An antibody-conjugated nanoparticulate biosensor for the in-vivo detection of circulating tumour cells in human blood system

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Background and Methods
Currently existing techniques for the isolation of circulating tumor cells in blood (CTC) have experimental approaches. Nanoparticles as biosensors could allow a refinement of the method and possible clinical applicability of the in vivo CTC-isolation in cancer patients. The Nanodetector consists of nano-gold particles loaded on a medical wire (GILUPI Inc., Potsdam). This gold covered medical wire with hydrogel surface allows a specific covalent binding of the anti-EpCAM-AK (epithelial cell adhesion molecule [EpCAM]) (Figures 1-2). The wire is inserted in the patient’s vena cubitalis for thirty minutes (Figures 3-4). CTC identification criteria are cytokeratin positive (CK), CD45 negative (CD45−) and Phenol, 4-[5-(4-methyl-1-piperazinyl)[2,5'-bi-1H-benzimidazol]-2'-yl]-triethylamine (nuclear stain) positive (Hoechst 33258`).

Results
In the first preclinical step, blood samples from 47 prostate cancer patients of different stages and 12 control patients (BPH) were studied ex vivo. These blood samples were used in the fluid dynamic system (Fig. 5). The characterization of isolated cells was carried out on the RT-PCR with regard to the marker PSMA, PSA, EGFR and cytochemical level on the detection of EpCAM expression and the control CD45-staining. We isolated an average of 32.92 CTC/7.5ml in blood samples of hrPCA patients and 3.41 CTC/7.5ml in blood samples of locally limited PCA patients (Table 1). Currently, the clinical application of an anti-EpCAM antibody conjugated nano-scaled detector (ND) for the in vivo isolation of CTCs from peripheral blood of patients with prostate cancer is being tested at our clinic. The first in-vivo use of this system for isolation of CTCs in the blood circulation (vena cubitalis, 30 minutes) show very promising results (Figures 6, 7, 8, 9).

Conclusions
Isolation and enumeration of CTCs from the blood of prostate cancer patients of different stages using an EPCAM-conjugated Nano-detector is possible with high efficiency. The clinical performance of this system in in settings of primary cancer diagnosis as well as therapeutic monitoring and prediction of prognosis continues to be of great interest and is currently being evaluated in this ongoing clinical trial.