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Short Review

Oncostatic-Antibody Complexes in Chemotherapy

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Summary. *Cancer-chemotherapeutic agents are not selective in their action against cancer cells. An approach to increasing the efficacy of currently available antitumor drugs is the binding of selected chemotherapeutic agents to antibodies that do posses specific affinity for the tumor cells.*

In this short review consideration is given to some aspects of induction of antitumor antibodies, techniques for binding of various chemotherapeutic agents to antibodies, immunochemotherapy with oncostatic-antibody complexes and their mode of action, and future prospects of this form of therapy.

Introduction

Cytostatic and cytotoxic drugs are frequently used in the treatment of cancer. However, there are often limitations in their application as result of an accompanying systemic cytotoxicity. Not only tumor cells are attacked, but all proliferating cells, including those of bone marrow, lymphoid tissue, and gastrointestinal and genitourinary [19, 43] epithelium are damaged.

Several attempts have been made at enhancing the efficacy of existing antitumor agents. One succesful attempt is combination chemotherapy, which has been receiving increasing attention [1, 7, 34]. Another approach is the use of carriers for chemotherapeutic drugs. Macromolecules [5, 54, 55] and liposomes [26, 33, 40] have been used. A still more attractive concept is the binding of anticancer drugs to tumor-specific antibodies. Treatment of tumors with cytostatic or cytotoxic agents linked to antitumor antibodies offers the possibility of combining the chemotherapeutic potency of the agents with the 'homing' activity of the antibodies. For this reason methods have been developed to bind cytotoxic drugs to plasma proteins [32, 43, 53, 56, 59].

This idea of addressing a cytotoxic drug to a particular tissue (tumor) destination and posting it on a tissue-specific antibody has already existed for many years. Ehrlich [14] pointed out the possibility of using diphtheria toxin bound to antitumor antibodies as a 'magic bullet' against malignancies in 1900 [18]. More than 50 years later Mathé et al. [39] reported the first successful treatment of L1210-bearing DBA/2 mice. They used methotrexate linked by diazotization to the globulin fraction from a hamster anti-mouse L1210 leukemia serum. More recently, clinical succes with this immunochemotherapy has been described by Ghose and his co-workers [22, 25] and Oon et al. [50]. In these studies a 'complex' of chlorambucil and antitumor antibodies was used. This short review will assess some aspects of the (immuno)-chemotherapy with cytostatic-antibody complexes. Attention is given to the induction of the antitumor antibodies, techniques for the binding of various chemotherapeutic agents to the antibodies, therapeutic efficacy of drug-antibody complexes and their mode of action, and future prospects of this form of cancer therapy.

Antitumor Antibodies

The use of antibodies in this form of immunochemotherapy can give rise to enhancement of tumor growth [15], a problem that is difficult to obviate. Furthermore, there is a chance of anaphylactic reaction(s) during treatment. The latter problem can be minimized by using aggregate-free autologous antibodies obtained from the patient's serum [8, 29, 38] or antibodies eluted from the disected tumor tissue [28]. Although autologous antibodies seem to offer the best possibilities for 'loading' with chemotherapeutic agents, the antibody activity of these antibodies is often low.

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In most experimental models [12, 13, 16] and in the human situation [15, 22, 43] drug-antibody complexes were formed with xenogeneic antibodies. Surprisingly, such problems as anaphylactic reactions or tumor enhancement are only rarely mentioned [19, 42] in human studies.

In experimental systems allogeneic and xenogeneic antitumor sera, and in the human situation xenogeneic antisera have been raised by immunization. The influence of various factors, such as the method of tumor cell preparation, the use of adjuvants, doses and route of immunization, have been extensively reviewed by Praeger and Baechtel [45]. Whole tumor cells, homogenates, or purified tumor-associated transplantation antigen (TATA)-containing fractions can be used.

A significant disadvantage of the use of these antisera is that they have to be adsorbed extensively with normal tissues to prevent cross-reactions with nontumor tissues. The absorption procedures demand considerable amounts of normal tissues [10] and can result in low specific antitumor antibody activity [10, 43].

Nevertheless, antitumor antibodies are attractive carriers for various chemotherapeutic drugs, as they do show specific homing activity for the tumor tissue [18]. For the homing activity it is necessary that the antibodies bind specifically to the tumor antigens, but it is not necessary that they are cytolytic to be effective in this form of therapy [12, 13].

Oncostatics and Their Binding to Antibodies

Several types of drugs, such as the alkylating agents chlorambucil [10, 12, 16, 18, 27] and trenimon [36, 39], radionuclides [21, 46], the antibiotics daunomycin and adriamycin [30, 35], the antimetabolite methotrexate [7, 39], and diphtheria toxin [41, 42] have been coupled to antibodies and tested both in vitro and in vivo for their antitumor efficacy (recently reviewed [25]).

For various drugs suitable physical or chemical coupling methods to antibodies have been developed [54]. Problems encountered in the linking procedures are possible formation of drug aggregates [3] and possible decrease of reactivity of the drug [57] and antibody [12, 13] after complexing. Three well-described methods of binding drugs to antibodies are: (a) noncovalent or physical binding [2, 12, 13, 22, 27]; (b) covalent binding [7, 17, 36]; and (c) covalent binding with the help of intermediate carriers such as carbodiimide [48], glutaraldehyde [31], polyglutamic acid [50], or dextran [49]. The aim of all these methods is to prepare a stable drug-antibody conjugate. However, the probability of in vivo dissociation of the conjugate cannot be excluded [10, 47, 51]. On the other hand, the necessity for complexing the drug and immunoglobulin is still questionable [4, 47, 49, 54], as it has been shown that a mixture of chlorambucil and antibody or successive injection of drug and antibody can also be effective in suppression of tumor growth in the in vivo situation [9–11]. In vitro a mixture as well as a complex of chlorambucil and specific antibodies can also kill cultured tumor cells [51, 57].

Immunochemotherapy with Oncostatic-Antibody Complexes

Treatment of tumor-bearing mice with drug-antibody complexes can result in prolongation of the survival time of these mice compared with the survival time of the untreated mice [4, 10, 12, 16, 20, 22, 39]. Moolten et al. [41] conjugated (with gluturaldehyde) diphtheria toxin to anti-DNP antibodies and found that the complex restricted the growth of DNP-coated sarcoma cells in hamsters. Ghose et al. reported protection of mice against Ehrlich ascites carcinoma and EL4 lymphoma, obtained with a complex of chlorambucil and absorbed rabbit-anti-mouse tumor sera [18, 22].

Complete tumor eradication has also been obtained: Flechner [16] reported successful treatment of EL4 lymphoma-bearing C57BL mice with a complex of chlorambucil and antitumor antibody. Similar results have been described by Dullens et al. [12, 13], who also used chlorambucil linked to rabbit antitumor antibodies in the BALB/c Harding-Passey melanoma system. However, no prolongation of the survival of tumor-bearing mice was obtained after treatment in the DBA/2–SL2 lymphoma model.

Treatment of tumor-bearing animals with the drug or the antibody only might also result in some prolongation of survival, but the effect of the complex has been shown to be better in a majority of studies [6, 12, 16, 22, 23]. Whether the drug-antibody complex is more effective than a mixture of both components is still controversial [12, 13, 49, 50].

The number of clinical studies is limited. A few case reports of patients with neuroblastomas [44] or with melanomas [22, 25] achieving tumor regression after treatment with a chlorambucil-antitumor antibody complex have been published. Promising, although not conclusive, results have been reported by Newman et al. [43] and Everall et al. [15] after treatment of cancer patients with a therapy of antitumor antibodies in combination with various drugs.

So the question as to whether the drug and the antibody need to be linked or not still needs to be elucidated both in the experimental and in the human situation.

Conclusions and Future Prospects

Drug-antitumor antibody complexes can have tumor-inhibitory activity [12, 13]. The precise mechanism of in-

creased tumor inhibition by oncostatic drugs attached to antibodies still remains to be elucidated. It might be explained on the basis of (a) a synergism between drug and the antibody, (b) antibody-preferential localization of the drug on tumor cells, or (c) both [23, 24]. Drug effects on the cell membrane resulting in an increased susceptibility for the lytic action of antibody and complement have been described [37, 52, 58]. On the other hand, the transport of the drug across the cell membrane might be facilitated by antibody-induced capping of the drug-antibody complex followed by endocytosis [27]. In any case, there is experimental evidence that antibodies [24], as well as the drug-antibody complex [53], can reach the tumor burden, and both the antibody and the drug might be able to exert their cytotoxic action against the tumor cells. Nevertheless, there are several limitations on the use of drug-antibody complexes against cancer. The preparation of proper antitumor sera [60] is a problem, especially for clinical studies. Furthermore, the type of tumor against which this form of therapy can be used might also be important. It has been shown that treatment of mice bearing a tumor with a low growth rate with clorambucil-antibody complexes was effective. However, this form of therapy was not effective in mice bearing a lymphoma with high growth rate [12, 13]. In other words, it has to be investigated whether this form of immunochemotherapy is suited for all types of tumors or only for a selected group.

Finally, one must consider the fact that a drug with certain antitumor effecacy might react differently when used in combination with antitumor antibodies [60], either because the antibody prevents the drug of reaching the target site or because it causes steric hindrance. These above-mentioned problems, plus the possible introduction of macromolecules [55, 56] or liposomes [26, 33, 40] as alternative carriers (instead of antibodies) need further experimental and clinical study.

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