

Ultra-sensitive, tumor-informed ctDNA profiling in pembrolizumab-treated gastroesophageal cancer patients reveals longitudinal ctDNA kinetics



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BACKGROUND

Metastatic esophagogastric cancer (mEGC) is a lethal disease with poor long-term survival. Recent studies have established anti-PD-1 therapy in combination with chemotherapy as the standard of care for first-line therapy for mEGC. KeyLargo (NCT03342937 [1]) was a single-arm phase II study of pembrolizumab in combination with oxaliplatin and capecitabine in the first-line treatment of patients with HER2 negative mEGC. While high response rates were noted, not all patients received benefits, emphasizing the need for better biomarkers. Paired tumor biopsies and plasma were longitudinally collected, processed, and stored for optimal biomarker testing. In this retrospective study, we employed a novel, tumor-informed ctDNA approach for longitudinal disease monitoring and dynamic tumor evolution.

OBJECTIVES

Investigate the application of ultra-sensitive ctDNA assay and explore tumor molecule PPM as a predictive biomarker for the clinical benefit of anti-PDL-1 immunotherapy in combination with oxaliplatin and capecitabine in the first-line treatment of patients with HER2-negative mEGC.

METHODS

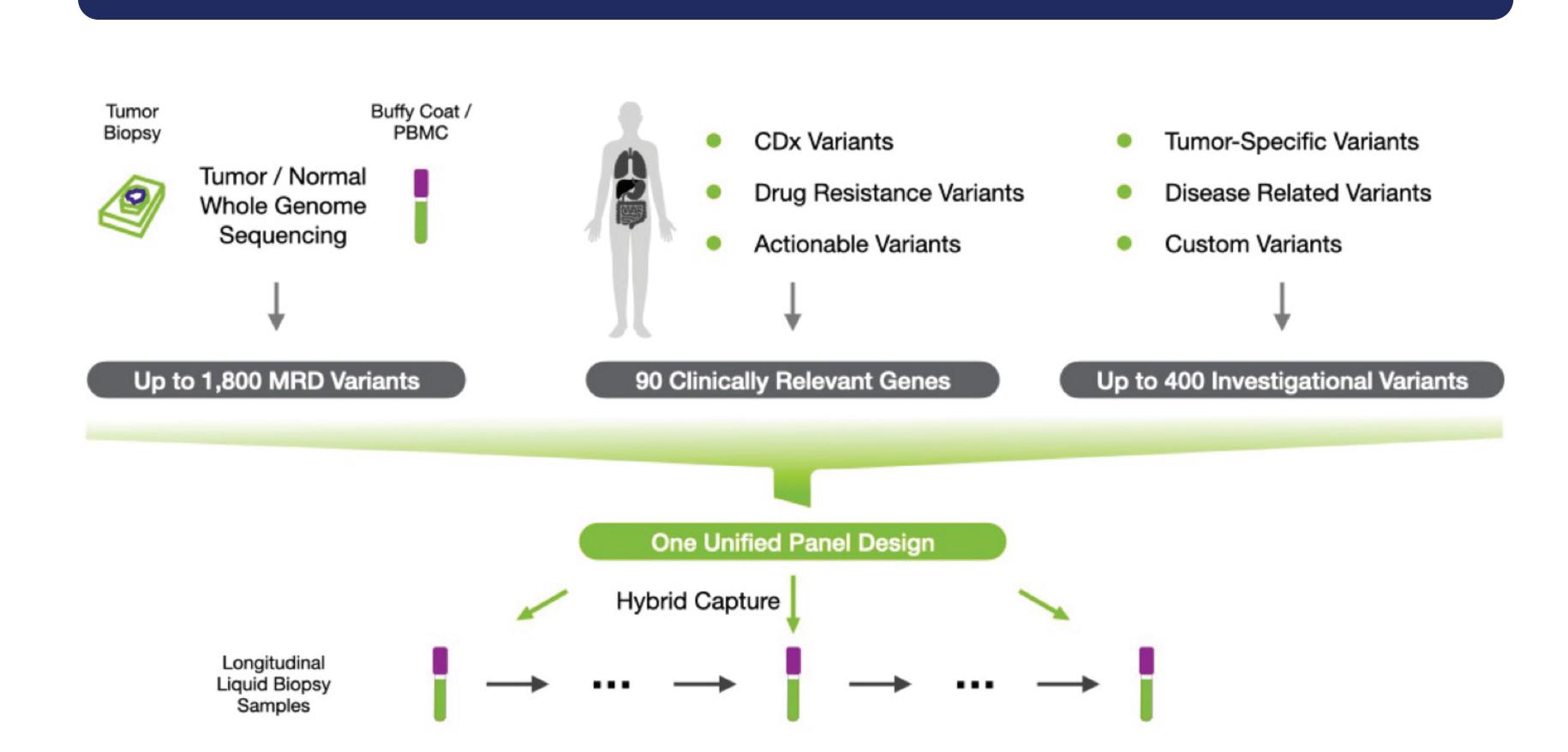


Figure 1. NeXT Personal® workflow

A total of 116 plasma samples from 18 patients have been retrospectively evaluated at baseline and during treatment. An ultra-sensitive, tumor-informed ctDNA approach was used to assess molecular/minimal residual disease (MRD) and evaluate tumor evolution. NeXT Personal® uses whole-genome sequencing of both tumor and normal samples to generate a personalized liquid biopsy panel for each patient consisting of up to 1,800 selected somatic variants, enabling ultra-sensitive detection down to 1-3 parts per million (PPM). Each bespoke panel also includes a tumor-agnostic set of 90 clinically relevant genes. NeXT Personal results were analyzed along with imaging assessments and clinical outcomes for each patient. Results are preliminary and descriptive in nature.

PATIENT CHARACTERISTICS

Thirty-six patients were enrolled between January 2018 and January 2020. Of 34 evaluable patients, 25 patients achieved a response (ORR= 74%), including 6 patients with a complete response (CR) and 19 patients with a partial response (PR). This interim analysis includes baseline tumor and plasma samples collected from 18 pts with 98 corresponding on-treatment (OT; up to cycle 35) plasma samples.

Patients, n (%)
60
30 (83)
6 (17)
10 (28)
19 (53)
7 (19)
29 (81)
16 (44)
9 (25)
4 (11)
4 (11)
11 (31)
24 (67)

Table 1. KeyLargo patient characteristics

Ultra-sensitive ctDNA detection

NeXT Personal detects a large spectrum of ctDNA-positive events, allowing for maximal sensitivity for low tumor fractions. The dynamic range of ctDNA-positive samples varied from 5.3 to 406,067 tumor molecules PPM (Figure 2). Baseline (red) and C1D8 (black) time points exhibited PPM than observed on-treatment time points. A significant number of ctDNA-positive detections were observed at low PPM levels, with 30% (20/65) of

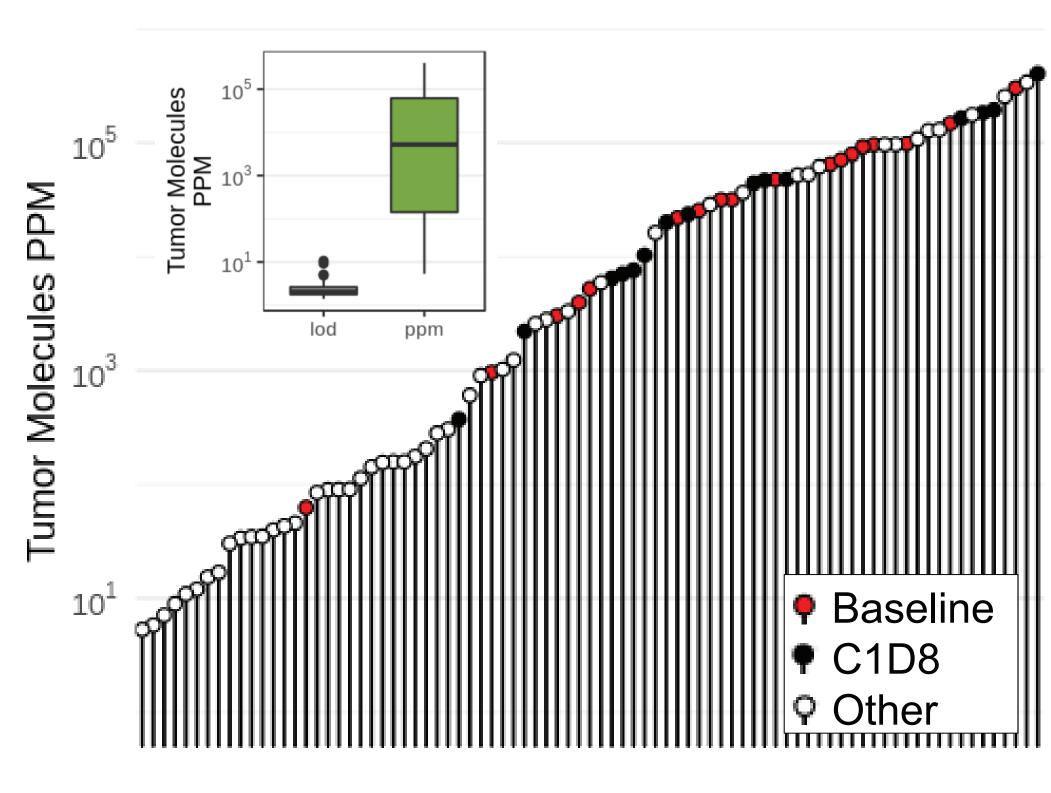


Figure 2. Tumor molecule dynamic range

positive, on-treatment detections occurring below 100 PPM and 23% (15/65) occurring below 50 PPM. Of the 36 pts who enrolled on KeyLargo, 32 pts had baseline tumor and longitudinal plasma samples available for testing. In this initial cohort of 18 pts, 18/18 (100%) were ctDNA-positive at baseline and after the first 30 days of treatment, with a median limit of detection of 2.05 (IQR=0.82).

RESULTS

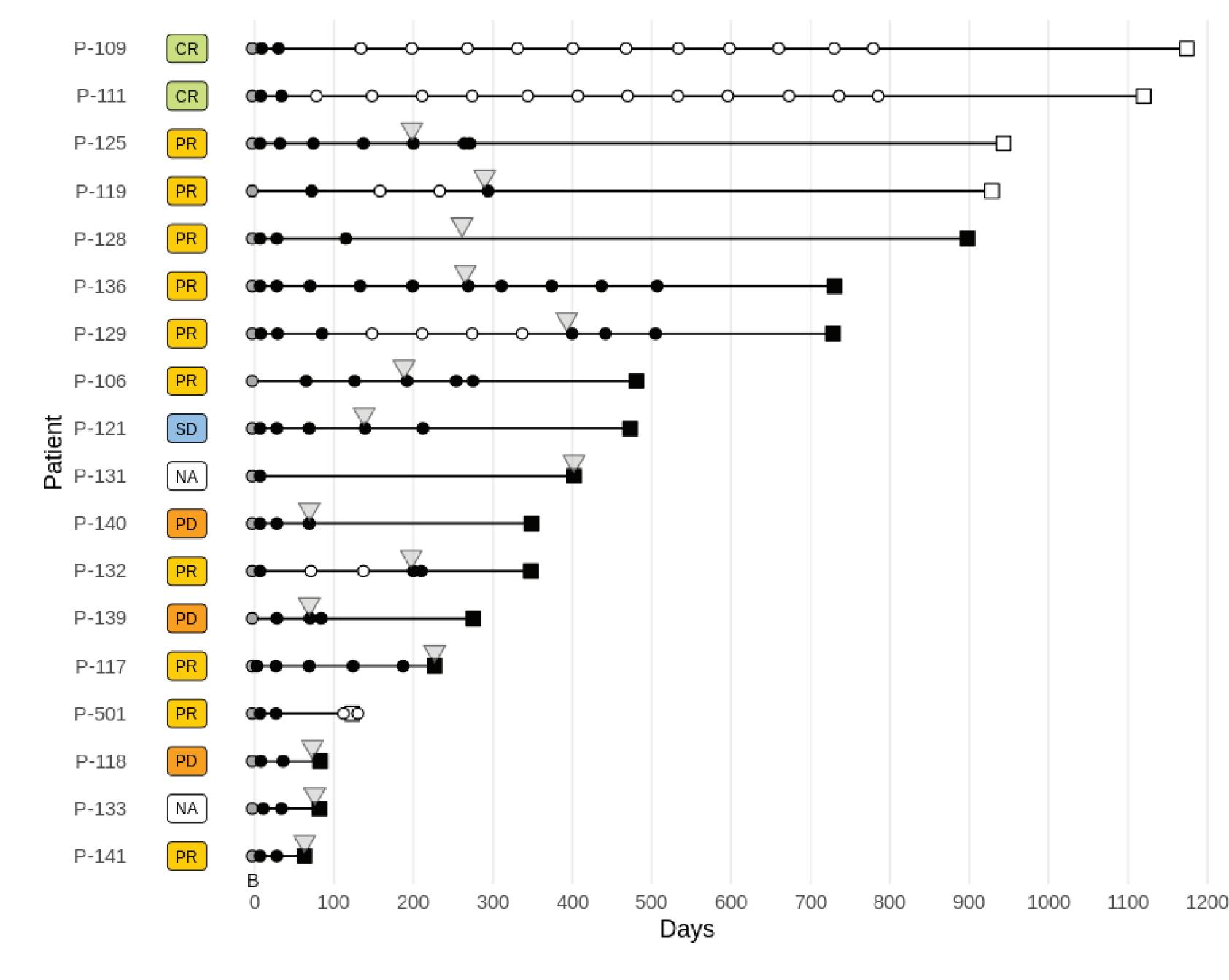


Figure 3. Longitudinal ctDNA detection

Two patients were CR, each with 21/21 (100%) ctDNA-negative plasma samples over approximately two years. One patient was stable disease (SD), and 3 patients were PD; all SD and PD patients were ctDNA-positive across all assessed time points (10/10; 100%) (Figures 3 & 4). For all patients ctDNA was detected before or at clinical progression.

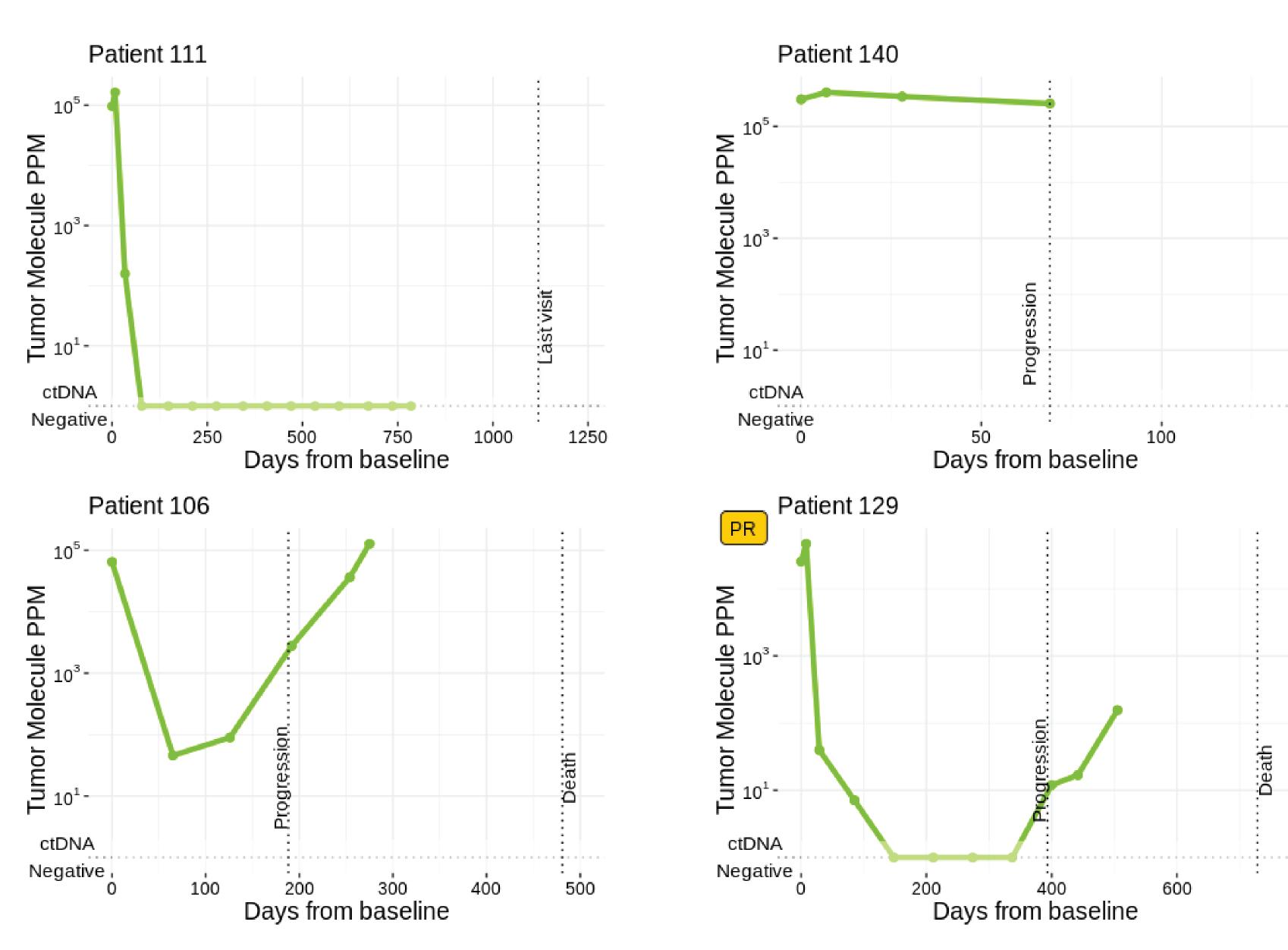


Figure 4. Individual patient profiles

RESULTS

Evolution of clinically actionable variants & tumor-specific variants under therapeutic pressure

Tumor-specific variants and clinically actionable variants for known drivers of therapeutic resistance were evaluated during treatment. This analysis shows the dynamic variant allele frequency changes in patient 106. TP53, CNN1, and APC mutations were identified early in treatment, while two mutations, RAF1 and PIK3CA, emerged near progression (Figure 5).

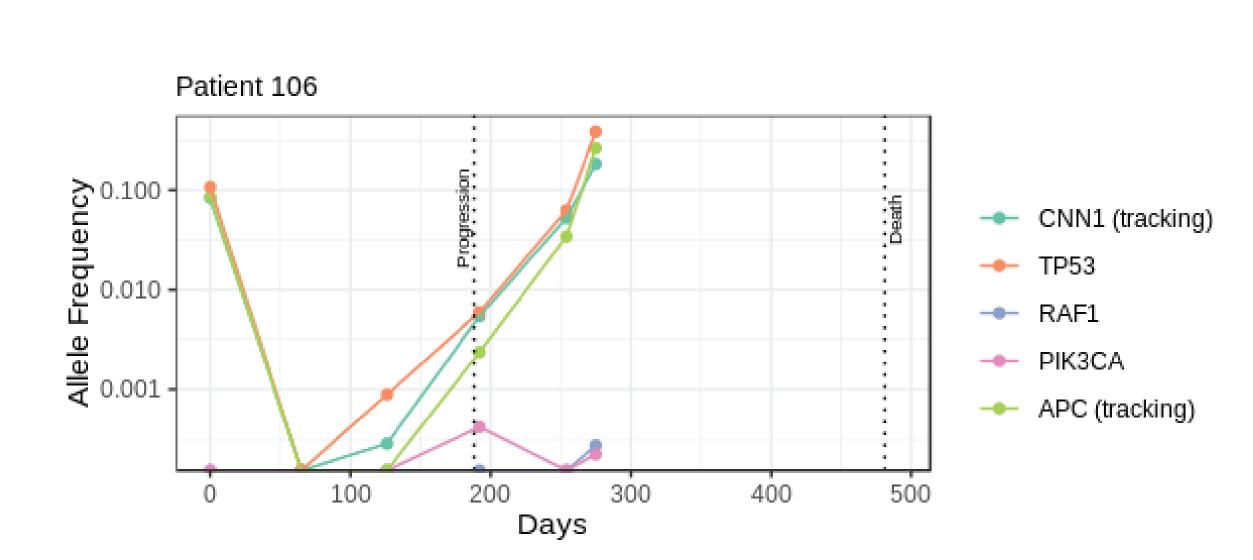


Figure 5. Patient-specific mutational evolution

ctDNA clearance predicts survival

OT samples were collected every three cycles (9 weeks). ctDNA clearance at any time point after 30 days correlated with progression-free survival (PFS; p=0.018, log-rank; Figure 6). Moreover, the ratio of tumor molecules between baseline and first on-treatment (rPPM) was significantly reduced in pts having a best overall response of PR/CR (98% mean rPPM) versus progressive disease (45% mean rPPM, p<0.007, Wilcox-test).

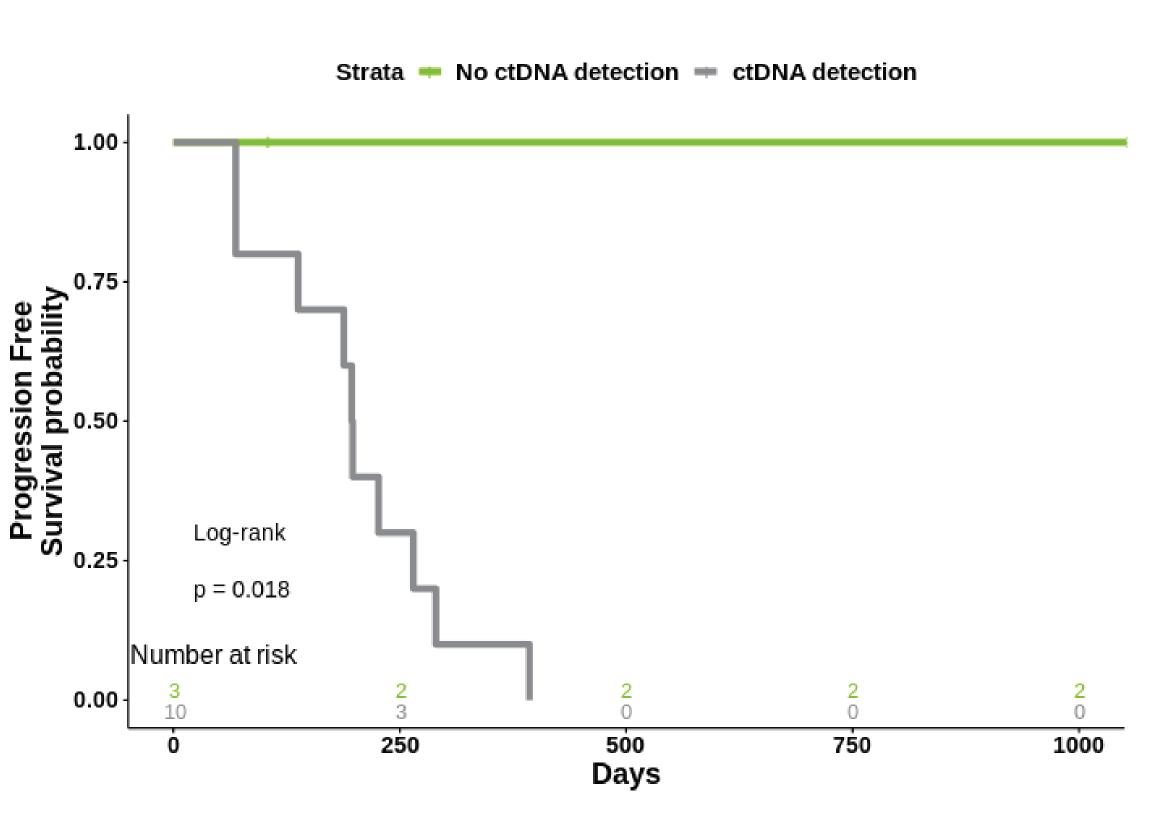


Figure 6. ctDNA clearance predicts PFS

CONCLUSIONS

- Low ctDNA levels are observed in late-stage disease, with positive ctDNA detections occurring as low as 5.3 PPM in this study.
- Detection down to low PPM levels correlates with the patients' best response.
- Tumor molecule PPM dynamics may be associated with patient response to immunotherapy.
- Novel, ultra-sensitive ctDNA detection provided accurate MRD information throughout treatment.
- Results suggest the clinical utility of tracking clinically actionable variants arising or changing in response to therapeutic pressure.
- Further analysis of this patient set is ongoing.

REFERENCES

¹KEYlargo: A phase II study of first-line pembrolizumab, capecitabine, and oxaliplatin in HER2-negative gastroesophageal adenocarcinoma. (https://ascopubs.org/doi/abs/10.1200/JCO.2021.39.3_suppl.228) Uronis *et al.* Journal of Clinical Oncology 2021 39:3_suppl, 228-228