abcam

Product datasheet

High-Sensitivity ChIP Kit ab185913

30 References 3 Images

Overview

Product name

Sample type

Species reactivity

Product overview

High-Sensitivity ChIP Kit

Tissue, Adherent cells, Suspension cells

Reacts with: Mammals

High-Sensitivity ChIP Kit (ab185913) is a complete set of optimized reagents to carry out a successful chromatin immunoprecipitation procedure in a high throughput format starting from mammalian cells or tissues. The highly specific and sensitive kit is suitable for selective enrichment of a chromatin fraction containing specific DNA sequences using various mammalian cell/tissues. The optimized protocol and kit components reduce non-specific background ChIP levels to allow capture of low abundance protein/transcription factors and increased specific enrichment of target protein/DNA complexes. The target protein bound DNA prepared with the High-Sensitivity ChIP Kit can be used for various downstream applications including PCR (ChIP-PCR), microarrays (ChIP-on-chip), and sequencing (ChIP-seq).

Starting Materials

Starting materials can include various tissue or cell samples such as cells from flask or plate cultured cells, fresh and frozen tissues, etc. In general, the amount of cells and tissues for each reaction can be 2×10^3 to 1×10^6 and 0.5 mg to 50 mg, respectively. For optimal preparation, the input amount should be 1-2 x 10^5 cells or 10-20 mg tissues since the enrichment of target proteins to genome loci may vary. For the target proteins that are low abundance transcription factors, the input amount should be 5-6 x 10^5 cells or 50 to 60 mg tissues.

Primers

The GAPDH primers provided with the kit are for the human sequence. If using the kit with a difference species, GAPDH primers for that species will need to be acquired.

Protein-DNA interaction plays a critical role for cellular functions such as signal transduction, gene transcription, chromosome segregation, DNA replication and recombination, and epigenetic silencing. Identifying the genetic targets of DNA binding proteins and knowing the mechanisms of protein-DNA interaction is important for understanding cellular processes. Chromatin immunoprecipitation (ChIP) offers an advantageous tool for studying such protein-DNA interactions. It allows for the detection of a specific protein bound to a specific gene sequence in living cells using PCR (ChIP-PCR), microarrays (ChIP-chip), or sequencing (ChIP-seq). For example, measurement of the amount of methylated histone H3 at lysine 9 (meH3-K9) associated with a specific gene promoter region under various conditions can be achieved through a ChIP-PCR assay, while the recruitment of methylated H3-K9 to the promoters on a genome-wide scale

Notes

can be detected by ChIP-on-chip or ChIP-sequencing. ChIP analysis requires that ChIPed DNA contains minimal background in order to reliably identify true TF-enriched regions. High background in ChIP is mainly caused ineffective wash buffers, insufficient cross-link reversal, inappropriate DNA fragment length, and residual RNA interference. To effectively capture TF/DNA complexes, which are often in low abundance, an ideal ChIP method requires having maximum sensitivity with minimized background levels. This method should also be able to enrich highly abundant protein/DNA complexes using a small amount of cells or tissues in a high throughput format. The High-Sensitivity ChIP Kit is designed to achieve these goals by maximizing sensitivity and minimizing non-specific background signals.

ChIP assay products and guides

Find more <u>ChIP assay / chromatin immunoprecipitation</u> resources and products, <u>ChIP antibody</u> products, and other <u>ChIP assay kits and related reagents</u>.

Tested applications

Suitable for: ChIP

Properties

Storage instructions

Please refer to protocols.

Components	24 tests	48 tests
1000X Protease Inhibitor Cocktail	1 x 30µl	1 x 60µl
8-Well Assay Strips (with Frame)	3 units	6 units
8-Well Strip Caps	3 units	6 units
Adhesive Covering Film	1 unit	2 units
Antibody Buffer	1 x 3ml	1 x 6ml
Anti-RNA Polymerase II	1 x 8µl	1 x 16µl
Blocker Solution	1 x 2ml	1 x 4ml
ChIP Buffer	1 x 6ml	1 x 12ml
DNA Binding Solution	1 x 7ml	1 x 14ml
DNA Elution Buffer	1 x 1ml	1 x 2ml
DNA Release Buffer	1 x 8ml	1 x 16ml
Enrichment Enhancer	1 x 55µl	1 x 110µl
F-Collection Tube	30 units	50 units
F-Spin Column	30 units	50 units
GAPDH Primer - Forward (20 μM)	1 x 8µl	1 x 16µl
GAPDH Primer - Reverse (20 μM)	1 x 8µl	1 x 16µl

Components	24 tests	48 tests
Lysis Buffer	1 x 14ml	1 x 28ml
Non-Immune IgG (1 mg/ml)	1 x 10µl	1 x 20µl
Proteinase K (10 mg/mL)	1 x 60µl	1 x 120µl
Rnase A	1 x 30µl	1 x 60µl
Wash Buffer	1 x 25ml	2 x 25ml

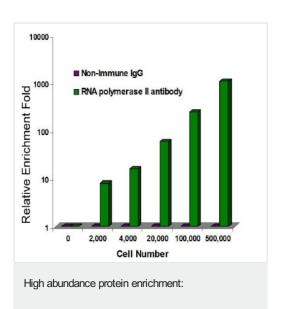
Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab185913 in the following tested applications.

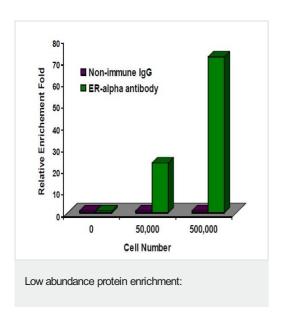
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ChIP		Use at an assay dependent concentration. The GAPDH primers provided with the kit are for the human sequence. If using the kit with a different species, GAPDH primers for that species will need to be acquired.

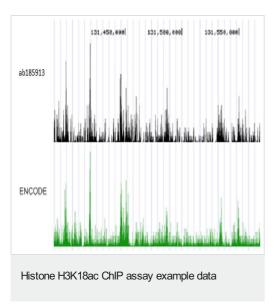
Images



Sheared chromatin isolated from different numbers of MBD-231 cells was used for ChIP-qPCR analysis of RNA polymerase II enrichment in GAPDH promoters using ab185913 and a quantitative PCR Fast Kit.



Sheared chromatin isolated from different numbers of MCF7 cells was used for ChIP-qPCR analysis of ER-a enrichment in TFF1 promoters using ab185913 and a quantitative PCR Fast Kit.



Histone H3K18ac ChIP assay was carried out using ab185913.

ChIP-Seg reads align with the same peak sites as ENCODE data.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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