ChIP-IT® Control qPCR Kit – Rat

Catalog No: 53028
Format: 5 rxns

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

Chromatin from a rat 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 Actb and Negative-1 qPCR primer sets.

To pass QC, the RNA pol II ChIP reactions should produce at least 5-fold enrichment with the Actb 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 Actb 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)
- Actb qPCR primer set (2.5 µM) (also sold as Catalog No. 71026)
- Negative-1 qPCR primer set (2.5 µM) (also sold as Catalog No. 71024)

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

<table>
<thead>
<tr>
<th>Component</th>
<th>Volume</th>
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</thead>
<tbody>
<tr>
<td>SYBR green Master Mix</td>
<td>5 µl</td>
</tr>
<tr>
<td>PCR primer mix (2.5 µM)</td>
<td>0.7 µl</td>
</tr>
<tr>
<td>Sterile water</td>
<td>1.3 µl</td>
</tr>
<tr>
<td>ChIP DNA</td>
<td>3 µl</td>
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<tr>
<td><strong>Total Volume</strong></td>
<td><strong>10 µl</strong></td>
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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 – Rat 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.