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Distinct roles of the Pumilio and FBF translational repressors during C. elegans vulval development

Claudia B. Walser, Gopal Battu, Erika Fröhli Hoier and Alex Hajnal

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We have discovered that the expression pattern of the PUF-8::GFP reporter zhEx61 shown in Fig. 2B-L on p. 3466 of this article, is based on an incorrect reporter construct that carries the insert in the reverse orientation. The corrected Fig. 2 below shows the PUF-8::GFP expression pattern that is observed with the correct PUF-8::GFP reporter zhEx274.1, which carries the insert in the correct orientation. Also shown is a corrected supplementary Fig. S1, in which a quantification of the expression pattern of zhEx274.1 is shown (A-C). Although the overall expression pattern observed with the PUF-8::GFP reporter zhEx274.1 is similar to that obtained with the reverse reporter zhEx61 shown in Fig. 2B-L of Walser et al. (2006), there are four differences in its expression, which are accounted for in the text changes detailed below. Also provided are new methods for the generation of zhEx274.1. These corrections do not change the overall conclusions of this paper. The page, paragraph and line numbers below refer to the PDF version of the article.

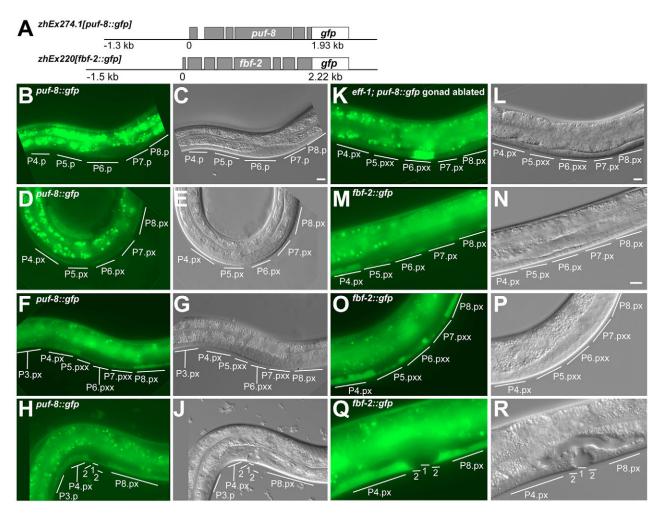


Fig. 2. PUF-8::GFP and FBF-2::GFP expression during vulval development. (A) Structure of the translational puf-8::gfp and fbf-2::gfp reporters. (B,D,F,H) Time-course analysis of PUF-8::GFP expression in the vulval cells from the L2 until the L4 stage with (C,E,G,J) the corresponding Nomarski images. For a semi-quantitative analysis of the expression patterns, see Fig. S1 in the supplementary material. (K,L) PUF-8::GFP expression in gonad-ablated eff-1(hy21) animals, and the corresponding Nomarski image. All VPC descendants showed PUF-8::GFP expression with a strong increase in the descendants of P6.p. Note that despite the extra round of cell divisions in P5.p and P6.p descendants of gonad-ablated eff-1 mutants, no vulval differentiation was observed. (M-R) FBF-2::GFP expression, and the corresponding Nomarski images, from the early L3 until the L4 stage. In all panels, anterior is to the left and ventral is to the bottom. Scale bars: 10 μm.

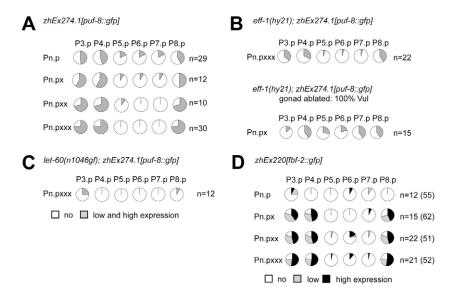


Fig. S1. PUF-8::GFP and FBF-2::GFP expression analysis. (**A**) Semi-quantitative time-course analysis of PUF-8::GFP expression in wild-type animals. The two daughter cells after the first cell division are termed Pn.px for all VPCs. The descendants of the second cell divisions of induced VPCs are termed Pn.pxx, and after the third round of cell divisions Pn.pxxx cells. Gray areas indicate the proportion of PUF-8::GFP-positive vulval cells, white areas the proportion of PUF-8::GFP-negative cells. (**B**) Analysis of PUF-8::GFP expression pattern in (top row) *eff-1(hy21)* mutants at the Pn.pxxx stage without gonad ablation and (bottom row) gonad-ablated *eff-1(hy21)* mutants at the Pn.px stage. Both conditions were analyzed in L4 larvae, but since VPCs in gonad-ablated animals are not induced to adopt vulval cell fates, they divide once and arrest at the Pn.px stage or occasionally divide a second time, as shown in Fig. 2K,L. (**C**) Analysis of the PUF-8::GFP expression pattern in *let-60(n1046gf)* L4 larvae at the Pn.pxxx stage. Since *let-60(n1046gf)*; *zhEx274.1[puf-8::gfp]* animals developed into sterile adults for unknown reasons, the *let-60(n1046gf)* were scored at the Pn.pxxx stage. (**D**) Semi-quantitative time-course analysis of FBF-2::GFP expression in wild-type animals. Only animals showing bright FBF-2::GFP expression in somatic tissues were used for the analysis. White indicates no FBF-2::GFP expression, grey low expression and black high expression.

Correction to the text on p. 3462, paragraph 6

Extrachromosomal transgenic arrays [transgenes; co-transformation marker; pBS: Bluescript (concentration in ng/µl)] were generated by microinjection of DNA into young adult worms (Mello et al., 1991):

Correction to the text on p. 3462, paragraph 7, line 6

...zhEx220[fbf-2::gfp; lin-48::gfp (100;50)], zhEx274.1[puf-8::gfp; lin-48::gfp (80;50)].

Correction to the text on p. 3464, paragraph 2, from line 4

PUF-8::GFP was expressed in various tissues including the hypodermis, the ventral cord motor neurons (not shown) and the vulval cells (Fig. 2B-J and see Fig. S1A in the supplementary material). Before vulval induction in L2 larvae, PUF-8::GFP was expressed in all six vulval precursor cells, although expression was more frequently observed in the distal VPCs (P3.p, P4.p and P8.p) than in the proximal VPCs (P5.p, P6.p and P7.p, Fig. 2B,C, and row with Pn.p cells in Fig. S1A in the supplementary material). After vulval induction in early L3 larvae, PUF-8::GFP expression persisted in the descendants of the 3° distal VPCs (P3.p, P4.p and P8.p), while expression faded in the 1° and 2° descendants of the proximal VPCs (P5.p, P6.p and P7.p, Fig. 2D-J, Fig. S1A in the supplementary material, rows Pn.px to Pn.pxxx).

Correction to the text on p. 3464, paragraph 3, from line 1

We hypothesized that PUF-8::GFP expression in the descendants of the distal 3° VPCs might persist because these cells fuse with the hyp7 hypodermis that also expresses PUF-8::GFP. To test if the expression of PUF-8::GFP in the descendants of the 3° VPCs is a consequence of their fusion with hyp7, we examined PUF-8::GFP expression in an *eff-1(hy21)* background, in which no cell fusions occur (Mohler et al., 2002).

Correction to the text on p. 3464, paragraph 3, from line 10

In most gonad-ablated *eff-1(hy21)* animals, PUF-8::GFP expression was observed in the VPCs and their descendants (Fig. 2K,L and see Fig. S1B in the supplementary material). Moreover, in *let-60 ras(gf)* animals, in which the distal VPCs frequently adopt the 1° or 2° induced cell fates, PUF-8::GFP expression was often absent in the distal VPCs and their descendants (see Fig. S1C in the supplementary material) (Beitel et al., 1990; Greenwald et al., 1983). We conclude that PUF-8::GFP is expressed in the descendants of VPCs that have adopted the uninduced 3° cell fate independently of their fusion with hyp7.

Correction to the text on p. 3466, paragraph 2, line 1

The expression of PUF-8::GFP in the distal 3° vulval cells raises the possibility that PUF-8 might regulate the competence of the distal vulval cells to respond to the inductive signal.

Correction to the text on p. 3469, paragraph 2, line 7

A PUF-8::GFP reporter transgene is expressed predominantly in the distal VPCs (P3.p, P4.p and P8.p) and their descendants that have adopted the 3° fate.

The authors apologise to readers for these mistakes and are grateful to Dave Hansen for discovering the error in the plasmid used to generate zhEx61.

Publisher's note: Although the mistake reported in this corrigendum has resulted in several corrections being made to Walser et al. (2006) and in an unusually lengthy corrigendum, we would like to reassure readers that expert opinion has confirmed that the minor changes in expression that are seen between the incorrect reporter *zhEx61* and the correct reporter *zhEx274.1* do not alter or affect the conclusions drawn by this paper.