

ChIP-IT[®] Control qPCR Kit – Human

Catalog No: 53026

Format: 5 rxns

Quality Control: ChIP-IT[®] Control qPCR Kit – Human is quality control tested in combination with Active Motif's ChIP-IT[®] Express Kit (Catalog No. 53008).

Chromatin from a human sample is fixed and sheared as described in the ChIP-IT Express manual. 5 µg of the prepared chromatin is used for each ChIP reaction. Positive control ChIP reactions are performed in triplicate using 2 µg RNA pol II antibody plus 2 µg Bridging antibody, while negative control ChIP reactions are performed in triplicate using 2 µg negative control antibody. The immunoprecipitated DNA and the Input DNA are used in qPCR with both GAPDH-2 and Negative-1 qPCR primer sets.

To pass QC, the RNA pol II ChIP reactions should produce at least 5-fold enrichment with the GAPDH-2 primer set as compared to the Negative-1 primer set. The negative control ChIP reaction should not show enrichment greater than half the RNA pol II enrichment with the GAPDH-2 primer set (Figure 1).

Contents:

RNA pol II mouse monoclonal antibody (0.2 µg/µl) (also sold as Catalog No. 39097)

Bridging antibody (1 µg/µl) (also sold as Catalog No. 53017)

Negative control mouse IgG (0.2 µg/µl)

GAPDH-2 qPCR primer set (2.5 µM) (also sold as Catalog No. 71006)

Negative-1 qPCR primer set (2.5 µM) (also sold as Catalog No. 71001)

qPCR Analysis

We recommend the following qPCR conditions as a starting point. Please optimize for your qPCR machine and master mix as needed.

SYBR green Master Mix	5 µl
PCR primer mix (2.5 µM)	0.7 µl
Sterile water	1.3 µl
ChIP DNA	3 µl
Total Volume	10 µl

95°C for 2 min

40 cycles as follows:

95°C for 15 sec

58°C for 20 sec

72°C for 20 sec

Storage and Guarantee: The ChIP-IT[®] Control qPCR Kit – Human components are shipped on dry ice. The negative control IgG antibody should be stored at 4°C, all other components can be stored at -20°C. This product is guaranteed for 6 months from date of receipt under the correct storage conditions. Aliquot the antibodies to avoid exposing to multiple freeze-thaw cycles.

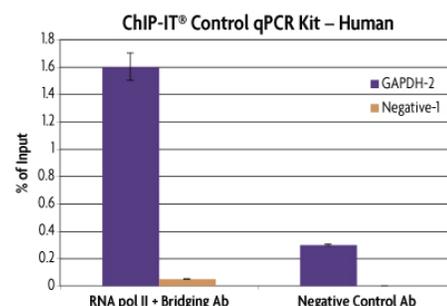


Figure 1: qPCR analysis of 5 µg human myeloma LP1 chromatin assayed using the ChIP-IT Control qPCR Kit – Human and ChIP-IT Express Kit.