Prostate cancer – CTC isolation with the GILUPI CellCollector®

Prostate cancer is the 2nd most common cancer in men with worldwide around 1.3 million new cases per year. With a wide set of treatment options available, the stratification of patients likely to benefit from a specific therapy is crucial. The isolation and characterization of circulating tumor cells (CTCs) is a promising tool to enable this personalized approach. This overview highlights an excerpt from the data obtained in more than 700 applications of the GILUPI CellCollector® (DC01).

CTC detection and clinical relevance

An EU-wide multicenter study aimed to analyze the incidence and clinical relevance of CTCs in non-metastatic prostate cancer patients. This study included a direct method comparison between different CTC isolation technologies. A meta-analysis of the published data regarding CTC positivity and CTC counts obtained with the CELLSEARCH® system and the GILUPI CellCollector® is shown in Figure 1. Furthermore, this study described a correlation of CTCs to clinical risk factors, suggesting a potential clinical relevance of CTCs as marker for minimal residual disease in high-risk prostate cancer.

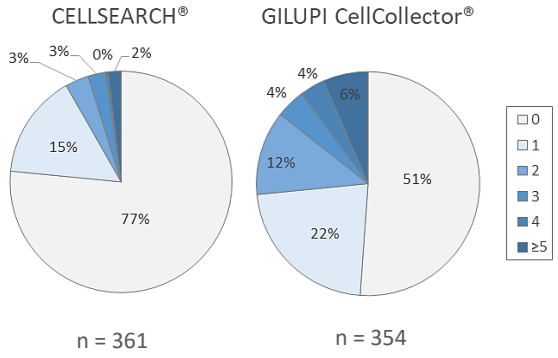

Figure 1: Comparison of CTC detection in non-metastatic high-risk prostate cancer patients with the CELLSEARCH® system and the GILUPI CellCollector®.

In addition, an *ex vivo* proof-of-concept study by Theil *et al.* found a significant difference in the overall survival between patients with ≤ 5 or >5 CTCs detected with the GILUPI CellCollector®.

CTC-based RNA profiling

Patient stratification based on mRNA analysis is one of the potential applications of CTCs. Multiplex gene expression profiling of CTCs isolated from non-metastatic high-risk prostate cancer patients and healthy donors was achieved with TRIzol™-mediated RNA isolation directly on the DC01 and subsequent RT-qPCR. Markou *et al.* observed heterogeneous gene expression in the isolated CTCs, but at least one of the 14 analyzed transcripts was increased in the majority of patients (71%, see Fig. 2A), which was not the case in the healthy control group. Moreover, it was observed that the molecular profile of the CTCs changed after treatment (Fig. 2B).

A

14 gene signature	High-risk PCa patients	Healthy volunteers
≥ 1 gene	71 %	22 %
≥ 2 genes	41 %	0 %
≥ 3 genes	15 %	0 %

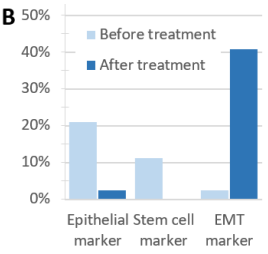

Figure 2: RNA profiling of CTCs from non-metastatic high-risk prostate cancer patients. (A) Percentage of patients expressing one or more of the 14 genes in their CTCs (PCa: n=105, Healthy: n=36). (B) Marker expression before and after treatment.

In situ padlock probe assay for AR-V7 detection

Detection of the androgen receptor splice variant 7 (AR-V7) in CTCs is currently a promising biomarker in castration resistant prostate cancer (CRPC), as it is associated with resistance to enzalutamide and abiraterone. El Heliebi *et al.* developed an *in situ* padlock probe assay that allows the quantitative analysis of AR-V7 expression on the single cell level. The conducted proof-of-concept study isolated CTCs from 62% of prostate cancer patients and detected therapeutically relevant AR-V7 in the majority of these cells (Figure 3).

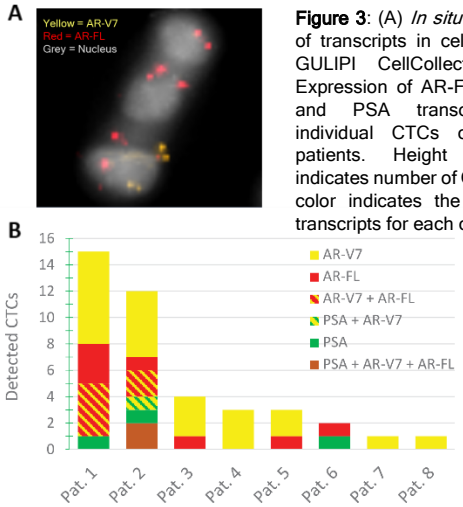

Figure 3: (A) *In situ* detection of transcripts in cells on the GULIPI CellCollector®. (B) Expression of AR-FL, AR-V7 and PSA transcripts in individual CTCs of CRPC patients. Height of bar indicates number of CTCs, the color indicates the detected transcripts for each cell.

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