Supplementary Table 1. **Rationale for the selection or omission of computational polyTE detection tools for this benchmark study and their relevance to human next-generation sequencing (NGS) data**. Extensive benchmarking was done on seven tools that were selected based on the criteria adopted in this study (see "Polymorphic TE detection tools" section). Additionally, four more previously not included polyTE detection tools were tested on the low coverage dataset. Other existing polyTE detection tools that were omitted from the benchmark are also listed along with the rationale of their omission. Briefly, tools that are not specialized for polyTE detection or requires specific TSDs were not included in the benchmark.

<table>
<tr><th colspan="4">PolyTE detection tools selected for benchmarking</th></tr>
<tr><th>Tool name</th><th>Rationale for selection</th><th>Tool's success</th><th>Relevance to human NGS data</th></tr>
<tr><td>MELT</td><td>All criteria</td><td>Success</td><td>High</td></tr>
<tr><td>Mobster</td><td>All criteria</td><td>Success</td><td>High</td></tr>
<tr><td>RetroSeq</td><td>All criteria</td><td>Success</td><td>High</td></tr>
<tr><td>Tangram</td><td>All criteria</td><td>Failure</td><td>High</td></tr>
<tr><td>TEMP</td><td>All criteria</td><td>Success</td><td>High</td></tr>
<tr><td>ITIS</td><td>Criterion #1 and #4</td><td>Success</td><td>Medium</td></tr>
<tr><td>T-lex/T-lex2</td><td>Criterion #1 and #4</td><td>Aborted</td><td>Medium</td></tr>
<tr><td>DD_DETECTION</td><td>Expanded set</td><td>Failure</td><td>High</td></tr>
<tr><td>Jitterbug</td><td>Expanded set</td><td>Failure</td><td>High</td></tr>
<tr><td>TE-Locate</td><td>Expanded set</td><td>Failure</td><td>Medium</td></tr>
<tr><td>TE-Tracker</td><td>Expanded set</td><td>Failure</td><td>Medium</td></tr>
<tr><th colspan="4">PolyTE detection tools omitted from the benchmarking</th></tr>
<tr><th>Tool name</th><th colspan="2">Rationale for omission</th><th>Relevance to human NGS data</th></tr>
<tr><td>GRIPper</td><td colspan="2">Detects non-reference gene copy insertion</td><td>High</td></tr>
<tr><td>TIGRA</td><td colspan="2">Breakpoint assembler, not an SV caller</td><td>High</td></tr>
<tr><td>TranspoSeq</td><td colspan="2">Requires paired tumor/normal WGS data</td><td>High</td></tr>
<tr><td>Tea</td><td colspan="2">Requires paired tumor/normal WGS data</td><td>High</td></tr>
<tr><td>TraFiC</td><td colspan="2">Requires paired tumor/normal WGS data</td><td>High</td></tr>
<tr><td>VariationHunter</td><td colspan="2">General purpose SV detection tool</td><td>High</td></tr>
<tr><td>HYDRA-SV</td><td colspan="2">General purpose SV detection tool</td><td>High</td></tr>
<tr><td>MetaSV</td><td colspan="2">General purpose SV detection tool</td><td>High</td></tr>
<tr><td>ngs_te_mapper</td><td colspan="2">Requires TSDs to be provided</td><td>Medium</td></tr>
<tr><td>RelocaTE</td><td colspan="2">Requires TSDs to be provided</td><td>Low</td></tr>
</table>

Supplementary Table 2. **Summary of algorithmic differences between the computational polyTE detection tools benchmarked in this study**. More detailed differences are listed in Supp. Table 3.

	Read mapping				Breakpoint estimation			Filtering criteria				Output features		
Tool	All DP reads	Treats SR & DP independently	SR searched after DP	Mobilome Aligner	Fragment size distribution	SR dependent	Holistic	Read depth - flanking	Read depth - site	Known TEs	Mapping quality	VCF file	Predicts TSD	Predicts zygosity
MELT	✓	✗	✓	Bowtie2	?	✗	✓	✓	✓	✓	?	✓	✓	✓
Mobster	✓	✓	✗	MOSAIK	✓	✗	✓	✗	✓	✓	✗	✗	✗	✗
RetroSeq	✓	✗	✓	Exonerate	✗	✗	✓	✓	✓	✓	✗	✓	✗	✓
Tangram	✓	✓	✗	MOSAIK	✓	✗	✓	✗	✓	✓	✗	✓	✗	✗
TEMP	✓	✗	✓	BWA	✓	✓	✗	✗	✓	✗	✗	✗	✗	NA
ITIS	✗	✓	✗	BWA	✗	✓	✗	✓	✓	✓	✓	✗	✗	✗

Supplementary Table 3. **Detailed Algorithmic differences between the computational polyTE detection tools benchmarked in this study**.

Tool	DP definition	SR definition	DP and SR search paradigm	Mobilome alignment tool	Cluster definition	Merging clusters	Breakpoint estimation	Filtering criteria
MELT	Information not available	Information not available	DPs were used to identify potential/ candidate TE sites SRs were used to identify breakpoints and TSDs	Bowtie2 with default parameters	Sites with at least 4 DP anchors clustered within 500bp of each other	Merges all DP and SR clusters from all BAM files (from 1KG project)	Unspecified type of model was built containing all available information for the candidate site. This model was then used to predict precise insertion site, strand, TSD, insertion sequence and length.	Based on: 1) minimum 4 supporting DPs 2) proximity to a reference TE 3) filter sites with depth of coverage outside 70-130% of the 100bp flanking region
Mobster	1) Orientation different from expectation or 2) Distance between pairs significantly different or 3) Reads mapping to different chromosome or 4) One read mapped, other not DP will have at least one uniquely mapping read referred to as the anchor read	Reads that map partially (clipped); will have one uniquely mapping anchor read and uniquely mapping unclipped part (anchor for SR)	DP and SR are searched independently Anchor reads tagged as unmapped or by the TE family their mate/clipping maps to	MOSAIK (hash size = 9; max mismatches = 10%, min length = 20 bp)	DP clusters 1) Anchors map to same strand 2) support the same TE family 3) have start position in proximity to each other SR clusters 1) Anchors belong to the same TE family or polyA/T stretch 2) same side clipping 3) clipped within a few bp of each other	Merge same family (or homopolymer) forward and reverse strand clusters First merge DP and SR independently, then proceed to merge the two Confidence assigned based on the number of clusters and orientations (5' and 3') that were merged	Breakpoints are estimated based on the inner borders of 5' and 3'clipped If clipped reads not available, inner borders of DP clusters are used for breakpoint estimation Else, estimated from insert size distribution and cluster length	Based on: 1) proximity (within 90bp) to a reference TE 2) user controlled read depth based filtering
RetroSeq	1) SAM flag 0x0002 unset, i.e., reads that are not proper pairs, or 2) One mate of the pair is unmapped Proper pairs are defined as reads whose pair maps within the expected distance	Partially mapped reads	Extracts DP in the beginning SR are only searched for breakpoint estimation step	Exonerate (80% min identity, 36 bp min length, mapping quality 30, local alignment with affine gap penalty, report best 5 results)	Forward and reverse orientation clusters created by the start position of the anchor reads Max gap 120bp between reads in a cluster	Uses bedtools window command to merge forward and reverse clusters	Excludes clusters with average read depth surrounding the cluster above a cutoff (def 200). Estimates using a set of parameters: 1) read depth of DP on both strands, 2) forward to reverse reads ratio at 5' and 3' of the putative breakpoint and 3) distance between last 5' and first 3' read.	Based on: 1) proximity (within 100bp of an Alu or within 200bp of an L1) to a reference TE Confidence for each genotype provided in the output VCF file

Supplementary Table 3. (Continued)

Tool	DP definition	SR definition	DP and SR search paradigm	Mobilome alignment tool	Cluster definition	Merging clusters	Breakpoint estimation	Filtering criteria
Tangram	Utilizes customized BAM format that contains both the genome and TE reference sequence alignment (No instruction provided on generating this alignment) DP are read pairs with one read mapping uniquely to the reference genome and the other mapping to the TE reference sequence	SR have one mate mapping uniquely while the other is either soft-clipped or unmapped The unaligned or soft-clipped reads are then realigned to both reference genome and reference TE sequences	DP and SR are searched independently	MOSAIK (Done before the process begins)	Clusters candidate read pairs using fragment center position; applies a customized nearest-neighbor algorithm for clustering Utilizes fragment length distribution Capable of handling multiple different libraries		DP – identifies pair of clusters spanning on the insertion from 3' and 5'. Leftmost position candidate insertion position SR – Performs fast local alignment to identify the breakpoint	Based on 1) supporting reads per insertion, 2) additional filtering if only DP support and 3) proximity to a reference TE
TEMP	One uniquely mapping read (anchor read) and the other read that maps to multiple distant locations or is unmappable	Reads that start in genomic sequence but are interrupted by transposon or non-contiguous genomic sequence Clipped portion which maps to the TE should be at least 7bp long	SR are looked for after DP DP identifies insertion regions, SR helps in breakpoint estimation	BWA-aln and BWA-sampe	Defines intervals such as they contain TE "junction in the beginning (and) at the end of the anchor read, and extending into the genome by the length of the average insert size" Reads supporting same TE type, same orientation and intervals that overlap by at least 1nt are clustered		Extends intervals in both directions to find overlapping SRs. Estimates breakpoint based on the clipped portion. In case of multiple locations, selects the one with highest support. When base estimate is not available, interval midpoint is taken as insertion position	Based on 1) Read depth
ITIS	One end mapped to the reference genome, other mapped to the TE	At least one end covers both reference and TE	DP and SR are searched independently but SR determines the genomic location	BWA	DP and SR that are in close proximity		Based on the SR	Based on 1) MAQ > 0 2) 2 < RD < 300 – around insertion 3) RD (DP/SR) > 2