Supplementary Materials

NANOG/NANOGP8 Localizes at the Centrosome and Is Spatiotemporally Associated with Centriole Maturation

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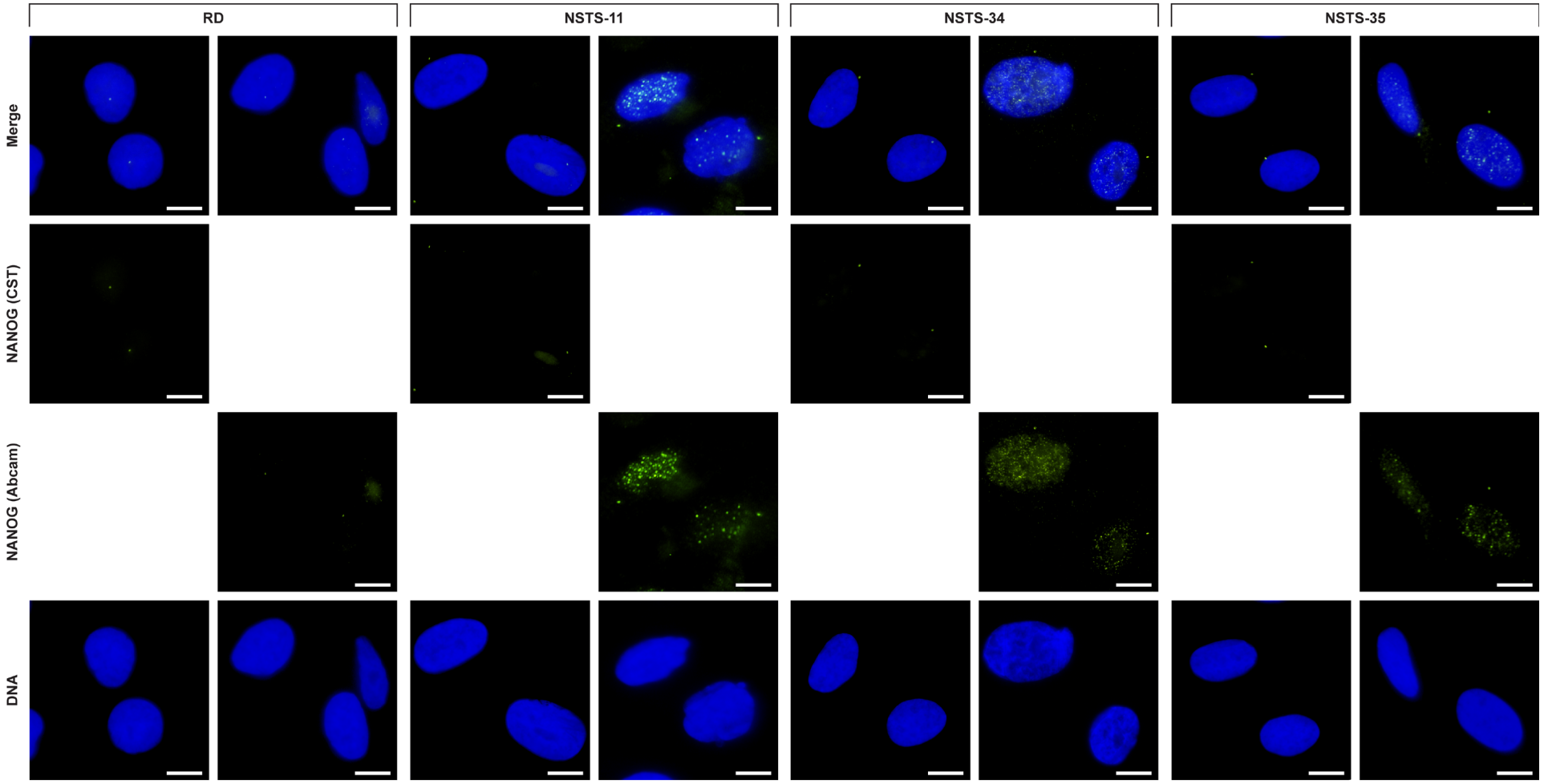

Figure S1. Unmerged images from Figure 1b. Localization of NANOG (green) was detected using two anti-NANOG antibodies (#4903, Cell Signaling Technology [CST] and #ab109250, Abcam). The nuclei were counterstained with Hoechst 33342 (blue). Scale bars, 10 μm.

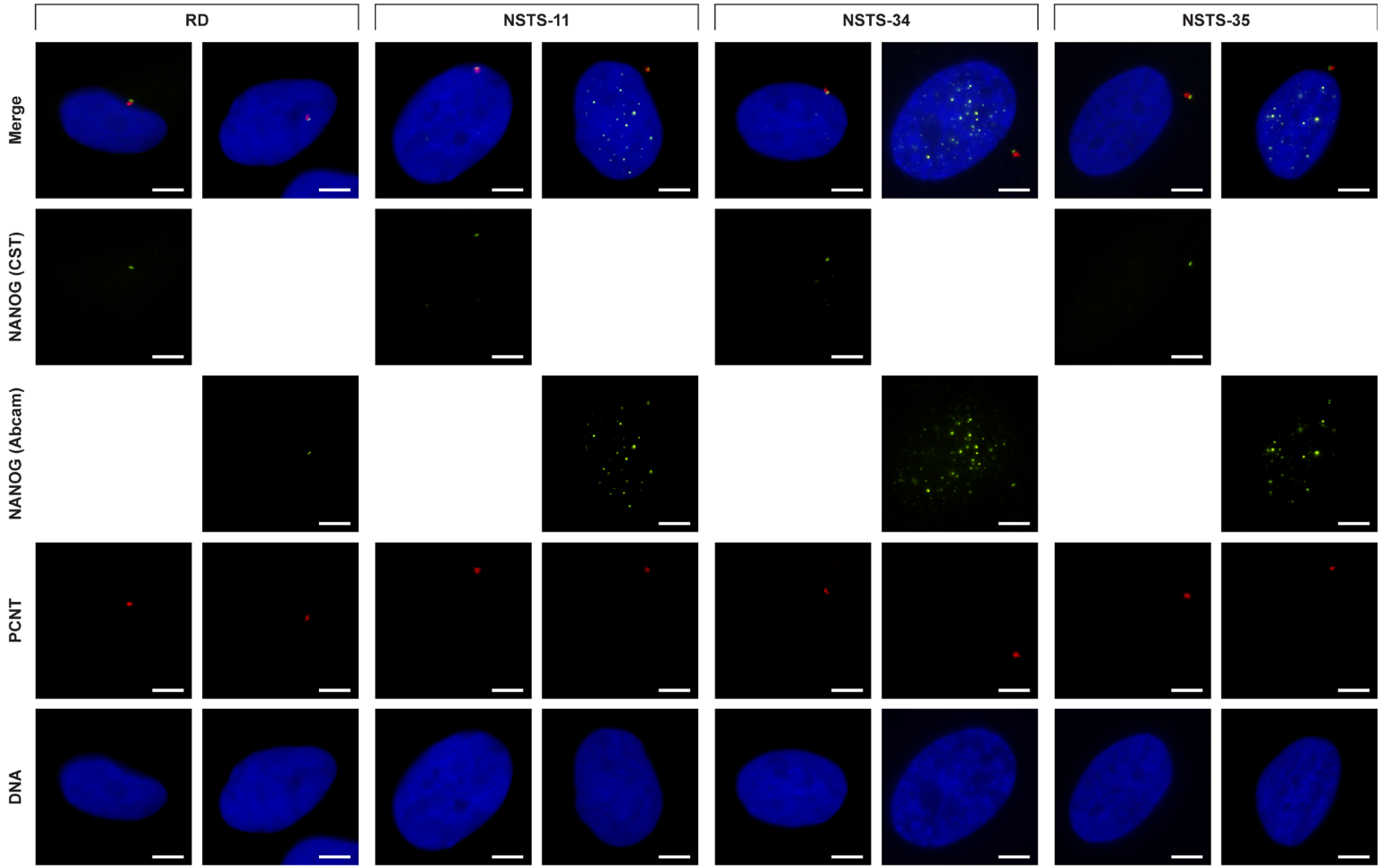

Figure S2. Unmerged images from Figure 3a. Localization of NANOG (green) was detected using two anti-NANOG antibodies (#4903, Cell Signaling Technology [CST] and #ab109250, Abcam) and anti-pericentrin antibody (PCNT; red). The nuclei were counterstained with Hoechst 33342 (blue). Scale bars, 5 μm.

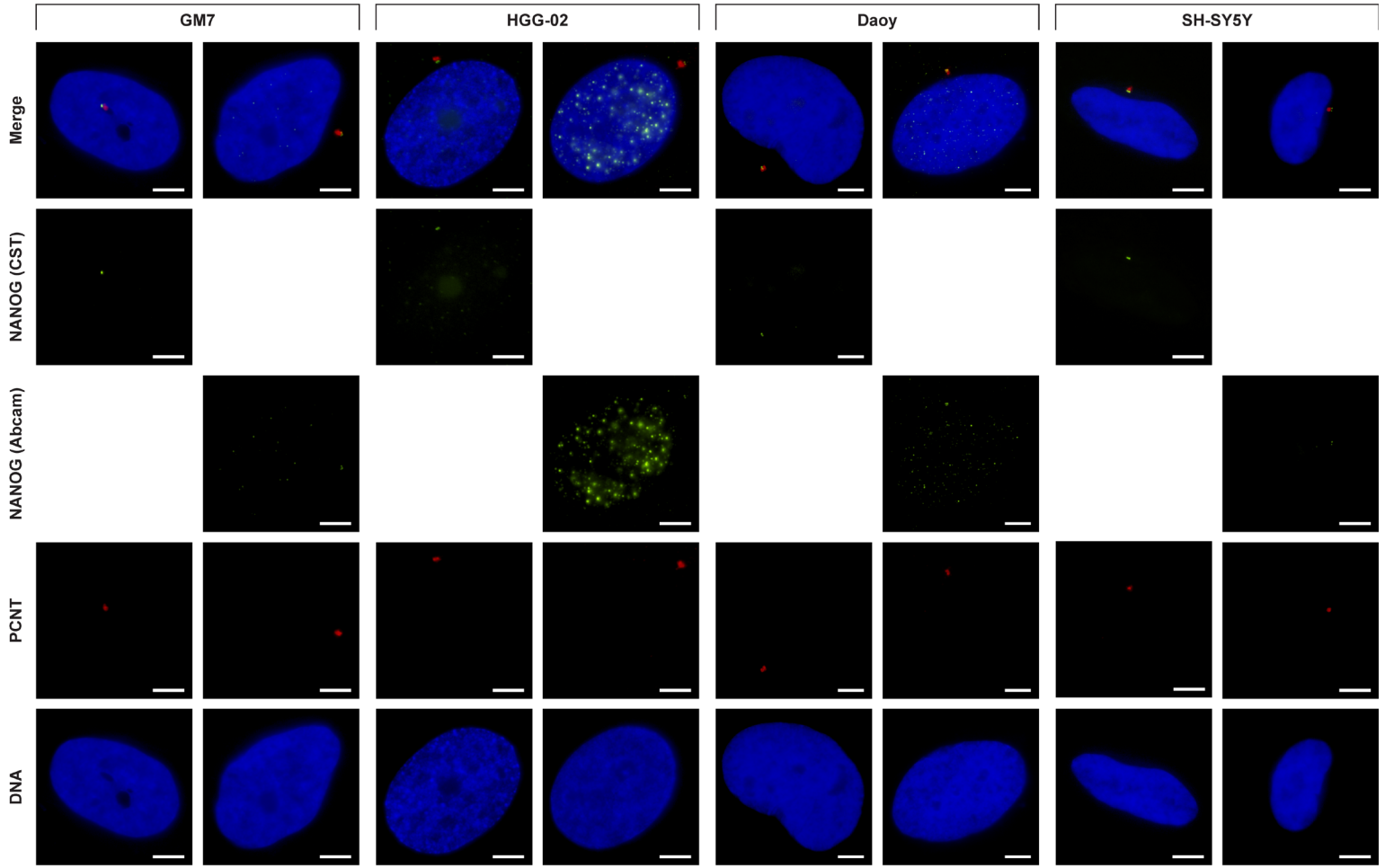

Figure S3. Unmerged images from Figure 3b. Localization of NANOG (green) was detected using two anti-NANOG antibodies (#4903, Cell Signaling Technology [CST] and #ab109250, Abcam) and anti-pericentrin antibody (PCNT; red). The nuclei were counterstained with Hoechst 33342 (blue). Scale bars, 5 μm.

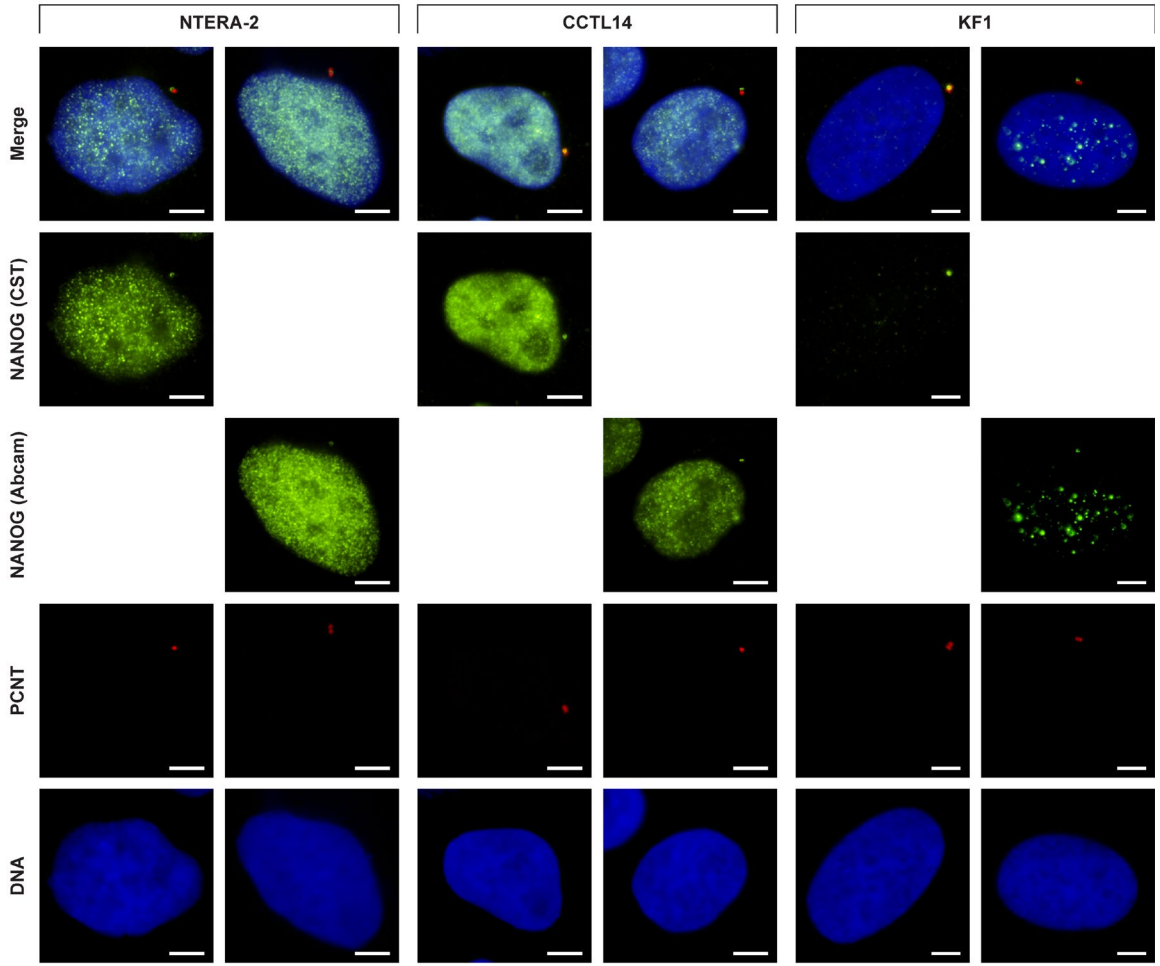

Figure S4. Unmerged images from Figure 3c and Figure 4. Localization of NANOG (green) was detected using two anti-NANOG antibodies (#4903, Cell Signaling Technology [CST] and #ab109250, Abcam) and anti-pericentrin antibody (PCNT; red). The nuclei were counterstained with Hoechst 33342 (blue). Scale bars, 5 μm.

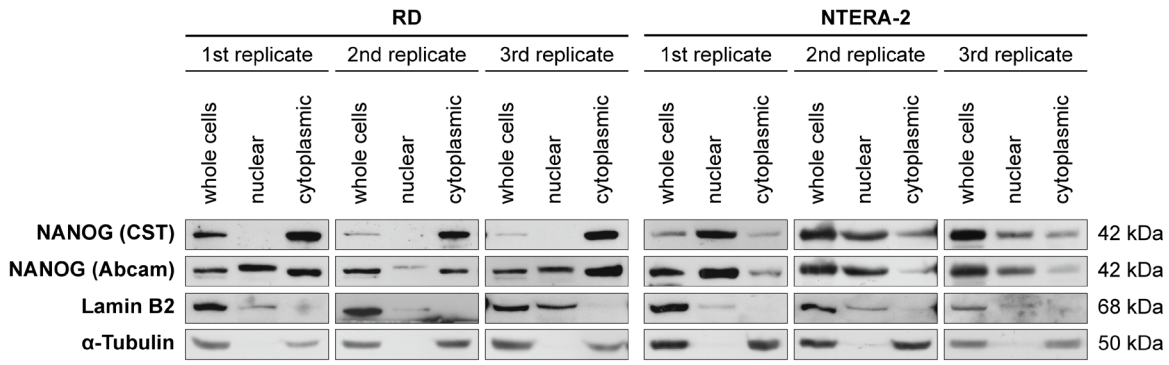

Figure S5. Western blot images of NANOG immunodetection in three biological replicates of the nuclear and cytoplasmic fractions of RD and NTERA-2 cells. Nuclear/cytoplasmic fractionation followed by immunoblotting was performed using two independent anti-NANOG commercial antibodies (#4903, Cell Signaling Technology [CST] and #ab109250, Abcam). Lamin B2 and α-tubulin served as controls of the nuclear fraction and cytoplasmic fraction purity, respectively.

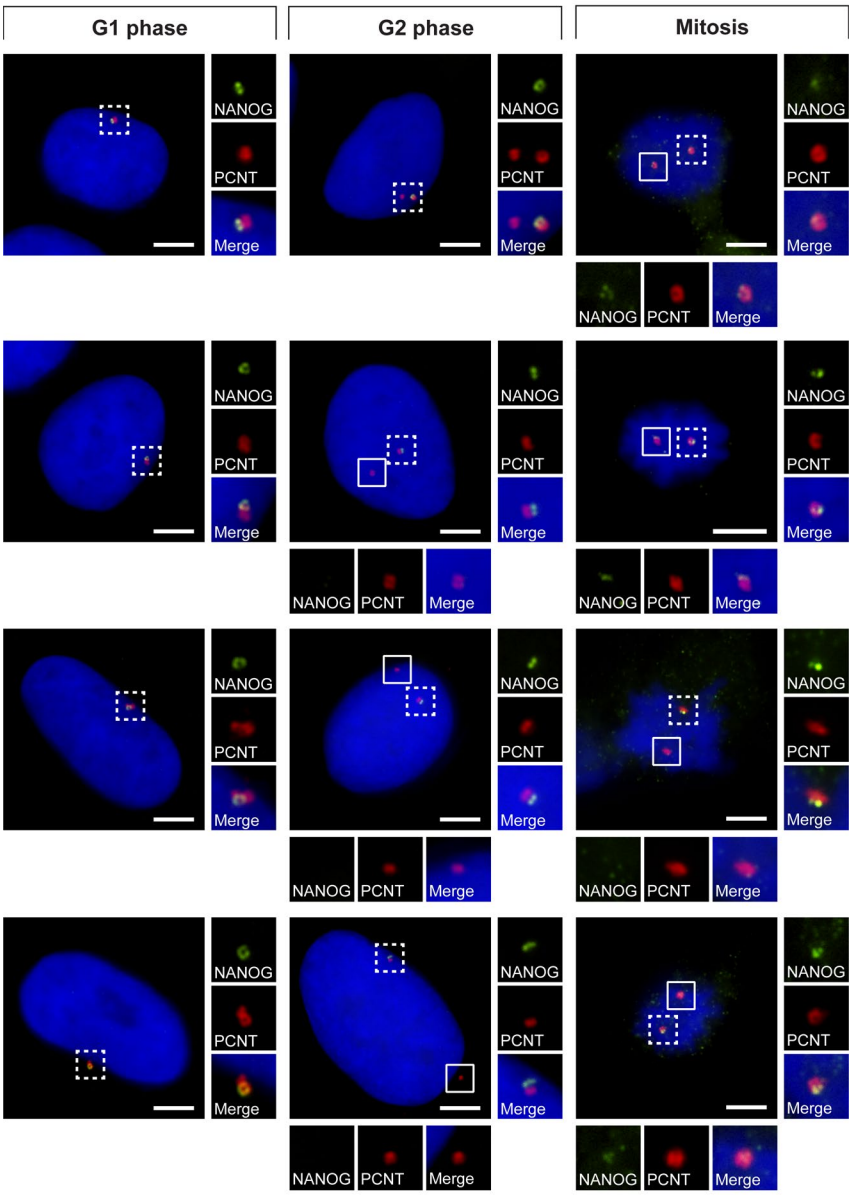

Figure S6. NANOG localization during the cell cycle. NANOG (green; #4903, Cell Signaling Technology [CST]) colocalized with one centrosome (pericentrin, PCNT; red) only during G1 and G2 phases but with both centrosomes (pericentrin, PCNT; red) during mitosis. The nuclei were counterstained with Hoechst 33342 (blue). For each image, regions of interest are indicated by the dashed and solid boxes, and the close-ups are provided on the right and below, respectively. Scale bars, 5 μm.

Table S1. Validation of anti-NANOG #ab109250 antibody specificity by mass spectrometry of immunoprecipitated NTERA-2 cells.

Accession	Gene	Gene symbol	Peptides Count	Spectra Count
P35580	Myosin-10	*MYH10*	45	144
Q13813	Spectrin alpha chain, non-erythrocytic 1	*SPTAN1*	31	70
Q01082	Spectrin beta chain, non-erythrocytic 1	*SPTBN1*	23	51
P35579	Myosin-9	*MYH9*	14	42
Q16643	Drebrin	*DBN1*	13	38
P11142	Heat shock cognate 71 kDa protein	*HSPA8*	11	41
Q8TEW0	Partitioning defective 3 homolog	*PARD3*	10	23
P05787	Keratin, type II cytoskeletal 8	*KRT8*	9	30
P10809	60 kDa heat shock protein, mitochondrial	*HSPD1*	8	20
P14866	Heterogeneous nuclear ribonucleoprotein L	*HNRNPL*	8	43
P25705	ATP synthase subunit alpha, mitochondrial	*ATP5F1A*	8	33
Q9UQ35	Serine/arginine repetitive matrix protein 2	*SRRM2*	8	12
O15164	Transcription intermediary factor 1-alpha	*TRIM24*	7	30
P05783	Keratin, type I cytoskeletal 18	*KRT18*	7	26
O00264	Membrane-associated progesterone receptor component 1	*PGRMC1*	5	13
P04843	Dolichyl-diphosphooligosaccharide--protein glycosyltransferase subunit 1	*RPN1*	5	11
P11021	Endoplasmic reticulum chaperone BiP	*HSPA5*	5	7
P60660	Myosin light polypeptide 6	*MYL6*	5	18
P78527	DNA-dependent protein kinase catalytic subunit	*PRKDC*	5	8
Q00839	Heterogeneous nuclear ribonucleoprotein U	*HNRNPU*	5	8
Q07157	Tight junction protein ZO-1	*TJP1*	5	9
Q92734	Protein TFG	*TFG*	5	22
P01870	Ig gamma chain C region		4	23
P04406	Glyceraldehyde-3-phosphate dehydrogenase	*GAPDH*	4	16
P08670	Vimentin	*VIM*	4	8
P09493	Tropomyosin alpha-1 chain	*TPM1*	4	28
P35221	Catenin alpha-1	*CTNNA1*	4	5
P43243	Matrin-3	*MATR3*	4	5
P61978	Heterogeneous nuclear ribonucleoprotein K	*HNRNPK*	4	8
P62805	Histone H4	*HIST1H4A*	4	32
Q9H444	Charged multivesicular body protein 4b	*CHMP4B*	4	14

Accession	Gene	Gene symbol	Peptides Count	Spectra Count
Q9NYL9	Tropomodulin-3	*TMOD3*	4	5
P05023	Sodium/potassium-transporting ATPase subunit alpha-1	*ATP1A1*	3	5
P23396	40S ribosomal protein S3	*RPS3*	3	9
P35613	Basigin	*BSG*	3	12
P46779	60S ribosomal protein L28	*RPL28*	3	5
P49411	Elongation factor Tu, mitochondrial	*TUFM*	3	7
P52272	Heterogeneous nuclear ribonucleoprotein M	*HNRNPM*	3	7
P56747	Claudin-6	*CLDN6*	3	10
Q07020	60S ribosomal protein L18	*RPL18*	3	9
Q07955	Serine/arginine-rich splicing factor 1	*SRSF1*	3	5
Q15393	Splicing factor 3B subunit 3	*SF3B3*	3	4
Q16629	Serine/arginine-rich splicing factor 7	*SRSF7*	3	13
Q2KHR3	Glutamine and serine-rich protein 1	*QSER1*	3	6
Q5PRF9	Protein Smaug homolog 2	*SAMD4B*	3	4
Q6NSW7	**Homeobox protein NANOGP8**	***NANOGP8***	3	6
	GKQPTSAENSVAK (NANOGP8)			
	QPTSAENSVAK (NANOGP8)			
	TVFSSTQLCVLNDR (NANOG, NANOGP1 and NANOGP8)			
O75569	Interferon-inducible double-stranded RNA-dependent protein kinase activator A	*PRKRA*	2	3
P04792	Heat shock protein beta-1	*HSPB1*	2	8
P06753	Tropomyosin alpha-3 chain	*TPM3*	2	11
P09496	Clathrin light chain A	*CLTA*	2	4
P17302	Gap junction alpha-1 protein	*GJA1*	2	8
P35232	Prohibitin	*PHB*	2	3
P36578	60S ribosomal protein L4	*RPL4*	2	5
P46776	60S ribosomal protein L27a	*RPL27A*	2	7
P46782	40S ribosomal protein S5	*RPS5*	2	5
P63244	Receptor of activated protein C kinase 1	*RACK1*	2	3
P78310	Coxsackievirus and adenovirus receptor	*CXADR*	2	8
Q00796	Sorbitol dehydrogenase	*SORD*	2	8
Q14257	Reticulocalbin-2	*RCN2*	2	5
Q15392	Delta(24)-sterol reductase	*DHCR24*	2	12
Q9BWH2	FUN14 domain-containing protein 2	*FUNDC2*	2	6