**Supplementary File 1: The detection for m5C in RNA.**

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| **Detection method** | **Advantages** | **Disadvantages** |
| BS-seq | Single nucleotide resolution;Primitive methylation patterns. | Influenced by the secondary structure of RNA;Disability for distinguishing m5C from other type of cytosine modifications,Causes RNA damage. |
| m5C-RIP-seq | Distinguishable between m5C and other types of cytosine modifications. | Insensitive to low-abundance m5C;Influenced by the secondany structure of RNA;Relatively low esolution;Highly dependent on antibody specificity. |
| miCLIP-seq | Specific detection of the m5C sites of NSUN2 methyltransferase. | Unable to detect m5C sites of other methyltransferases;Highly dependent on antibody specifity;Altered methylation patterns;Time-consuming; Expensive. |
| AZA-lP-seq | MethyItransferase-specific detection of m5C;Single nucleotide resolution. | Highly dependent on antibody specificity and 5-azaC binding efficiency;High toxiciticy of 5-azaC. |
| TAWO-seq | Single nucleotide resolution;Non-methylated cytosines have a low false positive rate of conversion. | Unstable conversion efficiency;Dependent on the oxidation efficiency of peroxotungstate. |
| Nanopore-seq | Raw DNA or RNA can be directly sequenced. | Expensive; High error rate;Sequencing signals probably affected by multiple nucleotides at the same time. |