BACKGROUND

• Changes in ctDNA levels during the first 6 weeks and immune-related gene expression signatures (GES) can predict outcomes in patients treated with anti-PD-1 therapy.
• PREDICT-ID is a GES identified from a large pan-cancer GE meta-analysis which has been shown to predict response and survival to immune checkpoint blockade better than other biomarkers.
• The earliest predictive time point in ctDNA kinetics for benefit to this therapy is unknown.

METHODS

• Sixteen patients with recurrent/metastatic head and neck squamous cell carcinoma treated with pembrolizumab in first line (N=11) or nivolumab in second line (N=5) were prospectively enrolled.
• Whole exome sequencing (WES) and RNA-seq were performed in parallel in an archival tumor.
• ctDNA was analyzed using a bespoke 16plex assay (Signatera) based on matched WES.
• RNA-Seq was aligned to hg39. Differential Gene Expression Analysis was performed with DESeq2.
• Gene Set Enrichment Analysis included different transcription factors, molecular/immune pathways and PREDICT-ID signature.

RESULTS

Figure 2. ctDNA kinetics in the 15 evaluable patients (one was screening failure as there was not enough tissue for WES). Despite ctDNA at baseline (BS) was quantified and treated in 12 patients samples in day 1 was mixed and day 3 in patient 1 did not pass QG. Patients highlighted with red square (13,14,15) have a similar pattern showing a continuous decline in ctDNA beyond day 2 (↓ΔctDNA). The remaining patients showed a tendency to increase or decrease in ctDNA from day 8 onwards (↑ΔctDNA). Median follow up is 18.5 months.

Table 1. Main characteristics of patients evaluable for ctDNA analysis and correlation with baseline ctDNA. The mean number of ctDNA molecules [MTD]/ml (MCTD) was 1.0. One patient was not evaluable due to not enough tissue for WES. There is no association between baseline ctDNA concentrations and response or any clinical characteristics.

Figure 3. Correlation between increase/stability vs decrease and progression free survival. Overall survival data are not mature and may explain the lack of differences in this endpoint. A decrease in ctDNA as early as day 8 post-treatment is predictive for longer PFS. Similar findings, are observed for clinical benefit (not shown).

Figure 4. Transcriptional changes analyzing different hallmarks in cancer. Patients with ↓ΔctDNA pattern have upregulation of the related pathways while those with ↑ΔctDNA pattern are enriched in oncogenic and mitotic spindle pathways.

Figure 5. PREDICT-ID transcriptomic signature is associated in our cohort with PFS but not with OS. Patients with PREDICT-ID High expression showed a larger PFS compared to those who are low. Overall survival data is still not mature and this may justify that we are not seeing any differences.

Figure 6. ctDNA kinetics may discriminate treatment outcomes in patients with PREDICT-ID High signature. All patients with PREDICT-ID Low have a ↑ΔctDNA pattern. Among those with PREDICT-ID High, 4 have ↑ΔctDNA and 4 have ↓ΔctDNA. There is a near statistically significant difference in PFS and OS. There is a significantly improved prognosis in these patients with PREDICT-ID high and ↓ΔctDNA compared to PREDICT-ID low. Further validation in larger cohorts is needed.

CONCLUSIONS

• A decrease in ctDNA by day 8 of anti-PD1 therapy is associated with clinical benefit and longer PFS.
• ctDNA kinetics may refine predictive value in PREDICT-ID High signature.
• Validations in larger pan-cancer cohorts are needed.