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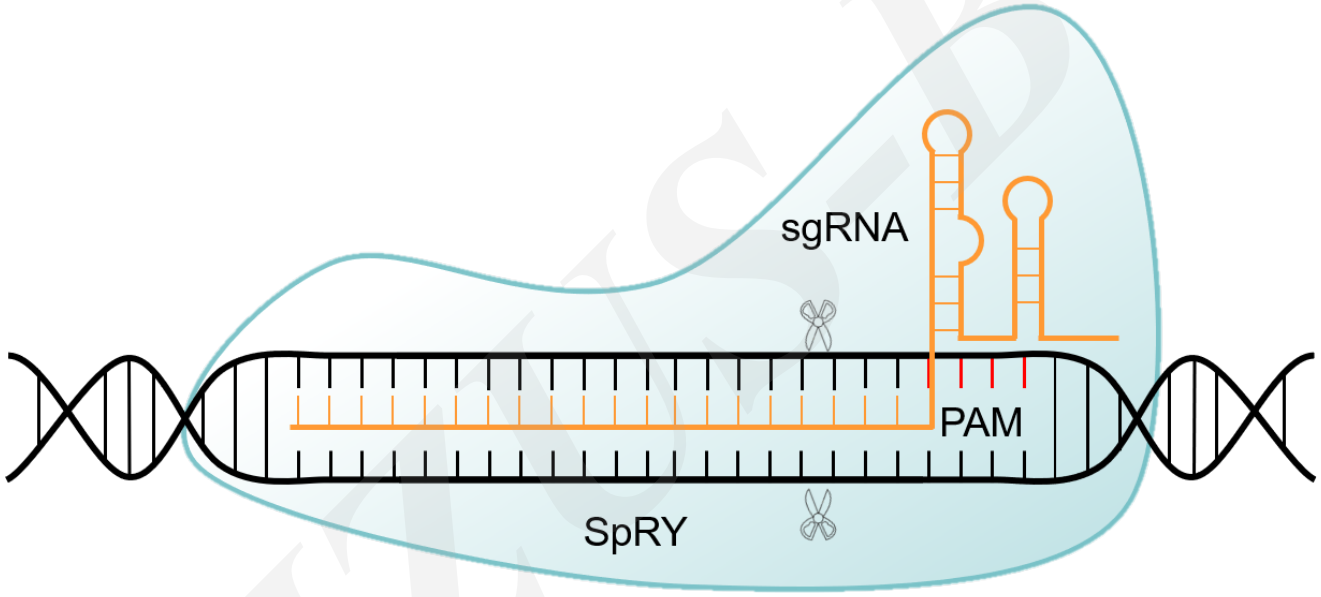
Can SpRY recognize any PAM in human cells?

Key words: CRISPR/Cas; SpRY; PAM; Recognize

Research Summary

This study identified the PAM preference of SpRY in human cells and summarized them as follows:

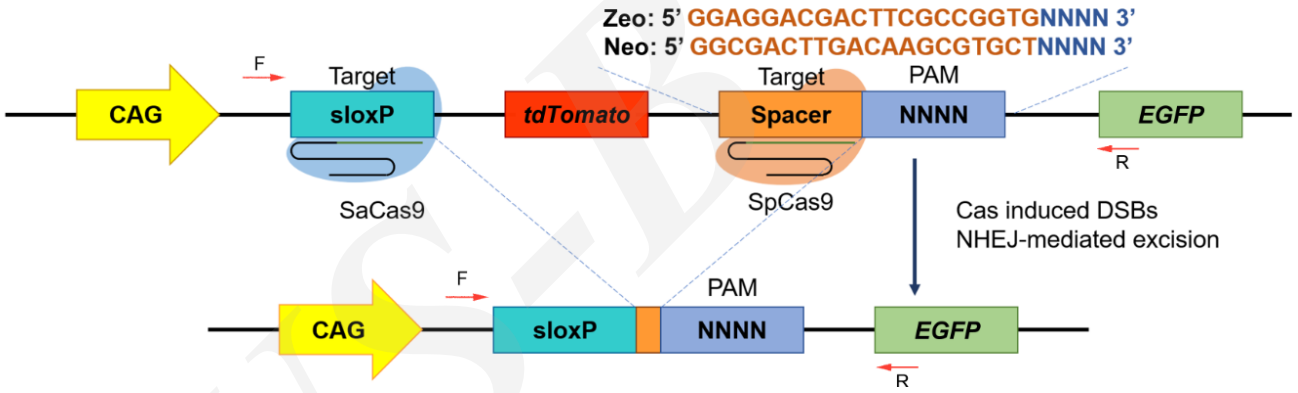


- **5’-NRN-3’, 5’-NTA-3’ and 5’-NCK-3’ could be considered as canonical PAMs for SpRY**
- **5’-NCA-3’ and 5’-NTK-3’ may be served as non-priority PAMs.**
- **5’-NYC-3’ are not recommended.**

Innovation points

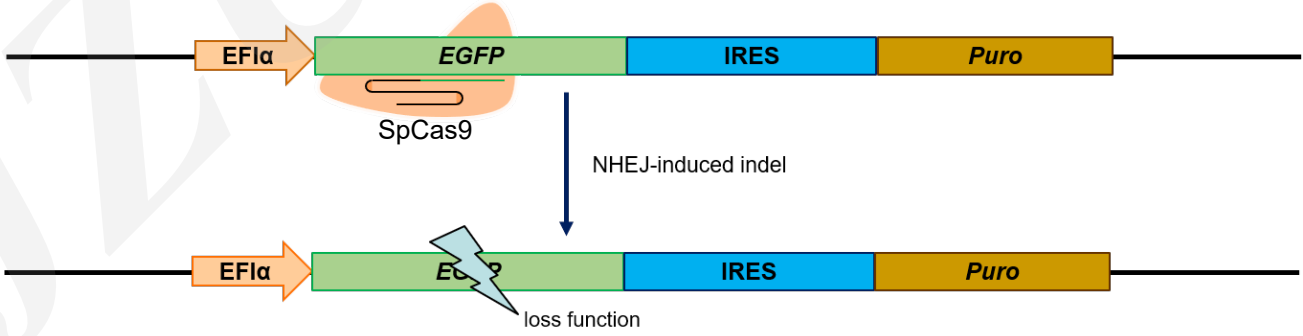
- **PAM-DOSE**

SpRY cleave the PAM library plasmid in the human cells. PAM preference can be identified with next-generation sequencing.



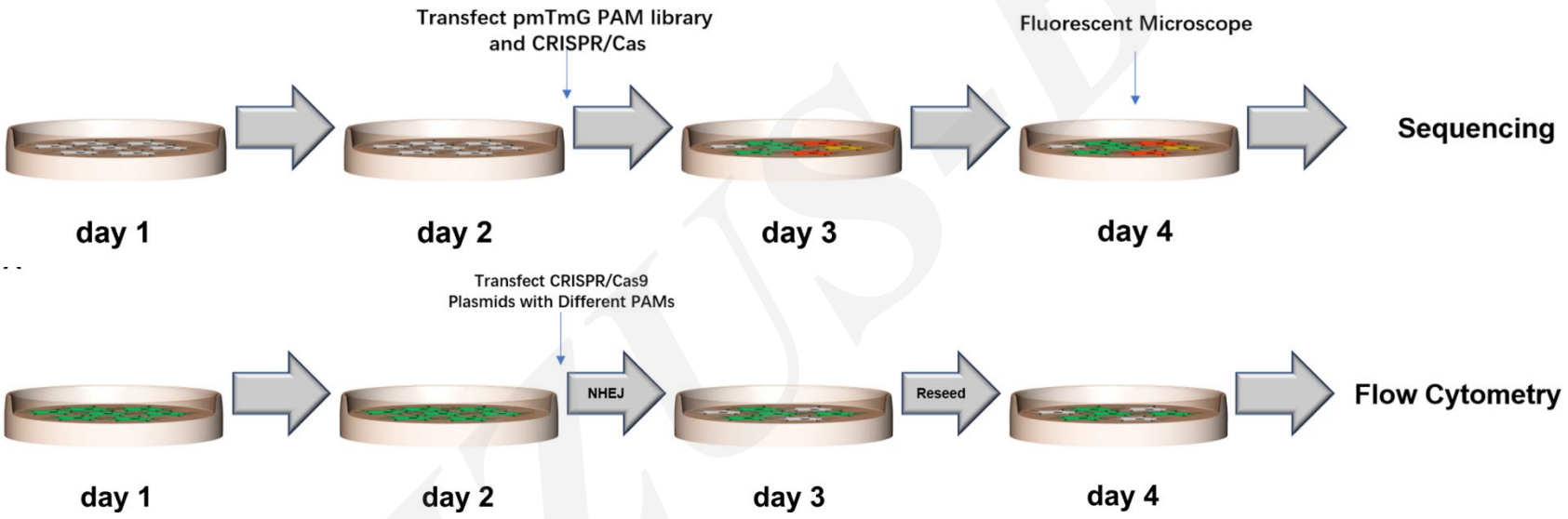
- **GFP-reporter system**

SpRY triggers frame-shift indel mutations in *EGFP* gene. The efficiency of target sequence with different PAM can be assessed via flow cytometry.



Innovation points

- **Identify the PAM preference in human cells without the purification of recombinant Cas proteins or *in vitro* transcription.**



- **Summarize the relative activity of SpRY with 5'-N<u>NN</u>-3'**

Table 1. Comparison of the relative activity of SpRY with 5'-N<u>NN</u>-3' PAMs between different studies.