The Function of L(3)mbt as a Reader of H4K20 Methylation

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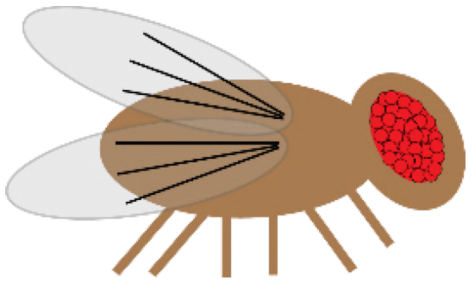
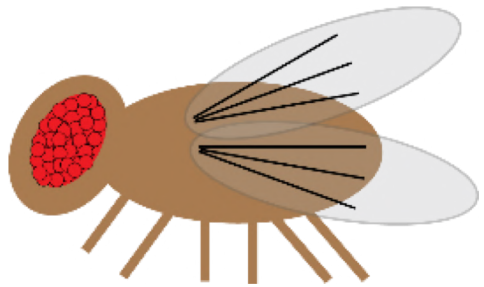
Duronio Lab

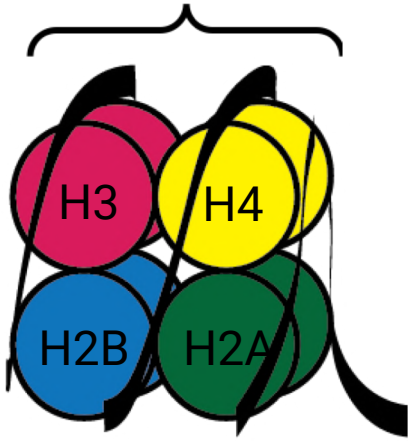
Celebration of Undergraduate Research, May 3rd



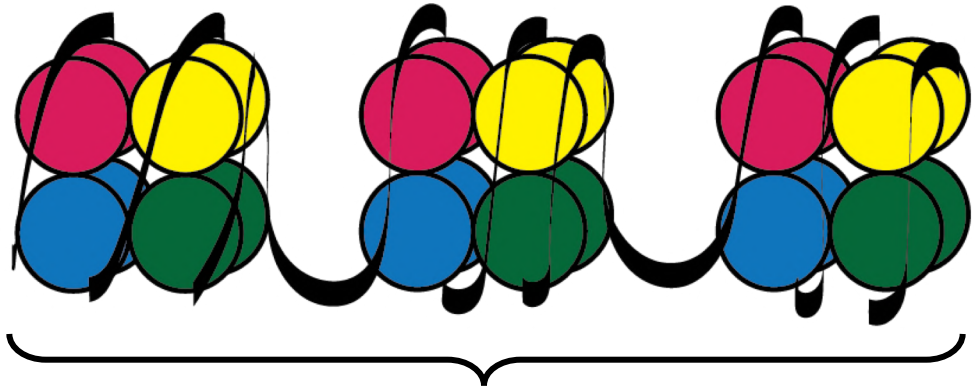


What is chromatin?

Nucleosome

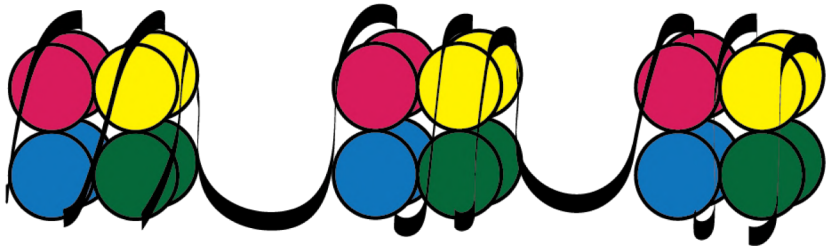


DNA wrapped around histone proteins

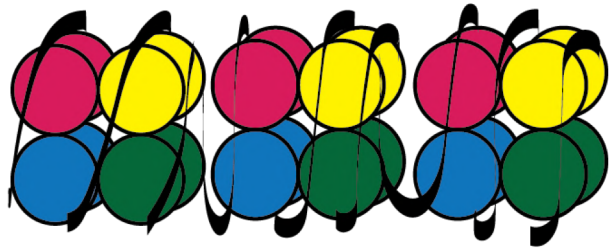


Chromatin

Euchromatin



Heterochromatin

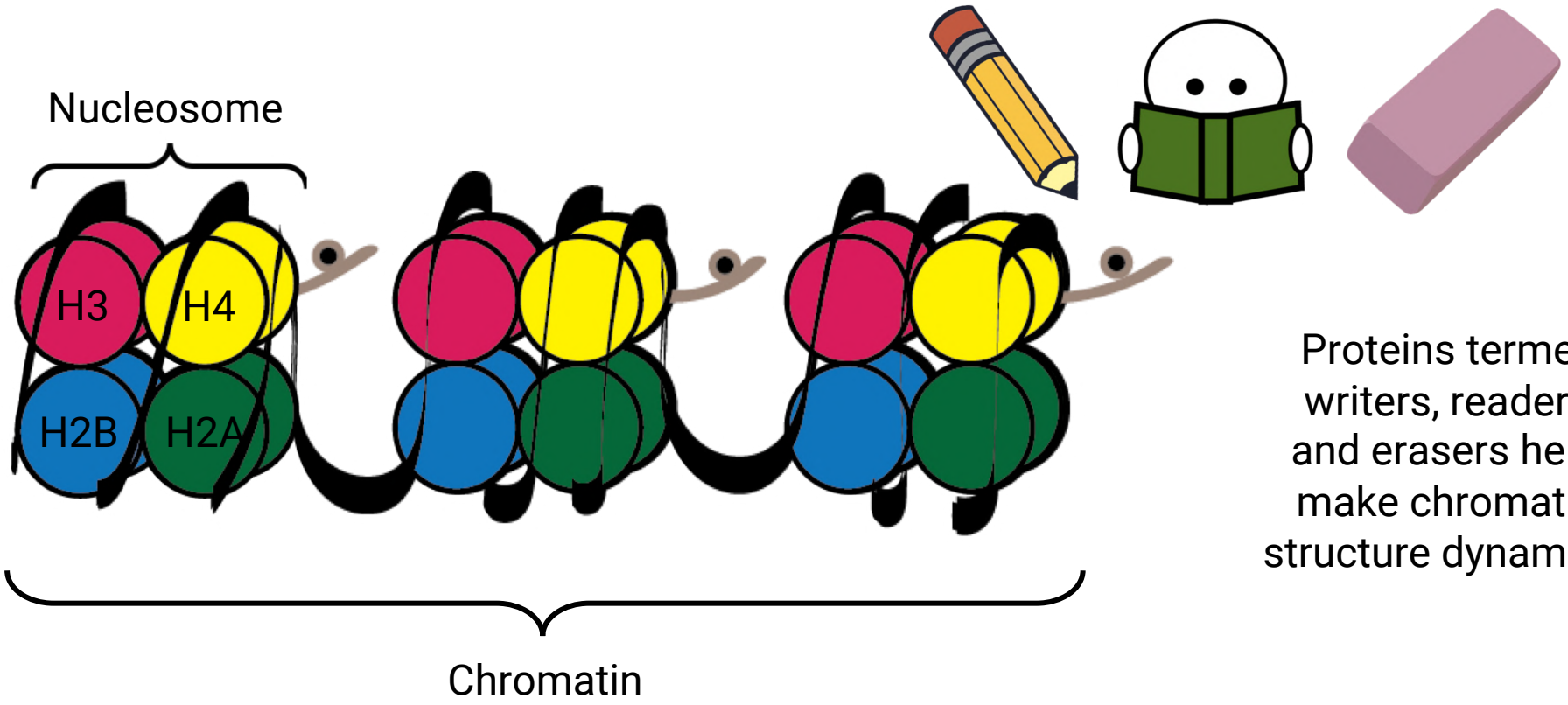


Can be spaced farther apart or closer together

Alabert et al.
Lachner et al.

What changes the organization of chromatin?

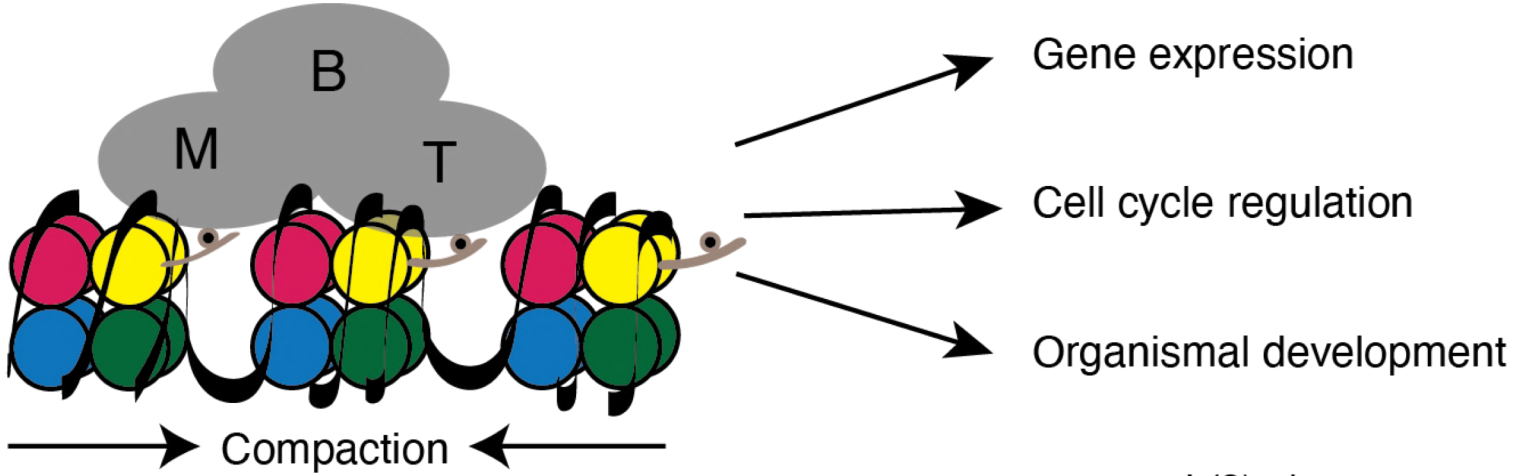
Histone tails can be post-translationally modified



Proteins termed writers, readers, and erasers help make chromatin structure dynamic

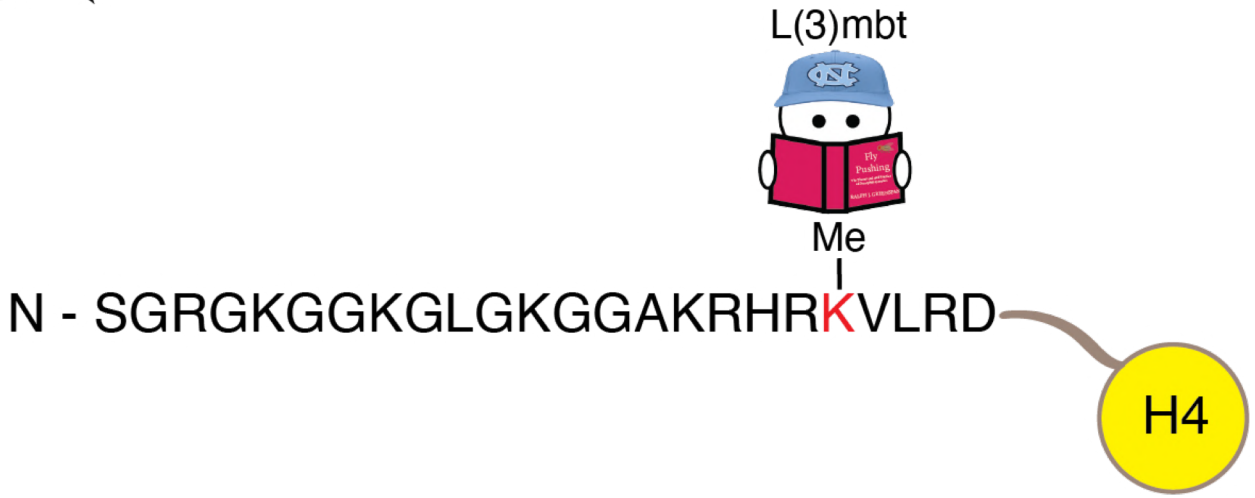
Alabert et al.
Lachner et al.

Lethal (3) Malignant Brain Tumor “reads” histone methylation *in-vitro,* but does its function depend on H4K20me-specific recruitment *in vivo*?



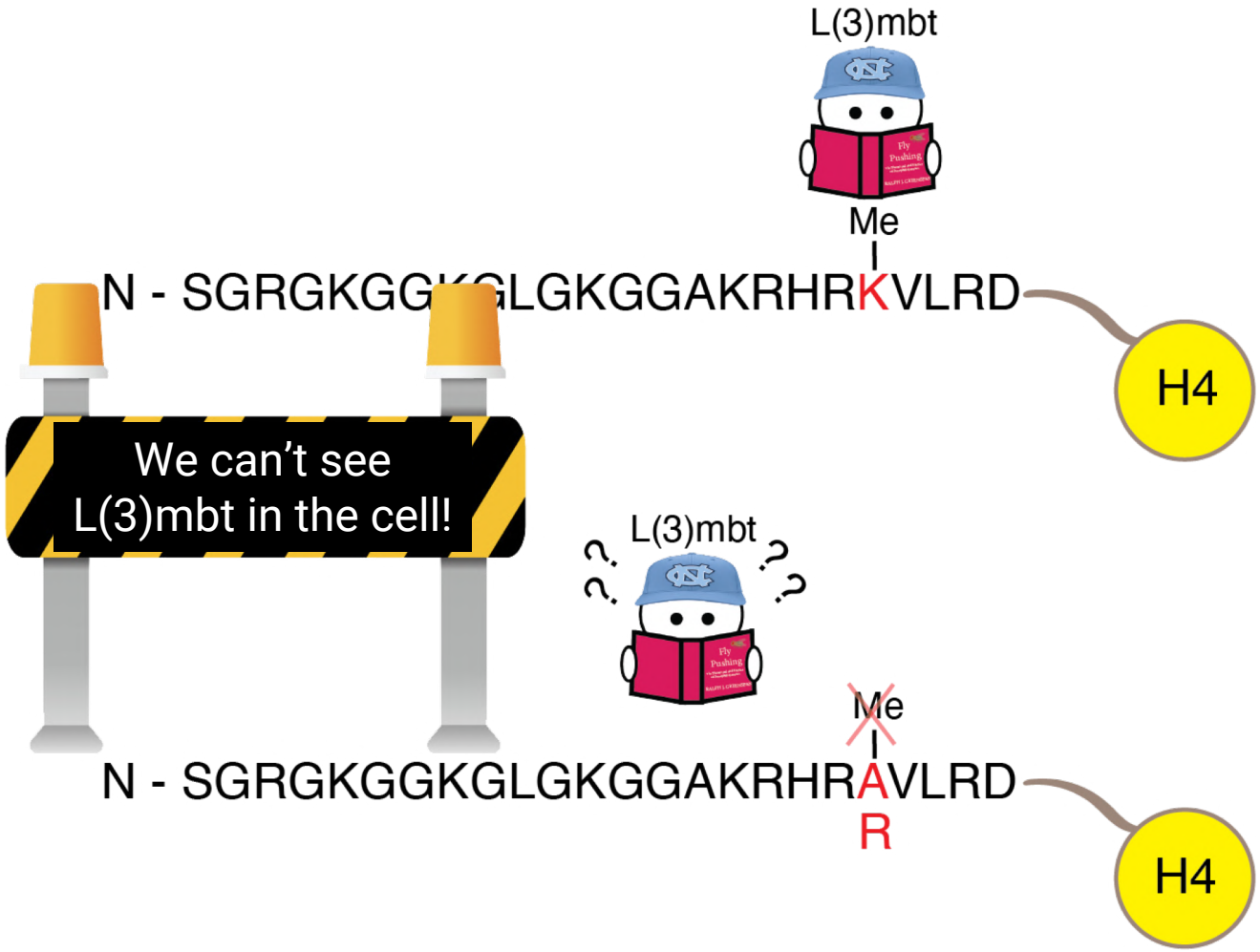
The human version of L(3)mbt has been implicated in a variety of cancers

L(3)mbt specifically binds histone H4 lysine 20 methylation (H4K20me) in the test tube, but this has never been seen in an organism

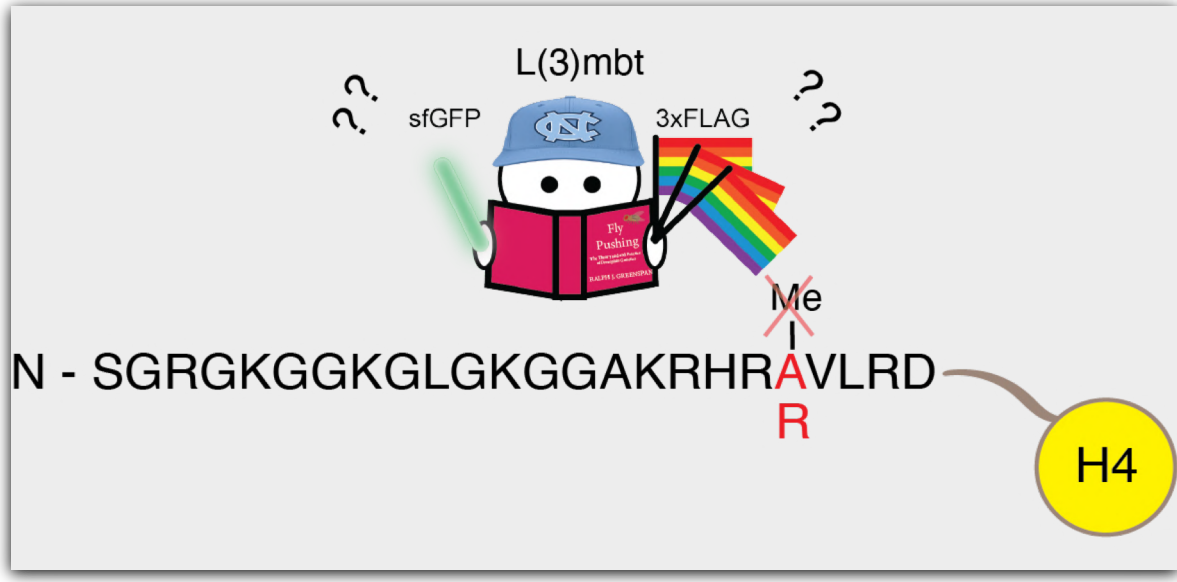


Bonasio et al.
Lu et al.
Blanchard et al.
Wismar et al.
Reader cartoon(s): Aaron Crain

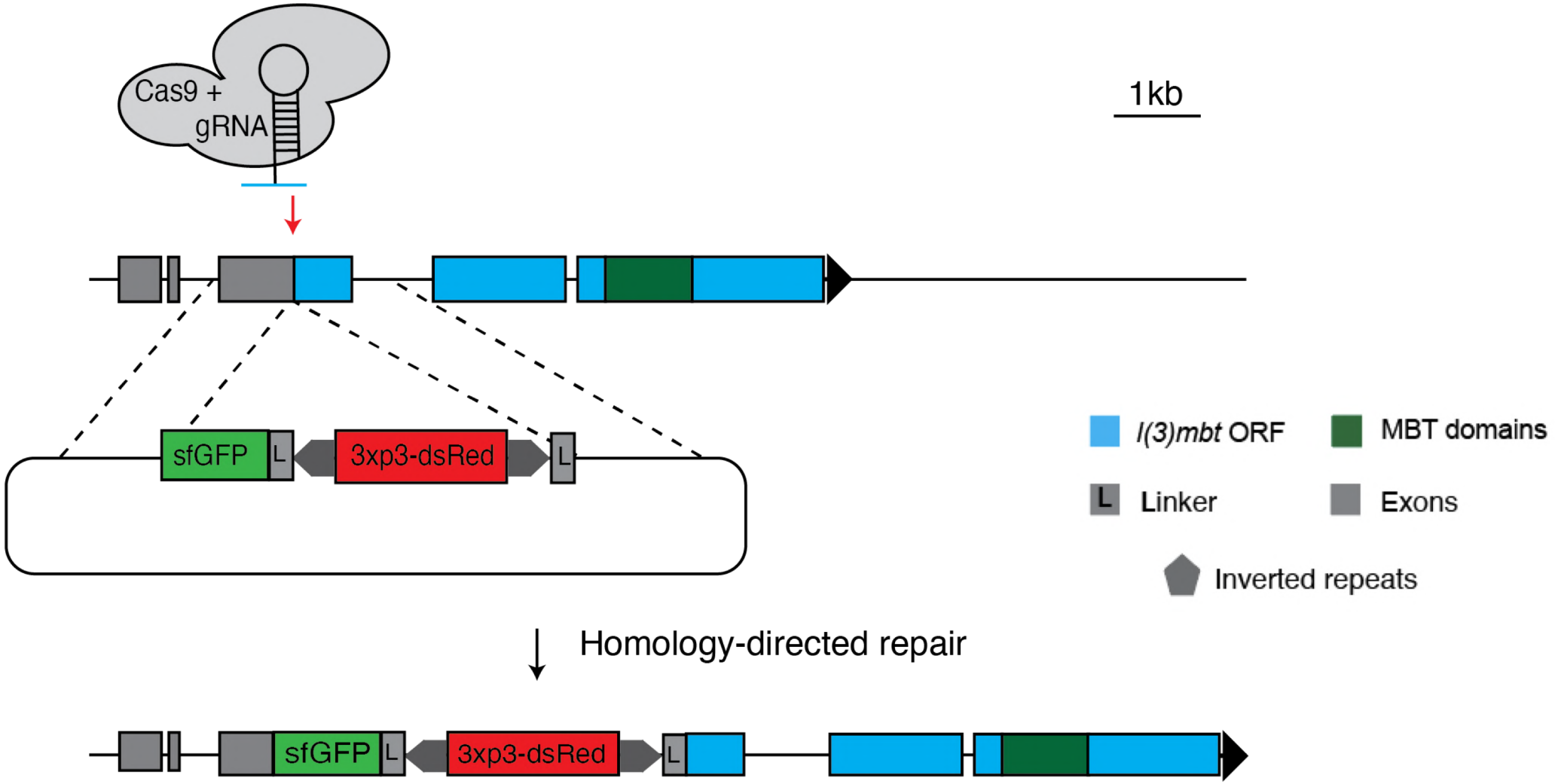
We hypothesize that L(3)mbt's chromatin binding will change if H4K20 cannot be methylated



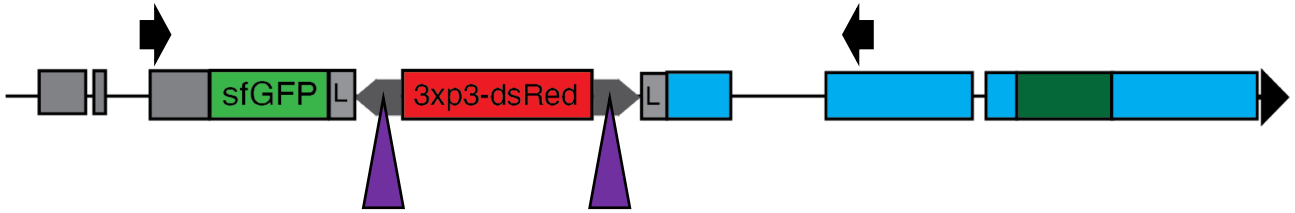
Solution: make tagged alleles!

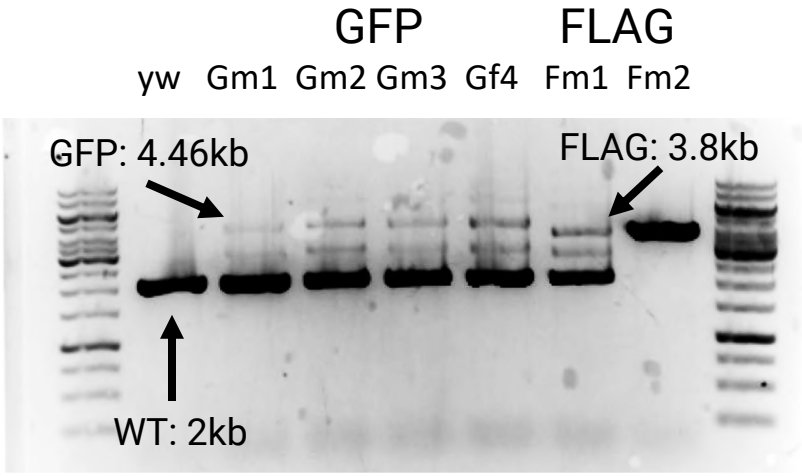


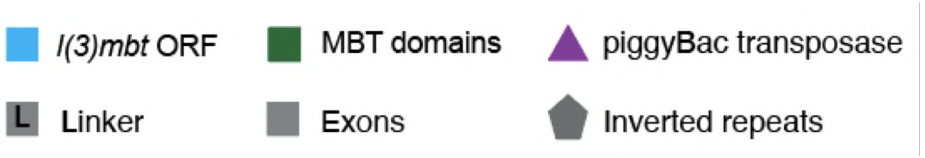
The Scarless Gene Editing System uses CRISPR to tag genes at the endogenous locus

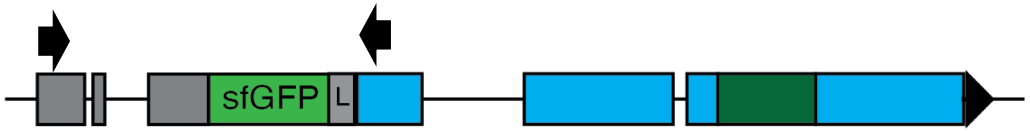


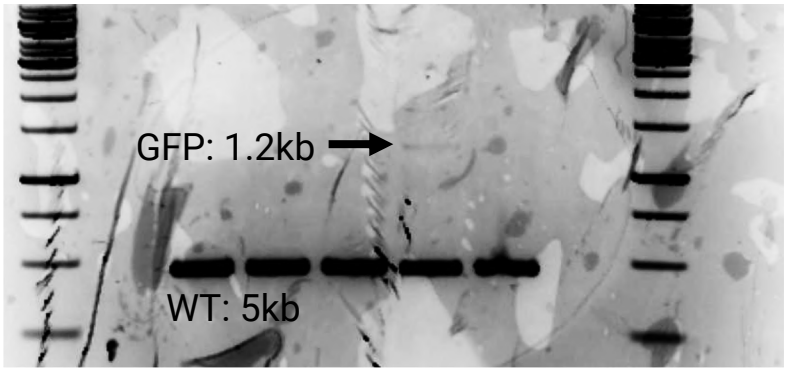
Scarless genotyping pre- and post-excision by piggyBac Transposase

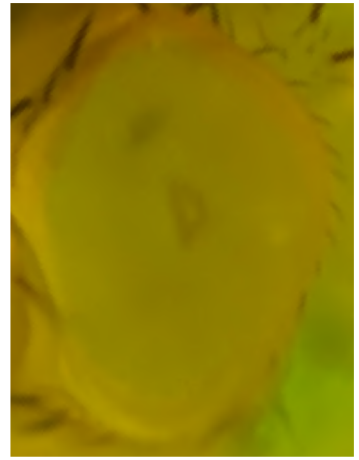




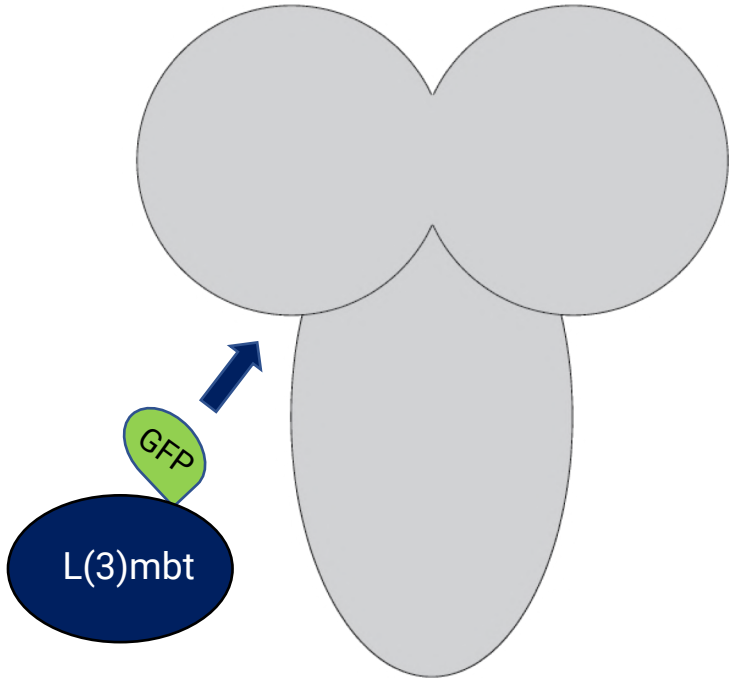








We can see endogenous GFP-L(3)mbt in third-instar larval brains



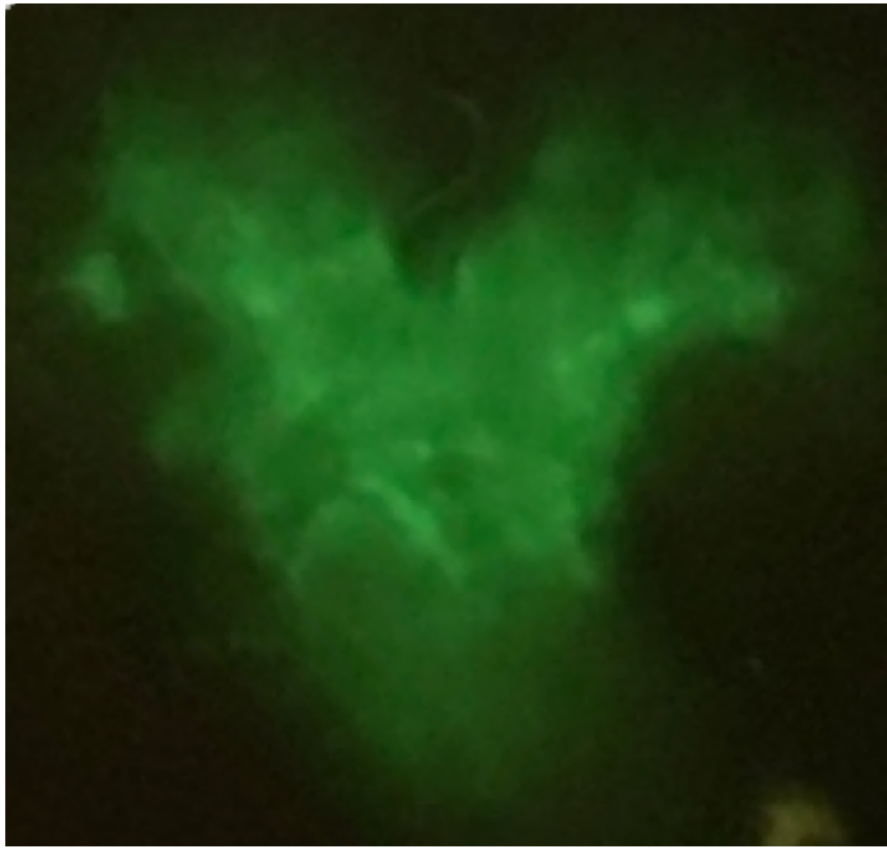
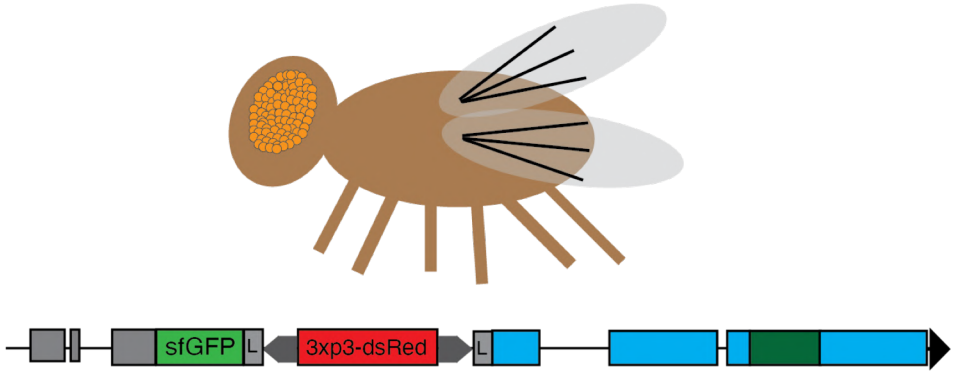
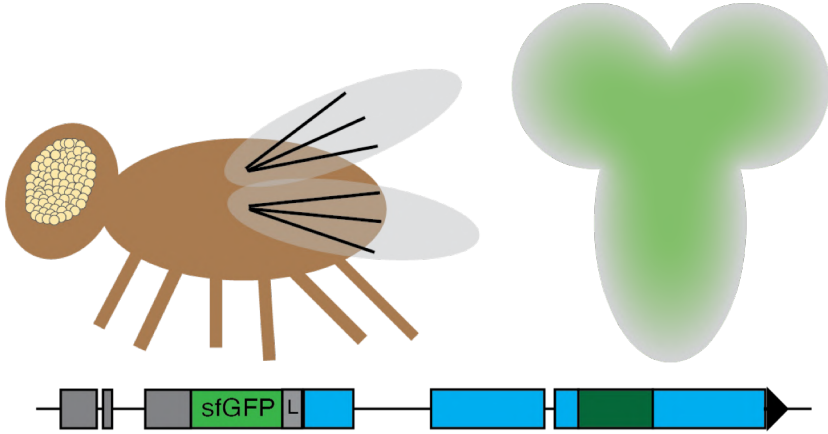
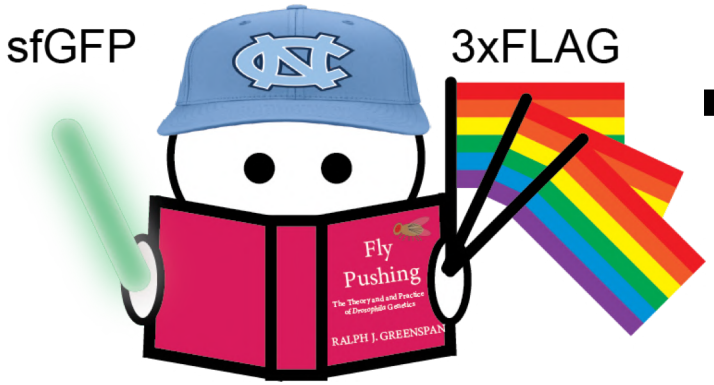


Image from Nikon

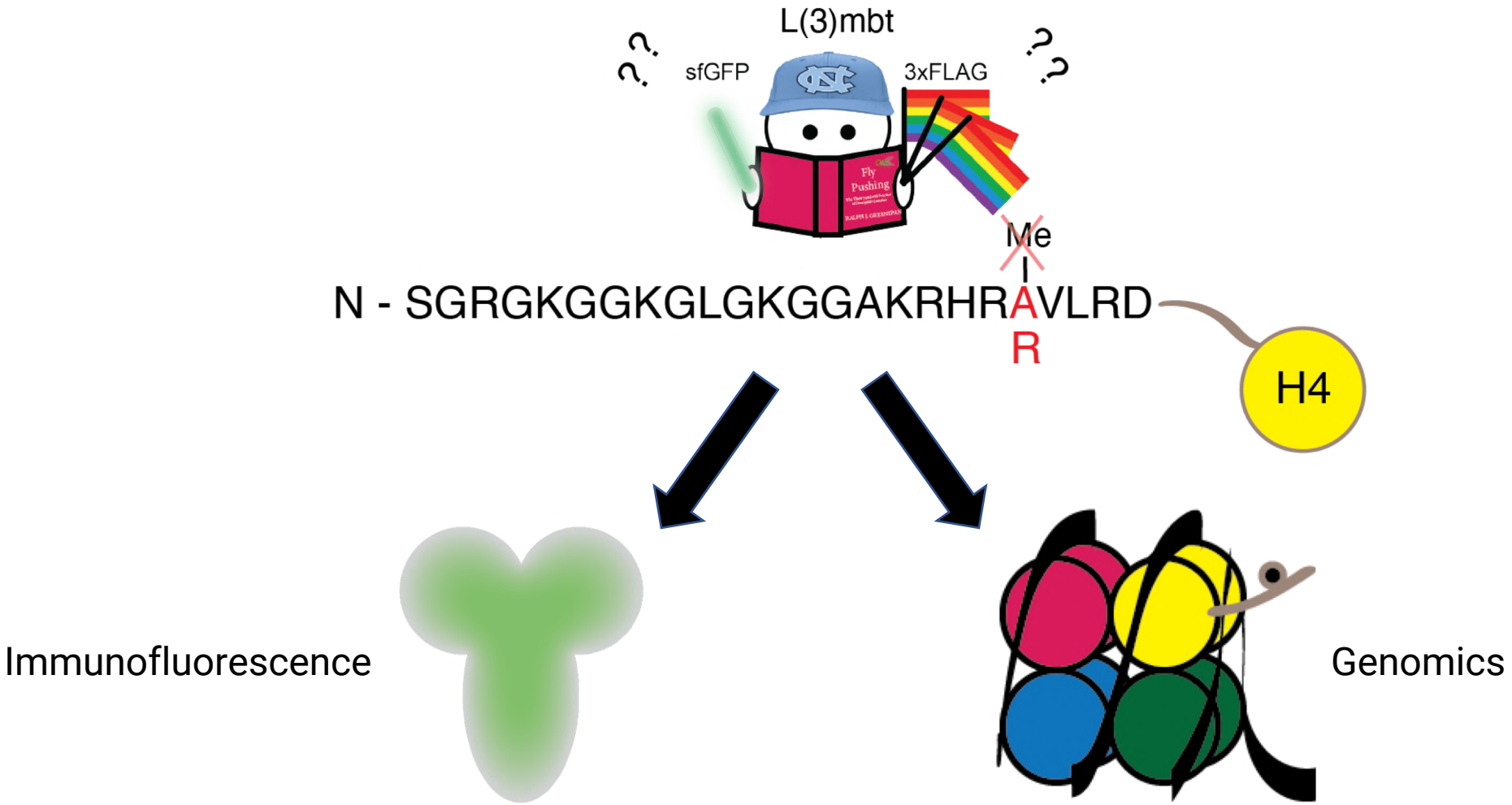
We can now visualize L(3)mbt in the cell and detect its presence via antibodies to GFP or FLAG







Will L(3)mbt's chromatin binding change in H4K20-mutant backgrounds?



Acknowledgements

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