

Claims

1. A method for analysis of methylation of ribonucleic acid (RNA) comprising the steps:

(i) contacting a sample comprising RNA with one or more antibodies which binds to methylated site(s) of RNA; wherein the methylated site(s) comprise at least one ribonucleotide base modified by one or more methyl groups;

(ii) photo-crosslinking the one or more antibodies to crosslink individual antibodies to the RNA molecule(s) to form RNA-antibody conjugates;

(iii) immunoprecipitating to separate the RNA-antibody conjugates;

(iv) treating the RNA-antibody conjugates with at least one exonuclease;

(v) removing the crosslinked antibodies from the RNA-antibody conjugates to release RNA; and

(vi) analysing the released RNA;

wherein the sample also comprises methylated RNA spike-in as a control.

2. The method according to claim 1, wherein the method further comprises ligating first adapter nucleic acid molecules to the 3' end of the RNA molecule(s).

3. The method according to claim 2, further comprising ligating second adapter nucleic acid molecules to the 5' end of the RNA molecule(s) after treatment with exonuclease.

4. The method according to any one of the preceding claims, wherein the exonuclease comprises a 5' to 3' exonuclease.

5. The method according to any one of the preceding claims, wherein analysing the RNA comprises reverse transcribing the released RNA to complementary deoxyribonucleic acid (cDNA) and analysing the cDNA.
6. The method according to any one of claims 2 to ~~35~~, comprising reverse transcribing the released RNA using an oligonucleotide molecule substantially complementary to the first adapter molecule to form single stranded complementary deoxyribonucleic acid (cDNA).
7. The method according to claim 6, further comprising amplifying the single stranded cDNA by polymerase chain reaction (PCR) to form double stranded cDNA (ds cDNA).
8. The method according to claim 7, comprising using a first primer substantially complementary to the first adapter molecule and a second primer substantially complementary to the second adapter molecule for the PCR amplification.
9. The method according to claim 7 or 8, further comprising analysing the ds cDNA.
10. The method according to claim 5 or 9, wherein analysing the cDNA or ds cDNA comprises sequencing and/or mapping the cDNA or ds cDNA.