Background The most important insights into mechanisms of tumour rejection, and how these could be exploited therapeutically, is likely to come from patients displaying the best responses. Those individuals with complete and sustained anti-tumour responses, without maintenance therapy, will provide the best evidence of genuine disease modification. Despite much speculation as to how these exceptional responses are generated there is increasing evidence for an immune basis which may be influenced by the tumour microbiome underlying the mechanism for sustained tumour rejection.

Methods To focus on individuals with complete and sustained metastatic disease resolution, we designed a pilot study, the Continuum Long-Term Survivor study, to evaluate patients with the best outcomes, where disease modification may have occurred. The study targeted only those patients (n=50) with a sustained (>5 years) complete clearance of metastatic cancers, without requiring maintenance therapy. Matched controls comprised patients unable to generate an initial response, or those who relapsed within 12 months. DNA extracted from tumour samples was analyzed by 16S BENCHMARK™ microbial amplicon sequencing (Diversigen) to profile the tumour microbiome, whilst the microbial composition of stool samples was determined using BoosterShot Shotgun Sequencing. In parallel mRNA expression from the tumour tissue was evaluated using NanoString’s PanCancer IO360 gene expression panel. From a homogeneous subgroup of bowel cancer long-term survivors and their matched controls, multiplex IHC using a panel of 8 immune markers to identify tertiary lymphoid structures (TLS) was performed on the tumour tissues and imaged using a PhenoImager (AkoyaBiosciences).

Results Results will be presented comparing tumour and gut-derived microbial species diversity between long-term vs. short-term survivor matched controls using alpha diversity estimates as well as differential abundance analysis. This will define the microbial species that are more likely to be associated with long-term survivorship. Characterization of the immune gene transcriptional patterns within the tumour microenvironment (TME) from long-term survivors vs controls will also be reported along with any observed differences from the multiplex IHC analysis of immune infiltrates in corresponding patient tissue. To explore any connections of the tumour-microbiome on the TME, an integrated analysis of the microbiome and tumour immune transcriptome will be presented.

Conclusions This study will reveal whether changes in TME influenced by the tumour microbiota may be important factors associated with long-term survivorship. Understanding the precise mechanisms of total tumour rejection in patients, and their evaluation may be game-changing in terms of design of new molecular, biological and immune therapies.

Ethics Approval This study was approved by the University of Surrey Ethics Board; approval number 266581.