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Methods to study RNA–protein interactions

Muthukumar Ramanathan^{1,3}, Douglas F. Porter^{1,3} and Paul A. Khavari^{1,2*} 

¹Program in Epithelial Biology, Stanford University School of Medicine, Stanford, CA, USA. ²Veterans Affairs Palo Alto Healthcare System, Palo Alto, CA, USA. ³These authors contributed equally: Muthukumar Ramanathan, Douglas F. Porter. *e-mail: khavari@stanford.edu

Supplementary Table 1. RNA library preparation steps

Method	Library preparation	References
HITS-CLIP /PAR-CLIP	Phosphatase› L3› kinase› gel› ppt› L5› RT› PCR	59,62
Variant CLIP 1	Phosphatase› L3› kinase› L5› gel› ppt› RT› PCR	64,73
Variant CLIP 2	Phosphatase› kinase› L5› L3› gel› ppt› RT› PCR	71,72
iCLIP	Phosphatase› L3› kinase(P32)› gel› ppt› RT› gel› ppt› circ› PCR	74
irCLIP	Phosphatase› L3› gel› ppt› RT› SA(L3-bio)› circ› ppt› PCR	76
eCLIP	Phosphatase› L3› gel› ppt or column› RT› exo› L3(DNA)› PCR	75
fCLIP	Phosphatase› ppt› kinase› ppt› gel› ppt› L3› gel› ppt› L5› RT› PCR	27
Monitored eCLIP	Phosphatase› L3› kinase› L5› gel› SA(L5-bio)› RT› L3(DNA)› SPRI› PCR	69
BrdU-CLIP	Phosphatase› L3› kinase› gel› ppt› RT (with BrdUTP)› IP(BrdU)› circ› PCR	77
GoldCLIP	Phosphatase› L3› ppt› RT› gel› ppt› circ› PCR	78

Supplementary Table 1. RNA library preparation steps. A sample of CLIP-seq protocols, and an outline of their library preparation steps. Manipulations of the protein (such as proteinase K extraction) are not noted, only manipulations of RNA and cDNA. L3: 3' adapter ligation to RNA. L3(DNA): 3' adapter ligation to DNA. L5: 5' adapter ligation. Phosphatase: removal of 3' phosphate. Kinase: addition of 5' phosphate. RT: reverse transcription. Gel: any protein or RNA gel, which may or may not include a transfer to a membrane. Ppt: precipitation. SA: Streptavidin pull-down of the indicated biotinylated molecule. Circ: circularization of DNA by a ligase. SPRI: SPRI beads clean-up. IP(BrdU): immunopurification of BrdU. Exo: exonuclease treatment.