in paraffin-embedded sections was performed using standard techniques. P53 and bcl-2 were detected by primary monoclonal mouse anti-human antibodies in conjunction with red APAAP stain. Antigen-expression of 3 × 100 tumor cells per sample was evaluated by light-microscopy.

Results: In only 3 of 18 EOC (17%) and 1 of 6 ENC (17%) 10–90% of tumor cells were positively stained for p53. In contrast, 10–90% bcl-2-expressing tumor cells were found in 17 of 18 EOC (94%) and 5 of 6 ENC (83%). Samples with high expression of p53 (i. e. >50%) were also highly positive for bcl-2, whereas all bcl-2 negative specimens were found to carry wild-type p53.

Conclusion: In a substantial proportion of advanced EOC and ENC, bcl-2 appears to be activated by p53- independent pathways.

549 PUBLICATION

CA125 mimicry by SCFV-fragments of the monoclonal anti-idiotypic antibody ACA125 for immunotherapy of ovarian cancer

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Purpose: The $F(ab')_2$ fragment of the murine monoclonal anti-idiotypic antibody ACA125 mimicking the tumor associated antigen CA125 is used as a vaccine for the induction of an anti-tumoral immunity in patients with ovarian carcinoma. We tried to generate a single-chain fragment (ScFv) composed of ACA125 heavy and light chain variable domains.

Methods: Heavy and light chain genes of antibody producing mouse hybridoma cell line were separately amplified and assembled into a ScFv gene with linker DNA by PCR. The ScFv gene was ligated into the phagemid vector pCANTAB5E, which allows the production of both, phage displayed and soluble ScFv. Transformed *E. coli* TG1 cells were infected with M13K07 helper phage to yield recombinant phage, which display ScFv fragments as a g3p fusion protein on the surface of the filamentous phage M13. The *E. coli* non-supressor strain HB2151 was infected with an antigen-positive phage clone, previously screened by ELISA, to express soluble ScFv fragments.

Results: Functional soluble ScFv binding to the idiotypic antibody OC125 $F(ab')_2$ could be detected in the bacterial periplasm by Western blot and ELISA. The variable heavy and light chain genes of the ACA125 ScFv fragment were further sequenced and compared with known antibody sequences.

Conclusion: ScFv-Fragments of an anti-idiotypic antibody mimicking CA125 serves as the basis for a site-directed mutagenesis of the CDR-Regions in order to improve the immunological reactivity of the anti-idiotype vaccine.

550 PUBLICATION

The value of neoadjuvant chemotherapy (NACT) after surgical-pathologic staging (SPS) vs. failed debulking (FD) for inoperable ovarian cancer (IOC) and subsequent interventional laparotomy (ILAP)

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Purpose: The role of a NACT for IOC (extensive peritoneal carcinomatosis and/or residuals (R) > 2 cm) after SPS and FD, respectively, was evaluated concerning remission, operability at ILAP und overall survival (OAS):

Methods: In a phase II multicenter study 64 patients with IOC after SPS or FD received a dose intensified NACT (Cisplatin 100 mg/m2/Treosulfan 5.000 mg/m2 i.v. + GCSF every 3 weeks, 3 cycles) with subsequent ILAP.

Results: To date, an ILAP was performed in 5 women with a FIGO-stage III (n = 24; 69%) und IV (n = 11; 31%) disease. A NACT was administered after SPS in 22 cases and after FD in 15. An R0 resection was possible in 53% of patients after SPS and in 27% after FD (p = 0.008). Remission (PR+CR) rates were 77% in the SPS and 76% in the FD group. The rate of PD und NC was comparable in the two groups.

Conclusion: Low toxicity of NACT (no WHO toxicity grade IV), good remission rates (76%) and operable findings at ILAP with the feasibility of optimal cytoreduction (R_0 43% and R_1 14%%) were achieved. The data also show a possible benefit in OAS in the SPS group. Final results are presently being evaluated.

551 PUBLICATION

High dose cyclophosphamide (C) and epirubicin (E) followed by cisplatin (P) and 5-fluoro-uracile (F) in patients with advanced epithelial ovarian adenocarcinoma (AEOC)

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There are data supporting a dose-response relationship for alkylating agents in OC. High-dose P is neurotoxic but high-dose C has not been explored in combination chemotherapy with Anthracyclins in AEOC. We have studied since 1992 a densified regimen with 4 courses at 2 wks interval of C (1200 mg/m²) and E (75 mg/m²), followed by 4 courses at 3 wks interval of P (75 mg/m²) and F (1 g/m²/d × 3 by protracted infusion). 29 stage IIIC–IV pts, with a median age of 53 y (range 35–73) were included. Other eligibility criteria were PS: 0–2, no cardiac nor renal contra indication to E or P. 55% of pts had >2 cm residual implants after primary surgery. CE courses were administered whatever the WBC count, without hematopoietic growth factor, but were delayed if febrile neutropenia. Toxicities differed in the 2 parts of treatment. Grade 4 neutropenia was more prominent after CE (70%), but neutropenic fever was unfrequent (14%). Grade 2–3 anemia and Grade 3 emesis were more frequent after FP (39 and 46% respectively). Clinical CR was achieved in 72% of pts. Among 19 pts with objective response assessed by second look laparotomy, 9 obtained pathological CR (47%). After a median follow up of 25 months, 17 pts have progressed and 12 died. Median progression free and overall survival were 13 and 28 months, respectively. These results are in the same range than those of C-P regimens, despite a lesser cumulative dose of P.

552 PUBLICATION

Stimulation of cytotoxicity and cytokine production in tumor – Associated macrophages from ovarian, breast and lung cancer patients

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Purpose: The role of tumor-associated macrophages (TAM) as potential effector cells for eradicating malignant cells is not vet entirely clarified. In the present study TAM were phenotypically and functionally characterized. The second part of our investigation was the activation of TAM by cytokines rhGM-CSF, rhIFN-γ or with the polyenzyme preparation Wobe-Mugos used in the treatment of cancer patients with malignant effusions.

Methods: TAM were isolated from malignant effusions of breast, ovarian and lung cancer patients by gradient separation and characterized, by the following parameters: surface epitopes (moAb 27E10, 25F9), respiratory burst activity, cytotoxicity and TGF-β production measured in culture supernatants by ELISA/RIA. Additionally mRNA of TGF-β was detected in TAM by in situ hybridization.

Results: Incubation of TAM with GM-CSF, IFN-γ and polyenzyme preparation Wobe-Mugos resulted in an augmentation of cytotoxicity. In contrast, GM-CSF, IFN-γ and Wobe-Mugos reduced the production of TGF-β by TAM, as verified by ELISA assay as well as by in situ hybridization. TGF-β is known as an immunosuppressive molecule.

Conclusion: Our studies show that the cytotoxic capacity of TAM obtained from malignant effusions of cancer patients can be increased by GM-CSF, IFN-γ and Wobe-Mugos. TGF-β release was reduced. Whether this observation is of therapeutical relevance has to be determined by further studies.

553 PUBLICATION

The integration of consolidation whole abdominal irradiation in the multi-modality management of advanced ovarian carcinoma: Long-term results

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57 patients (pts) with advanced ovarian carcinoma were treated by primary cytoreductive surgery followed by 6–11 courses of cisplatin (50 mg/m²) and adriamycin (50 mg/m²), second look laparotomy (SLL) (46 pts) and consolidation whole abdominal irradiation ± pelvic boost. Only 42/57 pts (74%) tolerated radiotherapy (RT) and were able to receive the full planned whole abdominal RT dose of 30 Gy. The median follow-up time is 39 months (range, 12–197 months). The following 5 and 10 year actuarial