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Supplemental Information

TRF2 Recruits RTEL1 to Telomeres in S Phase to Promote T-Loop Unwinding
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Figure S1

A

Figure S1. RTEL1 binds TRF2 in cell cycle dependent manner, Related to Figure 1. (A) Whole-cell extracts of control (-benz.) and benzonase-treated (+benz.) 293 FLAP-tagged RTEL1 cells were immunoprecipitated using anti-TRF1, -POT1, or -TPP1 antibodies. The immunoblots were probed with antibodies as indicated. Asterisks indicate the nonspecific cross-reactivity detected with anti-TPP1.
antibody. Input (5%) is shown on the right. (B) 293 NFLAP-tagged RTEL1 cells and (C) RTEL1v5 MEFs stably expressing Myc-TRF2 were either cultured asynchronously or were released from double-thymidine block (left panels) or thymidine plus nocodazole block (right panels). Cells were subjected to SDS-PAGE and progression through the cell cycle was monitored by immunoblotting with cell cycle markers as indicated. (D) Whole-cell lysates from RTEL1v5 MEFs at different cell cycle phases (asterisks in C) were immunoprecipitated using anti-V5 antibody and rabbit normal IgG served as a control for immunoprecipitation. Protein complexes were resolved on SDS-PAGE followed by immunoblotting with anti-Myc and -V5 antibodies. Input (5%) is shown on the right panel. (E) Synchronized RTEL1v5 MEFs derived from the same single experiment (asterisks in C) were subjected for in situ PLA assay to measure association of RTEL1 with TRF2. Dashed lines indicate nucleus outlines (as determined by DAPI in blue). Data is representative of two independent experiments.
Figure S2

**Figure S2.** RTEL1-TRF2 interaction does not require active DNA replication, Related to Figure 1. 293 HEK cells stably expressing Myc-TRF2 were incubated for 24 hours in the absence (untr.) or presence of low or high doses of DNA synthesis inhibitors (aphidicolin or hydroxyurea) as indicated on the top. Whole-cell lysates were subjected to immunoprecipitation with RTEL1 antibody or rabbit normal IgG as a control. Immunocomplexes were resolved on SDS-PAGE and followed by Western blotting analysis with antibodies against Myc and RTEL1. Input (5%) is shown on the right. The asterisks indicate unspecific bands detected by rabbit polyclonal anti-RTEL1 antibody.
Figure S3. Endogenous TRF2 interacts with C4C4 peptide, Related to Figure 2. (A) Whole-cell extracts from U2OS cells stably expressing an empty vector (ctrl), wild-type Myc-C4C4 peptide (WT), or mutant peptides Myc-C4C4^{R1264H} (R/H) and Myc-C4C4^{C1279A/C1282A} (C/A), were analyzed by immunoblotting with antibodies against Myc and TRF2. Tubulin served as a loading control. (B) Whole-cell extracts from A were reciprocally immunoprecipitated with anti-Myc or anti-TRF2 antibodies. Immunocomplexes were analysed by Western blotting with primary antibodies as indicated. Input (5%) is shown on the right.
Figure S4. Telomere dysfunction-induced foci (TIFs) in \textit{RTEL1}^{F/F} MEFs expressing mutant RTEL1, Related to Figure 3. Representative micrographs of the data shown in Figure 3F. \textit{RTEL1}^{F/F} MEFs transduced with empty vector (ctrl), wild-type V5-RTEL1 (WT), mutant V5-RTEL1^{R1237H} (R/H), or double mutant V5-RTEL1^{C1252A/C1255A} (C/A) were infected with control (Ad-GFP) or Cre-expressing (Ad-Cre-GFP) adenovirus. At 96 hours after infection cells were processed for 53BP1 and FITC-Tel repeat fluorescence. Dashed lines indicate nucleus outlines (as determined by DAPI; not shown). Insets represent 3X magnifications of the indicated fields. Scale bar is 10 µm.
Figure S5. Binding of the C4C4 motif of RTE1 to TRF2TRFH dimerization domain is metal-ion dependent, Related to Figure 5. (A) Western blotting of a scanning peptide array for the N-terminal 43 to 183 amino acids of human TRF2. The arrays were incubated either with GST or with human full-length RTE1 fused with GST and bound proteins were analyzed by immunoblotting with anti-GST antibody. The position of the N and C termini is shown, and the sequence of the region covering the RTE1-binding site is indicated. (B) TRF2TRFH recombinant proteins were incubated for two hours in the absence (-EDTA) or presence (+EDTA) of 50 mM of EDTA either with GST or GST-C4C4 fusion protein. Protein complexes were eluted and analyzed by SDS-PAGE followed by Coomassie brilliant blue staining. The input control in lanes 1 to 3 reflects 10% of the total amount of proteins used for the pull-down experiments.
Figure S6. Immunoblot analysis of cells expressing wild-type and mutant TRF2, Related to Figure 5. Whole-cell extracts from 293 FLAP-tagged RTEL1 cells (left panels) or RTEL1v5 MEFs (right panels) stably expressing an empty vector (ctrl), wild-type Myc-TRF2 (WT) or mutant Myc-TRF2\textsuperscript{I124D} (I/D) were resolved on SDS-PAGE and analyzed with antibodies as indicated.
Figure S7. Telomere dysfunction-induced foci (TIFs) \( TRF2^{F/-} \) MEFs expressing mutant TRF2, Related to Figure 6. Representative micrographs of the data shown in Figure 6H. \( TRF2^{F/-} \) MEFs transduced with empty vector (ctrl), wild type Myc-TRF2 (WT) or mutant Myc-TRF2\(_{1124D}^{I/-}\) (I/D) were infected with control (Ad-GFP) or Cre-expressing (Ad-Cre-GFP) adenovirus. At 96 hours after infection, cells were processed for 53BP1 and FITC-Tel repeat fluorescence. Dashed lines indicate nucleus outlines (as determined by DAPI; not shown). Insets represent 3X magnifications of the indicated fields. Scale bar is 10 µm.
**Supplemental Experimental Procedures**

**Cell lysis, Western blotting, and Immunoprecipitation**

Cells were rinsed twice with PBS, transferred to an ice-cold NET lysis buffer (50mM Tris (pH 7.2) 150 mM NaCl, 0.5% NP-40, 1x EDTA-free Complete protease inhibitor cocktail (Roche), 1x PhosSTOP phosphatase inhibitor cocktail (Roche)) and kept to lyse 10 minutes on ice. The cell lysates were then briefly vortexed and passed through a 23G syringe five times. The soluble protein fractions were collected after centrifugation at 16000 x g for 10 minutes at 4°C. Western blotting analysis was performed as described previously (Vannier et al., 2012). For protein immunoprecipitation, whole-cell extracts were precleared with protein G sepharose (Sigma-Aldrich), and one to two milligrams of precleared extract was incubated with antibodies as indicated in the text. Where specified, genomic DNA was removed by treatment with benzonase (Novagen; EMD Chemicals). Immunocomplexes were subjected to SDS-PAGE followed by immunoblotting using nitrocellulose membrane (GE Healthcare).

**Peptide array studies**

Peptide arrays were generated by automatic “pep-spot” synthesis and synthesized on Whatman 50 cellulose membranes using Fmoc-chemistry with the AutoSpot-Robot ASS 222 (Intavis Bioanalytical Instruments) as previously described (Horejsi et al., 2014). The binding of spot-immobilized peptides with purified recombinant GST-TRF2, GST-RTEL1, or H6-SUMO-C4C4-RING fusion proteins was carried out by incubating the membranes with 100 to 150 µg of recombinant protein as reported earlier (Thorslund et al., 2007; Ward et al., 2010).

**Antibodies**

The following primary antibodies against proteins were used: TRF2 (2645 and 13136; Cell Signaling, 56694; Novus Biologicals, 4A794; Millipore, H300; Santa Cruz, TRF21A; Alpha Diagnostic), TRF1 (ab10579; Abcam), Rap1 (A300-306A; Bethyl), TIN2 (SAB4200108; Sigma-Aldrich, NB600-1522; Novus Biologicals), CyclinD1 (2922; Cell Signaling), Cyclin E (HE12; Santa Cruz) phospho-HistoneH3-Ser10 (9701; Cell Signaling), GFP (ab290; Abcam, 11814460001; Roche), V5 (V8137; Sigma, ab27671; Abcam, 377500; Invitrogen), RTEL1 (NBP2-22360; Novus
Biologicals), POT1 (NB500-176; Novus Biologicals), TPP1 (H00065057-M02; Abnova), Myc (ab32, Abcam) GST (ab92; Abcam), 53BP1 (612523; BD Biosciences), PCNA (PC10; Santa Cruz), 6xHis (ab5000; Abcam). TRF2 (1254), TRF1 (1449), and Rap1 (1252) antibodies used in telomere ChIP were kindly provided by Titia de Lange.

Cell cycle synchronization
HEK 293 cells or RTEL1v5 fibroblasts expressing wild-type Myc-TRF2 were synchronized by the double-thymidine–block method as previously described (Whitfield et al., 2002), with minor modifications. Briefly, cells were treated with 2 mM thymidine (Sigma-Aldrich) for 18 hours, thymidine-free media for 9 hours to release the cells, and 2 mM thymidine was added to media for an additional 16 hours to arrest the cells at the G1 to S transition. Cells were washed twice with PBS and then released in fresh complete DMEM. Cells were analyzed at 70 minutes time intervals by immunoblotting and in situ PLA assay. For synchronization in mitosis, a thymidine-nocodazole block was used (Whitfield et al., 2002). Briefly, cells at confluence of 60% were treated with 2mM thymidine for 24 hours, washed twice in PBS, and released into complete DMEM for 3 hours. Next, cells were treated with 50 µg/mL of nocodazole (Sigma-Aldrich) for 15 hours, and the cells were washed twice with PBS and a fresh complete medium was added to the cell culture. Synchronized cells were analyzed at 90 minutes time intervals by Western blotting and in situ PLA with antibodies indicated in text.

Cell transfection and RNA interference
HEK 293 cells were transiently transfected using Lipofectamine 2000 transfection reagent (Life Technologies) according to the manufacturer's instructions. The lentiviral expression constructs sh-Rap1 in pLKO.1 vector backbone were purchased from Open Biosystems (Thermo Scientific). Sh-RNA sequences for Rap1 were (5'-AAACCCAGGGCTGCCTTGGAAAAG-3’) in sh1-Rap1 (5'-AAACCCAGGGCTGCCTTGGAAAAG-3’) in sh2-Rap1 and (5'-CCTAAAGTTAAGTCCCTGCTCTAGCGAGGGCGACTTACCTTGG-3’) in non-target control.
Expression vectors

Full-length human RTEL1 (1300 aa; NP_001269938.1) was generated by PCR followed by Gateway cloning (Invitrogen), starting with entry clones in pDONR221 and shuttled from entry clones into pHAGE-PGK-C-Flag-IRES-Puro lentiviral vector (a kind gift from Jianping Jin, University of Texas, USA). Full-length mouse RTEL1 (1273 aa; Q0VGM9.2) was synthesized de novo by GeneArt (Life Technologies). The RTEL1 cDNA was subsequently subcloned into the pBABEpuro vector polylinker downstream of the 5' LTR with the LTR acting as the promoter, allowing close to physiological levels of mRTEL1 expression. The human C4C4 domain fragment (corresponding to amino acids 1234-1293 of RTEL1) was generated by PCR and subcloned into pLPC-NMyc retroviral expression vector. The retroviral expression vector, pLPC-NMyc-TRF2, was kindly provided by Titia de Lange.

Site-directed mutagenesis

Single amino acid substitutions were performed with the following primers: human RTEL1<sup>R1264H</sup> – (5'-ctgtgacctccacagtgccagctgccgctgt-3') and (5'-agcagcctggcagtcttggaagtccag-3'), mouse RTEL1<sup>R1237H</sup> – (5'-ctctgaccttcacactgtgggccctgttg-3') and (5'-caacaggccccacagttgtaggtagacccagggc-3'), mouse TRF2<sup>I124D</sup> – (5'-cgggacttcaggcagggagctagcag-3') and (5'-tgcatgatgtccggcctgcctgaagtcccg-3'). Double mutants were generated with the following primers: human RTEL1<sup>C1279A/C1282A</sup> - (5'-gggctctcagggccgccagccccacagcctgcc-3'), mRTEL1<sup>C1279A/C1282A</sup> – (5'-ccagggctctcagggccgccagccccacagcctgcc-3'). Primers were designed with the QuikChange Primer Design Software (Agilent Technologies). Single mutants were generated using the QuikChange Lightning Site-Directed Mutagenesis kit (Agilent Technologies) and double mutants were created with the QuikChange Lightning Multi Site-Directed Mutagenesis kit (Agilent Technologies) according to the manufacturer’s instructions. The generated mutants were verified by sequencing to screen against spurious secondary mutations.

PNA FISH, CO-FISH, Q-FISH, and IF-FISH

Telomeric Peptide Nucleic Acid Fluorescence In Situ Hybridization (PNA FISH) was performed as described before (Lansdorp et al., 1996). Briefly, cells were treated with 0.2 µg/ml of colcemid for 90 minutes to arrest cells in metaphase. Trypsinized
cells were incubated in 75 mM KCL, fixed with methanol:acetic acid (3:1), and spread on glass slide. To preserve chromosome architecture better the slides were rehydrated in PBS for 5 minutes, fixed in 4% formaldehyde for 5 minutes, treated with 1 mg/ml of pepsin for 10 minutes at 37 °C, and fixed in 4% formaldehyde for 5 minutes. Next, slides were dehydrated in 70%, 85%, and 100% (v/v) ethanol for 15 minutes each and air-dried. Metaphase chromosome spreads were hybridized with telomeric FITC-TelC 5′-(CCCTAA)3-3′ PNA probe (Bio-synthesis) and slides were mounted using ProLong Gold antifade with DAPI (Life Technologies). Chromosome images and telomere signals were captured using Zeiss Axio Imager M1 microscope equipped with an ORCA-ER camera (Hamamatsu) controlled by Volocity 4.3.2 software (Improvision). For CO-FISH, cells were grown in medium supplemented with 10µM of BrdU (Sigma-Aldrich) for 18 hours and chromosome slides were prepared as described (Vannier et al., 2012). After treatment with 0.5 mg/ml RNAse A (Roche) for 10 minutes at 37°C cells were stained with 0.5 µg/ml Hoechst 33258 (Life technologies) in 2 × SSC for 15 minutes at room temperature and crosslinked by exposure to 365nm UV light (Stratalinker 1800; Agilent Technologies) for 30 minutes. The slides were digested with 10U/µl Exonuclease III (Promega) for 10 minutes at room temperature and sequentially incubated with TAMRA-TelG 5′-(TTAGGG)3-3′ and FITC-TelC 5′-(CCCTAA)3-3′ PNA telomere probes (Bio-synthesis) at room temperature for 2 h. Slides were mounted with ProLong Gold antifade containing DAPI (Life Technologies). Q-FISH on metaphases was performed as described earlier (Vannier et al., 2012). Telomere fluorescence signals were integrated from 50 metaphases and quantified by using the CellProfiler analysis software (The Broad Institute of MIT and Harvard, MA, USA). For IF-FISH, cells grown on #1.5 glass coverslips were fixed for 20 minutes in 2% (wt/vol) formaldehyde (Thermo Scientific) at room temperature and IF-FISH was performed as described previously (Vannier et al., 2012), using primary 53BP1 antibody (see antibodies section), anti-mouse Alexa Fluor 555 secondary antibody (Molecular Probes) and a FITC-TelC 5′-(CCCTAA)3-3′ PNA telomere probe (Bio-Synthesis). Slides were mounted with ProLong Gold antifade containing DAPI and images were acquired with an Olympus FLV1000 inverted microscope equipped with a 63X oil objective. Following acquisition, images were imported into ImageJ (NIH) and Adobe Photoshop CS5 for manual quantitation and processing.
Telomere circle (TC) assay
Cells grown at confluence between 70 to 80% were collected from two 10 cm dishes and extraction of genomic DNA for T-circle assay was performed as described previously (Munoz-Jordan et al., 2001). Total gDNA was digested by AluI/HinfI restriction enzymes and the TCA assay was performed as reported (Zellinger and Riha, 2007), with two essential modifications: (1) Phi29 DNA (Thermo Scientific) polymerization employed a mammalian telomere primer, and (2) Southern blotting membrane was hybridized to a γ[^32]P]-labelled (TTAGGG)$_4$ telomeric probe. Southern blot images were captured with Storm 840 scanner and the extent of[^32]P incorporation was quantified from the autoradiographs by ImageQuant TL software analyzer (Amersham Biosciences). The level of[^32]P incorporation obtained from the Phi29 negative control samples represented the background level, which was subtracted from the values obtained from the samples that contained the Phi29 DNA polymerase.

Telomere ChIP
Telomere ChIP assay was performed using the EZ-ChIP kit (17-371 Millipore) with minor modifications, which are outlined below. All reagents, unless otherwise indicated, were provided with the kit. $TRF2^{F/c}$ cells transduced with the indicated retroviral vectors were infected either with a control- or Cre-expressing adenovirus. At 96 hours post-infection cells were counted and 2x10$^7$ cells were crosslinked with methanol-free 1% formaldehyde (Thermo Scientific) for 12 minutes. Next, cells were quenched with glyceine (200mM final concentration), rinsed twice in ice-cold PBS, and pelleted. Chromatin was sheared using a sonicating waterbath (Bioruptor Diagenode), with 30 seconds on and 30 seconds off, for 20 total cycles. Two microliters of anti-TRF1, -TRF2, and -Rap1 antibodies was used for immunoprecipitation of protein/DNA complexes. Purified rabbit IgG was utilized as a negative control for immunoprecipitation. Subsequent washing, elution, reversal of crosslink, and DNA purification steps were performed according to manufacturer's instructions for the EZ-ChIP kit. DNA was boiled for five minutes and slot-blotted onto Hybond (Amersham Bioscience) positively charged nylon membrane. The membrane was incubated in freshly made denaturing solution (1.5M NaCl/0.5N NaOH) for 10 minutes followed by neutralization in 1M NaCl/0.5M Tris-HCl (pH 7.0) for 10 minutes. DNA was crosslinked to the membrane immediately by UV
crosslinking, then rinsed in 2X SSC. Telomere DNA was detected following hybridization with TTAGGG probe. Images were captured with Storm 840 scanner and quantified by ImageQuant TL software (Amersham Bioscience).

**DNA combing**

DNA combing was performed essentially as described in (Vannier et al., 2013). Briefly, \( RTEL1^{FF} \) mouse embryo fibroblasts expressing an empty vector, wild-type RTEL1, or mutants RTEL1 were infected with control- or Cre-expressing adenovirus. Cells were pulse-labeled with IdU/CldU for 20 minutes, each pulse. DNA fibres were extracted in agarose plugs and stretched on silanized coverslips with the molecular combing system (Genomic Vision). CldU was detected with rat anti-BrdU antibody (BU1/75, AbCys), followed by goat anti-rat coupled to Alexa 594 (A11007, Molecular Probes) and finally by chicken anti-goat coupled to Alexa 594 (A21468, Molecular Probes). IdU was detected with Mouse anti-BrdU coupled to FITC antibody (BD44, Becton Dickinson), followed by rabbit anti-mouse coupled to Alexa 488 (A11059, Molecular Probes) and finally by donkey anti-rabbit coupled to Alexa 488 (A21206, Molecular Probes). DNA fibres were captured with Zeiss Axio Imager M1 microscope equipped with an ORCA-ER camera (Hamamatsu) controlled by Volocity 4.3.2 software (Improvision). Representative images of DNA fibres were assembled from different fields of view and were processed as described in (Vannier et al., 2013).

**Generation of recombinant proteins**

Plasmid construct for expressing recombinant human C4C4-RING domain fragment (amino acids 1234 to 1293 of human RTEL1) was generated by PCR cloning using the full-length RTEL1 cDNA as a template. The PCR product was subcloned into pGEX-4T-2 (GE Healthcare) or pET-H6-SUMO (Life Technologies) expression vectors. Recombinant human GST-TRF2, GST-TRF2ΔB, and GST-ΔB constructs were provided by Titia de Lange. Recombinant TRFH dimerization domain, in pET28-H6-SUMO expression vector, was kindly provided by Ming Lei (University of Michigan, MI, USA). All recombinant proteins were expressed in *Escherichia coli* BL21 DE3 (Life Technologies). GST-tagged proteins were purified by affinity chromatography on glutathione Sepharose beads (GE Healthcare). Briefly, following
the induction of GST-tagged proteins with 0.1 mM isopropyl-β-d-thiogalactoside (IPTG; Sigma-Aldrich) for 16 h at 18°C, cells were harvested at 5000 x g for 10 minutes at 4°C, resuspended in lysis buffer (25 mM Tris (pH 7.2), 150 mM NaCl, 0.5% Triton X-100, and protease inhibitors,) and sonicated. Bacterial cell lysate was cleared at 16000 x g for 20 minutes, and the supernatant was incubated with glutathione Sepharose beads for 2h at 4°C. Bound proteins were eluted with 50 mM Tris (pH 8.0) containing 10 mM reduced glutathione. Protein purity and concentration were determined by Coomassie staining (Instant Blue; Expedeon) and Bradford assay (Biorad), respectively. The His-tagged proteins were purified on a nickel resin followed by gel filtration with AKTA explorer FPLC equipped with Superdex 200 column (GE Healthcare). Bacterial strains expressing His-tagged proteins were induced with 0.5 mM IPTG for 4 h at 30°C, bacteria were harvested, resuspended in lysis buffer (25 mM Tris (pH 7.2), 250 mM NaCl, 20 mM imidazole, and 1mM PMSF), and sonicated at 4°C. Lystes were centrifuged at 16000 x g for 30 minutes at 4°C and the supernatant was loaded onto a HisTrap FF (GE Healthcare) column equilibrated with the lysis buffer. The column was washed extensively with lysis buffer containing 20 mM imidazole and proteins were eluted using the buffer that contains the same components as in the wash buffer, except that the concentration of imidazole was 200 mM. The eluant was dialyzed against buffer containing 25 mM Tris (pH 7.2) and 250 mM NaCl in Slide-A-lyzer dialysis G2 cassette (Thermo Scientific) at 4°C. During the dialysis the buffer (approx. 2L) was changed twice to effectively remove the imidazole and detergent. Where applicable, 6xHis-Ulp1 SUMO protease (10U/1mg of proteins) was added upon dialysis to cleave the SUMO-His tag. After Ulp1 protease cleaved the SUMO fusion, the samples were loaded onto the HisTrap FF nickel column, to remove His-SUMO and His-Ulp1, and subsequently cleaved proteins were recovered from the unbound flow through fraction. Next, proteins were concentrated using Amicon Ultra (Millipore) filter units and proteins were subjected on gel filtration. Fractions containing proteins were evaluated by SDS-PAGE followed by Coomassie staining. Concentration of the purified proteins were examined by Bradford assay or from the absorbance at 280 nm based on the molar absorption coefficients determined from the Edelhoch relationship.
Statistical analysis
Statistical analyses were performed using GraphPad PRISM version 6.0a software (GraphPad Inc.). Statistical significance of data was assessed by 2-tailed Student $t$ test or one-way ANOVA unless noted otherwise. Data represent mean ± SD. $P > 0.05$ was considered not significant.

Supplemental references
Horejsi, Z., Stach, L., Flower, T.G., Joshi, D., Flynn, H., Skehel, J.M., O'Reilly, N.J., Ogrodowicz, R.W., Smerdon, S.J., and Boulton, S.J. (2014). Phosphorylation-dependent PIH1D1 interactions define substrate specificity of the R2TP cochaperone complex. Cell reports 7, 19-26.

Lansdorp, P.M., Verwoerd, N.P., van de Rijke, F.M., Dragowska, V., Little, M.T., Dirks, R.W., Raap, A.K., and Tanke, H.J. (1996). Heterogeneity in telomere length of human chromosomes. Human molecular genetics 5, 685-691.

Munoz-Jordan, J.L., Cross, G.A., de Lange, T., and Griffith, J.D. (2001). t-loops at trypanosome telomeres. The EMBO journal 20, 579-588.

Thorslund, T., Esashi, F., and West, S.C. (2007). Interactions between human BRCA2 protein and the meiosis-specific recombinase DMC1. The EMBO journal 26, 2915-2922.

Vannier, J.B., Pavicic-Kaltenbrunner, V., Petalcorin, M.I., Ding, H., and Boulton, S.J. (2012). RTEL1 dismantles T loops and counteracts telomeric G4-DNA to maintain telomere integrity. Cell 149, 795-806.

Vannier, J.B., Sandhu, S., Petalcorin, M.I., Wu, X., Nabi, Z., Ding, H., and Boulton, S.J. (2013). RTEL1 is a replisome-associated helicase that promotes telomere and genome-wide replication. Science 342, 239-242.

Ward, J.D., Muzzini, D.M., Petalcorin, M.I., Martinez-Perez, E., Martin, J.S., Plevani, P., Cassata, G., Marini, F., and Boulton, S.J. (2010). Overlapping mechanisms promote postsynaptic RAD-51 filament disassembly during meiotic double-strand break repair. Molecular cell 37, 259-272.

Whitfield, M.L., Sherlock, G., Saldanha, A.J., Murray, J.I., Ball, C.A., Alexander, K.E., Matousek, J.C., Perou, C.M., Hurt, M.M., Brown, P.O., et al. (2002). Identification of genes periodically expressed in the human cell cycle and their expression in tumors. Molecular biology of the cell 13, 1977-2000.

Zellinger, B., and Riha, K. (2007). Composition of plant telomeres. Biochimica et biophysica acta 1769, 399-409.