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Data Brief

SuperMethyl™ Max for low-input methylation profiling

Independently validated by Admera Health

admera health

Introduction

DNA methylation analysis at picogram-to-nanogram input levels requires high DNA recovery, minimal fragmentation, and low background conversion error to preserve biological signal.

Conventional bisulfite conversion methods can introduce DNA damage and loss, while enzymatic approaches may exhibit reduced conversion efficiency or increased background signal at low input levels, impacting accuracy and library complexity.

Here, we evaluate SuperMethyl™ Max, an ultra-mild bisulfite conversion workflow, and compare its performance to an enzymatic methylation method across matched input conditions.

Technical Characteristics of SuperMethyl™ Max

Ultra-mild Bisulfite Conversion

Reaction conditions minimize DNA fragmentation while maintaining efficient cytosine conversion.

High Conversion Efficiency Across Inputs

Consistent 99.8% C-to-T conversion observed from 100 pg to 50 ng gDNA.

Near Zero Background Signal

<0.05% unconverted cytosines detected at both CpG and non-CpG sites, indicating low false-positive rates.

Preservation of DNA Fragmentomics

Fragment size distributions indicate minimal degradation at all tested DNA input levels.

Consistent Performance Across Input Levels

Conversion efficiency and background signal remain stable across picogram to nanogram inputs.

2-3 hour Protocol, Compatible With Library Prep

Complete methyl conversion within 2-3 hours and compatible with both dsDNA and ssDNA library prep.

Applicable to:
cfDNA & liquid biopsy, FFPE-derived DNA, fresh and frozen tissue gDNA, single-cell samples

Compatible with downstream methylation readouts:
NGS, microarray, PCR, methylation-specific hybrid capture panels

Study Overview

SuperMethyl™ Max was evaluated against an enzymatic C-to-T conversion method across input DNA of 100 pg – 50 ng under matched library preparation, amplification, and sequencing conditions. See next page for results.

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Performance evaluation across low-input DNA (100 pg–50 ng)

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Study Design

Sheared human genomic DNA (100 pg – 50 ng) was used to evaluate performance across low input levels. SuperMethyl™ Max was compared to an enzymatic methylation method (NEBNext® Enzymatic Methyl-seq v2, NEB) under matched library preparation, amplification, and sequencing conditions. Performance was assessed by fragment size distribution, library yield, C-to-T conversion efficiency, and CpG-level unconverted cytosine (false positive) rates. Experimental workflow details are provided below.

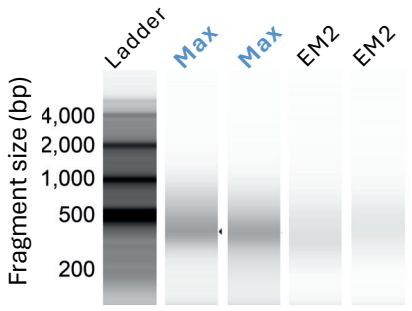
Experimental Workflows

Label	DNA sample	Library Preparation	Methyl Conversion	Amplification	Sequencing	Time
Max	100 pg - 50 ng sheared human gDNA	NEB Ultra II	SuperMethyl™ Max (Ellis Bio)	KAPA HIFI Uracil+ (Roche)	Illumina Platform	2-3 hours
EM2		NEBNext® Enzymatic Methyl-seq v2 (NEB)				1 workday

Amplification was performed using equivalent PCR cycling conditions across input levels.

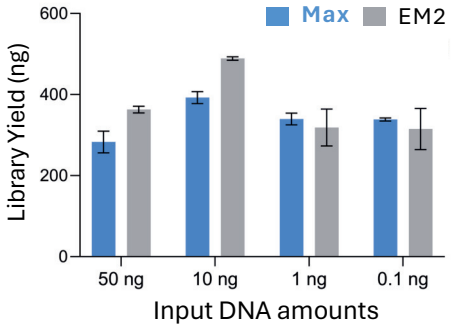
Results

DNA integrity is preserved



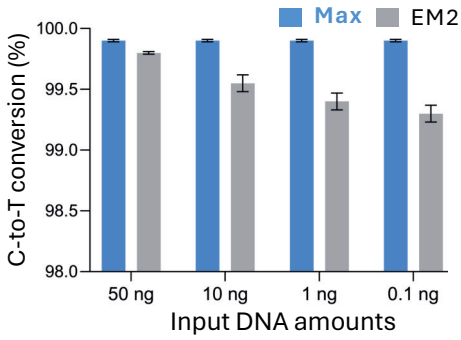
DNA integrity is preserved following ultra-mild bisulfite conversion across all input levels

Comparable library yield across input DNA levels



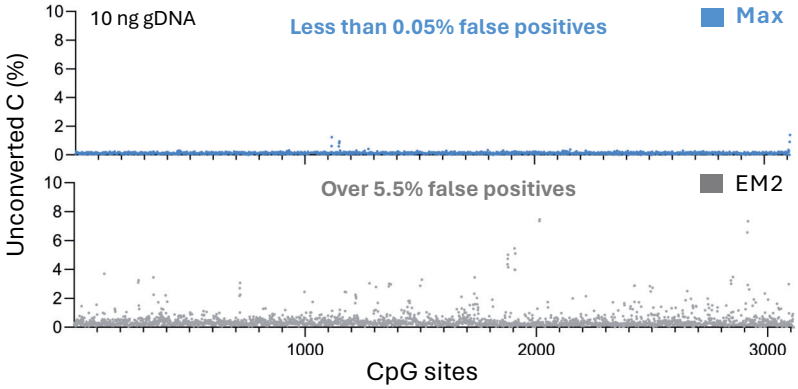
Library yield post-amplification is comparable between methods

Max 99.9% C-to-T conversion efficiency consistent across all inputs



Max conversion efficiency remains consistently high across decreasing input amounts

Near-zero false positives in the Max-treated samples



Max conversion demonstrates < 0.05% unconverted cytosines (false positives) compared to higher levels in enzymatic conversion

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