Supplementary Table S1. Primers used for the qPCR assays.

Gene Name	Accession Number	Primers
18S	LT631145.1	Forward GACCTGCTAAATAGTGACGCCATCCAC Reverse CGTGCGGCCCAGAACATCTAAG
EIF4A1	XM_022951886.1	Forward CACCAGGAATGGAACAGGACGGAG Reverse ACCGTATGCGTAGATGCCTCGGAG
Piwi-1	XM_022950268.1	Forward AGCCATCACATCAGCAAGGACCACAG Reverse CCGTTGATATTGTTGAGGCATCTGTG
Nanos-1	XM_022930859.1	Forward GTGTAGACAGCGTGGGATCGGATC Reverse TGACGTTTCAATGCACAGTTCAATGTC
Nanos-1-like	XM_022928001.1	Forward GCCACGACGTGACCGATTTGAC Reverse GAAAGGCCCATCTCCAAAGTGATCC
Tudor-5	XM_022924446.1	Forward TGGTAGGAGCTCAGTCGATGGCAG Reverse GGATAGAAGAAGCCCGCGAACATTC
Tudor-7	XM_022930561.1	Forward GTTGCAAGCGAGGCGGACAAAC Reverse TAGAGCTGCTGATCTCCCTACACCTGG
Boule	XM_022923693.1	Forward CTATCCTCACCCTCACCCTCACCCTAG Reverse TTTGTCGAGGATTTGGAGCTTGTGGAG
Sox-2	XM_022945978.1	Forward GCCCGCAGCAAAGACACCTATG Reverse CAACGGCATCATGAAGGCTTCAC
Myc-1	XM_022936348.1	Forward TAGACGAGGACAGTTGTTCACCTGCAC Reverse TCCACATGCAGTCTTGAATGAGTTTGG

Supplementary Table S2. Comparison of gene expression among naïve tissues, regenerating tissues, and growing edges: calculated p-values by repeated-measures ANOVA analyses.

Gene	Tests of Within-Subjects Effects (Significance)
Nanos-1	0.026
Nanos-1 like	0.019
Piwi-1	0.010
Sox-2	0.023
Boule	0.00002
Myc-1	0.02
Tudor-7	0.014
Tudor-5	0.001

Supplementary Table S3. P-values from LSD (Fisher's least significant difference) post hoc comparisons for significant differences among four biological states.

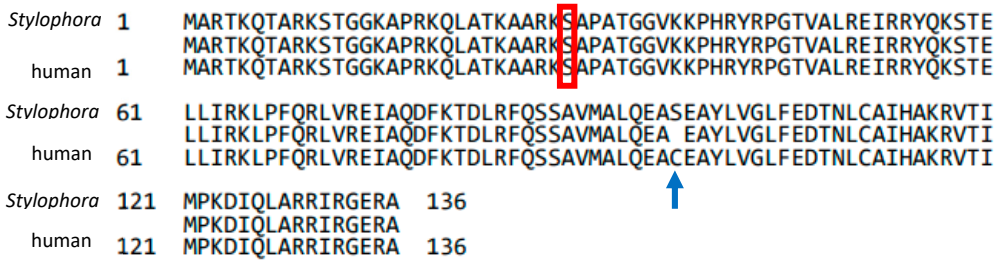
	RS Vs. REG	RS vs. FR	RS vs. TE	REG vs. TE
Nanos-1				0.049
Nanos-1-like	0.025	0.041	0.017	
Piwi-1	0.039	0.019	0.021	
Sox-2		0.039	0.045	0.04
Boule	0.003	0.0002	0.002	
Myc-1				0.002
Tudor-7		0.001		0.011
Tudor-5	0.018	0.021	0.003	

RS—resting state; Reg—10 hours post injury; FR—fully regenerated; and TE—tissue edge.

Supplementary Material S1. Specificity of antibody: p-histone H3 antibody (C-2): sc-374669, Santa Cruz.

This antibody is recommended for the detection of Ser28-phosphorylated histone H3 in several species, such as mouse and human species, using various methods, including immunofluorescence.

The sequence of *S. pistillata* H3 (XP_022792399.1) shares a remarkable 100% sequence identity with human histone H3.2 (NP_001005464.1) and *Mus musculus* histone H3.2 (NP_038576.1). It also shares a 99% sequence identity with *Mus musculus* histone H3.1 (NP_038578.2) and human histone H3.1 (NP_001363866.1, protein sequence comparison is shown in the scheme below).



The scheme illustrates the significant identity between the *S. pistillata* H3 protein and its human homologues. The same applies to the mouse homologues. The Ser28 residue is marked by a red rectangle, and the only difference in amino acid is indicated by a blue arrow. Given the similarity among human, mouse, and *S. pistillata* H3 proteins, especially with the sequences around Ser28 being completely identical, it is evident that the commercially used p-histone H3 antibodies should recognize the Ser28-phosphorylated histone H3 of *S. pistillata*.

Supplementary Material S2. Specificity of antibody: rabbit anti-*Bl*-Piwi polyclonals.

Methods: Protein extraction and Western blotting: Injuries were inflicted on coral 2D-spread tissue (Figures 1, 2a1-2) by tissue incisions made with a sharp knife. On the 4th day following the incisions, the tissues were re-injured using the same method. Two days after the second round of injuries (6 days from the onset), the tissues were flash-frozen in liquid nitrogen and stored at -70°C until processed. Naïve tissues were also collected for processing.

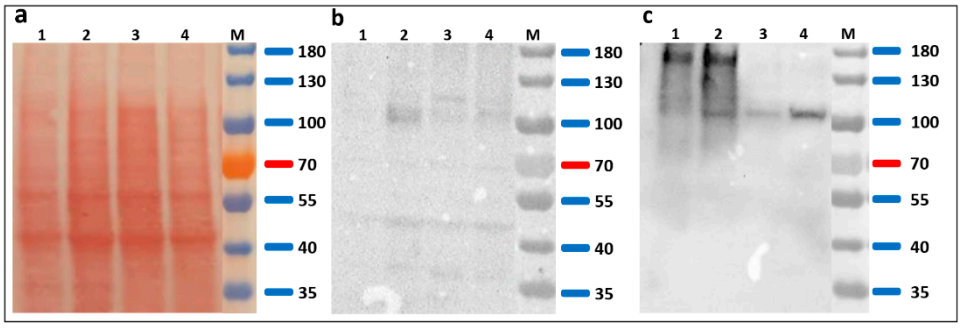
Cytoplasmic and nuclear protein fractions extraction: Six tissue samples from each treatment group (naïve or injured) were pooled together in 400 µl of cytoplasmic extraction buffer (10 mM HEPES, pH=7.9, 10 mM KCl, 1.5 mM MgCl2, 0.34 M sucrose, 10% glycerol, 1 mM DTT, Triton X-100 0.1%, protease inhibitor cocktail diluted 1:100 (cat. no. 5871, Cell Signalling Technology, Danvers, MA, US)) and vigorously shaken on ice for 30 minutes. The supernatant, containing the proteins of the cytoplasmic fraction, was collected following centrifugation (4 min, 1300g, 4°C), and the proteins of the nuclear fraction were extracted from the pellet using a nuclear solution (3 mM EDTA, 0.2 mM EGTA, 1 mM DTT, and protease inhibitor cocktail). The supernatant and cytoplasmic fractions were clarified by centrifugation (20 min, 20,000g, 4°C).

The proteins were separated on 10% SDS-PAGE gels (TGX™ FastCast™ Acrylamide Kit, 10% #1610173, Bio-Rad, Hercules, CA, US) and transferred to nitrocellulose membranes (Trans-Blot Turbo Transfer pack, cat. No. 1704159, Bio-Rad, Hercules, CA, US) using the Trans-Blot® Turbo™ Transfer System (Bio-Rad). The nitrocellulose membranes were stained with Ponceau S (cat. no. P7710, Sigma) to evaluate the transfer quality. Western blotting included membrane blocking performed using 5% skim milk in TBST. Afterward, the membranes were incubated overnight with either rabbit pre-immune serum or rabbit anti-*Bl-Piwi*, diluted 1:2000 in the blocking

solution. After washing (three times, 5 min each time in TBST), the blots were incubated for one hour with goat anti-rabbit IgG antibody, (H+L) HRP conjugate (cat. No. AP307P, Millipore, Burlington, MA, US), diluted 1: 10,000. The blots were washed (three times, 5 min, in TBST [Tris-HCl 0.2M, Tween-20 9mM, NaCl 1.5 M]) and immersed in an Immobilon Forte Western HRP substrate (cat. No. WBLUF0500, Millipore). Blot imaging was performed using the ChemiDoc XRS+ System (Bio Rad, Hercules, CA, US) and analysed using the Image lab software (Bio-Rad, Hercules, CA, US).

Results: The two *Stylophora pistillata* (coral) Piwi proteins (Piwi-like 1 accession no: XP_022806003.1, calculated MW 120 kDa, and Piwi-like 2 accession: XP_022780522.1, calculated MW 98.8 kDa) share a 48.5% protein sequence identity. The rabbit anti-*Bl-Piwi* was elicited against the polypeptide RDQSKARDFATKFNEVSRC [55], which shares 47% identity and 76% positives when compared to *S. pistillata* Piwi with accession: XP_022806003.1 and 44% and 56% similarities when compared to *S. pistillata* Piwi with accession no XP_022780522.1.

Western blot analysis (Figure below) reveals a specific band of about 104 kDa that is elevated in the cytoplasm of wounded tissues, in comparison to naïve or nuclear-enriched protein fractions. As the *Bl*-Piwi antibodies identify a specific protein whose size is in the expected range of the *S. pistillata* Piwi protein and its quantity in the cytoplasmic compartment of injured tissues is elevated, these results support the specificity of *Bl*-Piwi for *S. pistillata* Piwi and align with the immunohistochemical analyses shown in Figure 8.



Supplementary Figure S1. Western blot analysis of *Stylophora pistillata* Piwi protein using *Bl*-Piwi. Panel (**a**) shows Ponceau S staining of the blot; panel (**b**) shows staining with rabbit pre-immune serum; and panel (**c**) shows staining with *Bl*-Piwi antibody. Lane 1—nuclear-enriched extract from naïve tissues; lane 2–cytoplasm-enriched extract from naïve tissues; lane 3–nuclear-enriched extract from wounded tissues; and lane 4–cytoplasm-enriched extract from wounded tissues.