DNA

For all you seq...

DNA Rearrangements and Markers

RAD
PE RAD-Seq
Genome
Restriction sites
Restriction-site associated DNA marker generation (RAD)
Restriction digestion
Add barcoded adapters
Shear and size select
Add P2 adapter
Amplify
DNA

ddRADseq
Genome
Restriction sites
Double digest restriction-site associated DNA marker generation (ddRADseq)
Restriction digestion
Add barcoded adapters
Second restriction digest
Add P2 adapter
Amplify
DNA

2b-RAD
Genome
BsaXI restriction sites
Restriction-site associated DNA marker generation (RAD) with type IIB restriction endonucleases (2b-RAD)
Restriction digestion with type IIB restriction endonucleases e.g. BsaXI
Add restriction-site-specific adapters
Amplify
Add sample-specific adapters
Amplify
DNA

SLAF-seq
Genome
Restriction sites
Specific locus amplified fragment sequencing (SLAF-seq) for large scale genotyping
Digest with MseI
Add MseI adaptors
Digest with AluI
PCR with barcoded MseI primer
Purify and pool
Add sequencing adaptors
PCR
DNA

hyRAD
Genome
High quality DNA
Hybridization RAD (hyRAD) for degraded DNA
Prepare RAD-seq library and size select
Adaptor removal and labeling
Biotinylated probes
DNA to be captured
Shotgun library
Hybridize to probes
Streptavidin pull down
DNA

Rapture
Genome
Restriction sites
Restriction-site associated DNA marker generation (RAD) capture (Rapture)
Digest with restriction enzyme
Add well-specific barcodes
Well 1
Well 2
Pool wells and shear
Streptavidin pull down
Restriction enzyme digest
Plate 1
Plate 2
Library prep with plate barcodes
Pool
Hybridize Biotinylated capture bait
Streptavidin pull down
DNA

Dige-
nome-seq
Genome
Cas9 target
In vitro Cas9-digested whole-genome sequencing (Digenome-seq)
Deletion
Wild type
Insert
Target site digestion
Sequence
Align and determine sequence breaks

HydEn-seq
Genome
Ribonucleotides
Hydrolytic end sequencing (HydEn-seq) to reveal replicase- and strand-specific patterns of ribonucleotides in the genome
Alkaline hydrolysis (KOH)
2,3'-cyclic phosphate
5'HO
Phosphorylation (T4 PNK 3-phosphatase minus)
5'P-O
Ligate oligo with 5'-amino-terminated C6 spacer
Add sequencing primers, PCR and sequence
Map locations on the genome

Ribose-seq
Genome with ribonucleotides
Detect ribonucleotides embedded in DNA (Ribose-seq)
Fragmented genomic DNA
dA tailing
Adaptor ligation
Alkali treatment
Self-ligation by AtRNL
Degradation of linear ssDNA
Remove 2'-phosphate and PCR
DNA for sequencing

G4-seq
Genome
Potential G-quadruplex
Determine the location of potential G-quadruplexes in DNA (G4-seq)
Target sequence
Fragment and create library on flow cell
Hibridize sequencing primer
Read reference sequence
Denature and remove read treatment
Hibridize sequencing primer
Sequencing Primer
Self ligation by AtRNL
K+ or PDS
Add K+ or PDS to stabilize G4 regions
Polymerase stalls
Read stabilized sequence
Compare sequence reads 1 and 2

CAP-seq
CpG island
Methylated CpG
Unmethylated CpG
CXXC affinity purification plus deep sequencing (CAP-seq)
CXXC bound to nickel-charged sepharose beads
Hybridize to sepharose column
Elute unmethylated CpG enriched fragments
DNA

CPT-Seq
Genome
Contiguity-preserving transposition sequencing (CPT-seq)
Divide sample into 96 reactions
Indexed transposome reactions
EDTA Pool
Dilute and divide into 96 reactions
SDS
Indexed PCR primers
PCR
DNA

RC-Seq
Novel retrotrans-position events
Retrotransposon binding sites
RC-Seq: Retrotransposon capture sequencing
Genomic DNA
Fractionate
DNA fragments
Hybridize
Microarray with transposon binding sites
Read1
Read2
Sequenced fragment
Reference sequence
Transposon sites
Align
Exclusive retrotrans-poson
Novel retrotransposition events

TN-Seq
INSeq
Transposon
Transposon sequencing (TN-Seq) and insertion sequencing (INSeq)
Transposon
Inverted Mmel recognition site
Mmel recognition site
20bp
Mmel
Mmel digestion
Add adapters
PCR and sequence
Transposon insertion sites

TC-Seq
AID-dependent rearrangement
I-SceI site
+AID
Genomic DNA
-AID
Infect I-SceI
Sonicate, blunt and A-tail
Ligate linkers Cut I-SceI
Purification
Semi-nested PCR Linker cleavage
DNA

Ig-seq
Rep-Seq
MAF
CDR3 junction region
V D J Constant
DNA sequencing of immunoglobulin genes (Ig-seq), repertoire sequencing (Rep-Seq), and molecular amplification fingerprinting (MAF)
Extracted RNA
Reverse transcription
6N UID
Second strand synthesis
PCR
Purify
DNA

SSB-seq
Single-strand break
Map DNA single-strand breaks (SSB-Seq)
Genome DNA
DNA Pol I, dU-digoxigenin, and dNTPs
DNA isolation and random shearing
Immunoprecipitate with anti-digoxigenin antibody
DNA

BLESS
Double-strand break
Breaks labeling and enrichment on streptavidin and sequencing (BLESS)
Genome DNA
Biotinylated proximal primer
Ligate primer
DNA isolation and random shearing
Capture on streptavidin beads
Distal primer
Digest with I-SceI and PCR
DNA

DSB-seq
Double-strand break
Map DNA double-strand breaks (DSB-Seq)
Genome DNA
TdT and biotinylated-dUTP
DNA isolation and random shearing
Capture on streptavidin beads
DNA

GUIDE-seq
Oligo insert
dsOligo insert
Genome-wide, unbiased identification of DSBs enabled by sequencing (GUIDE-seq)
Genome DNA
dsOligo tag integrated in live cells
DNA isolation Random shearing
UMI
End repair Adaptor ligation
PCR
P5
Index 2
Index 1
P7
Oligo tag
dsOligo-specific amplification
DNA

EC-seq
V(D)J antigen receptor region
Excision circle sequencing (EC-seq)
Lymphocyte-specific recombination activating gene (RAG) recombinase, bound genome
Synapsis, cleavage and coding end hairpin formation
Resolved genomic coding junction
Liberated excision circle with resolved signal junction
Align read pairs to reference genome

Bubble-Seq
Libraries of restriction fragments that contain replication initiation sites (bubbles) in vivo
Bubble-containing fragment
Restriction digest
Cast fragments in trapping gel
Run gel
Recover bubble-containing plug
DNA extraction
Add sequencing primers
DNA

NSCR
Nascent strand capture and release (NSCR)
DNA replication bubble
RNA primer
gDNA
SNS
Denaturation and size-selection
Biotin
5'-biotinylation
Streptavidin pull-down
RNase I digestion
Isolate and amplify SNS
DNA

Repli Seq
Nascent DNA
BrdU
Repli Seq—to map temporally ordered replicating DNA
Run-on with analog
Sort cells and lyse
Bead coated with anti-BrdU antibody
Elute
End Repair
Purify
DNA

NS-seq
Nascent DNA
Nascent strand sequencing (NS-seq) to discover DNA replication origins and G4 structures
DNA replication origin
Lambda 5' to 3' exodeoxyribonuclease (λ-exo)
Digestion
Purify
Amplify
DNA

DNA Low-Level Detection

smMIP
Gene
Degenerate molecular tag
Single Molecule Molecular Inversion Probes (smMIPs) for detecting low frequency targets
Genomic DNA
Copy target sequence
Exonuclease
PCR amplification
Read1
Sample index
Read2
Random error
True variant
Align fragments from every unique molecular tag
Corrected sequence

MIPSTR
Short tandem repeat (STR)
Degenerate molecular tag
Targeted capture of STR loci by smMIPs (MIPSTR)
Targeted STR
Copy target STR
Amplify and sequence
Strain 1
Strain 2
Natural variation between individuals
Strain 1
Strain 2
Somatic variation within an individual

MDA
IMS-MDA
MIDAS
Genome
3' blocked random hexamer primers
Multiple displacement amplification (MDA). Immunomagnetic separation for targeted bacterial enrichment for MDA (IMS-MDA) Microwell displacement amplification system (MIDAS)
Hybridize primers
Phi 29
Synthesis
Nascent replication fork
Phi 29
Synthesis
S1 nuclease
Amplified DNA

MALBAC
Genome
27-bp common sequence
8 random nucleotides
Multiple annealing and looping-based amplification cycles (MALBAC)
Hybridize primers
Bst DNA polymerase
Denature
Hybridize primers
Partial amplicons
Cycles of quasilinear amplification
Synthesis
Denature
Template
Looped full amplicons
PCR
DNA

nuc-seq
SNES
Single cell genome
Single G2/M nucleus sequencing of cells in S phase (nuc-seq). Single nucleus exome sequencing (SNES)
Cell 1
Cell 2
Cell 3
Cell sorting from CVM distribution
Lyse cell
Nucleus
Phi 29
Limited amplification
Synthesis
S1 nuclease
DNA

OS-Seq
Gene
Target sequence
Fragment and add single adaptors
Single adaptor library
Oligonucleotide-selective sequencing (OS-Seq) captures and sequence gene targets on the flow cell
Target sequence
Adaptor sequence
Flow cell
Create target-specific oligos
Extend and Denature
Hybridize
Extend and Denature
Hybridize
Extend and Denature
Sequencing Primers
Sequence
Sequence reads 1 and 2

Safe-SeqS
Very rare mutation
Mutation
Safe-sequencing system is a unique molecular identifier (UMI) approach to detect rare variants (Safe-SeqS)
DNA
Shear
Randomly sheared ends serve as UMIs
Adaptor ligation
Amplify and solid phase capture
Sequence
Align sequences and determine actual ratio
True mutant

Duplex-Seq
Very rare mutation
Duplex sequencing detects rare mutations by sequencing and aligning both strands of the DNA
A mutation occurs on both strands
Add adaptors
P5
α
12 random base index
P7
Ligate and PCR
β
12 random base index
P7
P5
Sequence
Random error
True variant
Consensus
Create single strand consensus sequence from every unique molecular tag
Create duplex sequences based on molecular tags and sequencing primers
Rare variant

DR-Seq
Single cell
RNA
DNA
Genome DNA and mRNA sequencing (DR-Seq)
Single cell
Lyse cell
RT with barcoded primer
Ad-2 primer
Quasilinear amplification
2nd strand synthesis
PCR and Remove adaptors
Split samples
cDNA amplification
gDNA amplification
Sequence

G&T-seq
Single cell
RNA
DNA
Genome and transcriptome sequencing from a single cell (G&T-seq)
Cell suspension
Isolate single cell
Lyse cell
Streptavidin magnetic bead with mRNA capture primer
Separate the DNA and the RNA
On-bead transcriptome amplification with Smart-seq2
Whole genome amplification with MDA
Sequence
Align RNA and genome

Key

Yellow highlights indicate the target of the protocol

Bisulfite conversion
Cytosine
5-Methyl Cytosine
Uracil
5-Methyl Cytosine
Display methods on mobile device
Cytosine
5-Methyl Cytosine
5-hydroxymethylcytosine (5hmc)
5-formylcytosine (5fC)
5-carboxylcytosine (5caC)
Pyridostatin (PDS)
Digoxigenin
AI-mediated cyclization labeling in fC-CET
5fC
5fC-AI
5fC-AI-SH
EDTA

Epigenetics

BS-Seq
Bisulfite-seq
WGBS
Bisulfite conversion of genomic DNA (Bs-Seq) or whole-genome bisulfite sequencing (WGBS)
Methylated DNA
Shear DNA
Bisulfite conversion
Biotin
DNA

EpiGnome™
HELP-Seq
Bisulfite conversion of genomic DNA without shearing. HpaII tiny fragment enrichment by ligation-mediated PCR (HELP-Seq)
Methylated DNA
Bisulfite conversion
Converted single-stranded fragments
Random priming DNA synthesis
3' tagging
PCR
DNA

PBAT
Post-bisulfite adaptor tagging (PBAT)
Methylated DNA
Bisulfite conversion
First random priming
Biotin
Adaptor
Capture first strand on Streptavidin-coated magnetic beads
Streptavidin
Random primer 2
Adaptor
Second random priming
Generate second strand
Elution
DNA with adaptors

BSPP
Bisulfite sequencing with padlock probes (BSPP)
CpG island
Bisulfite conversion
Bisulfite-converted DNA
Padlock probe
MseI AluI MseI
Hybridize
Extension and ligation
Exonuclease digestion
PCR
End repair and adaptor ligation
DNA with adaptors

RRBS
scRRBS
Reduced representation bisulfite sequencing (RRBS-Seq). Single cell RRBS (scRRBS)
Methylated DNA
MspI digestion
Methylated regions
Methylated adapter
End repair and ligation
Bisulfite conversion
Converted fragments
PCR
DNA

BSAS
Bisulfite amplicon sequencing (BSAS)
Methylated DNA
Bisulfite conversion
Bisulfite-converted DNA
PCR with primers specific for bisulfite converted DNA
Amplicons
Transposome with adaptor
Tagmentation
DNA

Methyl-Seq
MRE-Seq
Methyl-seq and MRE-Seq use methyl-sensitive enzymes to identify methylation patterns
Methylated sites in the genome
Split sample
MspI
HpaII
Restriction enzyme digest
Sequence
Align sequences and determine undigested sites
Identified methylation site

EpiRADseq
Genome
Restriction sites
Double digest restriction-site associated DNA marker generation (ddRADseq) with a methylation-sensitive restriction enzyme ((EpiRADseq)
Restriction digestion
Add barcoded adapters
Second restriction digest with methylation-sensitive HpaII
Add P2 adapter
Amplify
DNA

T-WGBS
Tagmentation-based whole-genome bisulfite sequencing (T-WGBS)
Methylated DNA
Transposome with methylated adaptor
Tagmentation
Oligo with methylated adaptor
Displaced oligo
Displace oligo Hybridize methylated adaptor and gap repair
Bisulfite conversion
PCR
DNA

JBP1-seq
5hmc residues
J-binding protein 1 sequencing (JBP1-seq), for genome-wide profiling of 5-hydroxy-methylcytosine (5hmC)
Hydroxy-methylated DNA
Transposomes
Tagmentation
T4-βGT
Glucosylated 5-hmC
JBP1-magnetic bead pull down
PCR
DNA

Aba-seq
5hmc residues
AbaSI coupled with sequencing (Aba-seq) to map high-resolution hydroxymethylome (5hmC)
Hydroxy-methylated DNA
T4-βGT
Glucosylated 5-hmC
AbaSI
Biotinylated primers
Ligate
Fragment
Streptavidin magnetic bead pull down
DNA

TAmC-Seq
5mc residue
Tet-assisted 5-methylcytosine sequencing (TAmC-Seq)
5mC
5hmC
βGT
Glucosylation
TET
Oxidation
Oxidation
βGT-catalyzed UDP-6-N3 glucosylation
Biotinylation
Streptavidin pulldown and DTT cleavage
DNA

fC-Seal
5fc residue
A 5-formylcytosine-selective chemical labeling (fC-Seal) approach for genome-wide profiling of 5fC
βGT-catalyzed glucosylation
NaBH4
Convert 5fc to 5hmc
βGT-catalyzed UDP-6-N3 glucosylation
Biotinylation
Streptavidin pulldown and DTT cleavage
DNA

fC-CET
5fc residue
5fC based on selective chemical labeling of 5fC and subsequent C-to-T transition during PCR
Azido 1,3-indandione (AI)
DBCO-S-S-PEG3-biotin
Pulldown, NaOH and DTT
Adaptor ligation and PCR
Purify
DNA

CAB-Seq
5caC residue
Chemical modification-assisted bisulfite-sequencing (CAB-Seq) for 5caC detection
1-ethyl-3-(3-dimethylamino-propyl)-carbodiimide hydrochloride (EDC) chemistry
α-effosuorra
Nu
S-S
Linker
Biotin
5caC-S-S
Streptavidin pulldown and DTT cleavage
Control
5caC-S
Bisulfite treatment PCR amplification
Sequence

oxBS-Seq
5hmc residue
Oxidative bisulfite sequencing (oxBS-Seq) to map 5-methylcytosine and 5-hydroxymethylcytosine
Control
KRuO4
Bisulfite treatment PCR amplification
DNA

RedBS-Seq
caMAB-seq
5fc residue
Reduced bisulfite sequencing (redBS-Seq), to map 5-formylcytosine (5fC) in DNA
Control
NaBH4
Bisulfite treatment PCR amplification
DNA

fCAB-Seq
5fc residue
5fC chemically assisted bisulfite sequencing (fCAB-seq) method for the base-resolution detection of 5fC
Control
O-ethylhydroxylamine (EtONH2)
Blocked
Bisulfite treatment PCR amplification
DNA

MAB-seq
5fC/5caC residues
M.SssI methylase-assisted bisulfite sequencing (MAB-seq) to map 5fC/5caC
Control
M.SssI
Bisulfite treatment PCR amplification
DNA

RRMAB-seq
5fC/5caC residues
Reduced representation M.SssI methylase-assisted bisulfite sequencing (RRMAB-seq)
Digest with MspI
Add methylated adapters
Control
M.SssI
Bisulfite treatment PCR amplification
Bisulfite treatment PCR amplification
DNA

TAB-Seq
5hmc residue
TET-assisted bisulfite sequencing, (TAB-Seq) to map 5-hydroxymethylcytosine
βGT
Glucosylation
TET
Oxidation
Bisulfite treatment PCR amplification
DNA

MIRA
5mc CpG island
Methylated-CpG island recovery assay (MIRA)
Fractionate
MBD2B/MBD3L1 protein complex
Isolate on glutathione-coated beads
DNA purification PCR amplification
DNA

MeDIP-Seq
DIP-seq
Methylated DNA Immunoprecipitation (MeDIP-Seq). DNA immunoprecipitation followed by high throughput sequencing (DIP-seq))
Methylated DNA
Extract DNA
Fractionate Denature
Immunoprecipitate
DNA purification
DNA

hMeDIP-Seq
Hydroxymethylated DNA immunoprecipitation combined with next generation DNA sequencing (hMeDIP-seq)
Hydroxymethylated DNA
Extract DNA
Fractionate Denature
Immunoprecipitate
DNA purification
DNA

MBDCap-seq
Methyl-Cap-seq
MBD-Seq
MiGS
Methyl CpG binding domain-based capture and sequencing (MBDCap-seq). Capture of methylated DNA using the MBD domain of MeCP2 (Methyl-Cap-Seq). MBD-isolated Genome Sequencing (MiGS)
Methylated DNA
Extract DNA
Fractionate
MBD
Biotin
Streptavidin
Capture biotinylated MBD on Streptavidin coated magnetic beads
Elute with increasing salt concentration
DNA purification
DNA

BisChIP-Seq
ChIP-BS-seq
Bisulfite-treated chromatin immunoprecipitated DNA (BisChIP-seq) and ChIP-BS-seq), to correlate protein modifications with DNA methylation
DNA-protein complex with methylated histones and methylated DNA
Sonicate
Immunoprecipitate
Purify DNA and shear
Bisulfite conversion
DNA

Methidiumpro-pyl-EDTA (MPE)
DNA + Acylation
DNA
DIBO-biotin "click"
DNA
Biotin
Preparation of acylated RNA for biotin–streptavidin purification. DIBO, dibenzocycloextyne
Uridine diphosphate glucose (UDP-Glu)
Glucosylated 5hmc
N3-SGMC
Pyridine
Display methods on mobile device
Biotin

DNA-Protein Interactions

ChIP-Seq
ChIP-exo
HT-ChIP
Mint-ChIP
Chromatin immune precipitation (ChIP-Seq). High-throughput chromatin immunoprecipitation (HT-ChIP))
DNA-protein complex
Crosslink proteins and DNA
Sample fragmentation
Exonuclease digestion
Immunoprecipitate
DNA extraction
DNA

DNase-Seq
DNaseI-Seq
DNase I hypersensitive sites sequencing (DNase-Seq, DNaseI-Seq)
Active chromatin
DNaseI digestion
Isolate trimmed complexes
DNA extraction
DNA

DNaseI-SIM
DNase I simplified in-nucleus method (DNase I SIM) for plants
Cells
Lyse and centrifuge
Supernatant
Nuclei
Sort nuclei
Nucleus
DNaseI digestion
Terminate DNase I digestion
Polish ends
DNA extraction
DNA

MAINE-Seq
MNase-Seq
Nucleo-Seq
MNase-assisted isolation of nucleosomes (MAINE-Seq). Also Micrococcal nuclease sequencing (MNase-Seq)
Open chromatin
MNase digestion
Isolate trimmed complexes
DNA extraction
DNA

FiT-seq
Fixed-tissue chromatin immunoprecipitation sequencing (FiT-Seq)
FFPE-fixed chromatin
Deparaffination and rehydration
Heat 40˚C in SDS
Proteinase K digestion
Enzyme inactivation
Sonicate
Soluble extract
Immunoprecipitate
Cromatin elution
Purification
DNA

PAT-ChIP
Pathology tissue chromatin immunoprecipitation (PAT-ChIP)
FFPE-fixed chromatin
Deparaffination and rehydration
MNase digestion
Sonicate
Soluble extract
Immunoprecipitate and chromatin elution
Reverse crosslink and proteinase K digestion
Purification
DNA

X-ChIP
High resolution of mapping of in vivo chromatin associated proteins (X-ChIP)
Crosslink cells in vivo
Lyse cells
Cross-linked chromatin
MNase digestion
Sonicate
Soluble extract
Immunoprecipitate and DNA extraction
DNA

ORGANIC
Occupied regions of genomes from affinity-purified naturally isolated chromatin (ORGANIC)
Cell
Isolate nuclei
Isolated chromatin
MNase digestion
Soluble extract
Immunoprecipitate and DNA extraction
DNA

ATAC-Seq
Assay for transposase accessible chromatin (ATAC-Seq)
Open DNA
Tn5 Transposome
Insert in regions of open chromatin
Fragmented and primed
DNA purification Amplification
DNA

THS-seq
Transposome hypersensitive site sequencing (THS-seq)
Open DNA
Tn5 Transposome
Read primer
T7 promoter
Insert in regions of open chromatin
Purify
End fill-in and IVT amplification
dsDNA synthesis
Add barcodes and sequencing adaptors
DNA

CATCH-IT
Covalent attachment of tags to capture histones and identify turnover (CATCH-IT)
Cells starved of methionine
Add methionine analogue l-azidohomoalanine (AHA)
Isolate nuclei
Cycloaddition reaction
Biotin
MNase digestion
Streptavidin
Remove of H2A–H2B dimers and non-histone proteins
DNA extraction
DNA

MINCE-seq
Mapping in vivo nascent chromatin with EdU and sequencing (MINCE-seq)
Label with ethynyl deoxyuridine (EdU)
Chase with Thymidine
Newly-formed chromatin
Polymerase
Crosslink with formaldehyde
Click reaction to attach biotin
Sonication and MNase digestion
Streptavidin capture
DNA extraction
DNA

FAIRE-seq
Sono-Seq
Formaldehyde-assisted isolation of regulatory elements (FAIRE-Seq) and sonication of cross-linked chromatin (Sono-Seq)
Open DNA
Crosslink protein and DNA with formalin
Sonicate
Phenol extract and purify DNA from the aqueous phase
DNA

NOMe-Seq
Nucleosome Occupancy Methylome-sequencing (NOMe-Seq), a single-molecule nucleosome positioning assay
Open DNA
CpG dinucleotides Methylated CpG
GpC methyltransferase (M.CviPI) and S-Adenosylmethionine (SAM)
Bisulfite conversion BS-Seq
Control
Protected
Protected
I Protected methylated
I Unprotected methylated
M.CviPI
I Protected unmethylated
I Unprotected unmethylated

Histone methylation
ChIP-Seq of methylated histones (Histone methylation)
DNA-protein complex
Crosslink proteins and DNA
Sample fragmentation
Exonuclease digestion
Immunoprecipitate
DNA extraction
DNA

ChIPmenta-tion
Chromatin Immunoprecipitation with sequencing library preparation by Tn5 transposase (ChIPmentation)
Chromatin
Chromatin immunoprecipitation
Tn5 Transposase
Adaptor insertion
Fragmented and primed
DNA purification Amplification
DNA

Chia-PET
Chromatin interaction analysis by paired-end tag sequencing (ChIA-PET)
Sample fragmentation
Immunoprecipitate
Ligation
Restriction enzyme digestion
DNA

Hi-C
3-C
Capture-C
Chromatin conformation capture (3-C, Hi-C and Capture-C)
Crosslink proteins and DNA
Sample fragmentation
Ligation
PCR amplify ligated junctions
DNA

NG Cap-ture-C
Next-generation Capture-C (NG Capture-C)
Formaldehyde fixation
Restriction enzyme digestion
Ligation
De-crosslink and extract DNA
Sonicate to 200 bp fragments
Add indexed sequencing adaptors
PCR
Hybridize biotinylated capture bait
Streptavidin pull down
DNA

4-C
Chromatin conformation capture circular (4-C)
Crosslink proteins and DNA
Sample fragmentation
Ligation
Restriction digest
Self-circularization and Reverse PCR
DNA

UMI-4C
Targeted chromosome conformation capture (4C) with unique molecular identifiers (UMI-4C)
Crosslink proteins and DNA
HC DpnII digestion
Ligation
Reverse crosslink and proteinase K digestion
Sonicate
Single end adaptor ligation
Adaptor
US primer
DS primer
Universal primer
Index
Pad
Nested PCR amplification
DNA

5-C
Chromatin conformation capture carbon copy (5-C)
Crosslink proteins and DNA
Sample fragmentation
Ligation
LMA: Ligation-mediated amplification
DNA

PB-seq
Protein/DNA binding (PB-seq), to determine the binding energy landscape
DNA-protein complex
Purify DNA
Shear DNA
Hybridize with DNA-binding protein
Isolate protein-bound DNA
DNA extraction
DNA

Pu-seq
Origin of replication
Polymerase usage sequencing (Pu-seq)
Alkali treatment
Klenow reaction(+ random primer, dATP, dGTP,dCTP, dUTP)
Index 2
Index 1
Attach adaptors
Uracil DNA glycosylase-and DNA lyase (USER)
PCR and purify
DNA

Break-seq
Breakage at replication fork
Double-stranded break labeling to map chromosome breaks (Break-seq)
Break
DNA trapped in agarose gel
End-repair with dGTP, dCTP, dTTP, and biotinylated-dATP
Elution and fragmentation
Streptavidin magnetic bead pull-down
Index 2
Index 1
Attach adaptors
PCR and purify
DNA

LAM-HTGTS
Breakage in genome
Linear amplification-mediated high-throughput genome-wide sequencing (LAM-HTGTS)
Fragmentation by sonication
LAM-PCR with biotinylated primer
Streptavidin magnetic bead pull down
Adaptor ligation
Nested PCR
Enzyme blocking
Tagged PCR
Purify
DNA

HTGTS
Breakage in genome
Linear amplification-mediated high-throughput genome-wide sequencing (HTGTS)
Fragmentation by sonication
End repair
3' A addition
Adaptor ligation and PCR
Streptavidin magnetic bead pull-down
Nested PCR
Tagged PCR
Purify
DNA

SELEX
SELEX-seq
HT-SELEX
Transcription factor binding site
High-throughput systematic evolution of ligands by exponential enrichment (HT-SELEX)
Forward sequencing adaptor
Sequencing primer
Index for multiplex sample
14N with all possible combinations
Reverse sequencing adaptor
pGAWHISELEX
DNA binding region
Luciferase
Streptavidin binding peptide
Expression vector
Target sequence
Ligand
Fusion protein
Luciferase
Fusion protein immobilized in well
Matching ligand binds
Wash and elute
Recovered matching ligand
PCR and Sequence
Binding site

HiTS-FLIP
Protein binding site
High-throughput sequencing-fluorescent ligand interaction profiling (HiTS-FLIP)
Target sequence
Flowcell
Hybridize primer
Klenow
Synthesize second strand with unmodified nucleotides
Binding site
Add fluorescently labeled ligand
Hybridize
Elute with increasing stringency
Scan flowcell

DamID
DNA-protein interaction
DNA adenine methyltransferase (DAM)
Protein of interest
Fusion protein
DAM
Specific and non-targeted methylation
Split sample
Non-targeted methylation
DNA adenine methyltransferase interaction detection (DamID)
Create fusion protein
DpnI digestion
DpnII
PCR
DpnI digestion
Adaptor ligation
Unmethylated GATCs are cut by DpnII
Align sequences and determine differentially digested sites

MPE-seq
Methidiumpropyl-EDTA sequencing (MPE-seq)
Active chromatin
Isolated nuclei
MPE digestion
Add bathophenanthroline
DNA extraction
DNA

Chem-seq
Identify sites bound by small chemical molecules (Chem-seq)
DNA-protein complex with putative drug binding site
In vivo
In vitro
Crosslink
Add biotinylated compound
Harvest cells and fragment DNA
Streptavidin
Add biotinylated compound
Enrich DNA fragments
DNA extraction
DNA

Protein-Protein Interactions

PD-Seq
Protein target
PD-Seq identifies candidate cellular targets for proteins
Pyridine
Beads
Pyridine
Polyethylene glycol linker added
K2CO3
DMF
Immobilized protein
RNA from cells
Create phage display cDNA library
Hybridize
Wash
Repeat cycle three times
Protein-protein interaction
PCR phage insert
DNA

ProP-PD
PDZ-Seq
Protein target
Proteomic peptide-phage display (ProP-PD) to identify short linear motif (SLiM) interactions or PDZ domains (PDZ-Seq)
Identify C-terminal sequences
Create oligo library
Construct phage display library
Bait proteins immobilized on 96-well plate
Hybridize
Select phages against baits
Isolate and sequence
Peptide counts

Nucleic Acid Purification

Nucleic Acid Purification
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Nucleic Acid Purification
Nucleic Acid Purification
Nucleic Acid Purification
Nucleic Acid Purification
Nucleic Acid Purification
Nucleic Acid Purification

TruSeq PCR Free

Double-stranded DNA
Fractionate Size select
End repair Phosphorylate
A-overhang
Add Adaptors
Adaptor ligation
Product ready for cluster generation

TruSeq Nano

Double-stranded DNA
Fractionate Size select
End repair Phosphorylate
A-overhang
Add Adaptors
Adaptor ligation
Denature and amplify
Product ready for cluster generation

TruSeq Custom Amplicon

Double-stranded DNA
Region of interest
Custom Probe 1
Custom Probe 2
Add custom probes
Extension and ligation
Add sequencing primers
PCR
Product ready for cluster generation

TruSeq RNA

mRNA
polyA select
Fragment
Random hexamer
First and second strand synthesis

TruSeq Small RNA

Small RNA fragment
Ligate adaptors
3' Adapter
5' Adapter
Add primer
Reverse transcription
Denature and amplify
Product ready for cluster generation

TruSeq RNA Stranded

RNA
Random primer
Create cDNA
dTTP + dCTP + dATP + dGTP
dUTP + dCTP + dATP + dGTP
Create second strand cDNA
End repair Phosphorylate A-overhang
Adaptor ligation
Denature and amplify
Product ready for cluster generation

TruSeq RNA Access

Target
Pool stranded RNA-Seq libraries
Biotinylated target probe
Hybridize probes to targets
Capture on streptavidin magnetic beads
Elute
Product ready for cluster generation

TruSeq Targeted RNA Expression

Target
Total RNA
cDNA
Add custom primers
Hybridization
Extension-Ligation
Denature and amplify
Amplification
Product ready for cluster generation

Nextera Library Preparation

Transposase
DNA
Tagmentation
~300bp
Amplification
Product ready for cluster generation

Nextera Rapid Capture

Target
Denatured and pooled fragments from Nextera library
Biotinylated target probe
Hybridize probes to targets
Capture on magnetic beads
Elute
Product ready for cluster generation

Nextera Mate Pair

Transposase
Biotinylated junction adaptor
DNA
Tagmentation
Circularize
Fragment
Isolate biotinylated fragment
Adaptor ligation
Denature and amplify
Product ready for cluster generation

Sequencing by Synthesis

Adapter hybridizes to flowcell
Reverse strand synthesis
Forward strand
Reverse strand
Remove forward strand
Fold over and hybridize to second primer
Synthesize second strand
Bridge amplification
Thousands of molecules are amplified in parallel
Sequence primer
The reverse strand is cleaved and washed away
With each cycle, four fluorescently tagged nucleotides compete for addition to the growing chain. Only one is incorporated based on the sequence of the template.
The read product is washed away
Index 1 primer
Sequence Index1
The read product is washed away
Fold over and hybridize to first primer
Deblock P5 primer and add unlabeled bases
Sequence Index2
Synthesize second strand
The forward-strand is cleaved and washed away
Read 2 primer
The second read is sequenced

Moleculo

~10kb Sheared genomic DNA
End repair
Adapter ligation
Generate clonal pools
Amplify
Transposase
Tagmentation
~600bp
Add indices
Prepared fragments
Pool and purify

References

2b-RAD
3C
4-C
5-C
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ATAC-Seq
BisChIP-Seq
Bisulfite-seq
BLESS
Break-seq
BSAS
BSPP
BS-Seq
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CAB-Seq
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CAP-seq
Capture-C
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Dnase I SIM
DNaseI-Seq
DNase-Seq
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DR-Seq
DSB-seq
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fC-CET
fC-Seal
FIT-Seq
G&T-seq
G4-seq
GUIDE-seq
HELP-Seq
Hi-C
Histone meth.
HiTS-Flip
HMeDIP-seq
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hyRAD
Ig-Seq
IMS-MDA
INSeq
JBP1-seq
LAM-HTGTS
MAB-seq
MAF
MAINE-Seq
MALBAC
MBDCap-Seq
MBD-Seq
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