Project Title:	Exploring the molecular mechanisms of HERV-H in mediating chromatin high-order structure in human embryonic stem cells 研究 HERV-H 在胚胎幹細胞染色質高級結構調控中的分子機制
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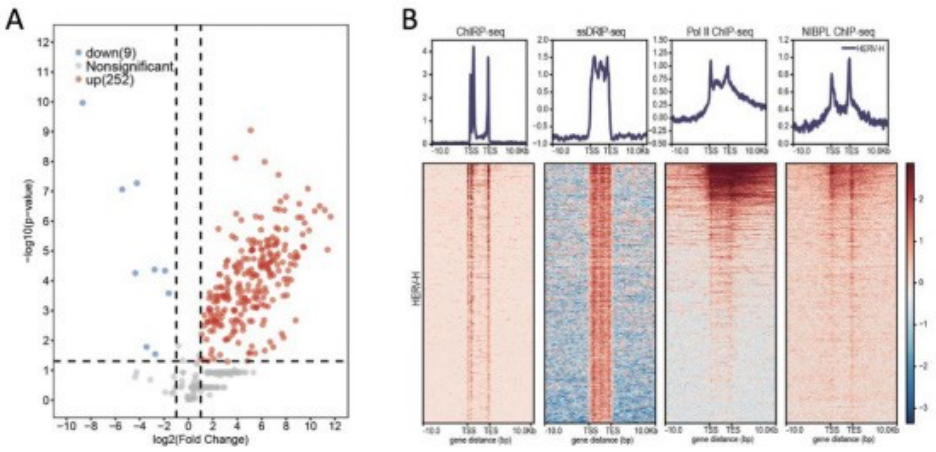
To study the role of lncRNA in difficult-to-transfect cell lines, hESCs, the team upgraded the CARPID 2.0 method by leveraging RNP-based delivery strategy, which can be widely used to explore the binding protein of endogenous lncRNA in living cells.

CARPID 2.0 method leverages ribonucleoprotein (RNP)-based delivery strategy to dissect the protein interactome of HERV-H RNA in living hESCs. 252 significantly associated proteins of HERV-H RNA were identified, the majority of which were nuclear-localised proteins and participated in chromatin remodelling (Fig. A). Notably, 177 out of 252 HERV-H RNAassociated proteins were also known as binding proteins of R-loop that had been reported to stall the translocation of the structural protein Cohesin. It was further confirmed that transcriptionally active HERV-H loci were highly enriched for R-loop structure.

Second, the team delineated the genomic binding loci of HERV-H RNA with ChIRP-seq and found predominant enrichment of HERV-H RNA in TAD boundaries (Fig. B). Two significant HERV-H RNA binding peaks at the transcriptional start sites (TSS) and transcriptional termination sites (TTS) were further observed, which coincided with the location of Rloop, Pol II, and NIPBL. All in all, the current data supports that HERV-H RNA likely participates in mediating loop extrusion by installing Cohesin protein complex.



(A): Volcano plot of HERV-H RNA-associated proteins identified by CARPID 2.0.
(B): Mapping of HERV-H RNA, R-loop structure, Pol II, and NIPBL localization on the HERV-H loci