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## Investigating how splicing factor homeostasis shapes transcriptomes in pluripotency and differentiation

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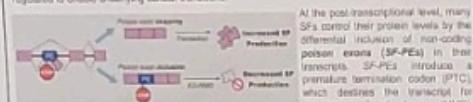
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### Background

Alternative splicing (AS) is a key contributor to cellular and tissue diversity, influencing developmental, oncogenic, and aging processes.<sup>1</sup> AS enables a single gene to give rise to a multitude of mRNA transcripts that can diversify the proteome when translated.

AS is regulated by splicing factors (SFs), which are RNA-binding proteins that activate or repress splicing in a dose-dependent manner. SFs are critical for organism development<sup>2</sup> and must be tightly regulated to ensure orderly cellular transitions.

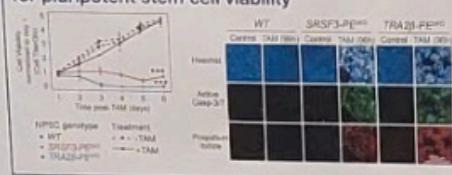


At the post-transcriptional level, many SFs control their protein levels by the differential inclusion of non-coding poison exons (SF-PEs) in their transcripts. SF-PEs introduce a premature termination codon (PTC) which destroys the transcript by nonsense-mediated decay (NMD). The resulting changes in SF levels then control splicing of downstream targets.

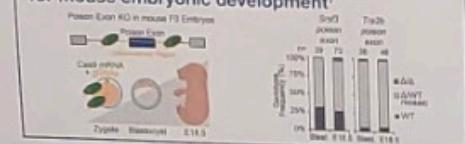
Work by us and others<sup>3,4</sup> has revealed that several SF-PEs are required for cancer cell survival. However, exactly how SF-PEs modulate splicing across pluripotent cellular states remains poorly characterized.

Multiple SF-PEs are highly conserved across species<sup>5,6</sup> and therefore, have been hypothesized to play an integral role in shaping cellular state and differentiation potential.

### SRSF3 and TRA2β poison exons are required for pluripotent stem cell viability



### SRSF3 and TRA2β poison exons are required for mouse embryonic development



### Conclusions & perspectives

- SFs maintain homeostasis via AS-NMD.
- Specific SF-PEs are required for human pluripotent stem cell and mouse embryonic viability.
- Overarching hypothesis: AS mediates pluripotent stem cell identity and ability to differentiate.
- Our findings will reveal how SF-PEs modulate transcription, safeguard pluripotency, and drive differentiation, with potential to advance cancer therapeutics and regenerative medicine.

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