

The GLIB technique for genome-wide mapping of 5-hydroxymethylcytosine.

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Public Summary:

5-Hydroxymethylcytosine (5hmC) is a newly discovered DNA base present at detectable levels in most mammalian cell types and tissues. It is generated by Tet-enzyme-mediated oxidation of 5-methylcytosine (5mC). 5hmC is important both because of its potential role in regulating gene expression and because it may be an intermediate in DNA demethylation. Here we describe a technique termed GLIB (glucosylation, periodate oxidation and biotinylation), which combines several enzymatic and chemical modification steps to attach biotin to 5hmC. Biotin-containing genomic DNA fragments are then enriched using streptavidin beads, eluted and sequenced. GLIB is capable of quantitatively tagging and precipitating fragments containing a single 5hmC molecule. Sample preparation and GLIB can be conducted in 2-3 d.

Scientific Abstract:

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