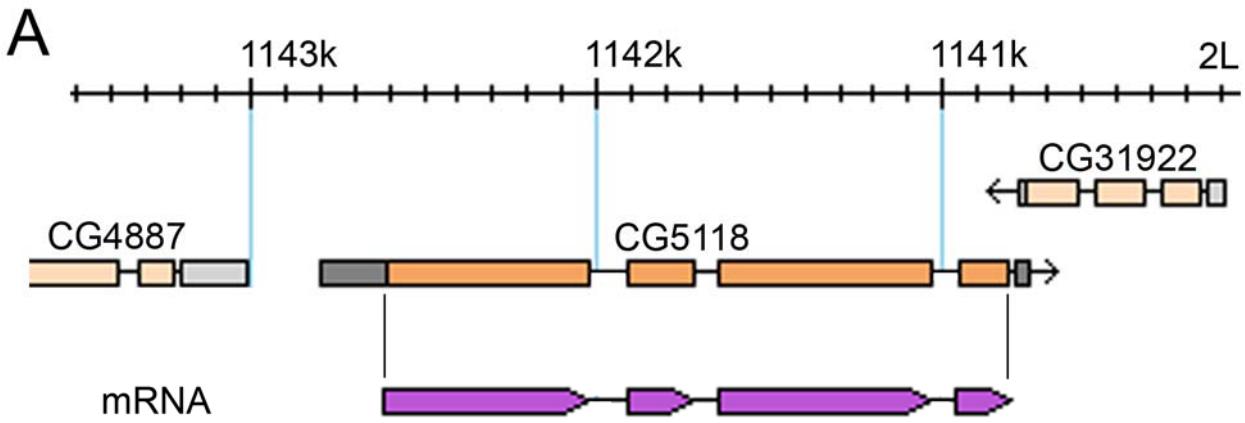

B

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  1  MKKVLLTKAQHIQPVVASSSDVLVIKPVGSLRVISISAPHSASTYIQRKS  50
 51  FQPVYEDIFKLLMKIPDKALTQRVIDAINESNNNDKVAISNHGKQCSCGV 100
101  QKVDSSTQTDWDTEETHMPGITNNSCEAKGNRHSHMTSSSSEPIKPPVDP 150
151  TKVPKKRGRKRNTCVPQVVKRSAAEMALKEREEKQLTPVITKRKKRDVTQ 200
201  AQNFESYQITNKTPVRRNSTLSDISFNSDELTRVENFISGNTEDRIIRIM 250
251  ANEFRKSHIMSEEGLLPIHEEILHGDVYGVKRQIFVCCHAKMDINELLTR 300
301  DGEDCLELALTNDTDTEIVSLILDARMMTDHLYENSNTALHLAVINHINI 350
351  ESIRLLLRRIDLNSLLLTNDDGYTVLHLAVRNNQFLVVEAILDSIDEREL 400
401  GETVYRRTLEAANPNEWDEKGFAKYYDRACERLELNKTLLKNRAHKRDVI 450
451  NASEARGGNPPLFYAVEGEQEHLCYFLLAHLADPDEENLSGHSPKSYHYE 500
501  YARTLRINLKVARVMEKVISILNT                            524
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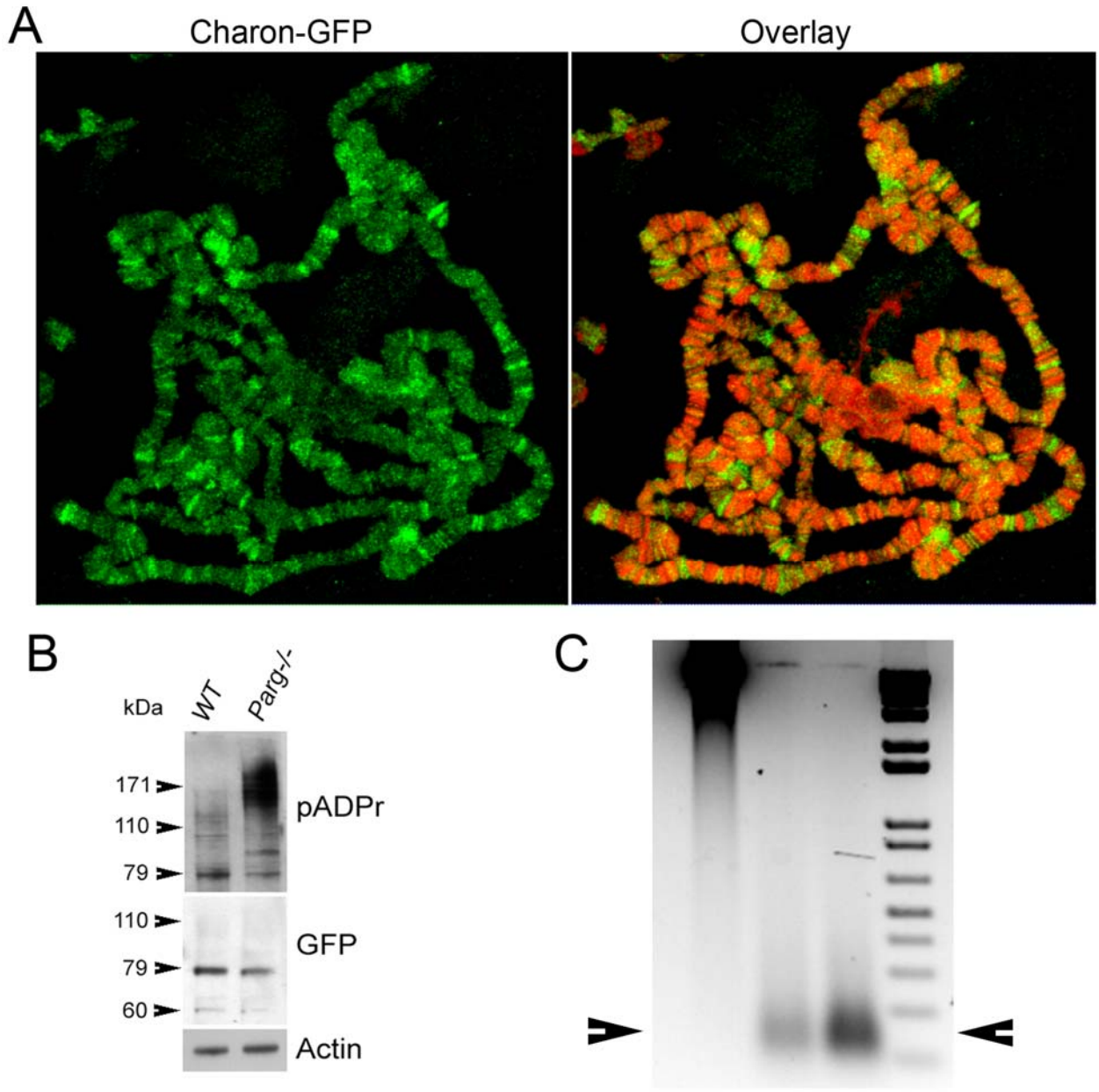
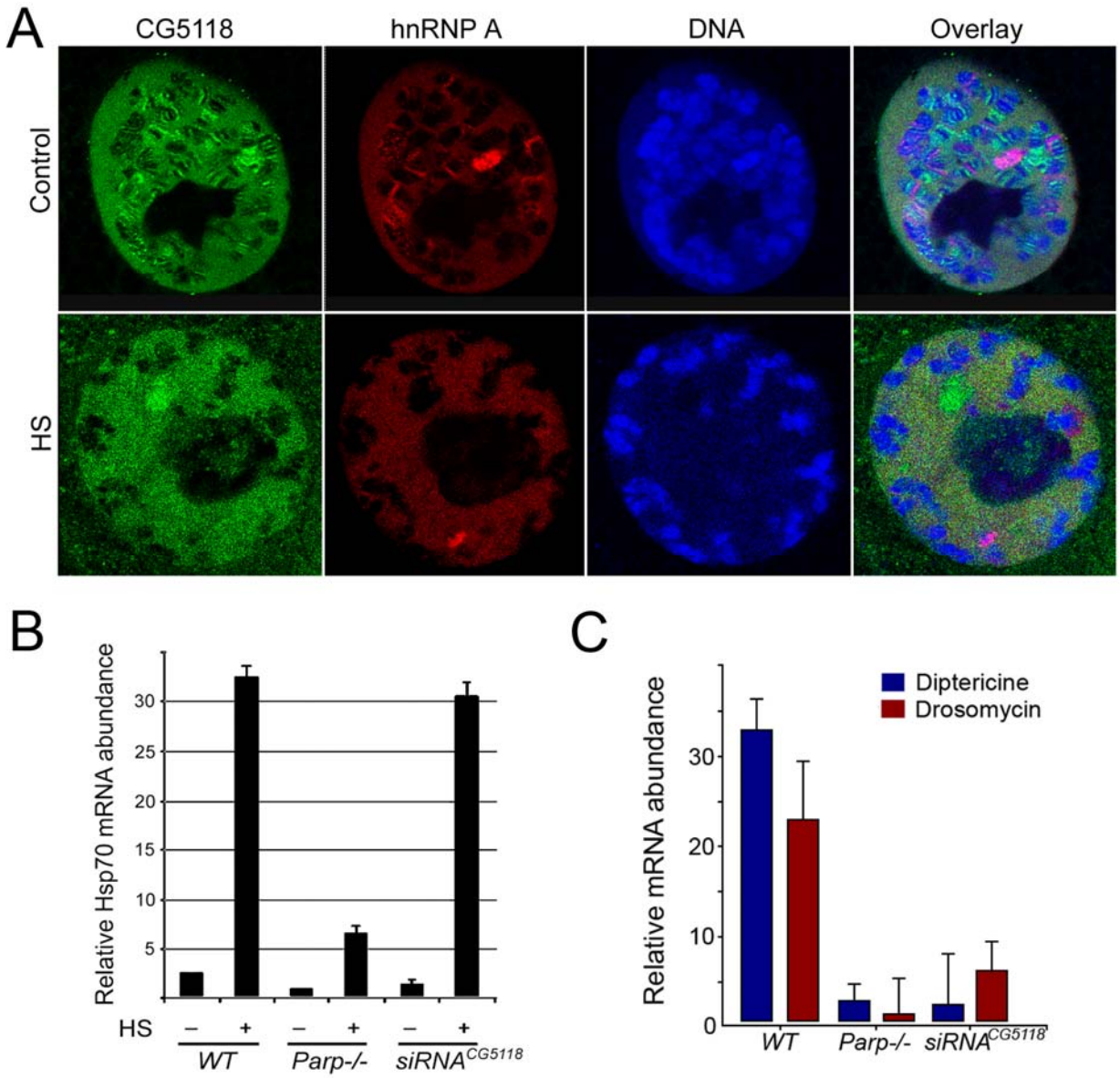
Supplemental Figure S1. The characterization of *Charon* locus in *Drosophila melanogaster*. A. Structure of *Charon* locus in *Drosophila* genome based on Flybase [26]. **B.** Sequence of *Drosophila* Charon protein. NLS sequences are shown in Red. Ankyrin repeats are shown in blue.

Supplemental Figure S2. Evolutionary conservation for Charon protein in *Insects*. Multiple sequence alignments of Charon family members: Lucilia - *Lucilia cuprina* (KNC31104.1)*;* CG5118 - *Drosophila melanogaster* (CG5118)*;* Musca - *Musca domestica* (101889410)*;* Ceratitis - *Ceratitis capitata* (101454591)*;* Bactrocera - *Bactrocera cucurbitae* (105210874).



Supplemental Figure S3. Charon is a nuclear chromatin associated protein. A. Charon protein labels ~ 400 loci in *Drosophila* chromatin: *Drosophila* larvae expressing Charon-GFP were cultured for 60hrs with *E.coli* bacteria; salivary glands were dissected from third instar larvae, squashed and stained with monoclonal anti-GFP (green) antibodys; DNA was detected using propidium iodide (red). **B.** Charon is not covalently modified with pADPr. Total nuclear protein extracts from animals expressing Charon-GFP wild type (WT) and *parg*$^{27.1}$ mutants were subjected to Western-blot analysis using ant-GFP, anti-pADPr antibodies. Proteins which covalently modified with pADPr demonstrate super-shift on Western-blots in *parg* mutants. Band corresponding to Charon-GFP does not demonstrate this. Anti-Actin antibody was used as a loading control. **C.** Quality control of MNase treated mononucleosomal chromatin for Co-IP and ChIP. Digested chromatin was checked on agarose gel after different time intervals of incubation with MNase.



Supplemental Figure S4. Charon represents a third PARP-1 dependent pathway. A. Charon protein is not required for PARP-1-dependent hnRNP-A1 pathway: Charon-GFP (green) is not colocalized with a key component of PARP-1 dependent RNA-fate-regulating pathway, hnRNP A1 (red) genomewide in steady state conditions (top). Upon heat shock hnRNP A1 remains bound to chromatin, while Charon relocalizes to soluble nucleoplasm. **B.** *Charon* is not required for PARP-1-depednent *hsp70* gene transcription. Quantitative RT-PCR experiment is shown. The amounts of PARP-1 dependent hsp70 mRNA was detected using specific primers before (-) and after (+) 30 minutes heat shock in wild type (WT), *parp-1* mutants and *Charon* knockdowns. **C.** Expression of endogenous *Drosomycin* and *Diptericine* genes was detected after 20 hours infection in *wild type (WT)*, *Parp-1*^{C03256} mutant, and *siRNA*^{CG5118}-*expressing* third instar larvae using quantitative RT-PCR.