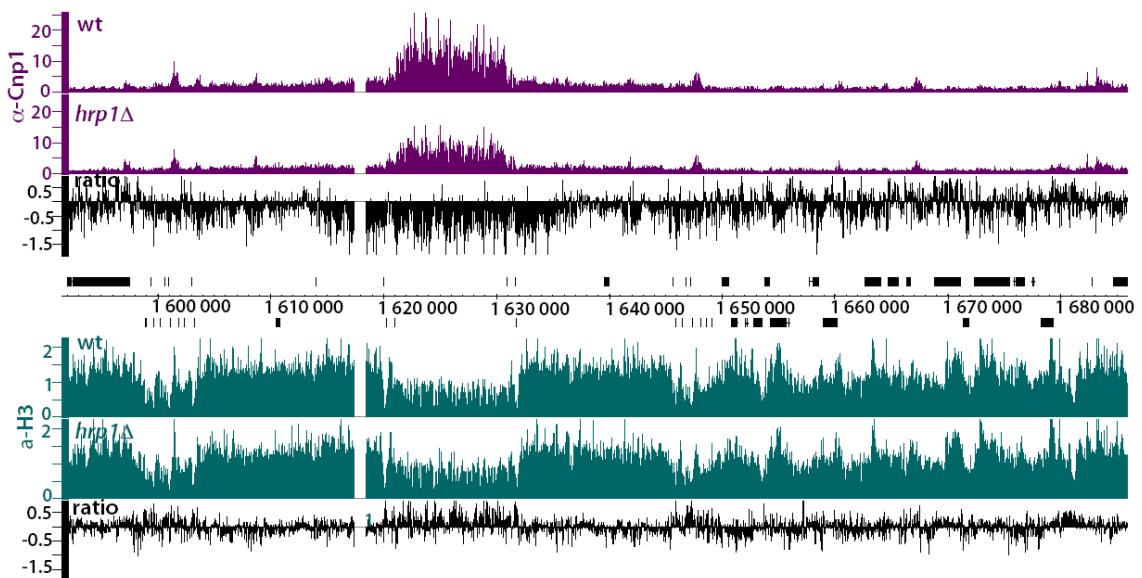
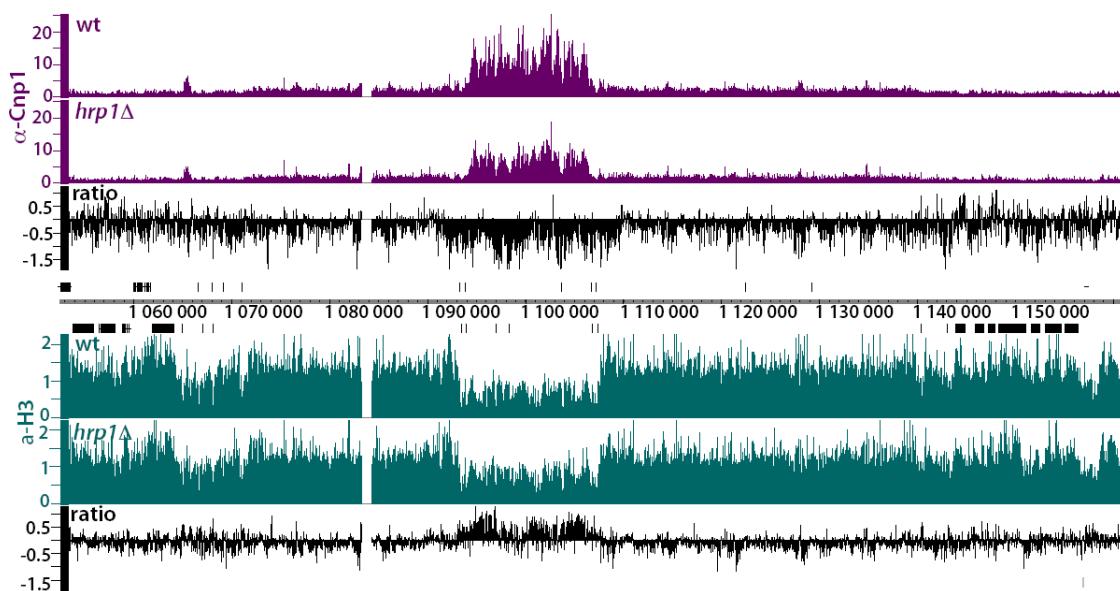
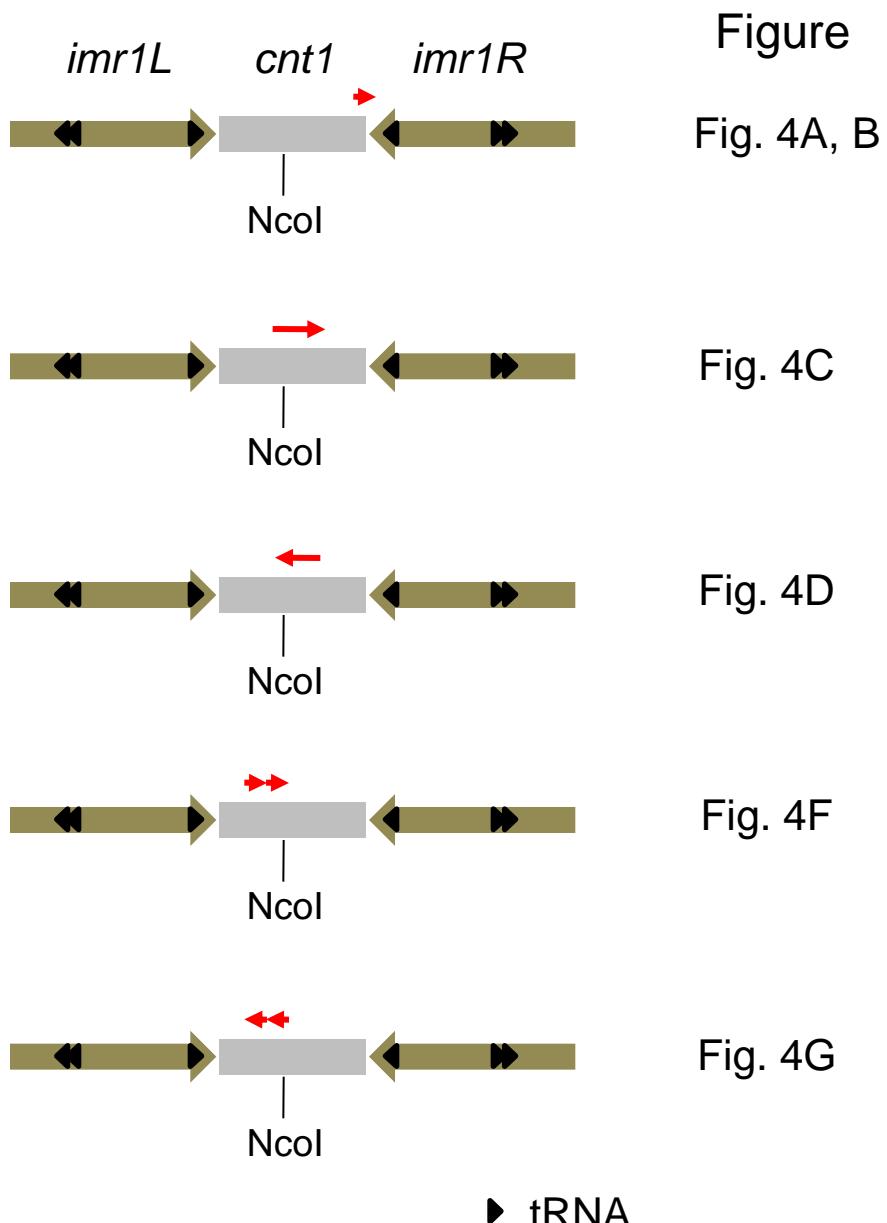


A**centromere 2****B****centromere 3****Supplemental Fig. S1**

The chromosome physical map shows ChIP-chip binding profiles for CENP-A^{Cnp1} (purple) and H3 (green) for centromere 2 (A) centromere 3 (B) in wild-type and *hrp1Δ* (as indicated) cells at 30°C. The relative ratios of CENP-A^{Cnp1} and H3 are indicated (black).

Location and orientation of Northern probes (red arrow)

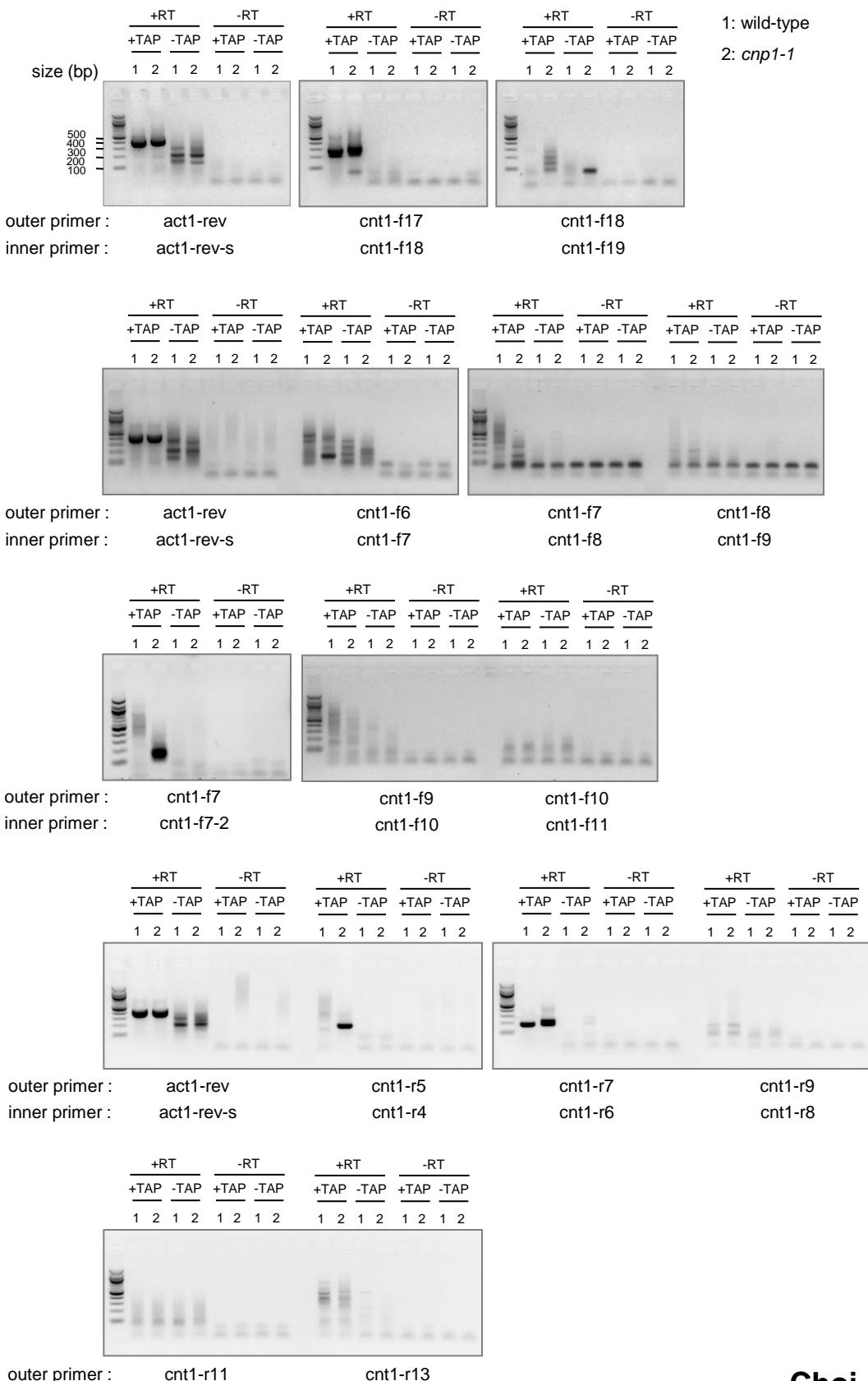


Supplemental Fig. S2

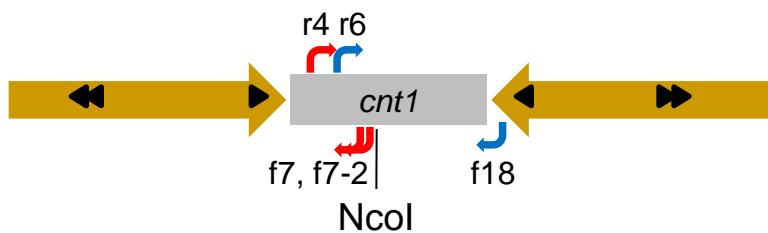
Schematic of RNA probes used in Northern analysis. Location and orientation of probes are indicated by red arrows. Probes used in Fig. 4F and Fig. 4G were generated by *in vitro* transcribing a mixture of two relatively short DNA fragments with equal amount in order to maximize production of full length RNA.

A

5'-RACE-PCR analysis to detect transcription start sites within *cnt1*



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Supplemental Fig. S3

B**centromere 1**red: transcription starts in *cnp1-1* onlyblue: transcription starts in wild-type and *cnp1-1***C**

wild-type, f18:

AATTATACTAGCCCTAAGAA

cnp1-1, f18:AATTATACTAGCCCTAAGAA
ATAACCACAAATCAACGATT*cnp1-1*, f7:

ATAACGCCGCCGGTCGCCAA

cnp1-1, f7-2:

AGACATTACCAACAAAAAGA

cnp1-1, r4:

GAACTAAAAGTAATCAATAC

wild-type, r6:

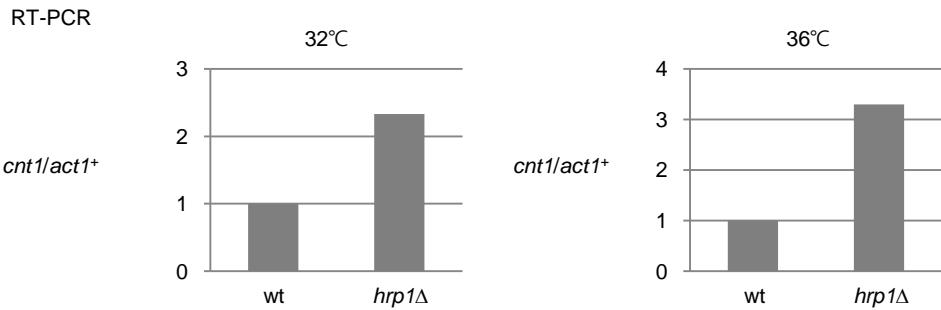
ATGAAATACACACCACTCAC

cnp1-1, r6:

ATGAAATACACACCACTCAC

Supplemental Fig. S3

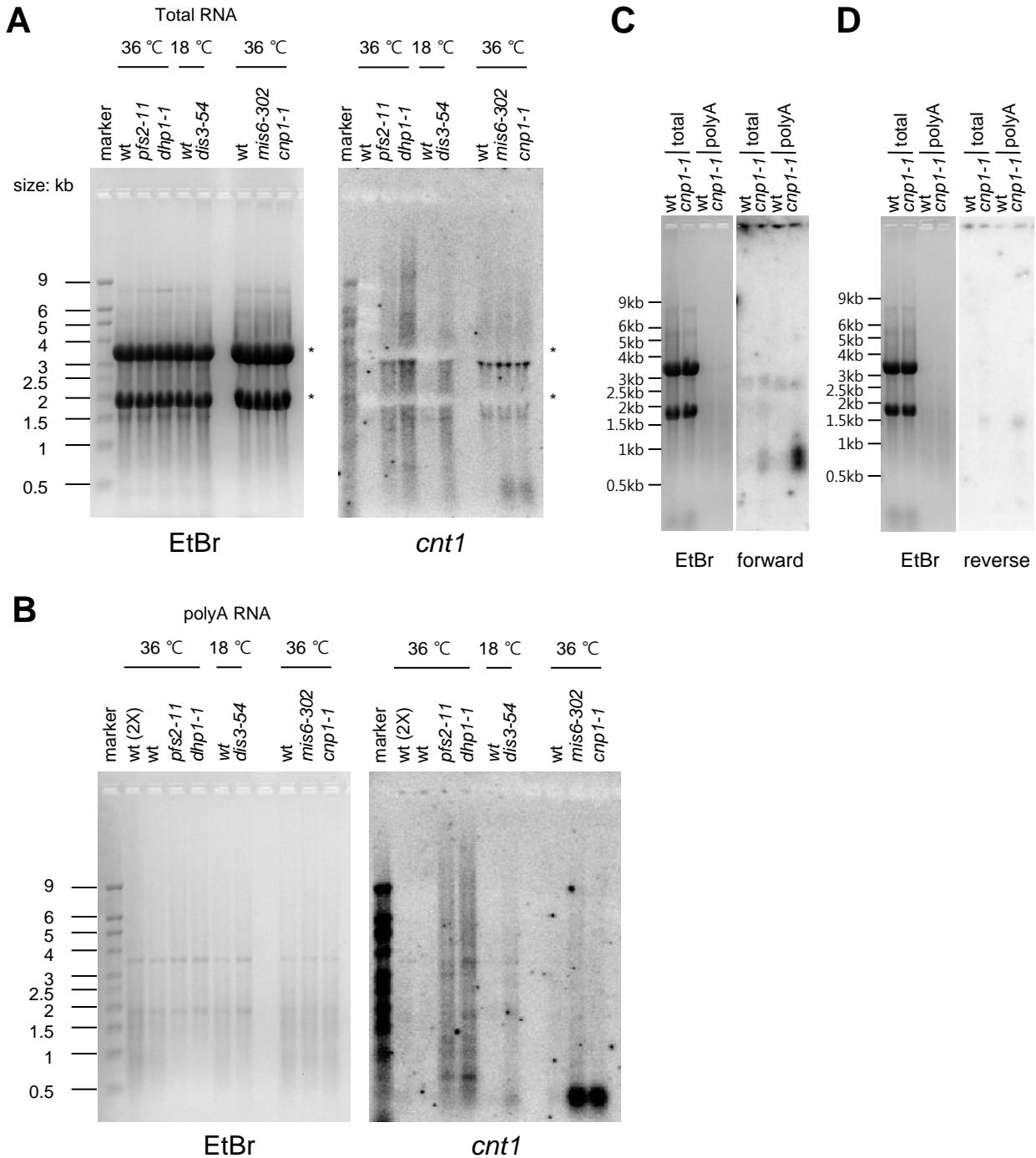
Detection of transcription start sites (TSS) within *cnt1*. (A) RNA ligase mediated (RLM) 5'-RACE-PCR was performed on polyA-selected RNA extracted from wild-type or *cnp1-1* grown at 36°C. -TAP shows that the product from +TAP reaction is specific to the 5' ends of decapped RNA. Outer and inner (nested) PCR Primers specific for *cnt1* or *act1⁺* were described in Supplemental Table 1B. (B) Schematic of representation of transcriptional start sites determined by 5'-RACE-PCR in wild-type and *cnp1-1* cells with utilized inner primers corresponding to each TSS indicated. (C) 20 bp sequences of each TSS in wild-type and *cnp1-1*.



Supplemental Fig. S4

Loss of Hrp1 causes accumulation of central domain transcripts. RT-PCR analysis to measure levels of central domain transcripts in wt and *hrp1* Δ grown at 32°C or 36°C. Levels of *cnt1* transcripts were calculated relative to *act1*⁺ control by qRT-PCR. Bars in the graph indicate fold increase relative to wt.

Choi et al., Supplemental Fig. S4



Supplemental Fig. S5

Images of Ethidium Bromide (EtBr) stained gels used in Northern analysis (inverted). (A) An additional information to Fig. 4A. Asterisk: position of rRNA bands which interfere with hybridization. (B) An additional information to Fig. 4B. Purity of poly(A) RNA after oligo-dT selection is monitored by loss of rRNA bands after EtBr staining. (C,D) Additional informations to Fig. 4C,D.