

## Figure S1. Structure of the *rsu-1* locus.

The *rsu-1(tm6690)* mutant harbors a 778-bp deletion at the 5'-end of the locus.



## Figure S2. Depletion of Ras<sup>LET-60</sup> and RSU-1 suppresses cytoplasmic protrusions in *rsu- 1(tm6690)*.

- (A) Representative confocal images of Pegl-15::GFP;Ras<sup>let-60</sup>(RNAi);rsu-1(RNAi). Scale bar, 10 μm.
- (B) Percentage of vulval muscle showing cytoplasmic protrusions in *rsu-1(RNAi);Ras<sup>let-60</sup>(RNAi)*.



**Figure S3. Depletion of Ras**<sup>LET-60</sup> **suppresses the multivulva in** *let-60(n1046)*. Ractio of different vulva worm in *let-60(n1046)* and *let-60(n1046);Ras*<sup>*let-60</sup>(RNAi*).</sup>





- (A) Representative confocal images of mCherry::RSU-1;GFP::PINCH<sup>UNC-97</sup> and mCherry::RSU-1;Integrin<sup>PAT-3</sup>::GFP in body-wall muscles. Scale bar, 10 μm.
- (B) Representative TEM images of body-wall muscle in control and *rsu-1(tm6690)*. Scale bar, 10 and 5 μm.



## Figure S5. Depletion of RSU-1 does not change the focal adhesion sites in vulval muscle.

- (A) Representative confocal images of mCherry::RSU-1 and mCherry::RSU-1;*rsu-1(RNAi)*.
  Representative confocal images of GFP::PINCH<sup>UNC-97</sup>, GFP::PINCH<sup>UNC-97</sup>;*rsu-1(RNAi)*.
  Scale bar, 10 μm.
- (B) Representative confocal images of Integrin<sup>PAT-3</sup>::GFP, and Integrin<sup>PAT-3</sup>::GFP;*rsu-*1(tm6690). Scale bar, 10 μm.



Figure S6. The morphology of vm1 muscle cells in L4 stage and adult stage.

Representative confocal images of Pegl-15::GFP and Pegl-15::GFP;rsu-1(tm6690) in L4 stage and adult stage. Scale bar, 10 µm.