

We are TWINS!!

Epigenetics Secret Garden

Why are we so different ?



ZYMO RESEARCH
The Beauty of Science is to Make Things Simple™

THE
Epigenetics
COMPANY™



騰達行

UNIMED unimed.com.tw

台北 TEL (02)2720-2215
新竹 TEL (03)6684-586
台中 TEL (04)2463-3591

嘉義 TEL (05)2844-162
台南 TEL (06)2890-665
高雄 TEL (07)3470-143



免付費客服專線
0800-211-819



各大品牌誠摯推薦



Infinium and GoldenGate assay systems



Agilent Technologies

SureSelect Methyl-seq Target Enrichment System



MassARRAY EpiTYPER



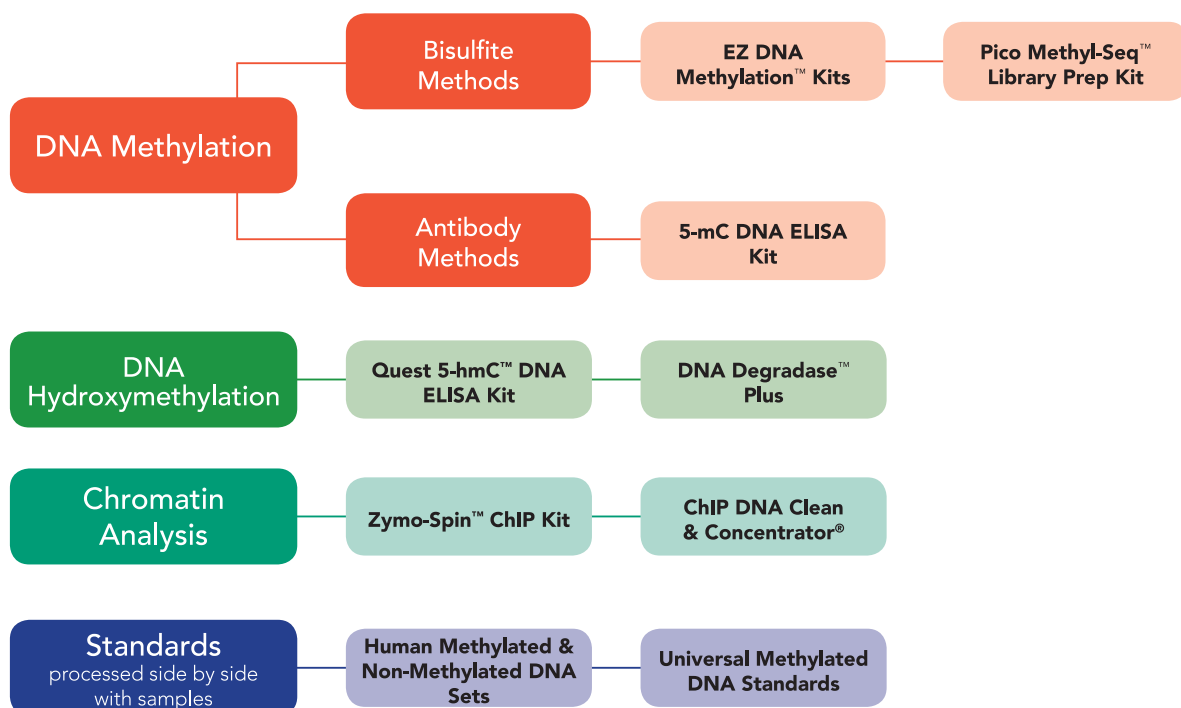
ZYMO RESEARCH

品質保證



NimbleGen SeqCap Epi Enrichment System

Quick Guide of Popular Epigenetics Products



Bisulfite Conversion-The Gold Standard

EZ DNA Methylation™ Kit

- DNA Input: 100 pg - 2 µg (optimal input : 200 - 500 ng) 。
- 彈性選擇：可使用 Direct 或 Lightning 系統，大幅縮短操作時間 。
- 高產量的 bisulfite-converted DNA 適用 PCR、MSP、array、NGS 等。

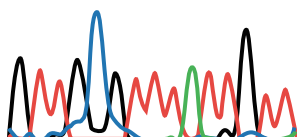
亦有 RNA Methylation 試劑
能方便並準確的分析
RNA methylation 。

R5001/R5002

1 2 3 4 5 6 7 8 9 10 11 12 13 14 15

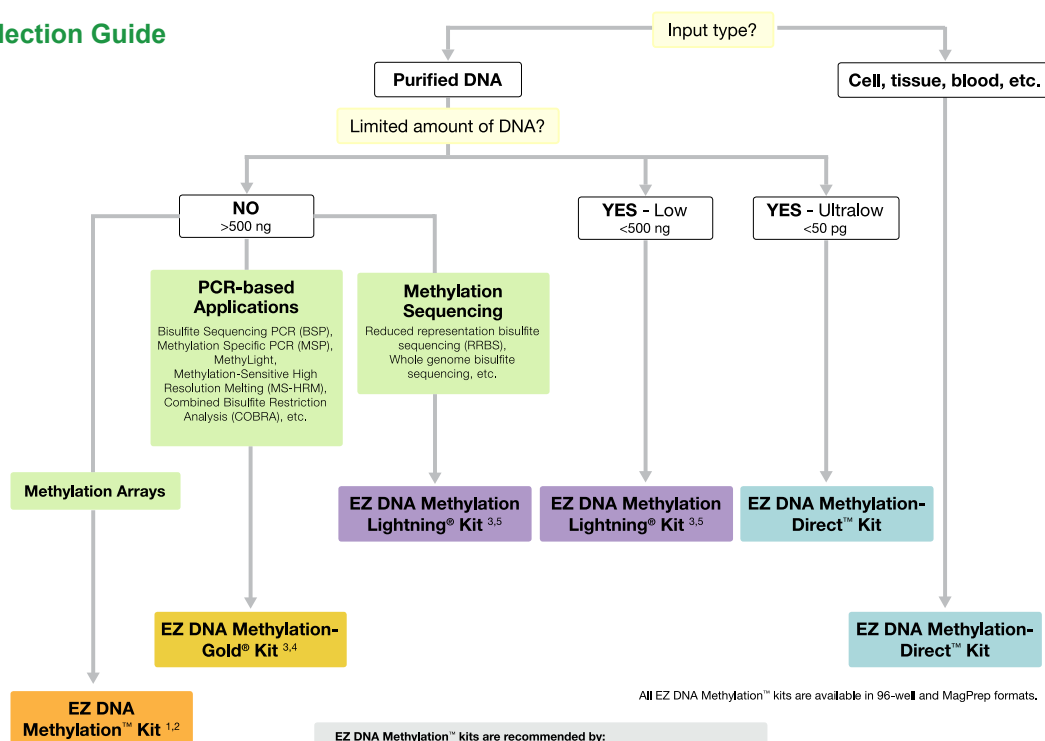
Original DNA with methylated C^mpG ➤ G T T G C^mG C T C A C T G C C

DNA Sequencing after CT conversion ➤ G T T G C G T T T A T T G T T



利用 bisulfite conversion，將 non-methylated cytosine 轉換為 uracil；純化後的 DNA 經 PCR 放大並定序，uracil 會轉換為 thymine，因此此反應又稱為 CT conversion 。

Selection Guide



EZ DNA Methylation™ kits are recommended by:

- 1 Illumina Infinium® and GoldenGate® assay systems
- 2 Sequenom MassARRAY® EpiTYPER®
- 3 Epicentre EpiGnome™ Methyl-Seq kit
- 4 Agilent Technologies SureSelect™ Methyl-Seq Target Enrichment System
- 5 Roche NimbleGen SeqCap Epi Enrichment System

品名	品號	包裝	適用樣本	CT conversion 效率	DNA 回收率	操作時間
EZ DNA Methylation™ Kit	D5001	50 Rxns	500 pg-2 ug DNA	> 99%	> 80%	12 - 16 hrs
	D5002	200 Rxns				
EZ DNA Methylation-Gold™ Kit	D5005	50 Rxns	500 pg-2 ug DNA	> 99%	> 75%	3 hrs
	D5006	200 Rxns				
EZ DNA Methylation-Lightning Kit	D5030	50 Rxns	100 pg-2 ug DNA	> 99.5%	> 80%	1.5 hrs
	D5031	200 Rxns				
EZ DNA Methylation-Direct™ Kit	D5020	50 Rxns	DNA(≥ 50 pg), cells(≥ 10), blood, tissue, FFPE	> 99.5%	> 80%	4 hrs
	D5021	200 Rxns				

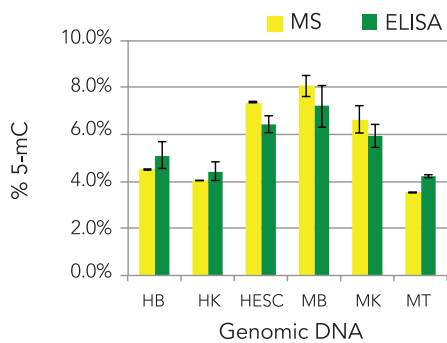
Sensitive 5-mC and 5-hmC Quantification

5-mC and 5-hmC ELISA Kits

- 利用 ELISA 偵測 5-mC、5-hmC 含量，適合高通量的實驗。
- 靈敏度：≥ 0.5% 5-mC、≥ 0.02% 5-hmC per 100 ng input DNA。
- 只需三小時即可完成實驗。

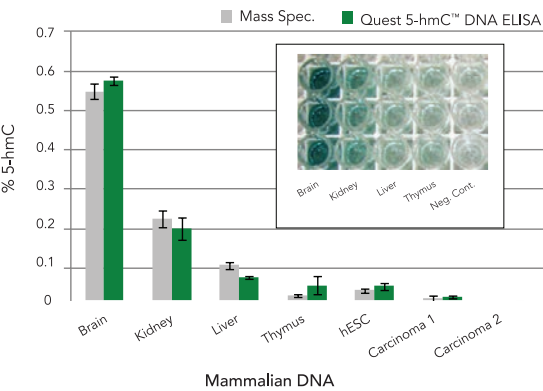
利用 5-mC ELISA/5-hmC ELISA 定量不同樣本 DNA 的 5-mC/5-hmC，其結果與用 mass spectrometry (MS) 分析的結果一致

Specific Quantitation of 5-mC DNA



The 5-mC DNA ELISA Kit can quantify 5-mC in numerous DNA samples with close correlation to LC-MS analysis. Percent 5-mC was calculated for the following genomic DNA samples: human brain (HB), human kidney (HK), human embryonic stem cell (HESC), mouse brain (MB), mouse kidney (MK), and mouse testes (MT). The percent 5-mC detected in DNA samples by 5-mC DNA ELISA Kit (ELISA) strongly correlates to mass spectrometry (MS) data of 5-mC found in the respective gDNA sample.

Specific Quantitation of 5-hmC DNA



5-hmC Quantification. Percent 5-hmC in mammalian DNA samples quantified by mass spectrometry or Quest 5-hmC DNA ELISA Kit. Inlaid image represents relative amounts of 5-hmC in triplicate gDNA samples.

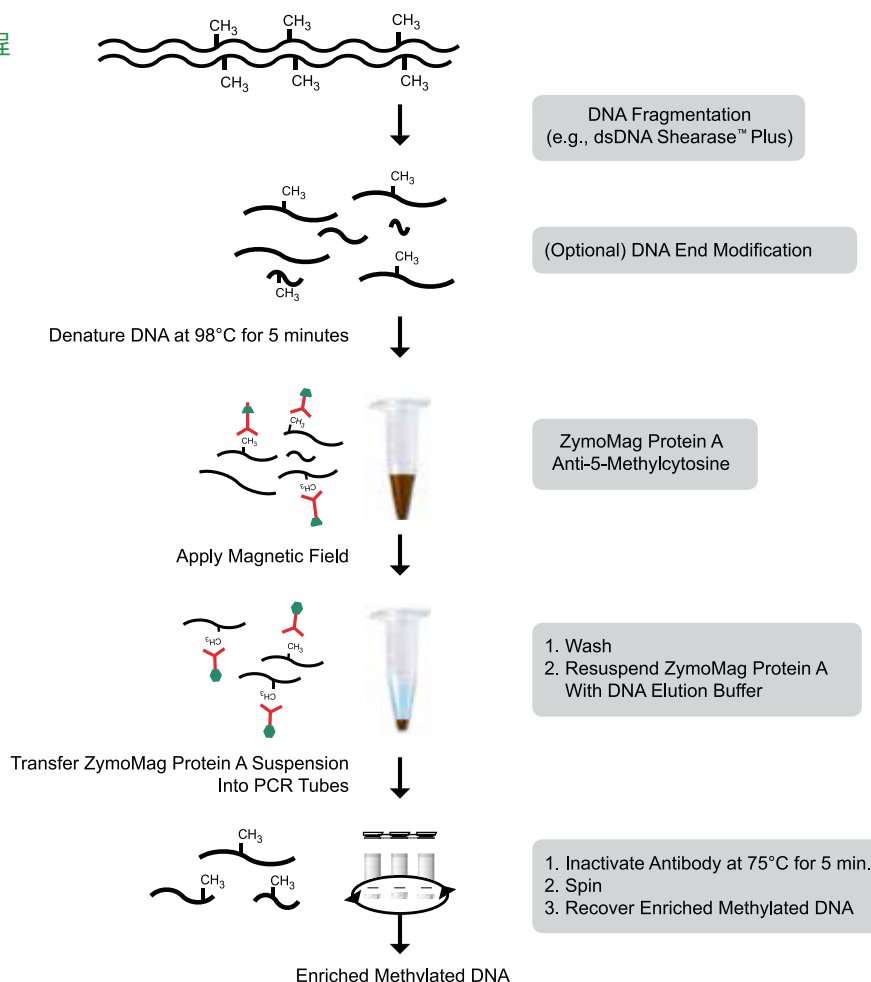
品名	品號	包裝
5-mC DNA ELISA Kit	D5325	1 x 96-well
	D5326	2 x 96-well
Quest 5-hmC™ DNA ELISA Kit	D5425	1 x 96-well
	D5426	2 x 96-well

Enrichment of 5-mC -containing DNA

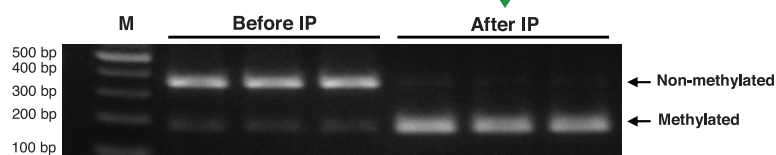
Methylated-DNA IP Kit

- 利用高度專一性的 **anti-5-methylcytosine monoclonal antibody** 抓取甲基化 DNA。
- 附有 **control DNA** 及 **primers**，方便監控實驗流程。
- 回收的 DNA 可用於 **PCR**、**whole-genome amplification**、**ultra-deep sequencing**、**microarray** 等等。

操作流程



IP 後有效的濃縮 methylated DNA



Salmon sperm genomic DNA "spiked" with non-methylated/methylated control DNA was processed with the kit and the eluted DNA was amplified by PCR using the supplied control primers. Digestion of the amplicons with Nco I produced two 175 bp fragments for the methylated DNA control or one 350 bp fragment for the non-methylated DNA control.

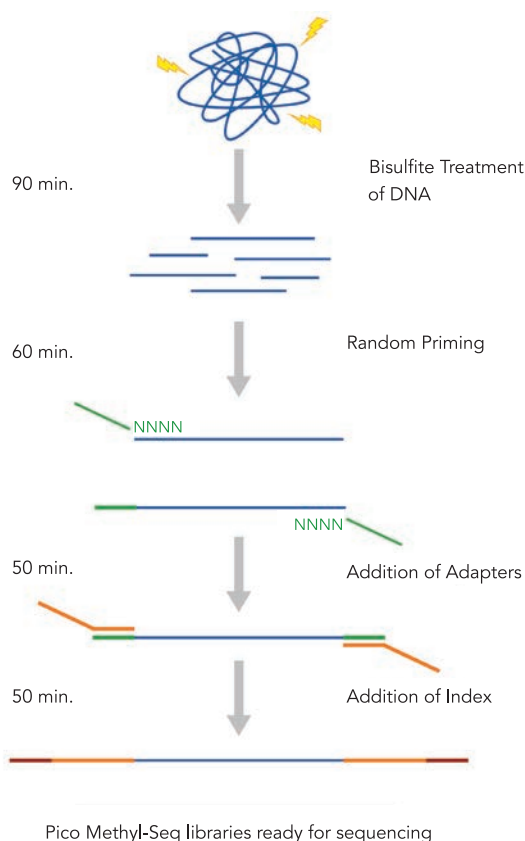
品名	貨號	包裝
Methylated-DNA IP Kit	D5101	10 Rxns

Post-Bisulfite Library Preparation

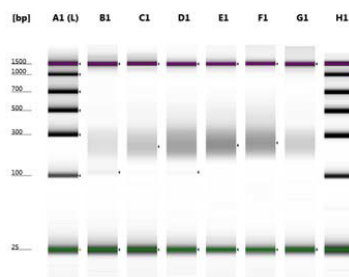
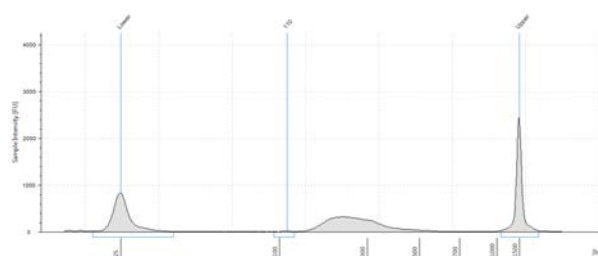
Pico Methyl-Seq™ Library Prep Kit

- 適用樣品來源：FFPE、ssDNA，只需 10 pg DNA。
- 利用 random primer 在進行過 bisulfite treatment 的 DNA 片段做 PCR 放大片段，進而製成 WGBS (Whole Genome Bisulfite Sequencing) library。
- 完整套組，從 bisulfite conversion 到 library preparation 一應俱全。
- 優化的操作流程，Ligation-free 和 gel-free，僅需六小時。
- 適用於 Illumina's sequencing platform：HiSeq、MiSeq。

操作流程



製備高品質的基因庫



(A) Electropherogram of a Pico Methyl-Seq (WGBS) library prepared using 100 pg of human gDNA. (B) Gel of libraries prepared from varied amounts of human gDNA. Fragments ranged between 150 and 350 bp.

品名	品號	包裝
Pico Methyl-Seq™ Library Prep Kit	D5455	10 Preps
Pico Methyl-Seq™ Library Prep Kit	D5456	25 Preps

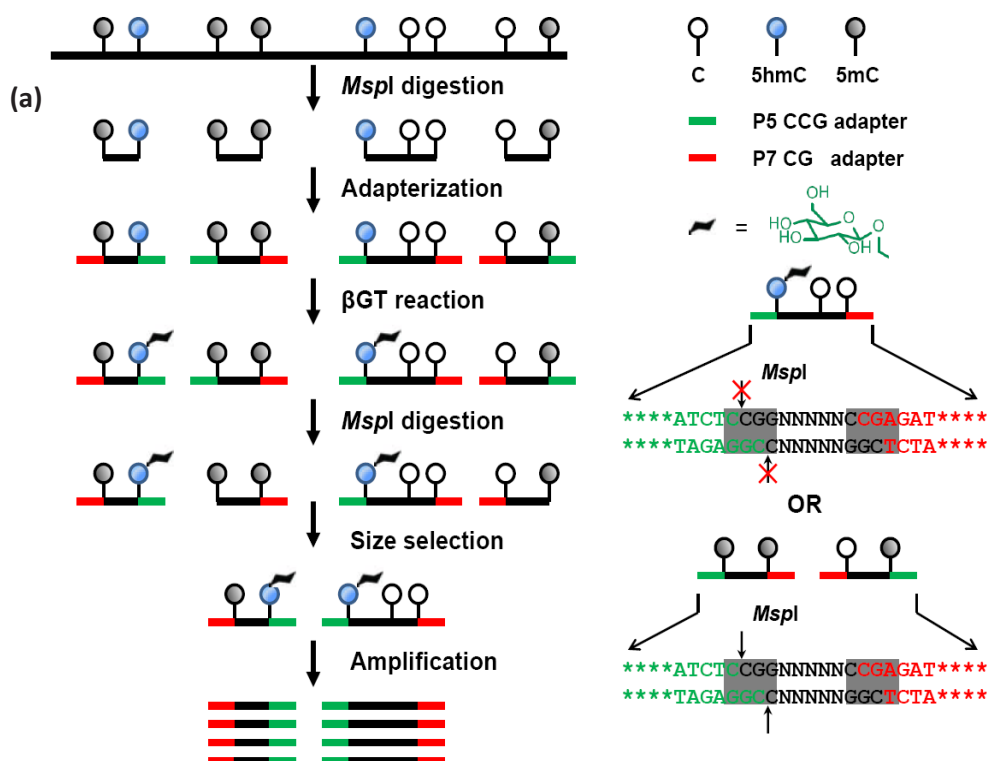
5-hmC Library Preparation

RRHP™ 5-hmC Library Prep Kit

- 利用 **Msp I** 的特性，會水解無 **glucosylated-5-hmC** 的片段，而 **glucosylated-5-hmC** 會阻擋 **Msp I** 辨認位，即可建構成 **library**。
- 樣品 ≥ 100 ng low input DNA。
- 可以得到 **strand-specific mapping of 5-hmC DNA** 資訊。
- 適用於 **Illumina's sequencing platform** : HiSeq、MiSeq。

β -glucosyltransferase (β -GT) 在 5hmC 上做標記，保護其不被 **Msp I** 水解。

操作流程



品名	品號	包裝
RRHP 5-hmC Library Prep Kit	D5450	12 preps
	D5451	25 preps

Region Specific 5-mC Analysis

OneStep qMethyl™

- 利用多種 **MSREs (Methylation Sensitive Restriction Enzymes)**，辨認 **CpG 序列**。
 - **non-methylated CpG**：MSREs 可切斷 dsDNA，qPCR Ct 值比 **reference** 高
 - **methylated CpG**：MSREs 無法切斷 dsDNA，qPCR Ct 值與 **reference** 相同
- 不需進行 **bisulfite conversion**，可用 qPCR 定量 DNA 甲基化程度。
- 適合快速篩選 **single-, multi-locus DNA methylation**。
- 附有 **control DNA 及 primers**，方便監控實驗流程。

Specifications

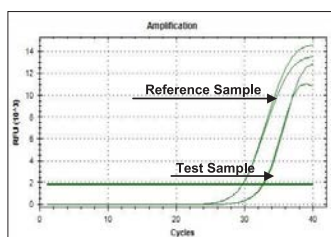
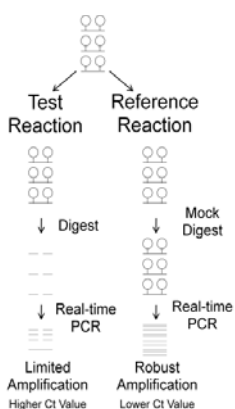
Format..... 96-Well Plate
 DNA Input.....20 ng in 5 µl
 Processing Time.....~4 hours



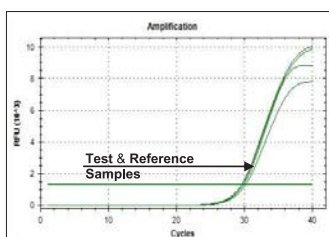
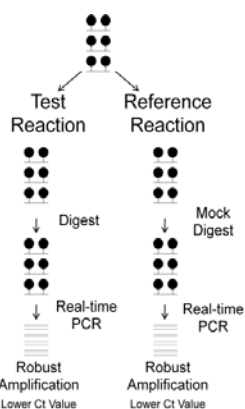
Simple, One Step
全程只需 4 hr
 降低誤差及污染的可能，
 快速準確偵測 DNA 甲基化

操作流程

Non-methylated DNA Input



Methylated DNA Input



- 1 準備 non-methylated 或 methylated DNA sample。
- 2 取一部份 DNA 進行 MSREs digestion，此為 test reaction；另一部份 DNA 則不進行 MSREs digestion，為 reference reaction。
- 3 利用自行設計的 primer set，進行 qPCR (可使用 SYBR green 或 Taqman probe 定量)。
- 4 若為 non-methylated DNA：
 $Ct_{Test} > Ct_{Ref}$
 若為 methylated DNA：
 $Ct_{Test} \approx Ct_{Ref}$ 。

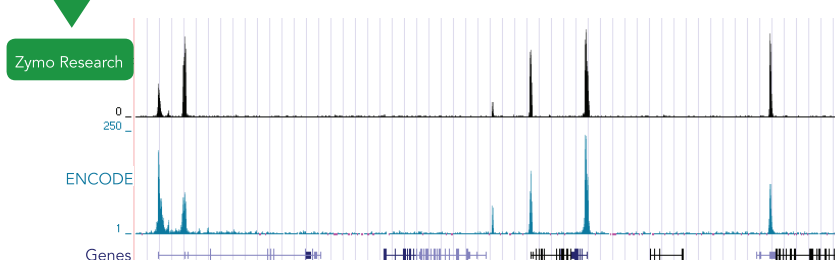
品名	貨號	包裝
OneStep qMethyl™ (with SYTO 9 dye for qPCR)	D5310	1 x 96 well
OneStep qMethyl™ -Lite (without SYTO 9 dye)	D5311	1 x 96 well

Chromatin Analysis

Zymo-Spin™ ChIP kit

- 完整的試劑組，從 ChIP 到純化出高品質 ChIP DNA 一應俱全。
- 僅需 6 μ l 即可回溶出高品質 ChIP DNA。
- 後續可應用於 ChIP-qPCR, ChIP-Seq 等實驗。

可獲得高品質的 ChIP DNA



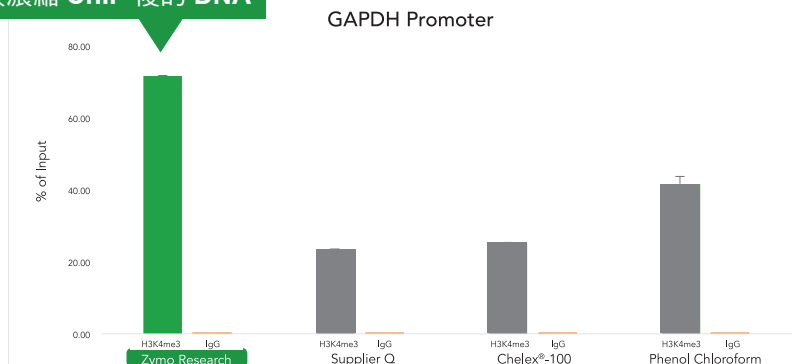
ENCODE Quality ChIP Workflow : Browser tracks depicting H3K4me3 ChIP-Seq assay using the Zymo-Spin™ ChIP Kit. Peaks overlap the same sites identified at the Broad Institute of MIT and Harvard as part of the ENCODE project.

ChIP DNA Clean & Concentrator™

- 快速，僅需 2 分鐘就能從 ChIP sample 純化高純度 DNA。
- 可濃縮 DNA，回收體積僅需 6 μ l。
- 高品質 ChIP DNA，後續可應用於 PCR, arrays, sequencing 等實驗。



高效率回收濃縮 ChIP 後的 DNA



ChIP DNA Purification Comparison : ChIP assays were performed with HeLa cells using ChIP-grade anti-H3K4me3 and rabbit IgG antibodies. Both total and immunoprecipitated chromatin were reverse cross-linked and recovered using either the ChIP DNA Clean & Concentrator® (included in the Zymo-Spin™ ChIP Kit), DNA recovery kit from Supplier Q, Chelex®-100 protocol or phenolchloroform extraction. The amount of ChIP DNA was determined using qPCR with primers specific to the GAPDH promoter. ChIP DNA enrichment is graphed as % input.

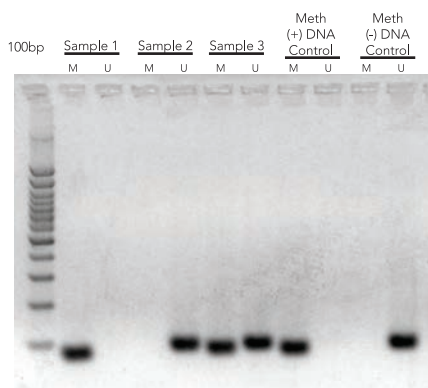
品名	品號	包裝
Zymo-Spin™ ChIP Kit	D5209	10 preps
	D5210	25 preps
ChIP DNA Clean & Concentrator™ Kit (Uncapped)	D5201	50 Preps
ChIP DNA Clean & Concentrator™ (Capped)	D5205	50 Preps

Accurately Quantitate DNA Methylation Epigenetic Standards

- non-methylated 及 methylated human DNA 做為 negative 和 positive control 。
- 監控 bisulfite conversion 效率和實驗流程 (primer design, bisulfite PCR and sequencing reactions) 。
- 確準定量，利用 non-methylated 及 methylated human DNA 互相調配，建立 calibration curves 。

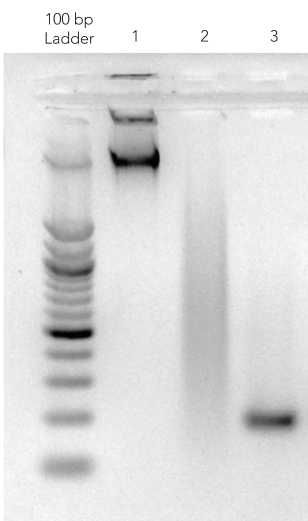
利用 DNA 標準品評估 bisulfite 效率和 primer 的設計

Ideal Controls for BSP and MSP Analysis



Example MSP experiment using MSP designed primers for RASSF1. Sample 1 is positive for a Methylated Template. Sample 2 is positive for a Non-Methylated Template and Sample 3 contains Methylated and Non-Methylated Templates. MSP experiment also shows proper controls: Meth (+) DNA Control D5014-2 Human Methylated DNA, Meth (-) DNA Control D5014-1 Human Non-methylated DNA. 2% Agarose Gel, 130V for 35 mins. M = Methylated Template, U = Non-Methylated Template

Assess Bisulfite Conversion Efficiency and Primer Design



Gel electrophoresis depicting genomic DNA, bisulfite converted genomic DNA and genomic DNA amplified with bisulfite-specific primers. Lane 1 – Input DNA: Universal Methylated Human DNA Standard (D5011). Lane 2- Bisulfite converted Universal Methylated Human DNA (D5011) using EZ-DNA Methylation Direct (D5020). Lane 3 – Universal Methylated Human DNA (D5011) bisulfite converted and amplified with supplied hMLH1 control primers.

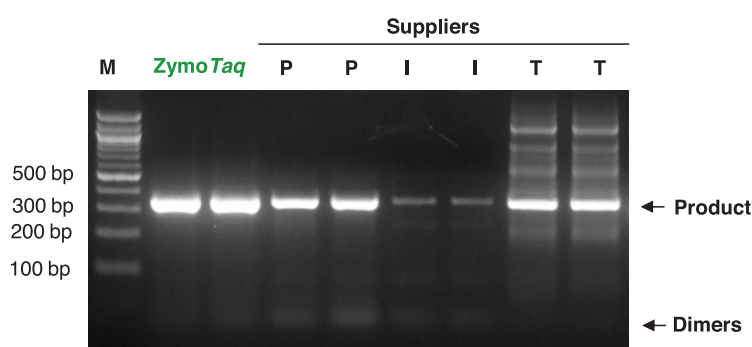
品名	品號	包裝
Human Methylated & Non-Methylated (WGA) DNA Set (DNA w/ primers)	D5013	1 set
Human Methylated & Non-Methylated DNA Set (DNA w/ primers)	D5014	1 set
Universal Methylated Human DNA Standard	D5011	20 Rxns
Universal Methylated Mouse DNA Standard	D5012	20 Rxns

Enzymes

ZymoTaq™ DNA Polymerase

- **Hot-start** 型式，可減少非專一性的產物。
- 適用於 **bisulfite-converted DNA** 後續 **PCR** 反應。

使用 **ZymoTaq** 能得到最專一且大量的 **PCR** 產物



PCR products of immunoprecipitated, methylated DNA vary depending on the hot-start polymerase used.

dsDNA Shearase™ Plus

- 可隨機產生 **dsDNA** 片段。
- 依 **enzyme** 濃度調整 **DNA** 片段的大小。
- 適用於 **library construction**, **Next-Generation sequencing** 和 **methylated DNA immunoprecipitation (MeDIP)**。

DNA Degradase™ & DNA Degradase Plus™

- 簡單省時，只要單一個酵素作用。
 - **DNA Degradase™** 可以水解 **DNA** 成單一個 **nucleotide**
 - **DNA Degradase Plus™** 可以水解 **DNA** 成單一個 **nucleoside**
- 後續適用於 **HPLC**, **TLC**, 和 **LC-MS**。

品名	品號	包裝	品名	品號	包裝
ZymoTaq™ Polymerase	E2001	50 Rxns	DNA Degradase	E2016	500 U
	E2002	200 Rxns		E2017	2000 U
dsDNA Shearase™ Plus	E2018-50	50 U	DNA Degradase Plus	E2020	250 U
	E2018-200	200 U		E2021	1000 U

