

# 正井 久雄 研究室 研究概要

2022 年 4 月 14 日

## 研究テーマ

- (1) ゲノム複製メカニズムの普遍性と多様性の解明と複製システムの進化
- (2) 核内クロマチン構造・配置の制御と複製の時空間プログラム制御機構
- (3) 細胞内グアニン 4 重鎖構造核酸の形成メカニズムと存在様式の解明とその生物学的意義の解明
- (4) 複製障害に対する細胞応答機構と、種々の生体ストレス応答経路とのクロストークの解明
- (5) 複製因子、複製チェックポイント因子などを標的とした新規な創薬戦略の開発
- (6) 複製因子の個体の分化発生・制御における機能の解明

これに加えて、最近、京都大学医学部 准教授から当研究室に赴任された笹沼博之博士は、乳がんの発生機序の研究を行なっておられます。興味のある方は別途説明しますのでご連絡ください。

## 研究活動のまとめ

ゲノム DNA 複製は、細胞の増殖・分化において中心的な役割を果たす。ゲノムは細胞の増殖に伴い、正確に、高速に、秩序正しく複製されなければならない。この過程に異常が生じると、がん細胞や老化細胞に見られるゲノムの遺伝的不安定性（変異、染色体の欠損や再編成など）を引き起こすことは容易に想像できる。実際、最近の研究から、内的・外的な原因による複製障害に対する適切な細胞応答の破綻が、初期がん細胞の遺伝的変化の主要な要因になっていることが示されている。

ゲノム複製に関して、3 個の重要な事象が知られている。

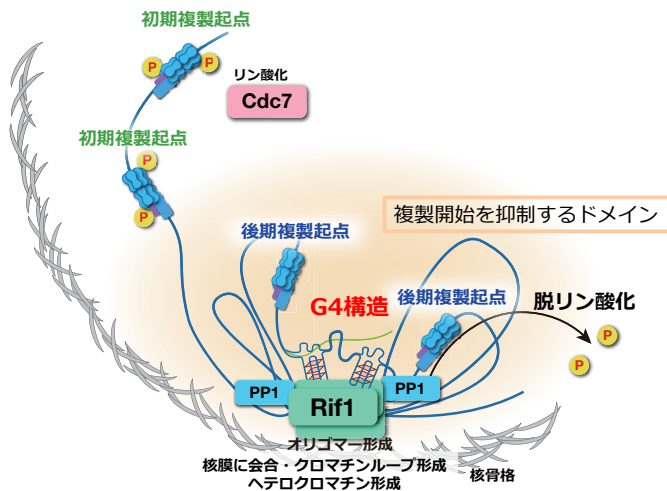
- ① ゲノム複製は細胞周期に一度のみ起こり、細胞分裂と厳密に連動する。
- ② ゲノム複製は、空間的、時間的な制御の元、すべてのゲノムの複製を完了する。またこの過程はエピゲノム情報の維持や、変化とも連動する。
- ③ 種々の要因で起こる DNA 複製の停止に細胞が迅速に応答し、ゲノム安定性を維持する。

上記の事象のうち、①と③の破綻は、直ちにゲノム変化、細胞の癌化に直結する。②は、DNA 複製のみならず、ゲノムの修復、変動、機能発現（遺伝子発現）

などとも関連し、生物の可塑性、適応性の基盤となる。私達は、これらに関連する問題を解決するために、主に以下の4個の研究課題を推進している。

### 1) 染色体 DNA 複製の開始とその時空間的制御の分子機構

真核細胞ゲノム複製では多くの部位から複製は開始する。また複製の時間的タイミングや、核内での複製部位の局在は、細胞型特異的な制御下にあり、染色体の核内配置、高次構造やエピゲノム情報などにも影響される(図1)。



Kanoh, Y. et al. (2015) *Nat. Struct. Mol. Biol.* 22: 889-897  
 Toteva, T. et al. (2017) *Proc. Natl. Acad. Sci. USA.* 114: 1093-1098.

図1 Rif1による染色体ドメインの形成のモデル: Rif1は多量体を形成し複数のG4構造を有するDNAに結合し、核膜近傍に染色体ループ形成を促す。これにより複製や転写の制御ドメインの形成を促進する可能性がある(加納ら、*Nature Structure Molecular Biology*, 2015より)。

### 意義の解明

これまでの研究からグアニン4重鎖など非B型DNA構造が複製開始や染色体の高次構造構築などにおいて重要な役割を果たすことを明らかにしてきた(図1および図2)。特に、グアニン4重鎖は、ゲノム上および転写されたRNA上に非常に頻繁に存在すると想像されており、ゲノム機能の未知の情報を担う可能性が

### 2) 特殊DNA構造の生物学的

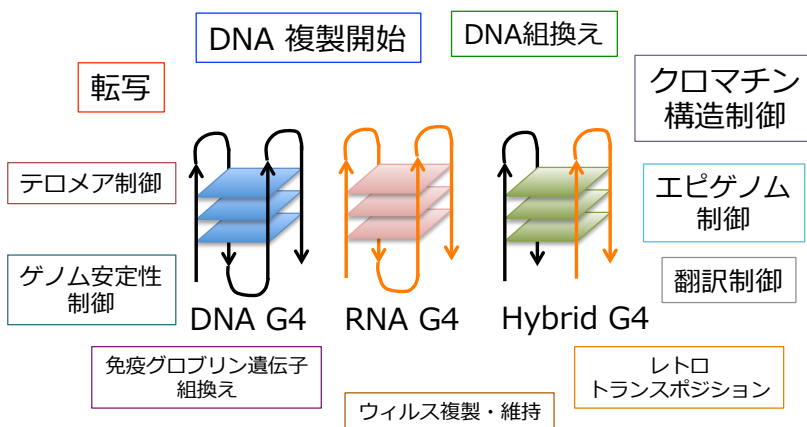


図2 G4(グアニン4重鎖)の多様な機能: G4は多様な形態でDNAおよびRNAあるいはDNA-RNA hybrid上に形成され、核酸が関与する多種多様な反応を制御する。

ある。

### 3) 複製ストレスに対する細胞応答の分子機構の解明とその新規制癌戦略への応用

複製障害は、がん化や老化のもっとも直接的な原因となる。従って、複製ストレスによる、細胞応答機構の解明は、がん化、老化の根本的メカニズム解明に必須である (図3)。さらに私達は最近複製ストレス応答が、浸透圧、酸化、温度、最近感染、低酸素など種々の生体ストレスとクロストークしていることが明らかとなり、これらの生体ストレスがゲノム安定性に及ぼす影響を明らかにする (図4)。

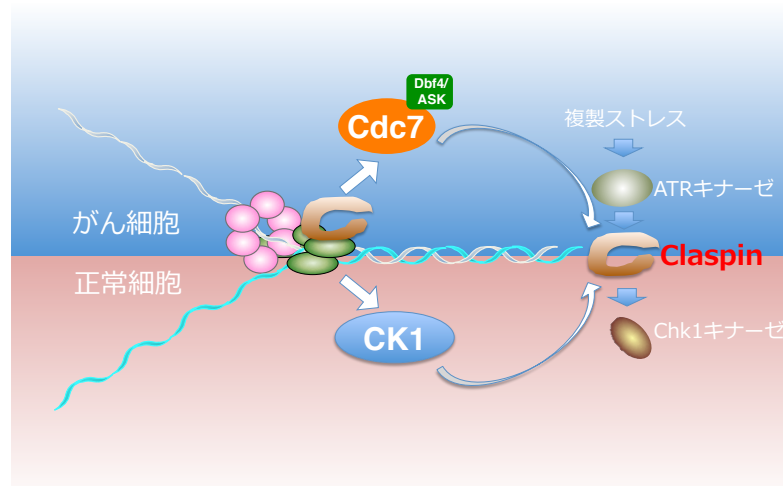


図3 がん細胞と正常細胞の DNA 複製停止に対する細胞応答の違い：複製が予期せずに停止すると細胞は、図の右側に記載される複製チェックポイント反応を誘導する。Claspin はこの反応経路で、複製停止の信号を伝えるために重要な役割を果たす。がん細胞と正常細胞は、異なるタンパク質を用いてこの経路を活性化する。すなわちがん細胞は Cdc7 キナーゼを用いるが、正常細胞では主に CK1 キナーゼに依存する。この違いを用いて、Cdc7 を標的としてがん細胞のみを選択的に除去する治療戦略が可能となる。実際に Cdc7 を制癌剤の標的として現在新規制がん剤の開発を進めている。

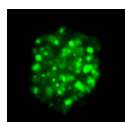
## がん細胞の発生

### 種々の生体ストレス

(栄養、温度、酸化、浸透圧、低酸素、細菌感染など)

複製障害[前癌状態]  
(DNA損傷シグナル)

核の中に現れた  
DNA損傷のfoci



DNA損傷応答機構の崩壊

腫瘍：悪性転化

### 図4 がん細胞の発生：

種々の生体ストレスは複製障害を誘導する可能性がある。複製障害は一時的にDNA損傷をゲノム上に誘導するが、修復される。しかしDNA損傷応答機構が崩壊すると細胞は腫瘍化する。

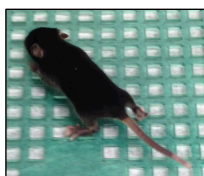
#### 4) 複製因子の個体レベルでの機能と疾患への関与

複製因子が、個体の発生、臓器・組織の発生・機能において担う役割を、臓器・組織特異的ノックアウトマウスや、遺伝子改変動物の表現型の解析に基づき明らかにする(図 5)。

多様な生物種と、手法を用いることにより、ゲノム複製のメカニズムの多様性と普遍性を解明し、さらに、核酸の形態が担う、ゲノムに隠された未知の機能シグナルの生物学的意義を解明する。得られた成果を基盤として、がんなどの疾患の予知、予防、診断、治療の新規戦略を開発する。

生後 1 2 日

コントロール

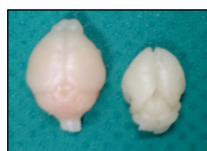


Cdc7 cKO



脳

Control Cdc7 cKO

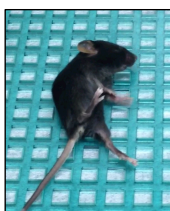


生後 1 8 日

コントロール



Cdc7 cKO



脳

Control Cdc7 cKO

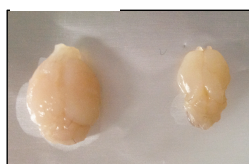


図 5 Cdc7 (f/+) *Nestin<sup>Cre</sup>* マウスは脳の形成異常、成長遅延を示す：神経幹細胞において Cdc7 をノックアウトすると、マウスは誕生するが、生後の脳の形成異常、成長遅延を示し、生後 20 日までで死亡する。Cdc7 の活性化サブユニット ASK の同様なノックアウトにおいても類似の表現型が観察された。

#### 原著論文(2006 年以降)

1. Ogino, K. and \*Masai, H. (2006) "Rad3-Cds1 mediates coupling of initiation of meiotic recombination with DNA replication: Mei4-dependent transcription as a potential target of meiotic checkpoint." *J. Biol. Chem.* 281,1338-1344.
2. Tanaka, T. and \*Masai, H. (2006) "Stabilization of a stalled replication fork by concerted actions of two helicases." *J. Biol. Chem.* 281, 3484-3493.
3. Sasaki, K., Ose, T., Tanaka, T., Mizukoshi, T., Ishigaki, T., Maenaka, K., Masai, H. and \*Kohda, D. (2006) "Crystallization and preliminary crystallographic analysis of the N-terminal domain of PriA from *Escherichia coli*."

*Biochim. Biophys. Acta.* 1764, 157-160.

4. Kitamura, R., Sekimoto, T., Ito, S., Harada, S., Yamagata, H., **Masai, H.**, Yoneda, H. and \*Yanagi, K. (2006) "Nuclear import of Epstein-Barr Virus Nuclear Antigen 1 mediated by NPI-1 (Importin  $\alpha$ 5) is up- and down-regulated by phosphorylation of the nuclear localization signal for which Lys379 and Arg380 are essential."  
*J. Virol.* 80, 1979-1991.

5. Ogino, K., Hirota, K., Matsumoto, S., Takeda, T., Ohta, K., Arai, K. and \***Masai, H.** (2006) "Hsk1 kinase is required for induction of meiotic double-stranded DNA breaks without involving checkpoint kinases in fission yeast."  
*Proc. Natl. Acad. Sci. USA* 103, 8131-8136.

6. Hayashida, T., Oda, M., Ohsawa, K., Yamaguchi, A., Giacca, M., Locksley, R.M., **Masai H.\*** and Miyatake, S.\* (2006) "Replication initiation from a novel origin identified in the Th2 cytokine cluster locus requires a distant conserved non-coding sequence." (\*cocommunicating authors)  
*J. Immunol.* 176, 5446-5454.

7. \***Masai, H.**, Taniyama, C., Ogino, K., Matsui, E., Kakusho, N., Matsumoto, M., Kim, J-M., Ishii, A., Tanaka, T., Kobayashi, T., Tamai, K., Ohtani, K., and Arai, K. (2006) "Phosphorylation of MCM4 by Cdc7 kinase facilitates its interaction with Cdc45 on the chromatin."  
*J. Biol. Chem.* 281, 39249-32961. **(This paper was selected as "JBC paper of the week" and was featured in the cover of December 22 issue of JBC.)**

8. Yoshizawa-Sugata, N. and \***Masai, H.** (2007) "Human Tim/Timeless-interacting protein, Tipin, is required for efficient progression of S phase and DNA replication checkpoint."  
*J. Biol. Chem.* **282**, 2729-2740

9. Tanaka, T., Mizukoshi, T., Sasaki, K., Kohda, D. and \***Masai, H.** (2007) "*Escherichia coli* PriA protein: Two modes of DNA binding and activation of ATP hydrolysis."  
*J. Biol. Chem.* **282**, 19917-19927.

10. Sasaki, K., Ose, T., Okamoto, N., Maenaka, K., Tanaka, T., \***Masai, H.**, Saito, M., Shirai, T. and D. Kohda (2007) "Structural basis of the 3'-end recognition of a leading strand in stalled DNA replication forks by PriA."  
*EMBO J.* **26**, 19917-19927

11. Kim, J-M., Kakusho, N., Yamada, M., Kanoh, Y., Takemoto, N., and \***Masai, H.** (2008) "Cdc7 kinase is required for Claspin phosphorylation in DNA replication checkpoint."  
*Oncogene* **27**, 3475-3482.

12. Sasanuma, H., Hirota, K., Fukuda, T., Kakusho, N., Kugou, N., Kawasaki, Y.,

- Shibata, T., **Masai, H.**, and \*Ohta, K. (2008) "Cdc7-dependent phosphorylation of Mer2 facilitates initiation of yeast meiotic recombination."  
*Genes & Dev.* **22**, 398-410.
13. Sakaue-Sawano, A., Kurokawa, H., Morimura, T., Hanyu, A., Hama, H., Kashiwagi, S., Fukami, K., Imamura, T., Ogawa, M., **Masai, H.** and \*Miyawaki, A. (2008) "Spatio-temporal dynamics of multicellular cell cycle progression."  
*Cell* **132**, 487-498. (Featured on the cover of the issue; "Exceptional" evaluation in F1000)
14. Kakusho, N., Taniyama, C. and \***Masai, H.** (2008) "Identification of stimulators and inhibitors of CDC7 kinase *in vitro*."  
*J. Biol. Chem.* **283**, 19211-19218.
15. You Z. and Masai H. (2008) "Cdt1 forms a complex with MCM and activates its helicase activity."  
*J. Biol. Chem.* **283**, 24469-24477.
16. Shimmoto, S., Matsumoto, S., Hayano, M., Yokoyama, M., Noguchi, E., Russell, P. and \***Masai, H.** (2009) "Interactions between Swi1-Swi3, Mrc1 and S phase kinase, Hsk1 may regulate cellular responses to stalled replication forks in fission yeast."  
*Genes to Cells* **14**, 669-682.
17. Tanaka, H., Kubota, Y., Tsujimura, T., Kumano, M., Masai, H. and \*Takisawa, H. (2009) "Replisome progression complex links DNA replication to sister chromatid cohesion in *Xenopus* egg extracts."  
*Genes to Cells* **14**, 949-963.
18. Yoshizawa-Sugata, N. and \***Masai, H.** (2009) "Roles of human AND-1 in chromosome transactions in S phase."  
*J. Biol. Chem.* **284**, 20718-20728.
19. Tanaka, T., Yokoyama, M., Matsumoto, S., Fukatsu, R., You, Z. and \***Masai, H.** (2010) "Fission yeast Swi1-Swi3 complex facilitates DNA binding of Mrc1."  
*J. Biol. Chem.* **285**, 39609-39622.
20. Kundu, L.R., Kumata, Y., Kakusho, N., Watanabe, S., Furukohri, A., Waga, S., Sekia, M., **Masai, H.**, Enomoto, T., and \*Tada, S. (2010) "Deregulated Cdc6 inhibits DNA replication and suppresses Cdc7-mediated phosphorylation of Mcm2-7 complex."  
*Nucleic Acid Res.* **38**, 5409-5418.
21. Takeishi, Y., Ohashi, E., Ogawa, K., **Masai, H.**, Obuse, C. and Tsurimoto, T. (2010) "Casein kinase 2-dependent phosphorylation of human Rad9 mediates the interaction between human Rad9-Hus1-Rad1 complex and TopBP1."  
*Genes Cells* **15**, 761-771.
22. Furuya, K., Miyabe, I., Tsutsui, Y., Paderi, F., Kakusho, N., **Masai, H.**, Niki, H. and

\*Carr, A. M. (2010) "DDK phosphorylates checkpoint clamp Rad9 and promotes its release from damaged Chromatin."

*Mol. Cell* 40, 606-618.

23. Day, T.A., Palle, K., Barkley, L.R., Kakusho, N., Zou, Y., Tateishi, S., Verreault, A., **Masai, H.** and \*Vaziri, C. (2010) "Cdc7-Mediated Rad18 Phosphorylation Directs the Accumulation of DNA Polymerase  $\eta$  at Sites of Stalled Replication."

*J. Cell Biol.* 191, 953-966.

24. Matsumoto, S., Shimmoto, M., Kakusho, N., Yokoyama, M., Russell, P. and \***Masai, H.** (2010) "Hsk1 kinase and Cdc45 regulate replication stress-induced checkpoint responses in fission yeast."

*Cell Cycle* 9, 4627-4637.

25. Kitamura, R., Fukatsu, R., Kakusho, N., Cho, Y-S., Taniyama, C., Yamazaki, S., Toh, G-T., Yanagi, K., Arai, N., Chang, H-J. and \***Masai, H.** (2011) "Molecular mechanism of activation of human Cdc7 kinase: Bipartite interaction with Dbf4/ASK stimulates ATP binding and substrate recognition."

*J. Biol. Chem.* 286, 23031-23043.

26. Hayano, M., Kanoh, Y., Matsumoto, S., Kakusho, N. and \***Masai, H.** (2011) "Pre-firing binding of Mrc1 defines the early-firing origins which are selectively hyper-activated upon loss of fork stabilizing factors in fission yeast."

*Mol. Cell. Biol.* 31, 2380-2389. (**"Recommended" evaluation in F1000**)

27. Uno, S and \***Masai, H.** (2011) "Efficient expression and purification of human replication fork-stabilizing factor, Claspin, from mammalian cells: DNA binding activity and novel protein interactions."

*Genes to Cells*, 16, 842-856.

28. Matsumoto, S., Hayano, M., Kanoh, Y. and \***Masai, H.** (2011) "Multiple pathways can bypass the essential role of fission yeast Hsk1 kinase in DNA replication initiation."

*J. Cell Biol.* 195, 387-401. (**"Must Read" evaluation in F1000**)

29. Hayano, M., Kanoh, Y., Matsumoto, S., Shrahige, K. and \***Masai, H.** (2012) "Rif1 is a global regulator of timing of replication origin firing in fission yeast."

*Genes and Development* 26,137-150. (**"Exceptional" evaluation in F1000; Highlighted in A-IMBN Research**)

30. Yamazaki, S., Ishii, A., Kanoh, Y., Oda, M., Nishito, Y. and \***Masai, H.** (2012) "Rif1 protein is a key regulator of the genome-wide DNA replication timing in human cells."

*EMBO J.* 31, 3167-3177. (**Highlighted in Commentary; highlighted in A-IMBN Research**)

31. Barkley, L.R., Palle, K., Durando, M., Day, T.A., Gurkar, A., Kakusho, N., Li, J., **Masai, H.**, \*Vaziri, C. (2012) "c-Jun N-terminal Kinase (JNK)-Mediated Rad18 Phosphorylation Facilitates Pol $\eta$  Recruitment to Stalled Replication Forks." *Mol. Biol. Cell.* 23, 1943-1954.
32. Moriyama, K., Yoshizawa-Sugata, N., Obuse, C., Tsurimoto, T. and \***Masai, H.** (2012) "EBNA1-dependent recruitment of Orc on OriP of Epstein-Barr virus with purified proteins: Stimulation by Cdc6 through Its direct interaction with EBNA1." *J. Biol. Chem.* 287, 23977-23994.
33. Ito, S., Ishii, A., Kakusho, N., Taniyama, C., Yamazaki, S., Sakaue-Sawano, A., Miyawaki, A., and \***Masai, H.** (2012) "Mechanism of cancer cell death induced by depletion of an essential replication regulator." *PLoS One*, 7, e36372. **(Highlighted in A-IMBN Research)**
34. Uno, S., You, Z., and \***Masai, H.** (2012) "Purification of replication factors using insect and mammalian cell expression systems." *Methods*, 57, 214-221.
35. Oda, M., Kanoh, Y., Watanabe, Y., and \***Masai, H.** (2012) "Regulation of DNA replication timing on human chromosome by a cell-type specific DNA binding protein SATB1." *PLoS One* 7, e42375.
36. Miyoshi, T., Kugou, K., Yamada, S., Ito, M., Furuichi, M., Oda, A., Hirota, K. and **Masai, H.** and \*Ohta, K. (2012) "A central coupler for recombination initiation linking chromosome architecture to S-phase checkpoint." *Mol. Cell* 47, 722-733.
37. Suzuki, T., Tsuzuku, J., Hayashi, A., Shiomi, Y., Iwanari, H., Mochizuki, Y., Hamakubo, T., Kodama, T., Nishitani, H., **Masai, H.** and \*Yamamoto, T. (2012) "Inhibition of DNA damage-induced apoptosis through Cdc7-mediated stabilization of Tob." *J. Biol. Chem.* 287, 40256-40265. **(Highlighted in A-IMBN Research)**
38. Yamada, M., Watanabe, K., Mistrik, M., Mailand, N., Lee, M-H., **Masai, H.**, Lukas, J. and \*Bartek, B. (2013) "ATR-Chk1-APC/C<sup>Cdh1</sup>-dependent stabilization of Cdc7-ASK(Dbf4) kinase complex is required for DNA damage bypass under replication stress." *Genes and Development* 27, 2459-2472.
39. You, Z., De Falco, S., Pisani, F.M. and \***Masai, H.** (2012) "MCM helicase interacts with primase and stimulates its priming activity." *PLoS One* 8, e72408



40. Aria, V., De Felice, M., Di Perna, R., Uno, S., **Masai, H.**, Syvaioja, J.E., van Loon, B., Hubscher, U., \*Pisani, F.M. (2013) "The Human Tim/Tipin Complex Directly Interacts with DNA Polymerase {epsilon} and Stimulates its Synthetic Activity." *J. Biol. Chem.* 288, 12742-12752.
41. Jeffery, D.C., Wyse, B.A., Rehman, M.A., Brown, G.W., You, Z., Oshidari, R., **Masai, H.**, \*Yankulov, K.Y. (2013) "Analysis of epigenetic stability and conversions in *Saccharomyces cerevisiae* reveals a novel role of CAF-I in position-effect variegation." *Nucleic Acids Res.* 41, 8475-8488.
42. Tanikawa, M., Wada-Hiraike, O., Yoshizawa-Sugata, N., Shirane A, Hirano M, Hiraike H, Miyamoto, Y., Sone, K., Ikeda, Y., Kashiya, T., Oda K, Kawana K, Katakura Y, Yano T, **Masai, H.**, Roy AL, Osuga, Y., \*Fujii, T. (2013) "Role of multifunctional transcription factor TFII-I and putative tumour suppressor DBC1 in cell cycle and DNA double strand damage repair." *Br. J. Cancer.* 109, 3042-3048.
43. Bellelli, R., Castellone, M.D., Guida, T., Limongello, R., Dathan, N.A., Merolla, F., Cirafici, A.M., Affuso, A., **Masai, H.**, Costanzo, V., Grieco, D., Fusco, A., Santoro, M., and \*Carlomagno, F. (2014) "NCOA4 Transcriptional Coactivator Inhibits Activation of DNA Replication Origins." *Mol. Cell* 55, 123-137.
44. D., Kakusho, N., You, Z., Gharib, M., Wyse, B., Drury, E., Weinreich, M., Thibault, P. Verreault, A., **Masai, H.** and \*Yankulov, K. (2015) "CDC28 phosphorylates Cac1p and regulates the association of Chromatin Assembly Factor I with chromatin." *Cell Cycle* 14, 74-85.
45. Kotaro Koiwai, Takashi Kubota, Nobuhisa Watanabe, Katsutoshi Hori, Osamu Koiwai and \*Hisao Masai (2015) "Definition of the transcription factor TdIF1 consensus binding sequence through genome-wide mapping of its binding sites." *Genes to Cells* 20, 242-254.
46. Zech, J., Godfrey, E.L., **Masai, H.**, Hartsuiker, E. and \*Dalgaard, J.Z. (2015) "The DNA-Binding Domain of *S. pombe* Mrc1 (Claspin) Acts to Enhance Stalling at Replication Barriers." *PLoS One* 10, e0132595.
47. Iguchi, T., Aoki, K., Ikawa, T., Taoka, M., Taya, C., Yoshitani, H., Toma-Hirano, M., Koiwai, O., Isobe, T., Kawamoto, H., **Masai, H.** and \*Miyatake, S. (2015) "BTB-ZF Protein Znf131 Regulates Cell Growth of Developing and Mature T Cells." *J. Immunol.* 195:982-993.
48. Yutaka Kanoh, Seiji Matsumoto, Rino Fukatsu, Naoko Kakusho, Nobuaki Kono, Claire Renard-Guillet, Koji Masuda, Keisuke Iida, Kazuo Nagasawa, Katsuhiko Shirahige, and \*Hisao Masai (2015) "Rif1 binds to G-quadruplexes and suppresses replication over long distances."

*Nature Struct. Mol. Biol.* 22, 889-897.

49. Zhiying You, Koji L. Ode, Haruhiko Takisawa, and \***Hisao Masai** (2016) "Characterization of conserved arginine residues on Cdt1 that affect licensing activity and interaction with Geminin or Mcm complex." *Cell Cycle* 5, 1213-1226.

50. Tanaka T, Nishito Y, \***Masai H.** (2016) "Fork restart protein, PriA, binds around oriC after depletion of nucleotide precursors: Replication fork arrest near the replication origin." *Biochem. Biophys. Res. Commun.* 470, 546-551.

51. Tanaka H, Muto A, Shima H, Katoh Y, Sax N, Tajima S, Brydun A, Ikura T, Yoshizawa N, **Masai H**, Hoshikawa Y, Noda T, Nio M, Ochiai K, \*Igarashi K. (2016) "Epigenetic Regulation of the Blimp-1 Gene (Prdm1) in B Cells Involves Bach2 and Histone Deacetylase 3." *J. Biol. Chem.* 291, 6316-6330.

52. \*Nonaka T, Suzuki G, Tanaka Y, Kametani F, Hirai S, Okado H, Miyashita T, Saitoe M, Akiyama H, **Masai H**, Hasegawa M. (2016) "Phosphorylation of TAR DNA-binding Protein of 43 kDa (TDP-43) by Truncated Casein Kinase 1 $\delta$  Triggers Mislocalization and Accumulation of TDP-43." *J. Biol. Chem.* 291, 5473-5483.

53. Chi-Chun Yang, Masahiro Suzuki, Shiori Yamakawa, Syuzi Uno, Ai Ishii, Satoshi Yamazaki, Rino Fukatsu, Ryo Fujisawa, Kenji Sakimura<sup>3</sup>, Toshiki Tsurimoto, \***Hisao Masai** (2016) "Claspin recruits Cdc7 kinase for initiation of DNA replication in human cells." *Nature Communications* 7:12135 doi: 10.1038/ncomms12135.

54. Matsumoto, S., Kanoh, Y., Shimmoto, M., Hayano, M., Ueda, K., Fukatsu, R., Kakusho, N., \***Masai, H.** (2017) "Checkpoint-independent Regulation of Origin Firing by Mrc1 through Interaction with Hsk1 kinase." *Mol. Cell. Biol.* Mar 17;37(7). pii: e00355-16. doi: 10.1128/MCB.00355-16. Print 2017 Apr 1. PMID: 28069740

55. Toteva, T., Mason, B., Kanoh, Y. Brøgger, P. Green, D., Verhein-Hansen, J. **Masai, H.** and \*Thon, G. (2017) "Establishment of expression-state boundaries by Rif1 and Taz1 in fission yeast." *Proc. Natl. Acad. Sci. USA.* 114, 1093-1098.

56. You, Z. and **Masai, H.** (2017) Potent DNA strand annealing activity associated with mouse Mcm2~7 heterohexamer complex." *Nucleic Acids Res.* 45, 6495-6506.

57. Irie T, Asami T, Sawa A, Uno Y, Hanada M, Taniyama C, Funakoshi Y, **Masai, H.**, and \*Sawa M. (2017) "Discovery of novel furanone derivatives as potent Cdc7 kinase inhibitors."

*Eur. J. Med. Chem.* 130, 406-418.

58. Moriyama, K., Yoshizawa-Sugata, N., and \***Masai, H.** (2018) "Oligomer formation and G-quadruplex binding by purified murine Rif1 protein, a key organizer of higher-order chromatin architecture."

*J. Biol. Chem.* 293, 3607-3624.

59. Iguchi, T., Miyauchi, E., Watanabe, S., **Masai, H.** and \*Miyatake. S. (2018) "A BTB-ZF protein, ZNF131, is required for early B cell development."

*Biochemical and Biophysical Research Communications*, 501, 70-575.

60. \***Masai, H.**, Kakusho, N., Fukatsu, R., Ma, Y., Iida, K., Kanoh, Y. and Nagasawa, K. (2018) "Molecular architecture of G-quadruplex structures generated on duplex Rif1 binding sequences."

*J Biol Chem.* 293, 17033-17049.

61. Kobayashi, S., Fukatsu, R., Kanoh, Y., Kakusho, N., Matsumoto, S., Chaen, S. and \***Masai, H.** (2019) "Both a unique motif at the C terminus and N-terminal HEAT repeat contribute to G4 binding and origin regulation by Rif1 protein."

*Mol Cell. Biol.* 39(4). pii: e00364-18. Appeared in the cover figure of the issue

62. \***Masai, H.**, Fukatsu, R., Kakusho, N., Kanoh, Y., Moriyama, K., Ma, Y., Iida, K., Nagasawa, K. (2019) "Rif1 promotes self-association of G-quadruplex (G4) by its specific G4 binding and oligomerization activities."

*Sci. Rep.*, 9(1), 8618.

63. Ito, S., Goto, H., Kuniyasu, K., Shindo, M., Yamada, M., Tanaka, K., Toh, G-K., Sawa, S., Inagaki, M., Bartek, B., and \***Masai, H.** (2019) "Cdc7 kinase stimulates Aurora B kinase in M-phase."

*Sci. Rep.*, 9(1):18622.

64. Yang, C-C., Kato, H., Shindo, M. and \***Masai, H.** (2019) "Cdc7 activates replication checkpoint by phosphorylating the Chk1 binding domain of Claspin in human cells."

*E-life*, 8. pii: e50796.

65. \*Kato H, Asamitsu K, Sun W, Kitajima S, Yoshizawa-Sugata N, Okamoto T, **Masai H**, Poellinger L. (2020) "Cancer-derived UTX TPR mutations G137V and D336G impair interaction with MLL3/4 complexes and affect UTX subcellular localization."

*Oncogene*. doi: 10.1038/s41388-020-1218-3. [Epub ahead of print] PubMed PMID: 32071397.

66. \***Masai, H**, Kanoh, Y, Kakusho, N, Fukatsu, R (2020) "Detection of cellular G-quadruplex by using a loop structure as a structural determinant."

*Biochemical and Biophysical Research Communications*, 531:75-83.

67. Fracassi A, Cao J, Yoshizawa-Sugata N, Toth E, Archer C, Groninger O, Ricciotti E, S-Y Tang, Handschin S, Bourgeois J-P, Ray A, Liosi K, Oriana S, Stark W, **Masai H**, Zhou R, Yamakoshi Y (2020) "LDL-mimetic lipid nanoparticles prepared by surface KAT ligation for in vivo MRI of atherosclerosis"  
*Chem Sci* in press

68. Kitajima S, Sun W, Lee KL, Ho JC, Oyadomari S, Okamoto T, **Masai H**, Poellinger L, Kato H. (2021) A KDM6 inhibitor potently induces ATF4 and its target gene expression through HRI activation and by UTX inhibition. *Sci Rep.* 11(1):4538.

69. \*Yoshizawa-Sugata N, Yamazaki S, Mita-Yoshida K, Ono T, Nishito Y, \***Masai H**. (2021) "Loss of full-length DNA replication regulator Rif1 in two-cell embryos is associated with zygotic transcriptional activation." *J Biol Chem.* 297:101367.

#### 英文総説(2008年以降)

1. Fujii-Yamamoto, H., Yamada. M., and \***Masai, H.** (2008) "Regulation of DNA replication factors by E2F in cancer and embryonic stem cells." in "Control of Cellular Physiology by E2F Transcription Factors." *Research Signpost* 209-221.

2. Ito, S., Taniyama, C., Arai, N. and \***Masai, H.** (2008) "Cdc7 as a potential new target for cancer therapy." *Drug News and Perspectives* 21, 481-488. **Featured on the cover of the issue**

3. Sawa, M., and \***Masai, H.** (2009) "Drug Design with Cdc7 kinase, a potential novel cancer therapy target." *Drug Design, Development and Therapy* 2, 255-264.

4. Toh, G.K., and **Masai, \*H.** (2009) "ASK" *UCSD-Nature Molecule Pages*, Published online: 25 Feb 2009 | doi:10.1038/mp.a000345.01 (Review)

5. \***Masai, H.**, Matsumoto, S., You, Z., Yoshizawa-Sugata, N. and Oda, M. (2010) "Eukaryotic DNA replication; where, when and how?" *Annual Rev. Biochem.* 79, 89-130.

6. \***Masai, H.**, Tanaka, T. and Kohda, D. (2010) "Stalled replication forks: Making ends meet for recognition and stabilization." *Bioessays* 32, 687-697. (Review)

7. Tanaka, T. and \***Masai, H.** (2010) "Bacterial primosome." In: Encyclopedia of Life Sciences (ELS). John Wiley & Sons, Ltd: Chichester. DOI: 10.1002/9780470015902.a0001048.pub2

8. Tanaka, T. and \***Masai, H.** (2010) "Bacterial replication fork: synthesis of lagging strand." In: Encyclopedia of Life Sciences (ELS). John Wiley & Sons, Ltd: Chichester. DOI: 10.1002/9780470015902.a0001049.pub2

9. \***Masai, H.** (2010) "FANCs regulate firing of DNA replication origins." *Cell Cycle* 9, 2494.
10. Vaziri, C. and \***Masai H.** (2010) "Integrating DNA replication with Trans-Lesion Synthesis via Cdc7." *Cell Cycle* 9, 4818-4823.
11. \***Masai, H.** (2011) "RecQL4: a helicase linking formation and maintenance of a replication fork." *J. Biochem.* 149, 629-631 (commentary)
12. Toh G-T. and \***Masai, H.** (2012) "Cdc7L1" *UCSD-Nature Molecule Pages*, Published online: 31 August 2012 | doi:10.6072/H0.MP.A003137.01 (Review)
13. \***Masai, H.** (2012) "Cdc7" *The Encyclopedia of Signaling Molecules* Springer Reference and Database Publishing
14. \***Masai, H.** (2012) "Dbf4" *The Encyclopedia of Signaling Molecules* Springer Reference and Database Publishing
15. Yamazaki, Hayano, M. and \***Masai, H.** (2013) "Replication timing regulation of eukaryotic replicons: Rif1 as a global regulator of replication timing." *Trends in Genetics.* 29, 449-460.
16. \***Masai, H.** (2013) "A personal reflection on the Replicon Theory: from R1 plasmid to replication timing regulation in human cells." *J. Mol. Biol.* 425, 4663-4672. (Review)
17. Matsumoto, S. and \***Masai, H.** (2013) "Regulation of chromosome dynamics by Hsk1 kinase." *Biochemical Society Transactions* 41, 1712-1719. (Review)
18. Yoshizawa-Sugata, N. and \***Masai, H.** (2014) "Cell cycle synchronization and flow cytometry analysis of mammalian cell." *Methods in Molecular Biology*, 1170, 279-293. (Review)
19. \***Hisao Masai** (2014) "ATM in prevention of genomic instability." *Cell Cycle*, 13, 882-883. (News and Views)
20. Renard-Guillet, C., Kanoh, Y., Shirahige, K., and \***Masai, H.** (2014) "Recent advances in temporal and spatial regulation of eukaryotic DNA replication: From regulated initiation to genome-scale timing program." *Seminars in Cell & Developmental Biology*, 30, 110-120. (Review)
21. Yamada, M., **Masai, H.**, and \***Bartek, J.** (2014) "Regulation and roles of Cdc7 kinase under replication stress." *Cell Cycle* 13, 1859-1866.
22. \***Masai, H.** (2015) "Building up the machinery for DNA replication." *Cell Cycle* 14, 3011-3012. (News and Views)

23. Naoko Yoshizawa, Satoshi Yamazaki, and \***Hisao Masai** (2015) "Rif1, a conserved chromatin factor regulating DNA replication, DNA repair and transcription." In *The Initiation of DNA Replication in Eukaryotes*, Springer
24. Hayano, M., Matsumoto, S. and \***Masai, H.** (2015) "DNA Replication Timing: Temporal and Spatial Regulation of Eukaryotic DNA Replication." in Fumio Hanaoka and Kaoru Sugasawa (Eds): *DNA Replication, Recombination, and Repair* (Springer)
25. \***Masai, H.**, Kanoh, Y., Moriyama, K., Yamazaki, S., Yoshizawa, N., and Matsumoto, S. (2017) "Telomere binding factors in regulation of DNA replication." *Genes & Genetic Systems*, Jun 30. doi: 10.1266/ggs.17-00008. [Epub ahead of print] (Review)
26. \***Masai, H.** (2017) "A novel p53-Cdc7 link induced by genotoix stress." *Cell Cycle*, 6, 735-736 (News and Views)
27. \***Masai, H.**, Yang, C-C., and Matsumoto, S. (2017) "Mrc1/Claspin: a new role for regulation of origin firing." *Current Genetics* 45, 6494-6506. (Review)
28. \***Masai, H.** Encyclopedia of Signaling Molecules, 2nd Edition : Dbf4 (2017)
29. \***Masai, H.** Encyclopedia of Signaling Molecules, 2nd Edition : Cdc7 (2017)
30. Moriyama, K., Lai, M.S. and \***Masai, H.** (2017) "Interaction of Rif1 Protein with G-Quadruplex in Control of Chromosome Transactions." *Adv Exp Med Biol* Vol. 1042, pp287-310. "DNA Replication, From Old Principles to New Discoveries" (Masai, H. and Foiani, M. Eds) Springer
31. Ghadiri, H. Alavi, S., Dabirmanesh, B., Moriyama, K., \*Khajeh, K. and **Masai, H.** (2018) "Study Break: Cell Timer/Cell Clock." *Iran Biomed J.* 22(6):360-1. Epub 2018 Sep 16. PMID: 30218996; PMCID: PMC6305818.
32. Tanaka, T. and \***Masai, H.** (2019) "Bacterial primosome." In: Encyclopedia of Life Sciences (ELS). John Wiley & Sons, Ltd: Chichester. In press
33. Masai, H. (2019) "For 60th birthday of BBRC: DNA replication factors outside S phase." *Biochemical and Biophysical Research Communications*, 520:685-686.
34. \***Masai, H.** and Tan, Z. (2020) "G-quadruplexes: tools, roles, and goals." *Biochemical and Biophysical Research Communications*, 531:1-2.
35. \***Masai, H.** and Tanaka, T. (2020) "G-quadruplex DNA and RNA: Their roles in regulation of DNA replication and other biological functions." *Biochemical and Biophysical Research Communications*, 531:25-38.
36. Alavi, S., Ghadiri, H., Dabirmanesh, B., Moriyama, K., Khajeh, K., and \***Masai, H.**

(2020) G-quadruplex binding protein Rif1, a key regulator of replication timing. *J. Biochem.* doi: 10.1093/jb/mvaa128. Epub ahead of print. PMID: 33169133.

37. \*Oki M, and \*Masai H. (2021) Regulation of HP1 protein by phosphorylation during transcriptional repression and cell cycle. *J. Biochem.* mvab040.