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The bispecific monoclonal antibody (biMAb) OC/TR combines the anti-ovarian-cancer-reactivity of the MOv18 monoclonal antibody (MAb) with the reactivity of an anti-CD3 MAb. Pre-clinical studies have indicated that this biMAb is able to redirect the cytolytic activity of T cells towards tumour cells, resulting in efficient tumour-cell lysis. To assess the clinical potential of systemic biMAb-based cancer therapy we initiated a study in ovarian-cancer patients. Five patients suspected of ovarian cancer received 125I-OC/TR F(ab')2 i.v. Unexpectedly, the first patient developed side effects (grade III-IV toxicity) starting 30 min after infusion (p.i.) of 1 mg of OC/TR F(ab')2. After approval of the Ethical Committee, the study was continued at lower dose levels (0.1 mg; 0.2 mg). However, at the 0.2-mg dose level similar side effects were observed. FACS analysis indicated that all peripheral T cells were coated with biMAb immediately following the infusion. The cytokines tumour necrosis factor-α, interferon-γ and interleukin-2 showed maximum serum concentrations 2 h p.i. Tumour uptake ranged from 0.8 to 1.9% ID/kg, resulting in tumour/background ratios of 3 to 8. Our results suggest that at higher antibody dose levels OC/TR F(ab')2 causes T-cell activation with acute release of cytokines. Only low doses of biMAb can be administered safely. Despite the interaction with T cells, OC/TR F(ab')2 preferentially localizes in tumours following i.v. administration, thus offering therapeutic perspectives.

A decade ago, redirecting effector cells towards tumour cells using bispecific monoclonal antibodies (biMabs) was proposed as a new concept for cancer therapy (Perez et al., 1985). In this approach, biMabs composed of an anti-tumour antibody on one hand and an anti-effector cell antibody on the other, are used to redirect the cytolytic activity of effector cells towards tumour cells. T-cell target recognition and cytolytic activity is restricted to major-histocompatibility-complex (MHC)-associated epitope presentation. In vitro studies have indicated that MHC restriction as well as epitope specificity of T cells can be circumvented with biMabs. With biMabs physically linking a T cell with a tumour cell, tumour-cell lysis could be obtained using either tumour cell lines or freshly isolated tumour cells (Perez et al., 1986; Pupa et al., 1988). In a number of pre-clinical studies, inhibition of tumour growth after loco-regional administration of pre-activated human peripheral-blood lymphocytes (PBL) retargeted with anti-tumour X anti-CD3 biMabs has been demonstrated (Titus et al., 1987; Van Ravenswaay Claassen et al., 1994).

Clinical experience with anti-tumour X anti-CD3 biMabs is limited and confined to locoregional administration of pre-activated PBL redirected with biMabs (Bolhuis et al., 1992; Kroesen et al., 1993). The first results of these studies indicated that locoregional administration can induce local clinical responses. However, with locoregional administration the growth of distant metastases was not controlled.

It remains to be investigated whether this approach can also be applied systemically. In mice with s.c. human Hodgkin lymphomas, i.v. administration of anti-tumour X anti-CD3 biMab and pre-activated human PBL resulted in tumour-cell lysis and cure (Renner et al., 1994). Furthermore, in a syngeneic mouse lymphoma model long-term survival and cure of the animals was demonstrated after a sole i.v. injection of biMAb (Brisbinck et al., 1991).

In vitro studies have demonstrated that, after one cytolytic cycle, clustered CD3/T-cell-receptor(TCR) complexes on the T cell can no longer transduce the lytic signal. Repeated addition of biMAb is required for binding newly expressed CD3/TCR complexes to re-engage the T cell in another lytic cycle (Blank-Voorthuis et al., 1993). This limitation of the cytolytic mechanism would indicate that administration of pre-coated T cells is a poor strategy, even if the retargeted T cells would find the target cell in vivo. Based on this, an approach in which the tumour cells are coated with biMAb seems preferable. In such an approach, it would be desirable to administer biMAbs systemically. The present study aimed to investigate localization in tumour, biodistribution and toxicity associated with the systemic administration of the biMAb OC/TR F(ab')2 in ovarian cancer patients.

Estimates on the toxicity related to i.v. administration of OC/TR F(ab')2 were based on the following observations and considerations. After repeated 1.p. infusions of redirected PBL, interleukine-2 (IL-2) and mg doses of OC/TR F(ab')2, only mild toxicity was observed (Bolhuis et al., 1992). F(ab')2 fragments of the biMAb were chosen to exclude other cytolytic mechanisms (antibody-dependent cell cytotoxicity, complement activation) than redirection of T cells. A monovalent anti-CD3 arm lacking an Fc part excludes cross-linking of CD3-TCR receptor complexes, therefore no activation of lymphocytes was expected to occur (Segal et al., 1991). Based on these considerations, no or at the most mild toxicity was expected after a single i.v. infusion of a low protein dose of OC/TR F(ab')2.

To visualize and quantify the biodistribution, the biMAb was radiolabeled. 125I was chosen as the most suitable radionucleide for this study because of its radiation characteristics and, compared with 111In and 90Y/Tc, its reduced tendency to accumulate non-specifically in liver, spleen, kidney and bone marrow.

A weighted antibody-dose escalation was proposed, since it was assumed that the biMAb uptake in tumour in terms of protein amount per gram of tumour would increase when a higher protein dose would be administered.

PATIENTS AND METHODS

Monoclonal antibody

The biMAb OC/TR (IgG1/IgG1) was obtained after fusion of the MOv18 hybridoma with spleen cells from a BALB/c mouse immunized with a human T-cell clone (Van Ravenswaay Claassen et al., 1993). The MOv18 MAb is a murine
antibody of the IgG1 immunoglobulin sub-class that recognizes a 38-kDa folate-binding protein expressed on the majority of ovarian carcinomas as well as on adenocarcinomas of the Fallopian tube, endometrium and cervix (Miotti et al., 1987; Campbell et al., 1991). Using immunoblotting and Northernblot techniques, reactivity with normal tissues was seen with ovarian, fallopian tube, kidney, lung, thyroid, and choroid plexus tissue (Weitman et al., 1992). No reactivity with bone marrow or peripheral-blood cells was observed (Miotti et al., 1987). The antibody preparations were prepared and supplied by Centocor Europe (Leiden, The Netherlands). The product was subjected to a wide range of quality-control tests, including tests for endotoxin and pyrogen contamination. The preparation was negative in the limulus amoebocyte lysate (LAL) test (sensitivity: 0.06 EU/mL).

Radiolabelling and quality control
F(ab')2 fragments of OC/TR were labelled with approximately 185 MBq (5 mCi) of 125I using the iodogen method (Fraker and Speck, 1978). All solutions and labware used were sterile and pyrogen-free. Briefly, the OC/TR F(ab')2 with 50 mM sodium phosphate (pH 7.2) was added to an iodogen-coated tube (10 µg/100 µl). Following 10-min incubation with 125I (370 MBq; 25 µl; Medgenix, Fleurus, Belgium), the reaction mixture was applied on a Sephadex G-25 column and eluted with PBS. The fractions containing the labelled antibody were pooled. Instant thin-layer chromatography was used to determine the presence of free 125I in the preparations. After labelling, the radio-immunoconjugates were subjected to a LAL test: these tests were negative as well.

Immunoreactivity of the preparations was assessed towards HeLa (human cervix-carcinoma) cells and towards Jurkat (CD3-positive human acute T-cell leukemia) cells, as described (Tibben et al., 1994). Data were plotted as described by Lindmo et al. (1984) and the immunoreactive fraction (IRF) was calculated from the Y-axis abscissa.

Patients
Patients enrolled in the study (n = 5) were suspected of having ovarian cancer and were scheduled for surgery. Patients had to be over the age of 18 years and had to have a life expectancy of at least 3 months. All patients with previous exposure to murine MAbs, known allergy for rodents, allergic diathesis, life-threatening infection, organ failure, evidence of recent myocardial infarction or diagnosis of a second malignancy were excluded. Written informed consent was obtained prior to study entry. Patient characteristics are summarized in Table I. All studies were conducted with the approval of the Internal Review Board of the University Hospital Nijmegen, The Netherlands.

Study design
Prior to the antibody injection, patients were evaluated by medical history and physical examination. An electrocardiogram, a chest radiograph, ultrasonography of the pelvis and X-ray-computed tomography of the abdomen (optional) were obtained.

A weighted protein-dose escalation was intended, starting at 1.0 mg. Three patients were planned to receive the same dosage. If no toxicity was observed, the study was to be continued at the next highest dose level. Endpoint of the study would be WHO-toxicity grade III or IV.

Patients received a single i.v. infusion of 125I-OC/TR F(ab')2. The radio-immunoconjugate was diluted in 5 ml of saline and infused over 30 min. In order to prevent thyroid radiation, patients were given 100 mg potassium iodide twice a day and 200 mg potassium perchlorate 4 times a day, orally, starting 4 hr before the infusion and continued for 3 days. Vital signs were measured frequently up to 8 hr post infusion (p.i.).

Blood samples were collected just prior to antibody administration and at various time intervals until surgery. Radioactivity in the blood as a percentage of the injected dose per gram (%ID/g) of blood was determined. The half-life of disappearance from the blood was calculated using non-linear least-square regression analysis.

Planar images of the chest, abdomen and pelvic region in anterior and in posterior views were obtained at approximately 4, 24 and 48 hr p.i. with a preset time of 5, 10 and 15 min respectively. Images were obtained with a single-headed gamma camera (Type Orbiter, Siemens, Hoffman Estates, IL) equipped with a medium-energy collimator. Planar views were recorded using the 159-keV gamma ray peak of 123I with a symmetric 15% window. To reduce bladder activity, patients were asked to void prior to imaging of the pelvic region.

The conjugate view-counting technique was used to quantify the activity uptake in some organs (Buijs et al., 1992). Briefly, region-of-interest(ROI) measurements of these organs were performed on anterior and posterior view images at all time points. After correction for background activity, the geometric mean of anterior and posterior images was calculated. The absolute uptake in an organ was calculated via comparison with whole-body counter measurements.

Surgery was performed at approximately 72 hr p.i. in all patients. The tumour status was carefully mapped. Ascites or peritoneal washings were collected. Suspected tissues were either removed or biopsied. All tissues obtained at operation were weighed and the amount of radioactivity of 123I was measured using a well-type gamma counter.

FACS analysis
Two-colour fluorescence-assistant-cell-sorter(FACS) analysis was performed to study the binding of OC/TR to white blood cells. Briefly, 100 µl samples of whole blood were incubated with anti-IgG1-phycocerytrin (PE) (50 µl; 10 µg/ml; Southern Biotechnology Associates, Birmingham, AL) and/or with anti-CD3-fluorescein isothiocyanate (FITC) (15 µl; Becton and Dickinson, Eten-Leur, The Netherlands). Control samples were incubated with species and isotype-matched control antibodies (PE or FITC-labelled). After washing (200 µl HBSS, 0.5% BSA, 0.1% NaN3) red blood cells were lysed with FACS-lysing solution (Becton and Dickinson, Eten-Leur, The Netherlands). The fluorescence of the lymphocytes was analyzed on a Coulter Epics Elite flow cytometer (Coulter, Hialeah, FL).

<table>
<thead>
<tr>
<th>Patient number</th>
<th>bMAB dose (mg)</th>
<th>Age (yr)</th>
<th>FIGO stage</th>
<th>Histology</th>
<th>Diff. grade</th>
<th>Side effects</th>
<th>Cyto- kines</th>
<th>RIS</th>
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<tr>
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<td>71</td>
<td>IIIc</td>
<td>serous papillary cystadenocarcinoma</td>
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<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
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<td>0.1</td>
<td>71</td>
<td>IIIc</td>
<td>serous papillary adenocarcinoma</td>
<td>III</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
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<td>66</td>
<td>Ia</td>
<td>endometrioid adenocarcinoma</td>
<td>II</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
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<td>56</td>
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<td>serous cystadenoma</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
</tbody>
</table>

NA, not applicable; -, negative; + , positive; + + , strongly positive.
Cytokines

Blood was collected in EDTA tubes and immediately put on ice. After centrifugation at 4°C, plasma was stored at −20°C until analysis. Plasma levels of the cytokines IL-2, tumour necrosis factor-α (TNF-α) and interferon-γ (IFN-γ) were determined using commercial immunoradiometric assays (IRMA) (IL-2 and IFN; Medgenix, Fleurus, Belgium) and an in-house radio-immuno assay (TNP).

Humoral anti-OC/TR response

Development of human anti-mouse antibodies (HAMA) was assessed using OC/TR F(αβ)2 in an in-house homologous sandwich-type IRMA. Briefly, OC/TR F(αβ)2-coated polystyrene wells were incubated with patients’ sera. After washing, the bound HAMA were traced with 125I-labeled OC/TR F(αβ)2.

RESULTS

Radiolabeling and quality control

The labeling efficiency of the radio-iodination of the antibody was between 42 and 78%. After gel filtration, ITLC immunoassay indicated that more than 95% of the 123I was protein-bound. The IRF was 40 to 52% for the anti-tumour arm of the biMAb and 76 to 84% for the anti-CD3 arm of the antibody.

Patients

Dose level 1.0 mg. Thirty minutes after the i.v. infusion of 1 mg of 123I-OC/TR F(αβ)2 the first patient experienced the following symptoms: chills and headache; nausea, vomiting and diarrhoea; fever up to 40°C in combination with hypotension; and fatigue. Symptomatic treatment consisted of administration of plasma substitutes, metoclopramide and paracetamol. At 48 hr p.i. the patient’s clinical condition was normalized.

According to the study protocol, the protein dose should be decreased if toxicity grade III (nausea and vomiting) or IV (fever in combination with hypotension) occurred. However, toxicity occurred at the lowest dose level. It seemed of scientific interest to further investigate the objectives of the study, and after approval of the Review Board the study was continued at lower dose levels, starting at 0.1 mg.

Dose level 0.1 mg. Three patients were injected with 0.1 mg 123I-OC/TR F(αβ)2. One of these patients developed fever up to 39°C from 8 to 10 hr p.i. No other side effects were observed.

Dose level 0.2 mg. One patient received 0.2 mg. This patient developed symptoms almost identical to those of the first patient: chills and headache; nausea and vomiting; hypotension in combination with fever up to 39.1°C. Again, plasma substitutes and paracetamol were given to reduce these symptoms. At 24 hr p.i., the patient’s clinical condition was normalized. Grade-IV toxicity (fever in combination with hypotension) had again occurred now at the 0.2-mg dose level. At this point the investigators decided to stop the study.

Pharmacokinetics

Blood clearance data are presented in Figure 1. The blood concentrations in terms of %ID/g did not appear to be related to the administered antibody dose. Therefore the data of all 5 patients were pooled. Clearance of 123I from the blood could be described by a bi-exponential model with mean half-life values of 1.0 ± 0.2 hr (range: 0.7–1.2 hr) for the distribution phase (T1/2α) and 18.1 ± 6.7 hr (range: 13.0–29.6 hr) for the elimination phase (T1/2β), which is in the normal range for a F(αβ)2 fragment (Buist et al., 1993). Mean cumulative excretion of the radiolabel in the urine was 63 ± 15% ID in 24 hr and 76 ± 16% ID in 48 hr.

Scintigraphy and dosimetric analysis of the images

On the images of 2 patients with ovarian carcinoma, the tumour was visualized (Fig. 2). With an antibody dose of 0.1 mg, tumour was imaged as early as 3 hr p.i. In the third patient, the ovarian-tumour deposit with a diameter of 3 mm was not visualized. This may have been due to activity in the urinary bladder. The presence of malignant tumour was surgically and histopathologically confirmed. The benign tumours of 2 patients (uterine myoma, and serous cystadenoma of the ovary) did not show elevated uptake.

High 123I uptake in the spleen was seen throughout the study in all patients. Drawing ROIs, uptake in various organs was calculated for all patients. The uptake in normal tissues did not appear to be related to the protein dose. Therefore the data of all 5 patients were pooled. Mean retention in organs expressed as %ID is presented in Table II. The uptake in the spleen was
Thyroid uptake was clearly visible at 24 hr and 48 hr p.i., the liver showed some elevated uptake. The uptake was approximately 30% ID at 4 hr p.i., whereas at 48 hr p.i. total bone-marrow uptake represented approximately 5% ID. Throughout the study, the spleen weighed 1.9 kg. Uptake in malignant tumour deposits varied from 0.1 to 0.6% ID/kg. The tumour-to-muscle ratio was 8, 3 and 3 for the patients with malignant tumours, whereas in the patients with benign tumours this ratio was 1.5 and 1 respectively.

**FACS analysis**

In all 5 patients, FACS analysis revealed that, at 10 min before the completion of the 30-min antibody infusion, virtually all CD4+ cells (CD4+ as well as CD8+) were coated with OC/TR F(\(\text{ab'}\))2 (Fig. 4). In the patients that received the higher protein dose levels (1.0 and 0.2 mg), the decrease of lymphocytes at later time points hampered accurate gating of large numbers of lymphocytes. Consequently, these FACS analyses are based on much lower lymphocyte numbers, and are less accurate.

Differential counting of the blood cells and the plasma indicated that less than 15% of the radioactivity was associated with the blood cells, indicating that the majority of the biMAb in the circulation was not cell-associated.

**White-blood-cell differentiation**

The white-blood-cell (WBC) differentiation showed relevant changes p.i. in 4 patients. In patient 01, this WBC counting was performed at only 3 time points (pre-infusion, and 24 and 48 hr p.i.). FACS analysis of the WBC of this patient semi-quantitatively showed a decrease of leucocytes at the time in between (pre-infusion—28 hr p.i.). In patient 03, the WBC differentiation did not change. In the other 3 patients, the absolute number of lymphocytes decreased 6 to 21 times, starting from half an hour p.i. (Fig. 5). At 48 hr p.i., the lymphocyte numbers had recovered to pre-infusion levels. The absolute number of granulocytes showed no consistent changes.

**Cytokines**

Only the patients who received the 1.0- and 0.2-mg antibody doses showed IL-2 concentrations above the detection limit of the assay shortly after the infusion. The maximum concentrations were measured 2 hr p.i. At 5 and 7 hr p.i., respectively, the IL-2 concentrations were still elevated, while in the next samples obtained the following day the concentrations had normalized to below the detection limit. Maximum serum levels of TNF-\(\alpha\) were also obtained 2 hr p.i. (Fig. 6a). The patients who received the higher doses (1.0 and 0.2 mg) demonstrated much higher TNF-\(\alpha\) peaks than those who received the lower dose (0.1 mg). The TNF-\(\alpha\) concentrations had normalized to pre-infusion levels at approximately 24 hr p.i. in all patients. In the higher-dose patients (1.0 and 0.2 mg), serum IFN-\(\gamma\) levels peaked simultaneously with IL-2 and TNF-\(\alpha\) (Fig. 6b). The concentrations of IL-2 and TNF-\(\alpha\) correlated well with the clinical symptoms: highest levels were obtained in the patients who experienced the most severe side effects (0.2- and 1.0-mg dose level). Moreover, at the 0.1-mg dose, the highest TNF-\(\alpha\) serum level was observed in the patient who experienced fever.

**HAMA response**

As early as 1 and 3 weeks p.i., respectively, elevated HAMA titers were detected in the serum samples of the patients who received the 1.0- and 0.2-mg antibody dose. One of the 3 patients receiving the 0.1-mg dose demonstrated apparent pre-existing HAMA, while no HAMA responses were observed in the other 2 patients.

**DISCUSSION**

The present study was performed to investigate the applicability of i.v. administration of the biMAb OC/TR F(\(\text{ab'}\))2. The toxicity and pharmacokinetics associated with the systemic administration of this biMAb and its potential to localize in
Lymphocytes of patient with ovarian carcinoma. Tumour-to-muscle ratios varied from ovarian carcinoma. Detection on the peripheral CD3+ cells. However, from 5 hr p.i. onwards OC/TR cannot be obtained with OC/TR F(ab')2 is the result of the ovarian-carcinoma-specific MOv18 reactivity of this biMAb. However, final proof on specific tumour uptake can be obtained only by comparing the tumour uptake of the biMAb OC/TR with that of a control biMAb with CD3 reactivity and an irrelevant other arm.

Several investigational methods provided information on the interaction of OC/TR F(ab')2 with CD3+ lymphocytes. Early planar images showed high uptake in spleen and bone marrow in all patients. FACS analysis revealed that during the i.v. infusion all peripheral CD3+ lymphocytes became coated with the biMAb. After completion of the infusion, the number of biMAb-coated lymphocytes decreased somewhat rapidly. In addition, the white-blood-cell differentiation showed a significant decrease in the number of lymphocytes, starting immediately after completion of the infusion. Taken together, these findings suggest that the biMAb-coated lymphocytes leave the circulation and traffic to the lymphoid organs. Kroesen et al. (1994) described a similar rapid decrease of BIS-1 F(ab')2 biMAb-coated lymphocytes in blood after i.v. administration in renal-cell-cancer patients. Their in vitro experiments indicated that this could not be attributed to internalization, supporting the hypothesis that T cells loaded with biMAb leave the circulation.

In 2 patients, side effects occurred as early as 30 min after the i.v. infusion of 1.0 and 0.2 mg OC/TR F(ab')2. Patients experienced chills, headache, nausea and vomiting, diarrhoea, hypotension and fever until approximately 20 hr p.i. The observed complex of symptoms suggested the acute release of cytokines after the infusion of OC/TR F(ab')2. Serum levels of IL-2, TNF-α and IFN-γ increased immediately after the infusion and reached maximum concentrations at approximately 2 hr p.i. The IL-2 and TNF-α levels correlated well with the clinical condition of the patients; the highest levels and the strongest increase were observed in the patients who experienced the most severe adverse reactions.

Similar symptoms were observed in renal-cell-cancer patients after i.v. administration of the biMAb BIS-1 F(ab')2 in combination with s.c. injection of IL-2 (Kroesen et al., 1994). In these patients, the TNF-α levels also peaked at 2 hr p.i. These findings suggest that the symptoms observed in the 2 studies are not the result of intrinsic characteristics of a particular MAb, but the consequence of the anti-CD3-x-anti-tumour-antibody construct.

A similar complex of symptoms has been reported after i.v. administration of the anti-CD3 MAb OKT3 in renal-trans-
plant patients (Abramowicz et al., 1989). The serum cytokine levels showed a pattern of prompt increase of both TNF-α and IFN-γ. It is thought that the acute symptoms are caused by cytokine release resulting from T-cell activation by the OKT3 MAb. In vitro studies indicate that intracellular clustering of the CD3/TCR complex is a prerequisite for effective T-cell activation with anti-CD3 MAb (Segal et al., 1991). However, this clustering can occur only when the biMAb is immobilized (e.g., via an intact Fc part or via an antibody-antigen interaction) (Segal et al., 1991). The anti-tumour MAb MOV18 is reactive with an epitope with very restricted expression on the CD3/TCR complex. It is therefore assumed that F(ab')2 fragments of the biMAb OC/TR cannot induce such clustering except in the presence of tumour cells. Theoretically, any cells expressing the MOV18 antigen could serve as a template for CD3/TCR-complex clustering. Assuming that a limited number of T cells would have to be activated to produce cytokines in concentrations able to cause the observed side effects, this could have happened in any tissue of the body expressing the MOV18 epitope.

It has been suggested that the presence of trace quantities of intact anti-CD3 in the biMAb preparations could induce T-cell activation (Weiner et al., 1994). In our study this appeared highly unlikely, since high-performance liquid-chromatography analysis of the OC/TR F(ab')2 preparation confirmed the absence of any intact IgG molecules.

Kroesen et al. (1994) proposed that the increase in serum cytokine levels may be the result of local activation of T cells at the site of the tumour after cross-linking of the biMAb with tumour antigens. However, we also observed the syndrome in a patient without any known malignant tumour lesion (patient number 05). In addition, T-cell activation occurred very early after injection, whereas localization of MAb's in tumours occurs somewhat later.

In conclusion, the concept of redirecting effector cells of the immune system towards tumour cells using bispecific antibodies is an attractive therapy concept for cancer patients. In potential, the biMAb OC/TR offers therapeutic perspectives, since the i.p. administration of this biMAb, in combination with pre-activated PBL, resulted in clinical responses. Toxicity of i.v. administered biMAb seems to be dose-dependent, with evident clinical symptoms at 0.2 and 1.0 mg (2 patients) and only sub-clinical changes in leucocyte differentiation counts at the 0.1-mg dose (3 patients). Based on our observations, the maximal tolerated dose after a 30-min i.v. infusion is 0.1 mg. Repeated injections with 0.1 mg or even lower doses of biMAb may circumvent the toxicity while still allowing sufficient tumour uptake. It is possible that i.p. administration may serve as a continuous systemic infusion of low doses of biMAb. Further studies are required to better understand the mechanisms involved in T-cell activation and the effects at the site of the tumour after systemic administration of the bispecific antibody OC/TR.

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