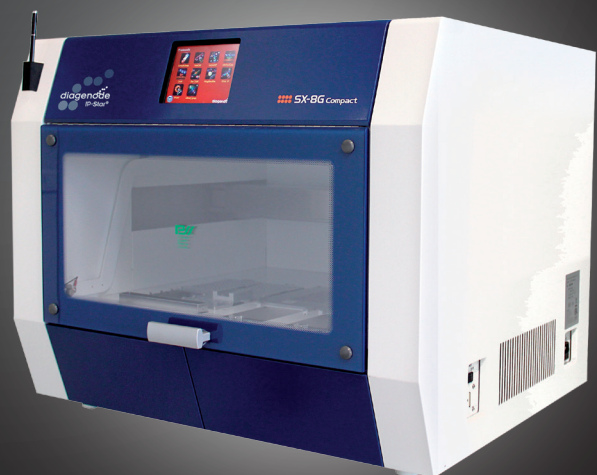


AUTOMATED SOLUTIONS FOR EPIGENETICS

IP-Star® Compact Automated System

- High resolution ChIP-seq and MeDIP-seq profiles
- Automated library preparation for Next-Generation Sequencing
- Reduces hands on time to just 30 minutes
- Reduces variability between operators and labs
- Ideal for low sample starting amounts



Standardize and streamline your epigenetic research

Diagenode IP-Star® Compact automates immunoprecipitation assays with increased reproducibility

Diagenode, the leading provider of complete solutions for epigenetic research, offers a variety of end-to-end systems to streamline DNA methylation and chromatin immunoprecipitation workflows. Central to this full offering is Diagenode's IP-Star® Compact, a simple yet robust automated bench-top instrument that standardizes different epigenetic applications (i.e. ChIP, MeDIP or MethylCap). Diagenode designed these automation assays to make epigenetic research accessible, reproducible, and consistent in every experiment.

The IP-Star® Compact replaces the numerous manual, error-prone steps of complex epigenetic applications with a reliable, highly consistent and automated process that requires minimal operator intervention. In addition, the IP-Star® Compact minimizes sample carryover, data variability, and costly errors. The platform offers full workflow support for epigenetic research, utilizing our complete kits and laboratory-validated protocols to rapidly deliver high-quality and consistent data.

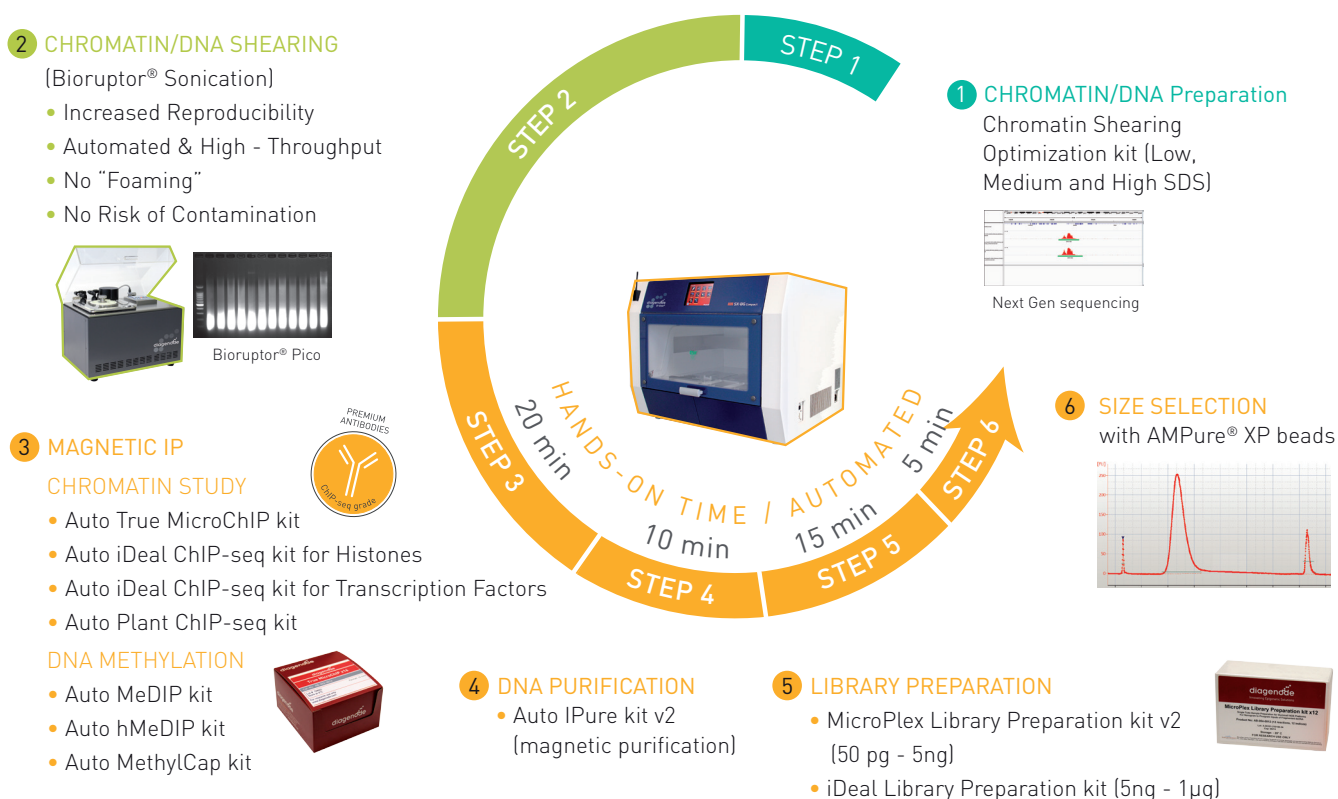


Figure 1. Diagenode provides a full suite of automated solutions for ChIP experiments.

For Step 1, we offer products to isolate nuclei and chromatin. Step 2 describes reproducible sample shearing with the Bioruptor® product line. In Step 3 and Step 4, the Diagenode IP-Star Compact provides error-free, walk-away automation for all your immunoprecipitation and antibody capture needs.

Walk-away automated system

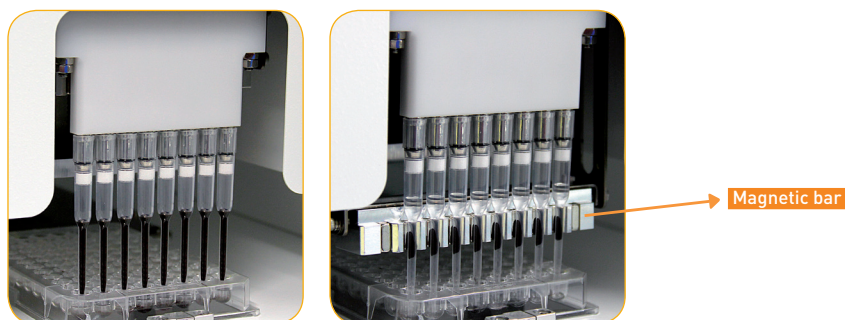
- Magnetic-bead technology for excellent reproducibility
- Open platform allows changes in protocol parameters
- Simple touch screen interface
- Minimal footprint
- Processing up to 16 samples
- Protocols optimal for 100 µl or 200 µl volumes
- Automated reagent dispensing
- Controlled temperatures via Peltier blocks

Overview of the Automated System



The Magtration® Technology

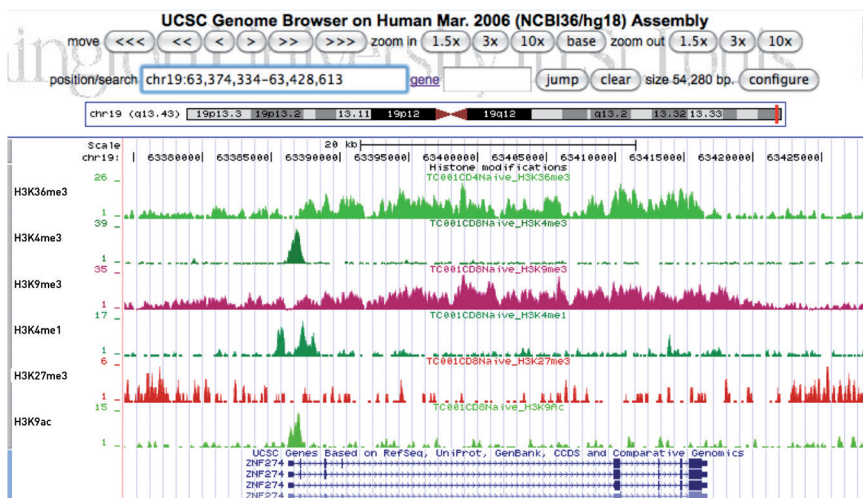
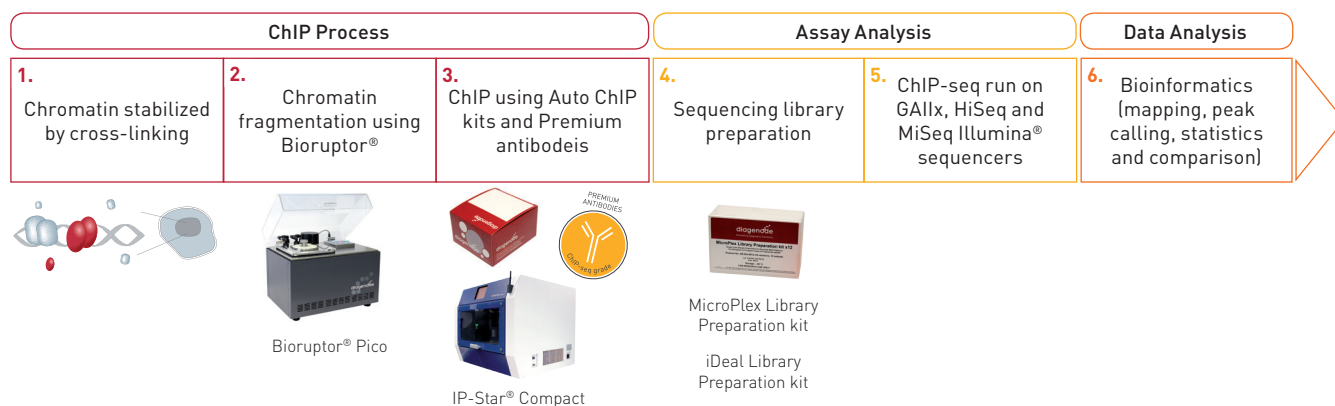
Diagenode's Automated System uses the principle of bead-based magnetic separation. Magnetic beads bound with chromatin or DNA are brought to the inner wall of the tip when a strong magnetic force is applied. This differs from other systems that collect the bound DNA on the bottom of a reaction well, resulting in cleaner assays and less carryover.



Automated Protocols

- Chromatin immunoprecipitation (ChIP)
- Methylated DNA Immunoprecipitation (MeDIP)
- Hydroxymethylated DNA Immunoprecipitation (hMeDIP)
- Methyl-binding domain protein capture assays (MethylCap)
- Sequential ChIP (Re-ChIP)
- Bisulfite conversion
- RNA immunoprecipitation (RNA-IP)
- DNA purification (IPure)
- Library preparation for NGS platforms

Successful implementation of automated ChIP-seq experiments on the IP-Star® Compact



Auto ChIP-seq assays were performed on the IP-Star® using primary human CD4 and CD8 T cells isolated by negative magnetic separation of peripheral blood mononuclear cells. Each sample was assayed using Diagenode's Auto Histone ChIP-seq kit reagents and 1 µg of input chromatin. Illumina Genome Analyzer libraries were prepared and the samples were sequenced at the UC Davis DNA Technologies Core Center.

Customer Feedback

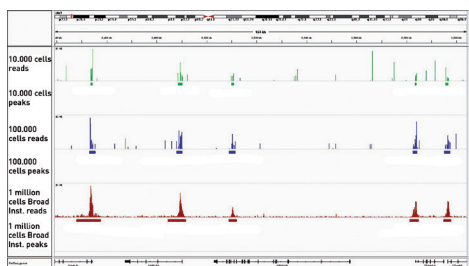
Not only does the IP-Star eliminate the problem of human variation associated with producing our samples, it also enables us to produce 1000-2000 ChIP-seq samples per year very reliably. The IP-Star reduces our processing time down from one day of manual work to just one overnight run with only 30 minutes of hands-on work. The IP-Star has made all our ChIPs consistent and the process completely reliable regardless of the operator or the time of day.

**Dr. John Lambourne, Postdoctorate Researcher
at the Innovation Centre, McGill University, Canada.**

ChIP kits

Auto True MicroChIP kit

- Compatible with 5,000-100,000 cells per IP
- Validated for histone antibodies
- Ideal for whole genome sequencing from low amount of chromatin when combined to the MicroPlex Library Prep kit

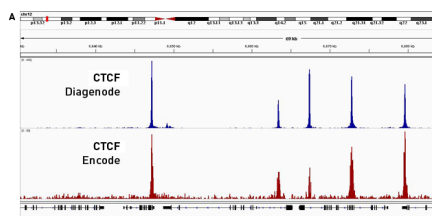


Library generation with the MicroPlex kit. ChIP assays were performed on 10,000 and 100,000 HeLa cells with Diagenode H3K4me3 antibody (0.25 µg/reaction). Libraries were made with the MicroPlex Library Preparation Kit. The generated libraries were then analyzed on an Illumina® HiSeq2000. Cluster generation and sequencing were performed according to the manufacturer's instructions.

Product	Cat. No.	Format
Auto True MicroChIP kit	C01010140	16 rxns
Auto True MicroChIP & MicroPlex Library Preparation Package	C01010141	16 ChIP + 12 library prep rxns

Auto iDeal ChIP-seq kit for Transcription Factors

- 4 millions cells per IP
- Validated for ChIP experiments with antibodies directed to transcription factors, epitope tagged transcription.



Chromatin Immunoprecipitation using chromatin from HeLa cells, the iDeal ChIP-seq kit for Transcription Factors and the Diagenode ChIP-seq-grade CTCF antibody. The IP'd DNA was subsequently analysed on an Illumina® HiSeq. Library preparation, cluster generation and sequencing were performed according to the manufacturer's instructions. This figure shows the peak distribution in a region surrounding the GAPDH positive control gene.

Product	Cat. No.	Format
Auto iDeal ChIP-seq Kit for Transcription Factors x24	C01010058	24 rxns
Auto iDeal ChIP-seq Kit for Transcription Factors x100	C01010172	100 rxns

Auto iDeal ChIP-seq kit for Histones

**BEST
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- 1 million cells per IP
- Validated for ChIP-seq and ChIP-qPCR experiments with antibodies directed to histone modifications
- Ideal for whole genome sequencing

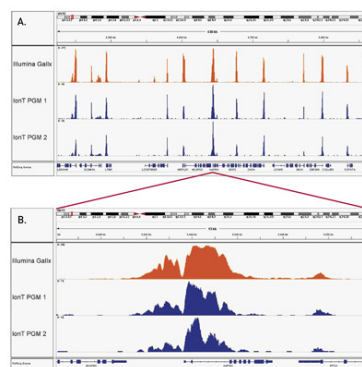


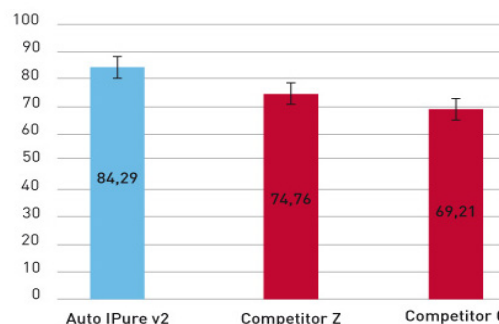
Image A shows a several hundred bp along chr12 with high similarity of read distribution despite the radically different sequencers.

Image B is a close capture focusing on the GAPDH that shows that even the peak structure is similar.

Product	Cat. No.	Format
Auto iDeal ChIP-seq kit for Histones x24	C01010057	24 rxns
Auto iDeal ChIP-seq kit for Histones x100	C01010171	100 rxns

Auto IPure kit v2

- Recovery of small amounts of DNA
- Straightforward protocol using magnetic beads
- Provides pure DNA for NGS downstream application



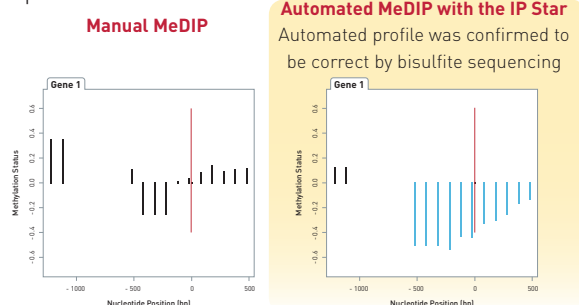
Sample I has been purified in duplicates using the IPure kit v2. The starting material (input) used ranges from 5 ng to 1 µg of DNA.

Product	Cat. No.	Format
Auto IPure kit v2 x100	C03010010	100 rxns

DNA Methylation kits

Auto MeDIP kit

- Compatible with 50 ng to 1 µg per IP
- Validated using Diagenode 5-mC monoclonal antibody 33D3 - Premium (Cat. No. MAb-081-100)
- Optimal for genome-wide screening of methylated CpGs

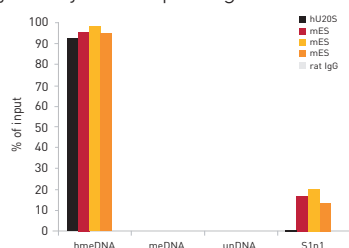


Methylation patterns were analyzed using MeDIP-on-chip in mouse wild type and mutant cell lines. The left panel shows the differential methylation patterns of Gene 1 using a manual MeDIP protocol. While the right panel shows results for automated MeDIP experiments on the IP-Star® Automated System.

Product	Cat. No.	Format
Auto MeDIP kit x16	C02010011	16 rxns
Auto MeDIP kit x100	C02010012	100 rxns

Auto hMeDIP kit

- Compatible with 1 µg per IP
- Validated using Diagenode's 5-hmC antibodies
- Includes control primer pairs for the assessment of the capture efficiency
- Optimal for genome-wide screening of hydroxymethylated CpG regions

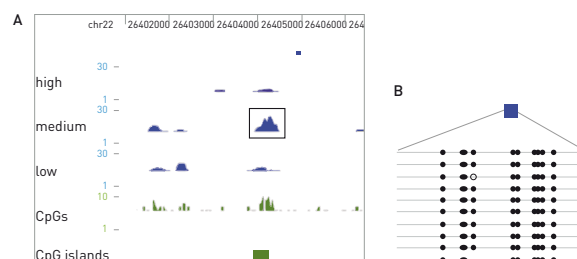


Unmethylated, methylated and hydroxymethylated spike DNA controls (included in the Auto hMeDIP kit) were used in the IP reaction together with 1 µg of genomic sheared DNA. Enrichments were assessed by qPCR using specific primer pairs for the unmethylated, methylated and hydroxymethylated DNA sequences. Sfi1 is a gene that has been identified as being hydroxymethylated using hMeDIP-seq. Human DNA from U20S was used as negative control DNA.

Product	Cat. No.	Format
Auto hMeDIP kit x16 (monoclonal rat antibody)	C02010033	16 rxns
Auto hMeDIP kit x16 (monoclonal mouse antibody)	C02010034	16 rxns
Auto hMeDIP kit x16 (polyclonal rabbit antibody)	C02010035	16 rxns

Auto MethylCap kit

- Compatible with 1 µg per reaction
- Validated using Diagenode's fusion MethylCap protein
- Allows fractionation for methylated DNA based on density of methylated CpG islands
- Ideal for any MBD-seq and MBD-qPCR experiments

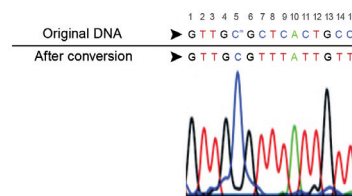


This figure shows MBD-seq results for one genomic region. Using MBD-seq, two methylated regions were detected in different elution fractions according to their methylated CpG density. (A) Low, Medium and High refer to different elution fractions with increasing salt concentration. (B) Methylated patterns of these two different regions were validated by bisulfite sequencing assay.

Product	Cat. No.	Format
Auto MethylCap kit	C02020011	48 rxns

Auto Premium Bisulfite kit

- Rapid bisulfite conversion of DNA - only 1 hour reaction time
- Simple workflow, 3 steps
- High-yields of converted DNA for methylation analysis
- Optimized for NGS, PCR, and more



DNA with methylated cytosine [Cm] at nucleotide position #5 was processed using the Premium Bisulfite Kit. DNA was PCR- amplified and then sequenced. The methylated cytosine at position #5 remains intact while the unmethylated cytosines at positions #7, 9, 11, 14 and 15 are completely converted into uracil following bisulfite treatment (detected as thymine following PCR).

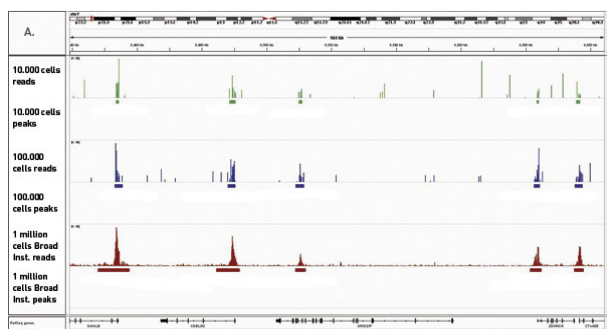
Product	Cat. No.	Format
Auto Premium Bisulfite Kit	C02030031	40 rxns

NGS Library Preparation kits

MicroPlex Library Preparation™ kit v2

- 3 times faster than TruSeq™ - only 2 hours
- 48 samples processed per run
- Picogram inputs (50 pg)
- High sensitivity ChIP-seq - low PCR duplication rate
- Great multiplexing flexibility with 48 barcodes (8 nt) included

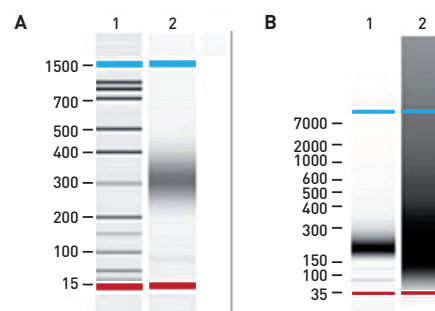
**BEST
SELLER**



Product	Cat. No.	Format
MicroPlex Library Preparation™ kit v2 x12 (12 indices)	C05010012	12 rxns
MicroPlex Library Preparation™ kit v2 x48 (12 indices)	C05010013	48 rxns
MicroPlex Library Preparation™ kit v2 x48 (48 indices)	C05010014	48 rxns

iDeal Library Preparation Kit

- DNA sample inputs from 5 ng to 1 µg
- Multiplexing capacity of up to 24 samples
- Only 3 hours from start to finish
- Ideal for use with our optimized ChIP-seq kits
- 32 samples per run



Product	Cat. No.	Format
iDeal Library Preparation Kit x24 (incl. Index Primer Set 1)	C05010020	24 rxns
iDeal Library Index Primer Set 2	C05010021	24 rxns

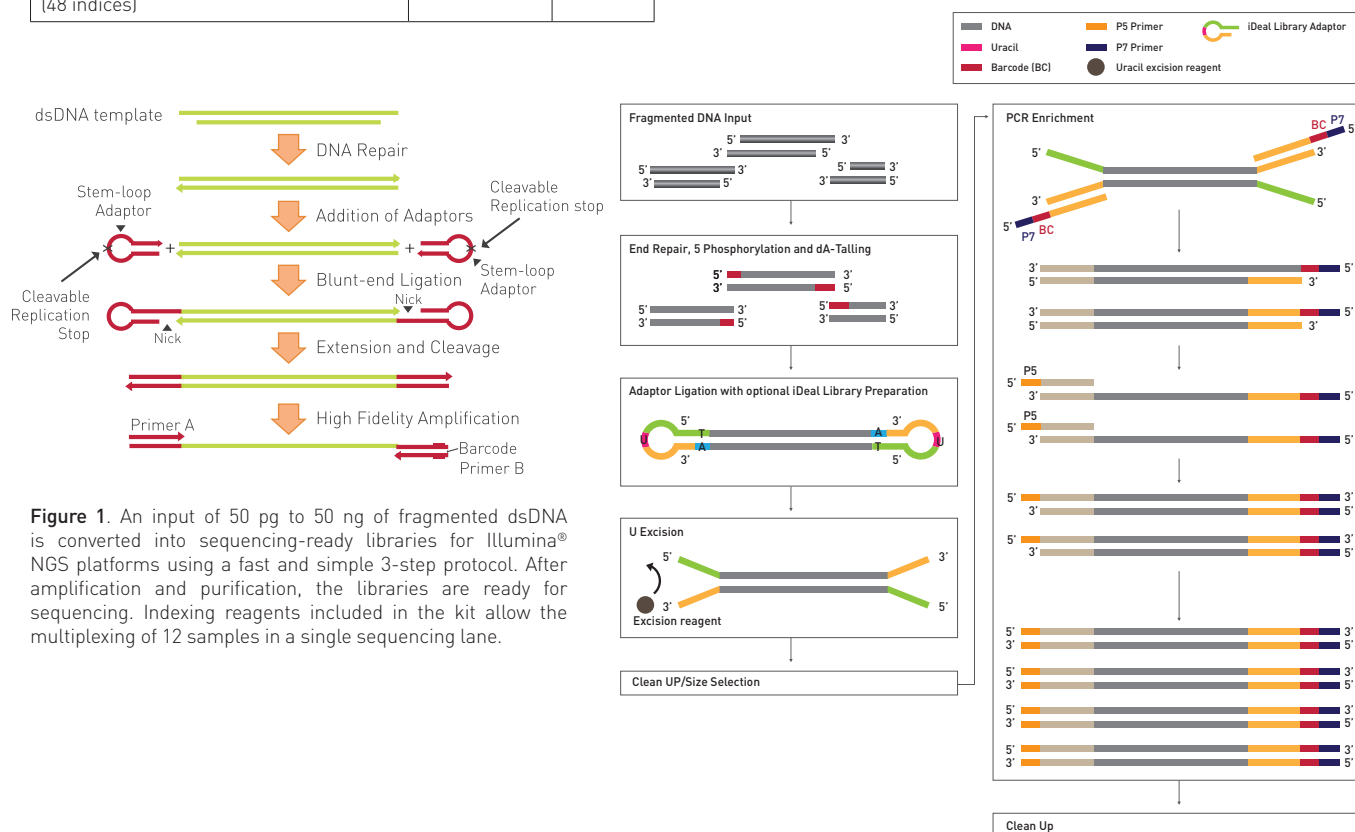


Figure 1. An input of 50 pg to 50 ng of fragmented dsDNA is converted into sequencing-ready libraries for Illumina® NGS platforms using a fast and simple 3-step protocol. After amplification and purification, the libraries are ready for sequencing. Indexing reagents included in the kit allow the multiplexing of 12 samples in a single sequencing lane.

Customer Feedback

The ability to produce libraries starting with **low input DNA** is primordial when using ChIP-seq. I am a ChIP-seq user and I always use the **MicroPlex kit** for library making from my immunoprecipitated DNA. Not only it's **quick** and requires as little as **50 pg** of input DNA but it also **never fails**. I've made over 100 libraries using the MicroPlex library preparation kit, and none of them failed. I use the Microplex kit for preparing libraries from immunoprecipitated DNA and from genomic DNA with low concentration.

As part of a genomics lab, I regularly prepare libraries from a large number of samples. The **IP-star®** is extremely useful for **high throughput library preparation**. I have prepared 200 genomic libraries in less than 1 month using the IP-star®, it has helped me **economize time and effort** and increase **reproducibility**. I have used the IP-star® for automation of ChIP as well; 16 samples can be processed in one run. In brief, it's a blessing to have this machine if you apply ChIP-seq to a large number of samples.

Zineb Rchiad, Microbial Genomics Laboratory, King Abdullah University of Science and Technology, Kingdom of Saudi Arabia.

References IP-Star® Compact

TE-7 cells

Kottyan LC, Davis BP, Sherrill JD, Liu K, Rochman M, Kaufman K, Weirauch MT, Vaughn S, Lazaro S, Rupert AM, Kohram M, Stucke EM, Kemme KA, Magnusen A, He H, Dexheimer P, Chehade M, Wood RA, Pesek RD, Vickery BP, Fleischer DM, Lindbad R, Sampson HA, Mukkada VA, Putnam PE, Abonia JP, Martin LJ, Harley JB, Rothenberg ME., **Genome-wide association analysis of eosinophilic esophagitis provides insight into the tissue specificity of this allergic disease.**, *Nat Genet.* 2014 Jul 13.

Plants - Arabidopsis

Ariel F, Jegu T, Latrasse D, Romero-Barrios N, Christ A, Benhamed M, Crespi M., **Noncoding Transcription by Alternative RNA Polymerases Dynamically Regulates an Auxin-Driven Chromatin Loop.**, *Mol Cell.* 2014 Jul 9.

Liver

Biswanath Patra, Lakshmi Kuttippurathu, Jan Hoek, Rajanikanth Vadigepalli, **Chronic ethanol intake leads to differential effects on the multivalent epigenetic state in liver [1120.1]**, *April 2014 The FASEB Journal vol. 28 no. 1 Supplement 1120.1.*

Plants – Medicago truncatula

Laloum T, Baudin M, Frances L, Lepage A, Billault-Penneteau B, Cerri MR, Ariel F, Jardinaud MF, Gamas P, de Carvalho-Niebel F, Niebel A., **Two CCAAT box-binding transcription factors redundantly regulate early steps of the legume-rhizobia endosymbiosis.**, *Plant J.* 2014 Jun 13.

Epithelial cells

Lim E, Rothenberg ME., **Demethylation of the Human Eotaxin-3 Gene Promoter Leads to the Elevated Expression of Eotaxin-3.**, *J Immunol.* 2013 Dec 9.

Cells: MCF7, MDAMB231

Dagdemi A, Durif J, Ngollo M, Bignon YJ, Bernard-Gallon D., **Histone lysine trimethylation or acetylation can be modulated by phytoestrogen, estrogen or anti-HDAC in breast cancer cell lines.**, *Epigenomics.* 2013 Feb;5(1):51-63. doi: 10.2217/epi.12.74.

Cell lines

Laget S, Miotto B, Chin HG, Estève PO, Roberts RJ, Pradhan S, Defossez PA. **MBD4 cooperates with DNMT1 to mediate methyl-DNA repression and protects mammalian cells from oxidative stress.** *Epigenetics.* 2014 Apr 1;9(4):546-56.

Human and chimpanzees peripheral blood

Wilson GA, Butcher LM, Foster HR, Feber A, Roos C, Walter L, Woszczek G, Beck S, Bell CG, **Human-specific epigenetic variation in the immunological Leukotriene B4 Receptor (LTB4R/BLT1) implicated in common inflammatory diseases.** *Genome Med.* 2014 Mar 5;6(3):19.

Embryonic stem cells

Veillard AC, Marks H, Bernardo AS, Jouneau L, Laloe D, Boulanger L, Kaan A, Brochard V, Tosolini M, Pedersen R, Stunnenberg H, Jouneau A. **Stable methylation at promoters distinguishes Epiblast Stem Cells from Embryonic Stem Cells and the in vivo epiblast.** *Stem Cells Dev.* 2014 Apr 16.

Human – prostate cancer

Ngollo M, Dagdemir A, Judes G, Kemeny JL, Penault-Llorca F, Boiteux JP, Lebert A, Bignon YJ, Guy L, Bernard-Gallon D, **Epigenetics of prostate cancer: distribution of histone H3K27me3 biomarkers in peri-tumoral tissue.**, *OMICS*, 2014-03-01, 18, 207-9.

Human – blood samples

Bell JT, Loomis AK, Butcher LM, Gao F, Zhang B, Hyde CL, Sun J, Wu H, Ward K, Harris J, Scollen S, Davies MN, Schalkwyk LC, Mill J; MuTHER Consortium, Williams FM, Li N, Deloukas P, Beck S, McMahon SB, Wang J, John SL, Spector TD. **Differential methylation of the TRPA1 promoter in pain sensitivity.** *Nat Commun.* 2014;5:2978. doi: 10.1038/ncomms3978.

Human – embryonic stem cells

Shah SN, Kerr C, Cope L, Zambidis E, Liu C, Hillion J, Belton A, Huso DL, Resar LM, **HMGAI Reprograms Somatic Cells into Pluripotent Stem Cells by Inducing Stem Cell Transcriptional Networks.**, *PLoS One*, 2012-11-15, 7, e48533.

Ordering information

Product	Cat. No.	Format
IP-Star® Compact Automated System	B03000002	1 unit

Visit us at one of Diagenode's demo sites or discover our Automated Systems by performing some assays with the help of our R&D and Technical Department.

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