

Supplementary material

Detailed next-generation sequencing (NGS) methodology. NGS was performed on tumor tissue using a 41-gene panel (catalogue no. LK105; Guangzhou Burning Rock Medical Laboratory Co., Ltd.) on the Illumina NovaSeq 6000 platform (Illumina, Inc.).

Raw data processing and quality assessment. Following completion of sequencing, the overall sequencing run was evaluated based on Q30, data yield and platform-specific performance metrics. Q30 refers to the proportion of bases with a base-calling error rate of $\leq 0.1\%$. Cluster density was also assessed.

Adapter sequences were removed and low-quality reads were filtered from the raw FASTQ files using one-loop-trimmer v1.1. Low-quality bases with a Phred quality score < 20 at the 5' end of reads were removed. A 30-bp sliding window was applied; when the mean Phred quality score within the window was < 25 , trimming was initiated, and reads with a length of < 50 bp were discarded.

BAM files generated from the initial alignment were processed using the GATK MarkDuplicates v4.0 module to identify and remove PCR duplicate reads.

Alignment and variant calling. Clean reads were aligned to the human reference genome GRCh37/hg19 using BWA-MEM v0.7.10. SAM/BAM files were generated and indexed using Sambamba v0.8.0. Local realignment in challenging regions was performed using ABRA2 v2.22. Single-nucleotide variants (SNVs) and insertions and deletions (INDELs) were called using VarDict v1.5.1, with falsepositive filtering performed by VarScan v2.4.3. Copy number variation (CNV) and structural variation (SV) calling were performed using BRCNV v4.3.0 and markSV v0.3.1, respectively. Variant annotation was performed using BrasAnnotation v4.3.0. The analytical tools used are summarized in Table SI.

Quality control (QC). Using BRQC or the projectspecific QC pipeline, the following quality metrics were calculated for the tumoronly sample: Mean effective sequencing depth (unique depth), coverage uniformity, Q30 percentage, mean sequencing depth (mean depth) and mapping ratio. The acceptance thresholds for the tumoronly branch are shown in Table SII. Only samples meeting these criteria were advanced to variant analysis.

Figure S1. Representative contrast-enhanced CT images of the metastatic lymph node before and after four cycles of FOLFIRI (5-fluorouracil, leucovorin and irinotecan) plus bevacizumab. (A) Axial CT image obtained in October 2021, showing an enlarged metastatic lymph node (maximum diameter, 21 mm; white arrow). (B) Oblique coronal CT image obtained in October 2021, showing the metastatic lesion involving the mid-ureter (white arrow indicates the metastatic lesion, with the ureter visible above it). (C) Axial CT image obtained in October 2021, showing mild dilation of the right renal pelvis. (D) Axial CT image obtained in January 2022, showing enlargement of the metastatic lymph node (maximum diameter, 32 mm; white arrow), consistent with progressive disease.

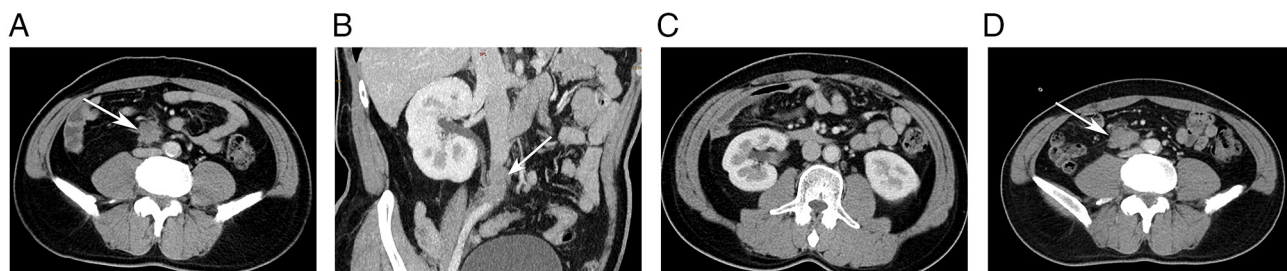


Figure S2. Representative contrast-enhanced CT images showing changes in omental thickening during the course of the ureteral fistula. (A) CT image obtained in June 2022, showing omental thickening (white arrow). (B) CT image obtained in September 2022, showing persistent omental thickening (white arrow). (C) CT image obtained in March 2023, showing resolution of omental fibrous density after improvement of the ureteral fistula. (D) CT image obtained in March 2023, showing the same resolution on a different slice.

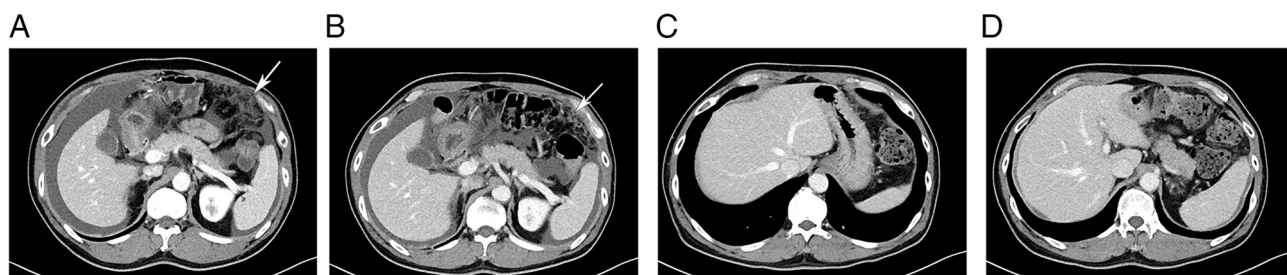


Figure S3. PET-CT findings in June 2024. (A) Axial CT image showing a small residual cystic lesion in the region of the ureteral fistula (white arrow). (B) Corresponding axial PET image showing focal uptake within the fistula sac (standardized uptake value, 145.5), consistent with urine accumulation. (C) Maximum-intensity projection PET image showing no other active lesions and no ascites. PET, positron emission tomography.

