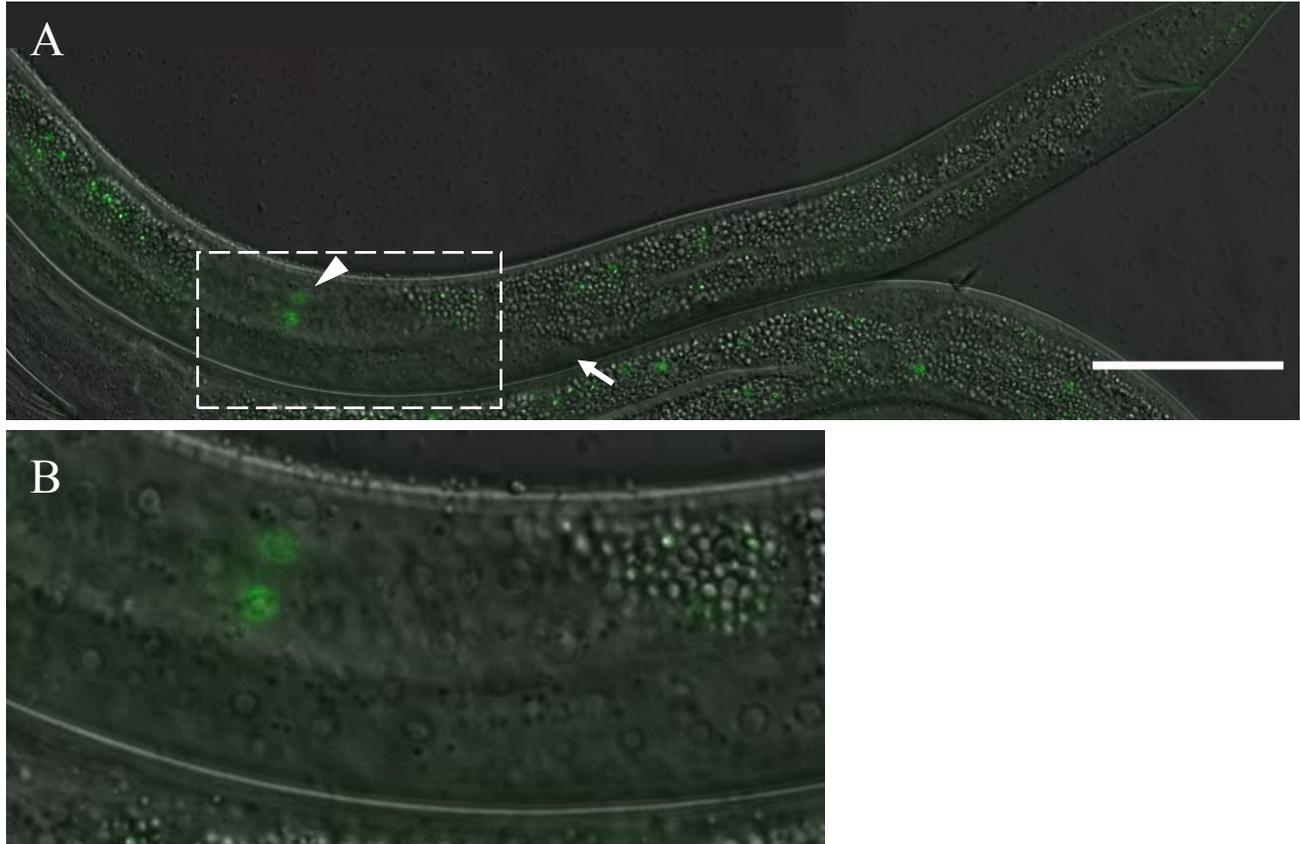


# FKH-6 is expressed in male gonadal cells during L3 larval development

Aaron Crosby<sup>1</sup>, and Mary B. Kroetz<sup>1\*</sup>

1. Department of Biological Sciences, University of South Alabama, Mobile, AL, USA

\* To whom correspondence should be addressed. E-mail: [marykroetz@southalabama.edu](mailto:marykroetz@southalabama.edu)



**Figure 1** FKH-6 is expressed in the male gonad during the L3 larval stage. Differential interference contrast (DIC) and fluorescence micrographs of FKH-6 expression in the male gonad. Arrow indicates distal end of gonad. Arrowhead indicates GFP-expressing nuclei. Boxed region in A is magnified in B. Bar, 50 μm.

## Description

FKH-6 is a forkhead transcriptional regulator that is necessary for proper gonadal development in *C. elegans*. A previous expression study using a transcriptional reporter showed that the *fkh-6* transcript was initially expressed in the gonad from mid-L1 larval stage until around the L1/L2 molt in both sexes (Chang *et al.* 2004). Transcriptional reporters and partial gene fusions indicated that FKH-6 was also expressed from the L3 larval stage through adult in the hermaphrodite gonad in the spermatheca and sheath cells (Hope *et al.* 2003, Chang *et al.* 2004). In this current study, the endogenous locus for *fkh-6* was C-terminally fused with GFP using CRISPR-Cas. In addition to recapitulating previously reported expression patterns, this new construct revealed expression of FKH-6::GFP in 2-4 male gonadal cells in the L3 larval stage. This is the first report of FKH-6 expression in males after the L1/L2 molt. The expression of FKH-6::GFP in the gonad of the L3 male is reproducible, appearing in 54 of the 58 male animals observed during this time period, but the expression is short-lived, and coincides with the period when the proximal region of the migrating male gonad is extending past the distal tip of the gonad. This work shows that FKH-6 is expressed in both sexes during the early mitotic period of the gonad in L1 as well as the late mitotic period of the gonad in L3.

12/31/2018 – Open Access

### Reagents

The *fkh-6::gfp* fusion was generated using CRISPR-Cas (Dickenson *et al.* 2015). Sequencing confirmed the integration of GFP at the 3' end of the *fkh-6* locus prior to the stop codon. The MBK9 strain (*fkh-6(ndz1[fkh-6::gfp::3xflag]) II; him-8(e1489) IV*) was used for imaging. Differential interference contrast (DIC) and fluorescent images were acquired on a Zeiss Axiovert 135 microscope using a Lumenera Infinity 3S-1UR monochrome camera and Infinity Analyze and Capture software.

### References

Chang W, Tilmann C, Thoemke K, Markussen FH, Mathies LD, Kimble J, Zarkower D. A forkhead protein controls sexual identity of the *C. elegans* male somatic gonad. *Development*. 2004 Mar;131(6):1425-36. Pubmed PMID: 14993191.

Hope IA, Mounsey A, Bauer P, Aslam S. The forkhead gene family of *Caenorhabditis elegans*. *Gene*. 2003 Jan 30;304:43-45. Pubmed PMID: 12568714.

Dickinson DJ, Pani AM, Heppert JK, Higgins CD, Goldstein B. Streamlined genome engineering with a self-excision drug selection cassette. *Genetics*. 2015 Aug;200(4): 1035-49. Pubmed PMID: 26044593.

**Funding** University of South Alabama faculty start-up funds

**Reviewed by** Jennifer Wolff

**Received** 11/29/2018, **Accepted** 12/31/2018. **Available** starting [WormBase](#) release WS271, **Published Online** 12/31/2018.

**Copyright** © 2018 by the authors. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International (CC BY 4.0) License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

**Citation:** Crosby, A; Kroetz, MB (1970). FKH-6 is expressed in male gonadal cells during L3 larval development. microPublication Biology. 10.17912/micropub-biology-000079