Dynamic Duo

Agilent Avida target enrichment

Agilent Avida Targeted Profiling Workflows Designed to "Duo" More

For the first time, experience the power of DNA and methylation in one target enrichment workflow. The Agilent Avida Duo workflow enables both DNA and methylation profiling of a single sample, with no sample splitting required. The novel, high-performance workflow offers multiomics capabilities and outstanding fidelity, turnaround time, and ease of use. Unleash the full potential of every sample with the Agilent Avida workflow—designed to "duo" more.

Assessing the genetic and epigenetic changes in liquid and tissue biopsies has become increasingly important for cancer detection, monitoring, and therapy selection. Now, Avida's innovative duo workflow empowers you to get more information from every sample, so you can keep pace with the ever-evolving cancer landscape. With Avida, go from extracting cell-free DNA (cfDNA) from a single sample to sequencing genetic and epigenetic targets in hours, not days.

## Optimized sample recovery unlocks maximum capabilities

Avida's proprietary workflow is purpose-built to maximize sample retention and recovery, ensuring the integrity and fidelity of your original sample is retained. With no fragment left behind, the Avida workflow and reagent kits are ideal for sensitive and innovative applications such as liquid biopsy and multiomics sequencing.

Avida Reagent Kits
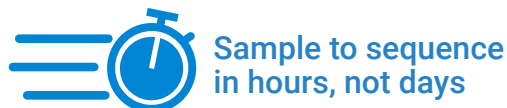
- Avida Duo Methyl Kit
- Avida DNA Kit
- Avida Methyl Kit
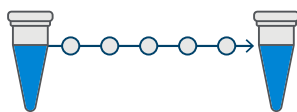
1. Optimize sample recovery

From initial library prep to indexing PCR, the Avida workflow is designed to minimize steps that lead to sample loss, such as sample transfers. And, Avida's novel interlocked hybrid capture chemistry is more efficient and faster than single, long biotinylated probes.

2. Maintain sample fidelity

Thanks to maximized sample recovery, the Avida workflow eliminates pre-capture PCR. This allows for capture of DNA and methyl targets from the original sample. As a result, sample complexity is maintained and bias is minimized.

Sample to sequence in hours, not days
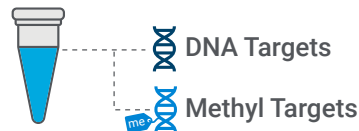
DNA Targets

Methyl Targets

3. Automate and streamline

The streamlined Avida workflow includes fewer sample transfers and amplification steps, combined with a more efficient hybridization method, dramatically reducing turnaround times and ensuring full automation capability.

4. Novel, multiomic application

The result? A sensitive, fast workflow ideal for low-input applications like cfDNA, and a novel, multiomic application. With the Avida Duo workflow, both DNA and methyl targets can be captured from a single sample without signal loss.

The Dynamic Duo | DNA + Methylation Enrichment

Experience the power of DNA and methylation in one target enrichment workflow with Avida Duo.

By maximizing sample recovery, the Agilent Avida Duo workflow enables DNA and methylation profiling from the same low input sample. The streamlined workflow is automation compatible so you can go from extracted sample to sequencing genetic and epigenetic targets in as little as 8 hours.
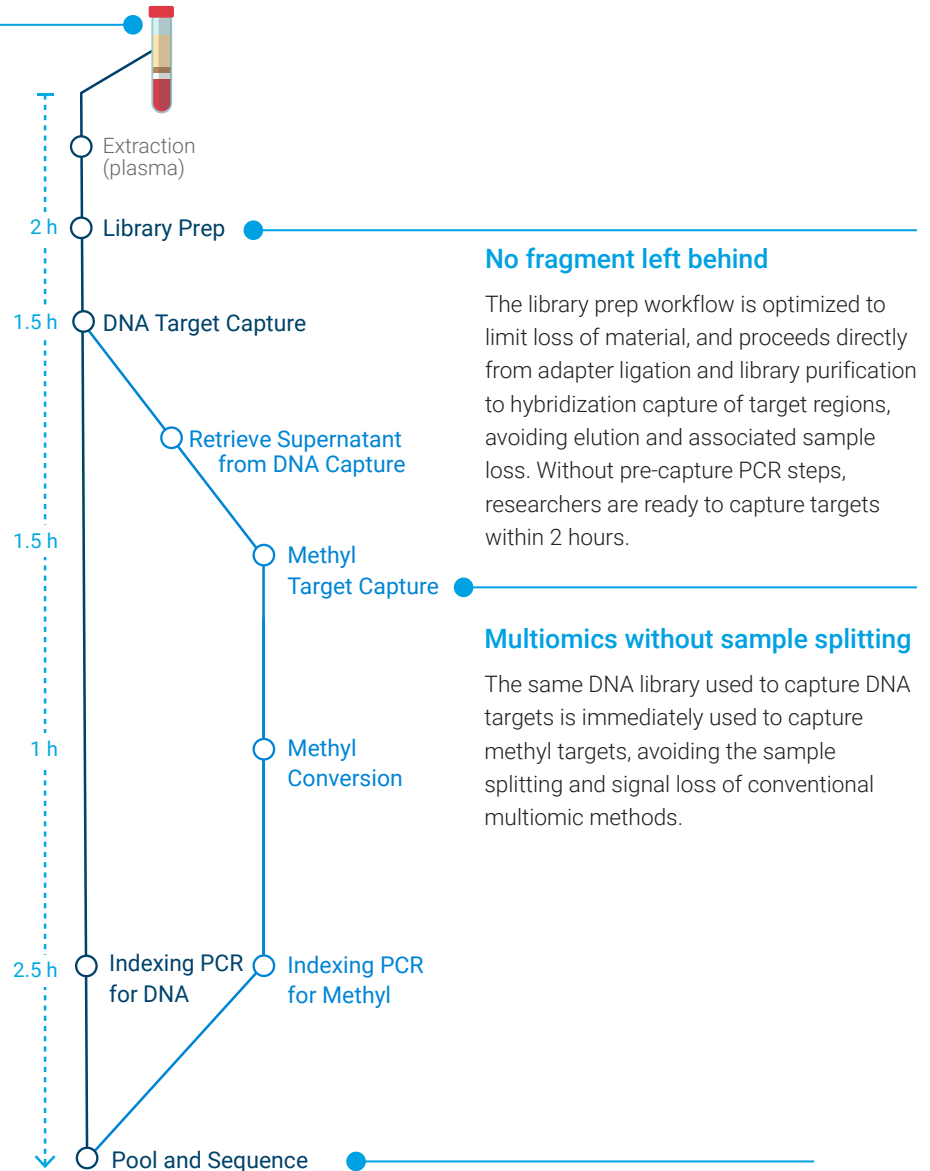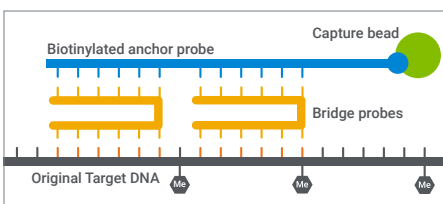
Applicable Kit

Avida Duo Methyl Kit

Optimized for sensitive sample types

Less required input makes the method ideal for analysis of difficult sample types like cfDNA and FFPE DNA from liquid and tissue biopsies where low input is typical.

Capturing more targets, more efficiently

A faster, more efficient hybridization reaction is made possible by a dual probe system. First, short bridge probes hybridize to target DNA. When more than one bridge probe hybridizes to the same target, they are stabilized by a biotinylated anchor probe, creating a "3D umbrella" probe system. Highly specific capture is achieved thanks to this synergetic probe structure.

Workflow

- Extraction (plasma)
- 2 h — Library Prep
- 1.5 h — DNA Target Capture
- Retrieve Supernatant from DNA Capture
- 1.5 h — Methyl Target Capture
- 1 h — Methyl Conversion
- 2.5 h — Indexing PCR for DNA / Indexing PCR for Methyl
- Pool and Sequence

No fragment left behind

The library prep workflow is optimized to limit loss of material, and proceeds directly from adapter ligation and library purification to hybridization capture of target regions, avoiding elution and associated sample loss. Without pre-capture PCR steps, researchers are ready to capture targets within 2 hours.

Multiomics without sample splitting

The same DNA library used to capture DNA targets is immediately used to capture methyl targets, avoiding the sample splitting and signal loss of conventional multiomic methods.

5 days	8.5 h
Total Time for DNA and Methyl NGS Profiling	Total Time for Avida Duo

Sensitive detection can lower sequencing costs

High sample recovery translates into high sensitivity — down to 0.0025% for methyl targets. Fewer reads required during sequencing can lower sequencing costs.
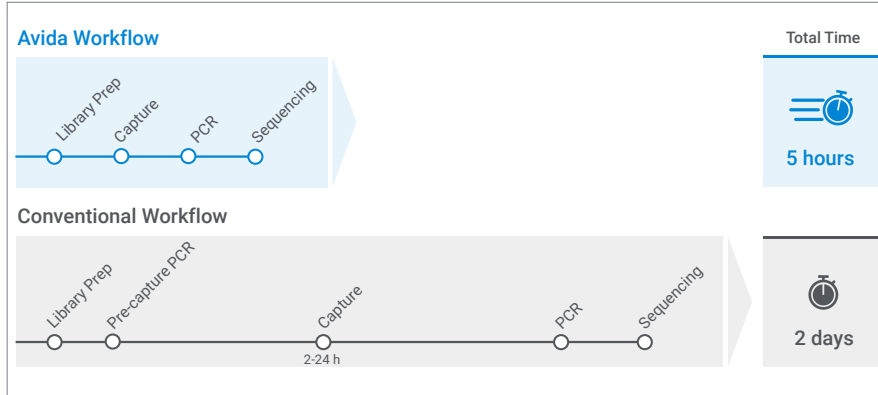
DNA Target Enrichment

Go from sample to sequencer in as little as 5 hours with Avida's DNA workflow.

By eliminating pre-capture PCR and enabling faster hybrid capture, the Avida workflow is streamlined for high-performance with low sample inputs, and can be used for a range of inputs and panel sizes.

Applicable Kits

Avida DNA Kit

Avida Duo Methyl Kit

Agilent Avida DNA Workflow

✓ **No pre-capture PCR**

✓ **4x faster hybrid capture**

✓ **Sample to sequencer in a single shift**

High-performance across input amounts

Maximizing sample recovery leads to more uniform coverage. In turn, high uniformity and percentage of on-target fragments allows for more efficient, lower-cost sequencing.

Sample Type	Sample Input	Coverage*	% on Target	% Mapped	Uniformity
cfDNA	3 ng	1010	81.3	98.4	100
cfDNA	1 ng	421	85.9	97.2	100
FFPE DNA	50 ng	1855	84.2	99.2	100
gDNA	100 ng	7980	88.7	99.2	100
gDNA	10 ng	1309	80.5	99.0	100

Figure 1: DNA performance for the Avida DNA Focused Cancer panel (27kb panel covering 14 actionable genes) was assessed for pooled normal cfDNA, cell line and normal FFPE samples. Data was averaged across at least 4 replicates. The sequencing amount was 20,000x and the total raw read was 3.5M.

Do it Together or Alone

In addition to the innovative Duo DNA + Methylation workflow, the Avida DNA and Avida Methylation workflows can be purchased separately. In combination or alone, the Agilent Avida targeted profiling workflows offer high performance and outstanding fidelity, turnaround time, and ease of use.

* Coverage metrics reflect the average number of unique reads per target after single-strand UMI deduplication (covering the center of the target). The average across replicates of the median of all target regions is reported.
**Uniformity reflects coverage uniformity across targets (fraction of target regions in which coverage is ≥ 0.2x the mean value)

Methyl Target Enrichment

Achieve superior recovery of cfDNA and 2-3x faster turnaround time with the Avida Methyl workflow.

By maximizing sample recovery, the Avida workflow enables native methylation targets to be captured before DNA conversion and amplification. In contrast, conventional workflows require pre-capture PCR to achieve the input requirements for methyl target capture. And, because PCR does not preserve methylation patterns, methylated DNA must be converted first. The innovative, streamlined Avida workflow makes probe design more straightforward, reduces bias from conversion and pre-capture PCR, and enables multiomic capabilities.

Applicable Kits

- Avida Methyl Kit
- Avida Duo Methyl Kit
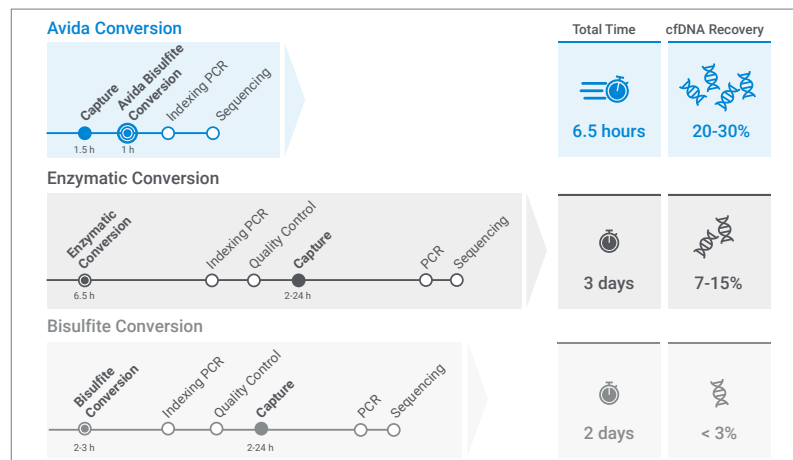
Agilent Avida Methyl Workflow

✓ Capture methylated targets before conversion and PCR

✓ 2-3x faster conversion

✓ Efficient, faster hybrid capture

✓ High DNA recovery

Unprecedented analytical sensitivity for methyl targets

Sensitivity to methylated DNA — down to 0.0025% in cfDNA background — is superior to conventional methods and SNV-based ctDNA detection.

Optimized conversion for more coverage

Combining pre-amplification capture with a proprietary, "soft" bisulfite conversion method reduces DNA damage compared to bisulfite conversion and reduces sample loss compared to enzymatic conversion, resulting in high coverage.
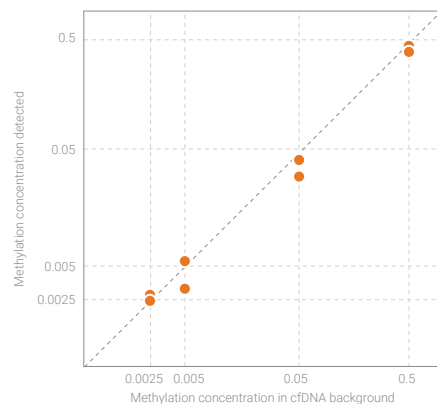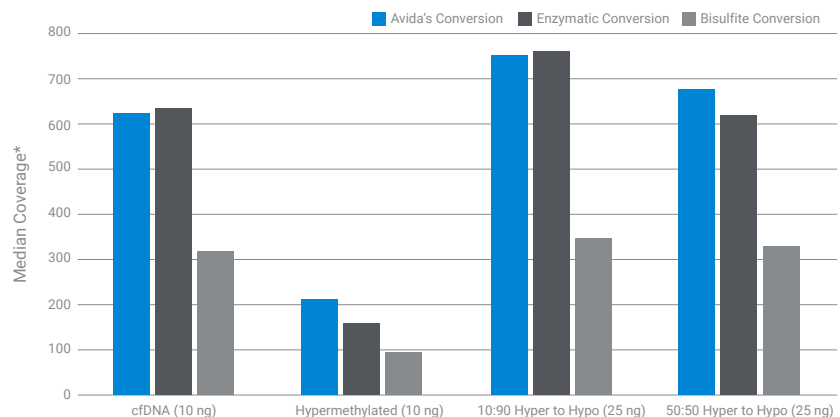
Figure 2. To assess analytical sensitivity, known spike-ins of 0.5%, 0.05%, 0.005%, 0.0025% and 0% of methylated DNA in cfDNA background were detected with a 200 DMR subset (60kb) of the Avida Methyl 3400 DMR Cancer panel. 10ng of Seraseq Methylation ctDNA was used as reference material, and methyl targets were sequenced on NextSeq1000 with 4-5 M paired-end reads per sample.*

Figure 3. To assess the efficiency of the Avida conversion, Human HCT116 DKO Methylated ('Hyper', Zymo D5014-2) and Non-methylated DNA ('Hypo', Zymo D5014-1) were spiked in at various percentages with in-house control cfDNA sample. All samples (at 10-25 ng input) were prepared using Avida library preparation and capture with a 200 DMR subset (60 kb) of the Avida 3400 DMR cancer panel. Varying conversion methods (traditional bisulfite and enzymatic conversion) were compared alongside Avida conversion. Each sample was sequenced with 10 M paired end reads per sample.*

* Beta version of the reagents were used to generate this data

The power of the Avida workflow can be paired with the high-performance panel of your choosing.

Agilent Avida DNA Focused, DNA Expanded, and Methyl 3400 DMR cancer panels are flexible, modular and automation-ready. DNA panels may be matched with the methyl panel for use with the Avida Duo workflow.

- DNA Focused Cancer Panel: Highly focused panel targeting 14 genes relevant to oncology research. This 27 kb panel provides either hotspot or full exon coverage to generate sequencing data on these actionable genes.

- DNA Expanded Cancer Panel: Medium sized panel targeting 105 cancer-associated genes with either hotspot or full exon coverage. This 345 kb panel balances content and sequencing depth requirements for efficient sequencing of liquid biopsy samples.

- Methyl 3400 DMR Cancer Panel: This 870 kb panel targets approximately 3400 differentially methylated region (DMRs) selected for their ability to discriminate solid tumor DNA vs non tumoral DNA, both from tissue and liquid.

Learn more:
www.agilent.com/en/product/next-generation-sequencing

Buy online:
www.agilent.com/chem/store

U.S. and Canada
1-800-227-9770
agilent_inquiries@agilent.com

Europe
info_agilent@agilent.com

Asia Pacific
inquiry_lsca@agilent.com

Agilent
Trusted Answers