with recombinant BCG expressing interferon-gamma. J Immunother. 2004 Mar-Apr;27(2):116-23.
7. Cervenakov I, Szoldova K, Mardiak J, Chovan D, Mala M, Slavov D. Alpha 2-b interferon and farmarubicin in the prophylaxis of recurrence of superficial transitional cell carcinoma of the urinary bladder. Bratisl Lek Listy. 2000;101(6):317-20.
8. Mitsumori K, Tsuchiya N, Habuchi T, Li Z, Akao T, Ohyama C, Sato K, Kato T. Early and large-dose intravesical instillation of epirubicin to prevent superficial bladder carcinoma recurrence after transurethral resection. BJU Int. 2004 Aug;94(3):317-21.
9. Kuroda M, Niijima T, Kotake T, Akaza H, Hinotsu S; 6th Trial of the Japanese Urological Cancer Research Group. Effect of prophylactic treatment with intravesical epirubicin on recurrence of superficial bladder cancer--The 6th Trial of the Japanese Urological Cancer Research Group (JUCRG): a randomized trial of intravesical epirubicin at dose of 20mg/40ml, 30mg/40ml, 40mg/40ml. Eur Urol. 2004 May;45(5):600-5. (Takahiro.yamaguchi@japan.pfizer.com)
10. Milonas D, Mickevicius J, Mickevicius R, Motiejunas A, Sukys D, Gudinaviciene I. [Treatment of superficial transitional cell bladder carcinoma. Long-term results of trial comparing transurethral resection alone and adjuvant chemotherapy with Doxorubicin] Medicina (Kaunas). 2002;38 Suppl 1:79-83.
11. Harris NM, Anderson WR, Lwaleed BA, Cooper AJ, Birch BR, Solomon LZ. Epirubicin and meglumine gamma-linolenic acid: a logical choice of combination therapy for patients with superficial bladder carcinoma. Cancer. 2003 Jan 1;97(1):71-8. (neilandjane@supanet.com)
12. Yu ZX, Weng ZL, Chen W, Zhang FY, Zhou XS, Li CD [Intravesical instillation of pirarubicin (THP) together with polyvinylpyrrolidone (PVP) in the prevention of postoperative recurrence of superficial bladder cancer]Ai Zheng. 2003 Apr;22(4):421-3. (yuzhix@wz163.com)
13. Huang JS, Chen WH, Lin CC, Liaw CC, Wang CH, Lan YJ, Lai CH, Liu JP. A randomized trial comparing intravesical instillations of mitoxantrone and doxorubicin in patients with superficial bladder cancer. Chang Gung Med J. 2003 Feb;26(2):91-7. (ruby800@cgmh.org.tw)
14. Colombo R, Brausi M, Da Pozzo L, Salonia A, Montorsi F, Scattoni V, Roscigno M, Rigatti P.Thermo-chemotherapy and electromotive drug administration of mitomycin C in superficial bladder cancer eradication. a pilot study on marker lesion. Eur Urol. 2001 Jan;39(1):95-100.
15. Colombo R, Da Pozzo LF, Salonia A, Rigatti P, Leib Z, Baniel J, Caldarera E, Pavone-Macaluso M. Multicentric study comparing intravesical chemotherapy alone and with local microwave hyperthermia for prophylaxis of recurrence of superficial transitional cell carcinoma. J Clin Oncol. 2003 Dec 1;21(23):4259-60. (renzo.colombo@hsr.it), (Renzco@tin.it)
16. Logothetis CJ, Dexeus FH, Finn L, Sella A, Amato RJ, Ayala AG, Kilbourn RG. A prospective randomized trial comparing MVAC and CISCA chemotherapy for patients with metastatic urothelial tumors , J Clin Oncol. 1990 Jun;8(6):1050-5.
17. Loehrer PJ Sr, Einhorn LH, Elson PJ, Crawford ED, Kuebler P, Tannock I, Raghavan D, Stuart-Harris R, Sarosdy MF, Lowe BA, et al , A randomized comparison of cisplatin alone or in combination with methotrexate, vinblastine, and doxorubicin in patients with metastatic urothelial carcinoma: a cooperative group study. J Clin Oncol. 1992 Jul;10(7):1066-73.
18. Mead GM, Russell M, Clark P, Harland SJ, Harper PG,

Cowan R, Roberts JT, Uscinska BM, Griffiths GO, Parmar MK. A randomized trial comparing methotrexate and vinblastine (MV) with cisplatin, methotrexate and vinblastine (CMV) in advanced transitional cell carcinoma: results and a report on prognostic factors in a Medical Research Council study. MRC Advanced Bladder Cancer Working Party, Br J Cancer. 1998 Oct;78(8):1067-75.
19. von der Maase H, Hansen SW, Roberts JT, Dogliotti L, Oliver T, Moore MJ, Bodrogi I, Albers P, Knuth A, Lippert CM, Kerbrat P, Sanchez Rovira P, Wersall P, Cleall SP, Roychowdhury DF, Tomlin I, Visseren-Grul CM, Conte PF, Gemcitabine and cisplatin versus methotrexate, vinblastine, doxorubicin, and cisplatin in advanced or metastatic bladder cancer: results of a large, randomized, multinational, multicenter, phase III study. J Clin Oncol. 2000 Sep;18(17):3068-77.
20. Sternberg CN, de Mulder PH, Schornagel JH, Theodore C, Fossa SD, van Oosterom AT, Witjes F, Spina M, van Groeningen CJ, de Balincourt C, Collette L; European Organization for Research and Treatment of Cancer Genitourinary Tract Cancer Cooperative Group. Randomized phase III trial of high-dose-intensity methotrexate, vinblastine, doxorubicin, and cisplatin (MVAC) chemotherapy and recombinant human granulocyte colony-stimulating factor versus classic MVAC in advanced urothelial tract tumors: European Organization for Research and Treatment of Cancer Protocol no. 30924. J Clin Oncol. 2001 May 15;19(10):2638-46.
21. Siefker-Radtke AO, Millikan RE, Tu SM, Moore DF Jr, Smith TL, Williams D, Logothetis CJ. Phase III trial of fluorouracil, interferon alpha-2b, and cisplatin versus methotrexate, vinblastine, doxorubicin, and cisplatin in metastatic or unresectable urothelial cancer. J Clin Oncol. 2002 Mar 1;20(5):1361-7;
22. Malmstrom PU, Rintala E, Wahlqvist R, Hellstrom P, Hellsten S, Hannisdal E. Five-year followup of a prospective trial of radical cystectomy and neoadjuvant chemotherapy: Nordic Cystectomy Trial I. The Nordic Cooperative Bladder Cancer Study Group.J Urol. 1996 Jun;155(6):1903-6.
23. Grossman HB, Natale RB, Tangen CM, Speights VO, Vogelzang NJ, Trump DL, deVere White RW, Sarosdy MF, Wood DP Jr, Raghavan D, Crawford ED. Neoadjuvant chemotherapy plus cystectomy compared with cystectomy alone for locally advanced bladder cancer. N Engl J Med. 2003 Aug 28;349(9):859-66. Erratum in: N Engl J Med. 2003 Nov 6;349(19):1880.
24. Millikan R, Dinney C, Swanson D, Sweeney P, Ro JY, Smith TL, Williams D, Logothetis C. Integrated therapy for locally advanced bladder cancer: final report of a randomized trial of cystectomy plus adjuvant M-VAC versus cystectomy with both preoperative and postoperative M-VAC. J Clin Oncol. 2001 Oct 15;19(20):4005-13.
25. Freiha F, Reese J, Torti FM. A randomized trial of radical cystectomy versus radical cystectomy plus cisplatin, vinblastine and methotrexate chemotherapy for muscle invasive bladder cancer. J Urol. 1996 Feb;155(2):495-9; discussion 499-500.
26. Neoadjuvant cisplatin, methotrexate, and vinblastine chemotherapy for muscle-invasive bladder cancer: a randomised controlled trial. International collaboration of trialists.Lancet. 1999 Aug 14;354(9178):533-40.
27. Sengelov L, von der Maase H, Lundbeck F, Barlebo H, Colstrup H, Engelholm SA, Krarup T, Madsen EL, Meyhoff HH, Mommsen S, Nielsen OS, Pedersen D, Steven K, Sorensen B. Neoadjuvant chemotherapy with cisplatin and methotrexate in patients with muscle-invasive bladder tumours. Acta Oncol. 2002;41(5):447-56 (lisa.sengelov@dadlnet.dk)
28. Shipley WU, Winter KA, Kaufman DS, Lee WR, Heney NM, Tester WR, Donnelly BJ, Venner PM, Perez CA, Murray KJ, Doggett RS, True LD. Phase III trial of neoadjuvant chemotherapy in patients with invasive bladder cancer treated with selective bladder preservation by combined radiation therapy and chemotherapy: initial results of Radiation Therapy Oncology Group 89-03. J Clin Oncol. 1998 Nov;16(11):3576-83.
29. Lorusso V, Pagliarulo A, Selvaggi FP, Durini E, Riccardi F, Comella P, Fiorillo C, De Lena M. Treatment of advanced urothelial carcinoma with M-VECA (methotrexate, vinblastine, epirubicin and carboplatin). J Chemother. 1996 Apr;8(2):154-8;
30. Bellmunt Molins J, Ribas A, Albanell J, Lorente Garin JA, de Torres Mateos JA, Morote Robles J, Lopez Palacios MA, Banus Gassol JM, Casado Cobo S, Eres N, Sole Calvo LA. [Preliminary results of a phase II randomized controlled trial comparing M-VAC and M-CAVI in patients with bladder cancer (T2-4 N0-1 M0)] Arch Esp Urol. 1996 Jun;49(5):465-72.
31. Patton SE, Hall MC, Ozen H., Bladder cancer., Curr Opin Oncol. 2002 May;14(3):265-72. (spatton@wfubmc.edu)
32. Hussain M, Vaishampayan U, Smith DC., Novel gemcitabine-containing triplets in the management of urothelial cancer. Semin Oncol. 2002 Feb;29(1 Suppl 3):20-4.
33. Hussain M, Vaishampayan U, Du W, Redman B, Smith DC. Combination paclitaxel, carboplatin, and gemcitabine is an active treatment for advanced urothelial cancer. J Clin Oncol. 2001 May 1;19(9):2527-33; (hussainm@karmanos.org);
34. Bellmunt J, Guillem V, Paz-Ares L, Gonzalez-Larriba JL, Carles J, Albanell J, Tabernero JM, Cortes-Funes H, Baselga J.Gemcitabine/paclitaxel-based three-drug regimens in advanced urothelial cancer. Eur J Cancer. 2000 Jul;36 Suppl 2:17-25 (bellmunt@hg.vhebron.es)
35. Edelman MJ, Meyers FJ, Miller TR, Williams SG, Gandour-Edwards R, deVere White RW. Phase I/II study of paclitaxel, carboplatin, and methotrexate in advanced transitional cell carcinoma: a well-tolerated regimen with activity independent of p53 mutation. Urology. 2000 Apr;55(4):521-5.
36. Edelman MJ, Meyers FJ, Houston J, Lauder I. Phase I trial of paclitaxel, carboplatin, and methotrexate with granulocyte colony-stimulating factor and leucovorin in advanced transitional cell carcinoma. Semin Oncol. 1997 Oct;24(5 Suppl 15):S15-68-S15-71.
37. Pectasides D, Mylonakis A, Antoniou F, Kostopoulou M, Triantaphyllis D, Papadopoulou M, Varthalitis J, Dimitriadis M, Athanassiou A.Chemotherapy with methotrexate, carboplatin, mitoxantrone (Novantrone) and vincristine (Oncovin) in transitional-cell urothelial cancer.Oncology. 1998 Mar-Apr;55(2):139-44.
38. Dreicer R, Manola J, Roth BJ, See WA, Kuross S, Edelman MJ, Hudes GR, Wilding G.iPhase III trial of methotrexate, vinblastine, doxorubicin, and cisplatin versus carboplatin and paclitaxel in patients with advanced carcinoma of the urothelium.Cancer. 2004 Apr 15;100(8):1639-45. (dreicer@cc.ccf.org)
39. Stadler WM., Gemcitabine doublets in advanced urothelial cancer.Semin Oncol. 2002 Feb;29(1 Suppl 3):15-9.
40. Slaton JW, Karashima T, Perrotte P, Inoue K, Kim SJ, Izawa J, Kedar D, McConkey DJ, Millikan R, Sweeney P, Yoshikawa C, Shuin T, Dinney CP. Treatment with low-

dose interferon-alpha restores the balance between matrix metalloproteinase-9 and E-cadherin expression in human transitional cell carcinoma of the bladder. Clin Cancer Res. 2001 Sep;7(9):2840-53.
41. Edland RW, Wear JB Jr, Ansfield FJ. Advanced cancer of the urinary bladder. An analysis of the results of radiotherapy alone vs. radiotherapy and concomitant 5-fluorouracil; a prospective randomized study of 36 cases. Am J Roentgenol Radium Ther Nucl Med. 1970 Jan;108(1):124-9.
42. Martinez-Pineiro JA, Gonzalez Martin M, Arocena F, Flores N, Roncero CR, Portillo JA, Escudero A, Jimenez Cruz F, Isorna S. Neoadjuvant cisplatin chemotherapy before radical cystectomy in invasive transitional cell carcinoma of the bladder: a prospective randomized phase III study.J Urol. 1995 Mar;153(3 Pt 2):964-73.
43. Simon R, Atefy R, Wagner U, Forster T, Fijan A, Bruderer J, Wilber K, Mihatsch MJ, Gasser T, Sauter G. [HER-2 and TOP 2A gene amplifications in urinary bladder carcinoma]Verh Dtsch Ges Pathol. 2002;86:176-83;
44. Bellmunt J, Hussain M, Dinney CP. Novel approaches with targeted therapies in bladder cancer. Therapy of bladder cancer by blockade of the epidermal growth factor receptor family.Crit Rev Oncol Hematol. 2003 Jun 27;46 Suppl:S85-104. (bellmunt@hg.vhebron.es)
45. Latif Z, Watters AD, Dunn I, Grigor K, Underwood MA, Bartlett JM. HER2/neu gene amplification and protein overexpression in G3 pT2 transitional cell carcinoma of the bladder: a role for anti-HER2 therapy? Eur J Cancer. 2004 Jan;40(1):56-63.
46. de Pinieux G, Colin D, Vincent-Salomon A, Couturier J, Amsellem-Ouazana D, Beuzeboc P, Vieillefond A. Confrontation of immunohistochemistry and fluorescent in situ hybridization for the assessment of HER-2/ neu (c-erbb-2) status in urothelial carcinoma.Virchows Arch. 2004 May; 444(5):415-9. Epub 2004 Mar 17;
47. Kraemer K, Fuessel S, Kotzsch M, Ning S, Schmidt U, Wirth MP, Meye A. Chemosensitization of bladder cancer cell lines by human telomerase reverse transcriptase antisense treatment.J Urol. 2004 Nov;172(5 Pt 1):2023-8.
48. Kraemer K, Fuessel S, Schmidt U, Kotzsch M, Schwenzer B, Wirth MP, Meye A Antisense-mediated hTERT inhibition specifically reduces the growth of human bladder cancer cells.Clin Cancer Res. 2003 Sep 1;9(10 Pt 1):3521-5.
49. Betensky RA, Louis DN, Cairncross JG. , Influence of unrecognized molecular heterogeneity on randomized clinical trials. J Clin Oncol. 2002 May 15;20(10):2495-9. (betensky@hsph.harvard.edu)
50. Yang AS, Monken CE, Lattime EC.Intratumoral vaccination with vaccinia-expressed tumor antigen and granulocyte macrophage colony-stimulating factor overcomes immunological ignorance to tumor antigen. Cancer Res. 2003 Oct 15;63(20):6956-61.
51. Jurincic-Winkler CD, Metz KA, Beuth J, Klippel KF. Keyhole limpet hemocyanin for carcinoma in situ of the bladder: a long-term follow-up study. Eur Urol. 2000;37 Suppl 3:45-9.
52. Tei Y, Matsuyama H, Wada T, Kurisu H, Tahara M, Naito K. In vitro antitumor activity of bropirimine against urinary bladder tumor cells.Anticancer Res. 2002 May-Jun;22(3):1667-71.
53. Chabalgoity JA, Dougan G, Mastroeni P, Aspinall RJ. Live bacteria as the basis for immunotherapies against cancer. Expert Rev Vaccines. 2002 Dec;1(4):495-505.
54. Secasan IS, Pop DI , Fighting HIV with HIV Med Hypotheses. 1998 Jan;50(1):39-42.
55. Secasan I, Pop DI , Secasan CC, Winning The Battle Against HIV-1: (MPTV-x , HAART-x) / (MPTV-x , Mega-HAART-x) - Couples Formed of a Multivalent-Polivalent-Therapeutic-Vaccine (MPTV-x) and Its Corresponding HAART-x , or Mega-HAART- Regimen , Respectively. HIV & AIDS Current Trends, 2004, Mediscript Ltd, London.
56. http://www.imnr.com : The Immune Response Corporation, Inc. web-site
57. http://www.therionbio.com/ The Therion Biologics Corporation, Inc. web-site
58. http://www.adventrx.com/products/antiv_eradicaide.htm
59. http://www.affymetrix.com/index.affx
60. Coulie PG. Human tumour antigens recognized by T cells: new perspectives for anti-cancer vaccines? Mol Med Today. 1997 Jun;3(6):261-8. (coulie@licr.ucl.ac.be)
61. http://www.fda.gov/cder/cancer/druglistframe.htm , An almost complete Listing of (FDA) Approved Oncology Drugs with "Approved Indications";

Author Information

Iosif Secasan
Department of Urologic Surgery, Spitalul Judetean Resita / The Hospital of Resita

Dan I. Pop
Data International SRL

Ciprian C. Secasan
Department of Microbiology / Department of Urologic Surgery, Spitalul Judetean Resita / The Hospital of Resita