Published in final edited form as:
Annu Rev Genet. 2008 ; 42: 587–617. doi:10.1146/annurev.genet.42.110807.091549.

The take and give between retrotransposable elements and their hosts

Arthur Beauregard[1,2], **M. Joan Curcio**[1,2], and **Marlene Belfort**[1,2,3]

1*Wadsworth Center, New York State Department of Health, Center for Medical Sciences, 150 New Scotland Avenue, Albany, NY 12208, USA*

2*School of Public Health, State University of New York at Albany, Albany, NY 12201−2002, USA.*

Abstract

Retrotransposons mobilize via RNA intermediates and usually carry with them the agent of their mobility, reverse transcriptase. Retrotransposons are streamlined, and therefore rely on host factors to proliferate. However, retrotransposons are exposed to cellular forces that block their paths. For this review, we have selected for our focus elements from among *t*arget-*p*rimed (TP) retrotransposons, also called non-LTR retrotransposons, and *e*xtrachromosomally-*p*rimed (EP) retrotransposons, also called LTR retrotransposons. The TP retrotransposons considered here are group II introns, LINEs and SINEs, whereas the EP elments considered are the Ty and Tf retrotransposons, with a brief comparison to retroviruses. Recurring themes for these elements, in hosts ranging from microbes to man, are tie-ins of the retrotransposons to RNA metabolism, DNA replication and repair, and cellular stress. Likewise, there are parallels among host-cell defenses to combat rampant retrotransposon spread. The interactions between the retrotransposon and the host, and their co-evolution to balance the tension between retrotransposon proliferation and host survival, form the basis of this review.

Keywords

TP and EP retrotransposons; host factors; DNA replication and repair; RNA surveillance

INTRODUCTION

Retrotransposons - what's in a name?

Retroelements, such as retroviruses and retrotransposons, move into genomes via RNA intermediates and most often carry with them the agent of their mobility, *r*everse *t*ranscriptase (RT). Retrotransposons are found in all three domains of life, bacteria, archaea and eukarya. Indeed, estimates are that at least 42% of the human genome comprises DNA that is derived from RNA via reverse transcription (116).

Retrotransposons have been classified in different ways (201), most recently into four groupings (63), which can be associated with two historic classes. The first class comprises *l*ong-*t*erminal *r*epeat (LTR) retrotransposons, so-called because of the presence of flanking repeat sequences (Figure 1A). LTR retrotransposons synthesize a *d*ouble-*s*tranded DNA (dsDNA) intermediate, using the element's RT and RNA as a template. The completed *c*omplementary DNA (cDNA) is then inserted into the host chromosome via a recombination

3Corresponding author – Phone: 518−473−3345 FAX: 518−474−3181 E-mail: belfort@wadsworth.org. Other email addresses: beaureg@wadsworth.org curcio@wadsworth.org.

event involving an associated integrase (or recombinase). Examples of LTR retrotransposons are the Ty elements of *Saccharomyces cerevisiae*, Tf elements of *Schizosaccharomyces pombe* and gypsy elements of *Drosophila melanogaster*. These retrotransposons serve as useful model systems of retroviruses, which also use an integrase-based DNA insertion mechanism. The second class comprises the non-LTR retrotransposons, unfortunately so-called for what they lack, namely terminal repeats, which in fact a minority of them do have (Figure 1B). Members of this class often encode endonucleases, and reverse transcribe a copy of their RNA template directly into the chromosome by a process termed *t*arget-*p*rimed *r*everse *t*ranscription (TPRT) (125). Examples of non-LTR retrotransposons are bacterial and organellar group II introns, R1 and R2 elements of arthropods, and mammalian *l*ong-*i*nterspersed *n*uclear *e*lements (LINEs) and *s*hort-*i*nterspersed *n*uclear *e*lements (SINEs) (retrotransposon architecture is represented in Figure 2).

We propose a revised nomenclature for these two groups, based on mechanism rather than structure. Thus, we will name non-LTR retrotransposons, for which priming occurs intrachromosomally, as *t*arget-*p*rimed TP retrotransposons (54). In contrast, we will name the LTR retrotransposons, for which cDNA synthesis occurs before integration, as *ex*trachromosomally-*p*rimed EP retrotransposons. This EP/TP classification scheme is in overall agreement with cladograms of the elements based on RT sequences (3,63). Most elements that were outliers in the original LTR/non-LTR classification will be accommodated in the TP/EP scheme. For example, the P*enelope-l*ike *e*lements (PLEs), originally discovered in *Drosophila virilis*, sometimes have LTRs and usually contain endonucleases. PLEs likely integrate by TPRT (65) and therefore would be considered TP retrotransposons. Another example is the D*ictyostelium i*ntermediate *r*epeat *s*equence (DIRS) retrotransposons. They have an RT more closely related to the EP than to TP retrotransposons, but they encode a tyrosine recombinase rather than the classic integrase, and they likely synthesize their cDNA independently of the chromosome (77,100). These elements would be considered EP retrotransposons. We urge our readers to consider whether they find these proposed nomenclature conventions more satisfying and useful than the historic names and if so, to adopt them.

The focus of this review

This review is confined to a handful of retrotransposons, to give the reader the essential information on the retromobility pathways of the elements, and to relate them adequately to host factors. The intention is not to survey retrotransposons, but rather to select a few, and provide enough detail to highlight similarities and differences that appear in the various battlegrounds between the elements and their phylogenetically distinct hosts. We have chosen from among the most thoroughly studied of the retroelement/host pairs: bacterial and, to a lesser extent, organellar group II introns, the mammalian LINE and SINE retrotransposons and yeast Ty and Tf elements.

Retrotransposons can be considered streamlined parasitic elements, in the sense that they need their host for replication, and carry only those molecular features that are required to multiply within genomes and to spread to others. A recent flourish of work describes how retroelements scavenge functions from their hosts, to complete their life cycles and sense their environments, adjusting their lifestyles accordingly. Responsiveness to their cellular milieu helps retrotransposons balance their conflicting needs of constraint to avoid harming the host, and promiscuity to achieve dissemination. While the element reacts to these contradictory needs, the host responds to the fitness penalty introduced by the retroelements by down-regulating or restricting expression of the retrotransposon. Study of this conflict has led to the nascent field of retrotransposon-host relationships. The adaptations and counter-adaptations between retroelement and host, and their co-evolution, form the focus of this review.

RETROTRANSPOSON ARCHITECTURE AND MOBILITY PATHWAYS

Group II introns in bacteria and organelles

Mobile group II introns are TP retrotransposons. Group II introns are of special interest because they are thought to be the progenitors of nuclear spliceosomal introns (37,99,177), and they are found in all three domains of life (57,58). These introns are ribozymes that catalyze the splicing of their flanking exons (23,136), and some are also highly mobile retroelements (13, 113,115). They move to cognate or ectopic sites, in processes termed retrohoming and retrotransposition, respectively, by means of an *i*ntron-*e*ncoded *p*rotein (IEP) that has RT activity (Figures 2 and 3).

Retrohoming—Retrohoming, which occurs at frequencies approaching 100%, has been studied in detail for two related yeast mitochondrial introns (aI1 and aI2) and two bacterial introns (Ll.LtrB and RmInt1) (13,115,129). Retrohoming occurs by a process in which the excised intron RNA reverse splices into one strand of a DNA target site and is then reverse transcribed by the IEP (Figure 3A & B), by TPRT.

This process is mediated by a *r*ibo*n*ucleo*p*rotein (RNP) complex that forms during RNA splicing and contains the IEP and the excised intron lariat RNA. Retrohoming of the aI1, aI2 and Ll.LtrB introns is dependent upon three activities of the IEP, in addition to catalytically active intron RNA (Figure 3A) (13, 114, 115). These are RNA maturase, to stabilize the catalytically active structure of the intron RNA for RNA splicing and reverse splicing; DNA endonuclease, for cleavage of the target DNA to generate a primer for reverse transcription; and RT, for making a cDNA copy of the intron RNA. After reverse-splicing of the intron into DNA (Figure 3A, step 1) and endonucleolytic cleavage of the second strand, downstream of the intron-insertion site (Figure 3A, step 2), full-length cDNA synthesis occurs by TPRT (Figure 3A, steps 3 & 4). The final steps of the process can diverge: whereas the Ll.LtrB intron retrohomes independently of RecA recombinase function (52, 137), the yeast introns can invoke homologous recombination (64).

Interestingly, many group II introns lack the endonuclease domain, and are thus unable to cleave target DNA; these introns likely use replication intermediates to prime cDNA synthesis. The best studied of the introns lacking endonuclease is RmInt1 of *Sinorhizobium meliloti* (130), an intron which targets primarily *s*ingle-*s*tranded DNA (ssDNA) of the nascent lagging strand at DNA replication forks in order to prime reverse transcription (Figure 3B) (129). After the initial steps in the pathway, the group II intron recruits multiple host functions to complete the process (Table 1).

Retrotransposition—Retrotransposition of the Ll.LtrB intron, a process which is at least 10^4-fold less efficient than retrohoming, was first studied in *Lactococcus lactis* (51). Not only is there a more relaxed sequence requirement for retrotransposition than for retrohoming, but also the intron targets ssDNA rather than dsDNA (Figure 3B); further it does not require endonuclease function, but rather uses the replication fork to prime cDNA synthesis (95). This pathway is similar to retrohoming of the RmInt1 intron (129), and to retrohoming of endonuclease-minus mutants of the Ll.LtrB intron (212). In contrast to retrotransposition in *L. lactis*, retrotransposition of Ll.LtrB in *Escherichia coli* has an overall target preference for dsDNA, indicating that not only the intron but also the host cell influences pathway selection (8,47).

LINES and SINES in mammalian systems

LINEs and SINEs are the most prevalent TP retrotransposons in mammalian systems. LINEs encode RTs, which mobilize both LINEs and SINEs, with the latter being nonautonomous

because they lack RT (Figure 2) (199). LINEs and SINEs are also found in other vertebrates (2,148), and some will be referred to, but our focus here will be on LINE-1 (L1), the most abundant human LINE. L1 is transcribed by RNA polymerase II to give a 6−7 kb mRNA with a poly(A) tail. The mRNA encodes two proteins, ORF1p, a chaperone protein (128), and ORF2p, which has RT and endonuclease activities (Figure 2) (66,133). Both proteins are required for retrotransposition (140,141). Interestingly, although L1s are often truncated at their 5′ ends, humans still contain ~100 active L1 elements (28,168). The most common human SINE is Alu, which is transcribed by RNA polymerase III as a ~300-nt noncoding RNA (7). L1 and Alu have accumulated to the point of constituting at least 17% and 11% of the human genome, respectively, with Alu present in excess of one million copies (116).

L1 retrotransposes by TPRT (50) (Figure 4), in a manner similar to group II introns (13) and the R2*Bm* TP retrotransposon of *Bombyx mori*, for which the mechanism has been extensively studied (62). After L1 transcription and ORF1p and ORF2p translation, retrotransposition proceeds when the ORF2p endonuclease nicks target DNA exposing a 3′-hydroxyl that serves as primer for reverse transcription. L1 endonuclease has preference for the consensus sequence 5′-TTTTA-3′, cleaving 5′ to the A, with a plethora of sites in the genome (49). Priming can be facilitated by alignment of the poly(T) DNA with the poly(A) tail of L1, upon which the RT reverse transcribes a cDNA copy of L1 into the host chromosome (66,151). Whether the nick on the top strand is catalyzed by ORF2p or by a host nuclease is not known. Also unclear is if removal of the L1 RNA template involves host proteins, because the element appears not to specify its own ribonuclease H (RNase H), but strand-displacement activity of ORF2p may suffice to remove the RNA. Second-strand cDNA synthesis of both R2*Bm* and L1 is likely to involve the RT, which has DNA-dependent DNA polymerase activity (112,151). Finally, different cellular repair functions appear to be recruited to complete the LINE integration process (74,188).

Ty and Tf retrotransposons in yeast

Infectious retroviruses, so far found primarily in vertebrates, are so closely related to EP retrotransposons that it can be difficult to assign newly unearthed elements to one class or the other based solely on phylogenetic information. EP retrotransposons comprise the most ancient families, and therefore it is generally assumed that retroviruses emerged from these progenitors by acquisition of an *env* gene, and the concomitant ability of virion particles to bud out of one cell and into another (105). However, the converse, that retrotransposons arise by intracellularization of retroviruses, is likely to occur as well. Indeed, a recent study indicates that mouse *i*ntracisternal *A p*article (IAP) retrotransposons are derived from a retrovirus that infected the germline, allowing the element to be transmitted vertically (158). Phylogenetic analysis places the IAP retrotransposon in the retrovirus family (Retroviridae), yet IAP is one of the most active and mutagenic retrotransposons in the mouse genome (87). A single active copy of an endogenous retrovirus, IAPE, which generates infectious extracellular viral particles, is the likely progenitor of IAP retrotransposons (158). Remarkably, deletion of the *env* gene from the IAPE retrovirus and substitution of a small region of the *gag* gene of IAPE with that of IAP results in robust retrotransposition activity.

Given this pendular evolutionary relationship, it is not surprising that retroviruses and EP retrotransposons utilize many of the same host trafficking pathways to complete their intricate journeys through the replication cycle (27,134). The yeasts *S. cerevisiae* and *S. pombe* are unique model systems for studying EP retrotransposons because these EP elements are the only family of transposable elements present in their nuclear genomes, and therefore are solely responsible for insertional mutations and formation of the retrogenomes. The *S. cerevisiae* genome contains five families of EP retrotransposons (Ty1−5), three of which, Ty1, Ty3 and Ty5, have been studied extensively. Ty1 and Ty5 retrotransposons are members of the Ty1/

copia family (Pseudoviridae), while Ty3 belongs to the Ty3/gypsy/BEL family (Metaviridae). Ty elements consist of two direct long terminal repeats flanking *gag* and *pol* ORFs or a single *gag-pol* ORF (Figure 2). *Gag* encodes the structural protein(s) of the cytoplasmic viral core particle or *v*irus-*l*ike *p*article (VLP), while *pol* encodes a polyprotein with four distinct activities: protease, integrase, RT, and RNase H. All activities of the *pol* polyprotein are required for retrotransposition (reviewed by 197). The *gag* and *pol* of Ty1 and Ty3 are overlapping, and Pol is expressed as a Gag-Pol polyprotein resulting from ribosomal frameshifting from *gag* into *pol*. The Ty replication cycle can be divided into the following steps (Figure 5): transcription and nuclear export; translation; VLP assembly, including encapsulation of Ty mRNA, and VLP maturation by protein processing; reverse transcription of Ty mRNA; and nuclear import, followed by integration of cDNA into the chromosome. The structure and replication cycle of Tf retrotransposons in *S. pombe* are similar to those of Ty retrotransposons.

HOST RNA PROCESSING FACTORS THAT FACILITATE RETROTRANSPOSITION

Group II Introns

Dependence of group II intron splicing on host factors—Splicing is required to generate the excised intron lariat that forms the RNP and invades DNA in the first step of retromobility. Although group II introns are autocatalytic, they require proteins to enable them to splice efficiently in cells (113). There are two basic varieties of proteins that facilitate splicing: maturases, which are usually intron-encoded, and scaffold the active structure of the intron; and chaperones, which are extra-intronic proteins that resolve poorly folded inactive structures until an active conformation is reached. Maturases and chaperones can collaborate to promote the activity of a single intron, but whereas maturases bind introns rather specifically, whereas chaperones are usually non-specific. By and large, chaperones have evolved the ability to help fold introns as a role secondary to some other specific cellular function. Although an active intron is required in the first step of retromobility, namely reverse-splicing of the DNA target, it is unclear if the same proteins that facilitate RNA splicing are required to drive the intron into a DNA target.

Genetic analyses of yeasts, algae and plants have revealed a large number of nuclear genes that are required for splicing of organellar group II introns. Although algae and plants have many such splicing factors (22), we will focus on *S. cerevisiae*, in which nuclear-encoded aminoacyl tRNA synthetases and Mrs2p are required to promote splicing of all four mitochondrial group II introns, aI1, aI2, aI5γ, and bI1 (82,113). Interestingly, Mrs2p, which is a Mg^{2+} transporter in the inner mitochondrial membrane, regulates the Mg^{2+} concentration that is responsible for group II intron splicing (79,202). Additionally, the nuclear DEAD-box protein, Mss116p is involved in splicing mitochondrial group I and all four group II introns, including those that require IEPs and other splicing factors for structural stabilization (91,174). CYT-19, a *Neurospora* DEAD-box ATPase, suppresses *mss116* mutants, suggesting a common and general mechanism of action throughout lower eukaryotes (91). Although there is debate on their precise role, these DEAD-box proteins are thought to function primarily as RNA chaperones, to accelerate, in ATP-dependent and -independent ways, structural rearrangements by unwinding short intron RNA helices and disrupting other structural kinetic traps (60,139, 183,191).

Other RNA-processing functions that stimulate group II intron retrohoming—The relationship between group II intron retromobility and the host has been studied most thoroughly for the Ll.LtrB intron of *L. lactis*. Ll.LtrB contains a functional promoter upstream of the IEP, LtrA, and this promoter is required for efficient LtrA expression to ensure splicing

and mobility (213). This intron is also functional in splicing and retromoblity in *E. coli*, and the endonuclease-dependent retrohoming pathway is mechanistically similar in *L. lactis* and *E. coli* (52,137) (Figure 3A). After reverse transcription, the intron RNA must be removed for synthesis of the second DNA strand using the cDNA as template. Unlike retroviruses and EP retrotransposons, group II introns and most other TP retrotransposons do not encode their own RNases H (126). *E. coli* has two RNases H, H1 and H2, of which only RNase H1 is required for retrohoming (181). RNase H1, the product of the *rnhA* gene, and the 5′−3′ exonuclease activity of Pol I, the product of the *polA* gene, are both likely involved in intron RNA degradation in *E. coli* (Figure 3A). Whereas the *rnhA* gene can complement an RNase H1 defect to restore retrohoming, it fails to complement a Pol I 5′−3′ exonuclease defect, suggesting distinct roles for RNase H1 and Pol I. This result can be reconciled in view of the different activities of these enzymes: RNase H1 endonucleolytically cleaves the RNA strand in an RNA/DNA hybrid, while Pol I is an exonuclease (108). RNA processing functions that negatively regulate group II intron retromobility will be described in the Silencing section.

LINEs and SINEs

Regulation of L1 RNA begins at the level of transcription and continues throughout complex patterns of RNA processing. Human L1 contains an internal pol II promoter in the 5′ UTR. This promoter drives transcription of the full-length L1 bicistronic mRNA (189), which encodes ORF1p and ORF2p. L1 is preferentially transcribed and retrotransposes most actively in the germ-line and in early development (26,194); and the suppression of such activity in somatic cells is at least partially regulated by tissue-specific transcription factors such as Yin Yang-1 (9), SOX family members (190), and RUNX3 (204). All of these transcription factors bind to regions of the 5′ UTR to regulate L1 expression and retrotransposition at the RNA level.

Ty and Tf retrotransposons

Five different genome-wide screens have been performed to identify stimulators or inhibitors, or both, of Ty1 or Ty3 retrotransposition (Table 2) (4,81,97,147,170). Hundreds of regulators of Ty1 and Ty3 moblity have been identified, and only about 40 have been found in more than one screen (134,147). Like the host factors required for the replication of the HIV-1 retrovirus, Ty1 and Ty3 host factors play roles in diverse cellular processes, from transcription and translation to endomembrane trafficking and nuclear import (27). Interestingly, screens for Ty1 and Ty3 stimulators also implicate many different components of mRNA processing and degradation pathways in the regulation of retrotransposition.

Dbr1, a 2′−5′ phosphodiesterase required for debranching of the intron lariat RNA formed in the process of mRNA splicing, was identified 16 years ago as a stimulator of Ty1 retrotransposition (38), and it was re-isolated in screens for both Ty1 and Ty3 stimulators. Notably, not only Ty1 and Ty3 retrotransposition, but also HIV-1 replication, require Dbr1. In all three systems, accumulation of cDNA is decreased when Dbr1 expression is reduced (101,166,205). However, there is a debate over the role of Dbr1 in the replication cycle of these retroelements. Hypothetically, VLPs of *dbr1* mutants harbor a lariat form of the Ty1 RNA in which the 5′ phosphate of the first nucleotide of Ty1 RNA forms a phosphodiester bond with the 2′ hydroxyl of a nucleotide in the 3′ LTR. Lariat formation could facilitate transfer of *m*inus-strand strong-stop (msss) cDNA (the first segment of cDNA synthesized) from the 5′ LTR to the 3′ LTR. Thus, Dbr1 is proposed to resolve the lariat so that RT can traverse the branch point, allowing minus-strand cDNA synthesis to resume (Cheng and Menees, 2004). Notably, HIV-1 cDNA synthesis is also blocked at a step subsequent to msssDNA synthesis when DBR1 expression is reduced (205). However, another laboratory was unable to detect the Ty1 RNA lariat in VLPs of *dbr1* mutants using several methods; moreover, they demonstrated that RT-PCR could provide artefactual evidence for the existence of a lariat form of Ty1 RNA (45).

Thus, the mechanism by which Dbr1 promotes Ty1, Ty3 and HIV-1 replication remains to be elucidated.

Several components of the deadenylation-dependent mRNA degradation pathway were isolated as stimulators of Ty1 and Ty3 retrotransposition in genome-wide screens (81,97). Deadenylation-dependent mRNA degradation, one of the major pathways of mRNA degradation in the cell, occurs when deadenylated mRNA is recognized by the translational repressors, Pat1 and Dhh1, and the decapping co-activator complex, Lsm1−7. Interaction of these factors with mRNA impedes the association of mRNA with the translation initiation complex and promotes sequestration of mRNA in dynamic RNP complexes known as mRNA processing bodies, or P bodies (Figure 5). The 5′ cap is removed by the Dcp1/Dcp2 decapping complex, leading to rapid degradation of the mRNA by the 5′−3′ exoribonuclease, Xrn1. P bodies are found in mammalian cells as well as in yeast, and the core components including Pat1, Dhh1, Lsm1−7, Dcp1/Dcp2 and Xrn1 are conserved in eukaryotes (reviewed in 43). Thus, it was hypothesized that the mRNA degradation pathway is involved in sequestering Ty3 mRNA away from translation so that it could be packaged into assembling VLPs. Accordingly, Ty3 VLP proteins and RNA are associated with core components of P bodies in cytoplasmic foci (15). Moreover, mutations in Ty3 Gag proteins that prevent packaging of the Ty3 mRNA in VLPs also block the association of Ty3 mRNA and proteins with P bodies (117).

The association of VLPs with P bodies is apparently not specific to Ty3, since VLPs produced from endogenous Ty1 elements are also found in association with P body proteins (Dutko, J., Kenny, A. & Curcio, M.J., in preparation). Perhaps, then, the association of Ty VLPs with P bodies promotes assembly of functional VLPs, but a major question remains: do deadenylation-dependent mRNA degradation pathway enzymes act directly on Ty RNA to promote retrotransposition (Figure 5)? Both capped and uncapped Ty1 mRNAs are found in VLPs (40,45), suggesting that Ty1 mRNA is the direct target of the Dcp1/Dcp2 decapping complex. Perhaps uncapping of Ty1 mRNA is necessary for msssDNA synthesis or strand-transfer during reverse transcription. However, this model does not provide an obvious explanation for the role of the 5′−3′ exonuclease, Xrn1, which is necessary for both Ty1 and Ty3 retrotransposition. Given that all retrotransposons need a mechanism of partitioning their transcripts between translation and association with RT, it is likely that this area of investigation will remain an important one for some time.

THE ROLE OF DNA REPLICATION AND/OR REPAIR

Group II Introns

Replication—The nature of group II intron mobility pathways makes it clear that after the initial stages of retrohoming and retrotransposition, the mobility intermediates become subsumed into cellular replication and repair pathways (8,47,181,212). Because the RT of group II introns has very low processive DNA-dependent polymerization activity (165), second-strand cDNA synthesis is presumed to be carried out by a host polymerase (Figure 3). In *E. coli*, the α catalytic subunit of the major replicative polymerase, Pol III, has been implicated as the enzyme recruited to synthesize the complement of the RT-generated cDNA (181). Furthermore, MutD (DnaQ), the ε 3′−5′ exonuclease proofreading subunit of the 10-subunit Pol III, is also required, suggesting that these two enzymes act in concert to promote intron mobility with high fidelity (44).

Other evidence in favor of group II intron mobility exploiting host functions in general and DNA replication in particular comes from group II intron distribution. The majority of these introns are found on multicopy genomes, other mobile genetic elements or plasmids, suggesting that the intron hitches a ride on the multi-copy replicon or exploits the dissemination apparatus of the mobile element on which it residues. In eukaryotes, group II introns occur exclusively,

and sometimes in great numbers, in mitochondrial and chloroplast genomes (23,46). In bacteria, roughly 35% of group II introns occur on plasmids or other mobile genetic elements like transposons, conjugative plasmids, integrons, insertion sequences and pathogenicity islands (57,107). Indeed, the two best studied bacterial group II introns, Ll.LtrB and RmInt1, were discovered on mobile elements. Ll.LtrB exists on the conjugative plasmid pRS01 and on the homologous integrated sex factor of different *L. lactis* strains (138,178), and RmInt1 was discovered in the insertion sequence IS*Rm2011−2* of *S. melliloti* (132).

Significantly, the mobility frequency of the Ll.LtrB intron is elevated in the presence of plasmid pRS01 (14), suggesting that either the conjugative transfer process or factors residing on pRS01 boost intron movement. Ll.LtrB retrotransposition in *L. lactis* is further linked to replication by an integration bias toward the template for lagging-strand DNA synthesis, where Okazaki fragments likely serve as primers for reverse transcription (95). The likelihood of the dependence on replication as a means for retrotransposition was further strengthened by the observation that a retrotransposing Ll.LtrB favors plasmid over chromosomal targets, likely reflecting the higher relative density of replication forks on plasmids, as targets for retrotransposition (94).

Group II introns that lack the endonuclease domain to cleave bottom strand target DNA, like RmInt1, retrohome by targeting ssDNA (129). The process is similar to retrotransposition of Ll.LtrB in *L. lactis* (95) (Figure 3B), with RmInt1 likely utilizing replication intermediates to prime cDNA synthesis by targeting the nascent lagging strand at DNA replication forks (129,212). Group II introns thus piggy-back on the replication or dissemination machinery, respectively, of multicopy genomes and transposons of their hosts.

DNA repair functions that stimulate retrohoming—In addition to a role for Pol III, the replicative polymerase, the repair polymerases, Pol II (*polB*), Pol IV (*dinB*) and Pol V (*umuDC*) have a substantial collective effect on retrohoming in *E. coli* (181). This observation suggests a need for polymerization across DNA lesions or DNA-RNA junctions (Figure 3A), consistent with the relaxed specificity of the enzymes, which assist the high-fidelity processive Pol III (152,192). These requirements for the repair polymerases suggest that the cell views the later steps of retrohoming as a DNA damage response.

RecJ, is a 5′−3′ ssDNA exonuclease that stimulates retrohoming. RecJ was postulated to be required following synthesis of a full-length intron cDNA for resection of the 5′ exon DNA, to allow the nascent cDNA to pair with the top strand (181). Whereas such a function might be achieved by DNA unwinding, none of the DNA helicases tested had a major facilitatory effect. Finally, *E. coli* DNA ligase is required, presumably to seal the nicks and generate the completed retrohoming product (Figure 3A).

LINEs and SINEs

DNA repair functions that stimulate retrotransposition—Whereas little is known about the relationship between replication and L1 retrotranspsosition, L1 elements have recently been inferred to interact with host factors involved in DNA repair, in what is the dawning of a new field. Mammalian L1s generate a *d*ouble-*s*trand *b*reak (DSB) during integration (72). DSBs in a cell elicit the DNA repair response and accordingly, the *a*taxia-*t*elangiectasia *m*utated (ATM) protein, a multifunctional serine/threonine protein kinase that regulates the cell's response to DSBs (179), is required for L1 retrotransposition (72). ATM is recruited to DSBs and activates repair by phosphorylation of a network of other proteins. Recent work has shown that components of the *n*on-*h*omologous *e*nd *j*oining (NHEJ) pathway maybe involved in LINE retrotransposition. Mutations of DNA-end binding protein Ku70, the 5′−3′ exonuclease Artemis, and DNA ligase IV, result in decreased retrotransposition frequencies of two structurally different LINEs, human L1 and zebrafish ZfL2−2, in chicken

cell lines. These same elements retrotranspose with reduced efficiency in HeLa cells deficient in DNA-dependent protein kinase, catalytic subunit (DNA-PKcs), which is presumed to propagate the signal of DSBs by phosphorylation of other required enzymes (Figure 4) (188).

A model has been proposed for LINE integration by TPRT using these components. Accordingly, after nicking of the first (bottom) strand, cDNA and second strand synthesis, the Ku complex binds to the 5′ end of the DNA/RNA heteroduplex, recruiting DNA-PKcs, Artemis exonuclease and DNA ligase IV. Since Artemis can digest both DNA and RNA, this exonuclease might account for the frequently 5′-truncated LINEs. Also implicated as an inhibitor of LINE integration is the ERCC1/XPF heterodimer, which has been postulated to act on 3′ flap DNA intermediates that arise as a result of cDNA synthesis (71). Once the ends are compatible, DNA ligase IV is proposed to ligate the junctions, much as bacterial ligase is required for group II intron retrotransposition in *E. coli* (188). Interestingly, ZfL2−2 is more dependent than L1 on the NHEJ proteins, suggesting that L1 relies on alternative repair pathways as well (74), with the differences possibly being attributable to the zebrafish elements lacking ORF1p, which are produced by L1s. This situation is again reminiscent of what is seen for group II introns, which use different repair pathways depending on activities encoded by the element: introns encoding endonuclease activities retrohome strictly by TPRT, whereas those that do not are subsumed into replication pathways (Figure 3).

Ty retrotransposons

There is a dearth of evidence for DNA repair functions that participate in Ty1 and Ty3 retrotransposition, although S-phase checkpoint pathway proteins Rad9 and Rad24 have a modest stimulatory effect on Ty1, and Rad24 promotes Ty3 retrotransposition (Table 2) (55, 97). However, more than 30 different proteins with diverse roles in genome maintenance repress Ty3 and/or Ty1 retrotransposition (29,97,118,119,147,157,170,171,187). In most instances, the absence of genome maintenance factors/Ty3 repressors results in elevated levels of Ty3 Gag protein and/or Ty3 cDNA (97). On the other hand, the absence of genome maintenance factors/Ty1 repressors increases Ty1 cDNA levels but not Ty1 RNA or Gag protein levels (29,118,119,157,170,171,187). Studies illustrating the effects on Ty1 mobility of mutations in individual DNA maintenance factors such as Est2 (the catalytic subunit of telomerase), Rad27 (a homolog of the human FEN-1 endonuclease), Ssl2 and Rad3 (TFIIH-associated helicases), Rad52 (a DSB repair protein) and Sgs1 (a RecQ-family helicase) suggest that diverse mechanisms are involved in modulating the level and the destination of Ty1 cDNA in genome-maintenance mutants (29,118,119,157,171,187). Three major mechanisms are discussed below.

DNA damage checkpoint pathways stimulate Ty1 retromobility—One explanation for the increased Ty1 cDNA and retromobility in a variety of different genome-maintenance mutants is that DNA lesions activate DNA-damage checkpoint pathways, which in turn stimulate reverse transcription of Ty1 cDNA. This was first observed in telomerase-negative *est2* mutants, in which the extent of telomere erosion is correlated with the induction of Ty1 cDNA accumulation (171). Ty1 cDNA levels in *est2* mutants are significantly reduced in the absence of checkpoint pathway proteins Rad9 or Rad24, implicating the DNA-damage checkpoint pathway in the elevation of cDNA levels. Notably, the DNA damage checkpoint pathway or the replication stress checkpoint pathway stimulates Ty1 retrotransposition in the absence of 19 additional genome maintenance factors (55). Ty1 VLPs in *rtt101* mutants, which have elevated levels of replication fork pausing, contain substantially increased amounts of mature RT and integrase, and RT activity in VLPs is also elevated. Thus, DNA lesions created in the absence of many different factors required for genome maintenance trigger S-phase checkpoint pathways to stimulate Ty1 RT activity.

Direct interaction of DNA repair proteins with Ty1 cDNA?—Studies of Ty1 transposition intermediates in *rad27*, endonuclease *ssl2* and *rad3* and helicase mutants indicate that Ty1 cDNA is stabilized in some genome maintenance mutants (118,119,187). Following exposure to an RT inhibitor, Ty1 cDNA levels decrease more slowly in these mutants than in wild-type strains, supporting a role for Rad27, Ssl2 and Rad3 in destabilization of cDNA. Rad27 possesses 5′-flap endonuclease and 5′−3′ exonuclease activities that are required to process Okazaki fragments during replication. Because Ty1 RNA:cDNA hybrids, as well as cDNA containing a DNA flap structure, are intermediates in transposition (70,88,89), Rad27 could interact directly with partially synthesized Ty1 cDNA intermediates, promoting degradation. It has been proposed that Ssl2 and Rad3 bind free Ty cDNA ends and facilitate cDNA unwinding, leading to degradation (118).

While the model that genome maintenance factors directly promote degradation of Ty1 cDNA fits with their roles in preserving genome integrity, this cannot be the whole story, because the half-life of Ty1 cDNA in wild-type strains is very long (93 to 252 minutes in different studies), and is similar to the doubling time of yeast cells (118,171,187). However, the synthesis of the plus strand of Ty1 cDNA has been reported to be inefficient, resulting in the accumulation of Ty1 RNA:cDNA hybrid molecules (88,153). Therefore, partially synthesized Ty1 RNA:cDNA molecules are probably converted to double-stranded cDNAs by host repair enzymes, and this process may be more efficient in *ssl2*, *rad3* and *rad27* mutants.

Multimeric cDNA arrays—Another mechanism by which Ty1 mobility is increased in genome maintenance mutants, exemplified by mutants in which the RecQ-like helicase, Sgs1, is deleted, is by the formation of multimeric Ty1 cDNA arrays. In *sgs1*Δ mutants, there is no increase in the frequency of integration; rather, each integration event consists of multiple cDNAs (29). It is not known if Sgs1 binds directly to Ty1 cDNA to suppress recombination between cDNAs. Multimeric Ty1 cDNA arrays can form before integration (200); alternatively, they may form in the process of repair of unstable recombination events (176). The latter explanation is consistent with the observation that complex multimeric Ty1 arrays are found at chromosomal breakpoints (134,195) and may well be responsible for boosting Ty1 mobility.

SILENCING

Group II introns

Cells mount a variety of counter-attacks on retroelements in response to the fitness penalties these invaders impose. However, group II introns, through their ability to splice, minimize the burden on the host and also the pressure to evolve specific silencing mechanisms: the introns can be viewed as self-silencing. Nevertheless, both RNases and DNases inhibit retrohoming and may well provide either a cellular response against movement of group II introns, or at least a signal to group II introns to live within the cell's means. RNase I and RNase E are degradative enzymes that inhibit retrohoming (48,181). They reduce the half-life of intron RNA and could act on either free or integrated intron RNA (Figure 3). RNase E is a component of a multiprotein complex, the RNA degradosome, which determines RNA stability (reviewed in 36). The degradosome is assembled on RNase E and is regulated by growth conditions, which modulate mRNA degradation. As a further focus of regulatory activity, RNase E is inhibited by *r*egulator of *r*ibonuclease *a*ctivity proteins, RraA and RraB (69,120). These regulatory influences provide multiple opportunities for group II intron movement to be synchronized with the cell's physiology, given that introns are sensitive to RNase E degradation, a process upon which various cellular controls impinge.

A DNA exonuclease, Exo III, the product of *xthA*, also inhibits retrohoming. Exo III's 3′−5′ exonuclease activity may degrade the newly synthesized cDNA and/or interfere with second-

strand synthesis, thereby inhibiting repair (Figure 3). Interestingly, Exo III is positively regulated by RpoS (KatF) (164), which is an alternative sigma factor that regulates RNA polymerase in stationary phase (163), possibly accounting for the reduction in retrohoming in stationary phase (Coros CJ and MB, unpublished). Thus, without any element-specific silencing mechanism, the group II intron activity can be inhibited in accord with the physiological status of the cell.

LINEs and SINEs

L1 retrotransposition occurs predominantly in germ and embryonic cells, and immortalized cell lines. In contrast, the elements appear silenced in most somatic cells, to avert the potentially catastrophic consequences of uncontrolled retrotransposition (reviewed by 33, 109). Maintaining their silence can be a matter of the element having acquired features that respond to the normal macromolecular machinery of the cell, or can result from cells mounting an array of active silencing responses.

The first examples of the self-limiting nature of LINES stem from the A-richness of the 6−7 kb transcript. Not only is it hypothesized that this feature sets limits on transcription elongation to reduce the overall transcription efficiency (84), but also that it leads to premature polyadenylation at internal poly(A) sites, suppressing L1 expression and retrotransposition (150). Another example is that although L1 has no introns, the transcript contains multiple 5′ splice donor and 3′ splice acceptor sites in both the sense and antisense strands. Splicing at these sites deletes functional sequences from L1 transcripts required for ORF1p and ORF2p translation, leading to decreased L1 retrotransposition (10,12). Active silencing responses to retrotransposons in mammalian cells include inhibition by methylation, RNAi and APOBEC3.

DNA Methylation—Mammalian cytosine methylation occurs predominantly in transposons, being the primary means of respression for mammalian transposons. Methyltransferase deficiencies activate retroelements in both mammalian (198) and plant systems (90). CpG-rich sequences in the 5′ UTR of L1 (but not Alu) are methylated and the L1 promoter is repressed by methyl-CpG-binding protein (MeCP2), which inihibits L1 mobility in cells in culture (207). Repression occurs in somatic cells, and to a lesser extent in cells that are hypomethylated, such as germ-line cells and in somatic human tumors (17,160). Furthermore, L1s are transcriptionally activated in germ cells of mouse knockout mutants lacking Dnmt3 methylase family proteins, further implicating methylation as a regulatory force in L1 expression (24, 102).

RNAi—RNA *i*nterference (RNAi) is widely viewed as a defense against foreign genetic elements such as viruses and transposable elements. RNAi is the process of post-transcriptional gene silencing in which double-stranded RNA is processed by the RNase III homolog Dicer to generate *s*hort *i*nterfering RNAs (siRNAs) (reviewed in 35). The resulting siRNAs are then incorporated into a complex called the *R*NAi-*i*nduced *s*ilencing *c*omplex (RISC), which guides the endonucleolytic cleavage of corresponding mRNAs (reviewed in 30). Recently, RNAi has also been invoked as a silencer of mammalian retrotransposons. Human L1s contain internal sense and antisense promoters in the 5′ UTR (184,189) and the L1 bidirectional transcripts can be processed to siRNAs that in turn suppress retrotransposition (182,203). Moreover, the L1 ORF1p colocalizes with the RISC complex in stress granules, suggesting that silencing by RNAi may be invoked under particular cellular circumstances (76).

DNA methylation and RNAi are in independent silencing pathways that can combine to form a powerful and redundant mechanism for keeping retrotransposons in check. PIWI is part of the Argonaute family of proteins, which are only expressed in germ cells and are key players in RNA silencing (93). Recently, the mouse PIWI homologues MILI and MIWI2 have been

implicated in repression of IAP and L1 retrotransposons, with methylation of the L1 5′ UTR being reduced in newborn mice defective in these proteins (1,34). Interestingly, impaired *de novo* methylation was observed in these cell lines, seemingly as a result of reduced *P*IWI *i*nteracting RNA (piRNA) expression (111). Furthermore, L1 expression was enhanced in the MILI- and MIWI2-null testes, which correlated with a reduction in DNA methylation in these mutant male germ cells.

The APOBEC3 family of cytidine deaminases—The APOBEC3 proteins were discovered to be a defensive network of proteins that restrict the replication of retroviruses (18,41). They have since been shown to be potent selective inhibitors of specific TP and EP retrotransposons. Indeed, it is likely that the APOBEC3 family evolved to maintain the integrity of genomes against endogenous retrotransposons, and that their effectiveness as antivirals is a fortuitous outcome of their attack on the insidious threat of endogenous retroelements.

Human APOBEC3 family members APOBEC3A, 3B, 3F, and to a lesser extent 3C, inhibit L1 retrotransposition. Cytidine deaminase activity is not required for retrotransposon silencing, and it is hypothesized that the APOBEC3 family of enzymes mediates cytoplasmic sequestration of L1 RNA and/or encoded proteins, or directly inhibits the activity of L1 ORFs (20,21,39,143,185). Interestingly, APOBEC3G inhibits Alu, but not L1 retrotransposition. This intriguing observation is thought to result from Alu RNA sequestration in cytoplasmic complexes that are inaccessible to the L1 retrotransposition machinery (42,92).

Ty and Tf retrotransposons

The only characterized mechanism of RNA-dependent gene silencing in *S. cerevisiae* is the recently described transcriptional regulation of Ty1 by an antisense noncoding RNA transcript, Ty1-RTL (16). Ty1-RTL is approximately 400 bp long, initiating within the *gag* ORF and continuing into the 5′ LTR. Synthesis of the Ty1-RTL is independent of that of the Ty1 sense transcript. Interestingly, the integrity of Ty1-RTL is required for silencing, indicating that the RNA may interact directly with the Ty1 promoter, although apparently through an RNAi-independent mechanism, since *S. cerevisiae* lacks RNAi pathways.

Recently, a mechanism of genome surveillance was described for retrotransposons in *S. pombe*. This pathway involves recruitment of CENP-B homologs, Abp1, Cbh1, and Cbh2, which have roles in formation of centromeric heterochromatin and chromosome segregation (96,146). These CENP-B homologs bind to Tf2 LTRs and also solo LTRs, and recruit histone deacetylases Clr3 and Clr6 that result in Tf2 silencing. The CENP-B proteins also organize retrotransposons into higher-order structures called Tf bodies (32). CENP-B surveillance also displays a type of immunity by blocking recombination, as exogenously provided Tf1 is prevented from integrating into the genome. The overall silencing mechanism could result from the recruitment of histone deacetylases by CENP-B proteins, resulting in a closed chromatin structure that could repress Tf2 transcription and prevent recombination. This observation is intriguing as CENP-B proteins themselves are derived from transposase originating from other transposable elements (106) such that the host fights fire with fire, using a derivative of one transposable element to regulate another (196). This surveillance system thus provides a fine example of the opportunistic interplay of the retrotransposon and its host.

GLOBAL AND ENVIRONMENTAL INFLUENCES

Group II introns

The nucleoid—Bacterial genomes and associated proteins are condensed into a structure called the nucleoid (159). The *E. coli* nucleoid was implicated in maintaining group II intron retrotransposition frequency, which is sharply reduced by deletions of the genes encoding

nucleoid proteins H-NS (*hns*) and StpA (*stpA*) (8). Although H-NS and StpA also regulate global gene expression, their affect is presumed to be nucleoid-specific, because they do not perturb retromobility levels into plasmids, which are excluded from the nucleoid (181). Although, there is an integration bias of retrotransposition in *E. coli* towards the origin (Ori) and terminal (Ter) macrodomains of the chromosome (47,211), this bias persists despite disruption of the nucleoid (8). Rather, bipolar localization may be attributed to the IEP being consistently occluded from the nucleoid, and located in the cytoplasmic spaces that are associated with the cellular poles (8,209). Additionally, some small molecules that influence retromobility levels may act at the level of the nucleoid, as described below.

Small molecules: polyphosphates, (p)ppGpp and cAMP—Recent work has identified five genes in *E. coli*, *gppA*, *uhpT*, *wcaK*, *ynbC*, and *zntR*, whose disruption leads to a more diffuse cellular distribution of the IEP, accompanied by a more uniform distribution of intron integrations around the genome (210). The common factor in these mutants is accumulation of intracellular polyphosphate, which was also shown to bind to LtrA and to alter distribution of other pole-localized basic proteins. Polyphosphates accumulate when cells undergo nutritional stress, or enter stationary phase, and their accumulation relies on high levels of (p)ppGpp (110,156). Particularly interesting is the relationship to ppGpp, which is elevated in a "stringent response" to amino acid starvation, as is retromobility (48). Conversely, specific mutations in *relA* and *spoT*, with reduced levels of ppGpp, are deficient in retrohoming, whereas suppressor mutations *rpoB** and *rpoC** in RNA polymerase (6) restore intron movement. Suppression suggests that the effect of ppGpp is mediated via RNA polymerase. Although not all ppGpp mutants depress retromobility, possibly due to the different host strains and assay systems used, the above studies provide a satisfying correspondence of regulatory effects mediated via phosphate metabolism and suggest that nutritional stress may be an activator of group II intron mobility (48,210).

Similarly, the transcription regulator cAMP, in complex with its receptor protein CRP, is activated by changes in the nutrient environment, particularly carbon source starvation. Group II inton mobility is depressed in a *cya* mutant, and restored when cAMP is provided exogenously (48). It is unclear in either the ppGpp or cAMP case if transcriptional regulation of the intron is direct or indirect. Examples of indirect targets might again be the nucleoid (8), or stress-induced error-prone polymerases (67), which enhance group II intron mobility as previously described (181). Regardless, with both cAMP and ppGpp signaling, rather than shutting down to preserve cellular energy stores, the intron can prepare itself for movement to other, perhaps safer genomic havens.

LINEs and SINEs

The global response of LINEs and SINEs to cellular stress, such as DNA-damaging agents and environmental carcinogens is of particular importance, given the prevalence of these elements in our genomes and that unchecked retrotransposition can lead to genomic instability and genetic disorders (11,104). Selected examples of cellular stress are exposure to UV light and ionizing radiation leading to increased expression of L1 in cultured rat cells (175) and also human L1 activation by benzo(*a*)pyrene, a polycyclic aromatic hydrocarbon carcinogen (186). Additionally, LINEs and SINEs, such as Alu, are upregulated following treatment of DNA-damaging agents such as cisplatin, which creates DNA intrastrand crosslinks, etoposide, a topoisomerase II inhibitor, and γ-radiation, correlating with an increase in cellular reverse transcriptase levels that may be post-transcriptional (11,162). Overall, these observations indicate that environmental stress increases the activity of normally silent LINEs and SINEs, thereby adversely affecting the health of mammals, including humans.

Ty and Tf retrotransposons

Over the last two decades, numerous studies have demonstrated that Ty retrotransposon transcription and mobility are regulated by a variety of environmental conditions and stresses, such as exposure to mating pheromone, DNA damage agents and ionizing radiation, as well as temperature extremes and nutrient deprivation (reviewed in 122). In general, environmental signals regulate Ty elements at a transcriptional level, through cis-acting enhancer elements that activate or repress not only Ty1 transcription, but also the transcription of adjacent genes. The Tf2 retrotransposon of *S. pombe* is upregulated by low oxygen levels (172). This up-regulation of Tf2 targeting is initiated by the oxygen-dependent transcription factor Sre1 (193). By binding to the Tf2 LTR, Sre1 can directly activate Tf2 transcription and cDNA mobilization under low oxygen conditions. Moreover, Sre1 binding to Tf2 LTRs is responsible for the upregulation of neighboring genes (172). Hence, regulation of both these EP retrotransposons by environmental cues can increase diversity in the genome-wide transcriptional response to stress, by altering the expression of genes that reside in their vicinity. Consequently, the target specificity of EP-retrotransposons has important consequences for the ability of cells to respond to stress; not surprisingly, target-site specificity is highly regulated by the host cell (**Sidebar 1**).

Importantly, Ty5 target specificity was shown to be environmentally regulated by stress. The targeting domain (TD) of Ty5 integrase interacts with silent information regulator protein, Sir4, to tether the integration complex to the target, by an interesting example of molecular mimicry (**Sidebar 2**). Phosphorylation of the TD is required for interaction with Sir4, to confer site selectivity to heterochromatin, while the absence of phosphorylation causes Ty5 to integrate more randomly throughout the genome. TD phosphorylation is reduced during nutritional stress, which suggests Ty5 target specificity changes in response to nutrient availability as an adaptive response to environmental challenge (56). The response of retrotransposons to varying stresses is important in that these stresses increase both the frequency of retrotransposition and change integration patterns. These responses may be beneficial to both the retrotransposon and the host: the element proliferates, causing genome rearrangements that lead to changes in gene expression that could benefit the host in times of stress.

OVERVIEW

Retrotransposon trash? or treasure?

There has been a decades-long debate on whether transposons are selfish, parasitic intruders or altruistic guests, providing benefit to the host. Increasingly, it is being concluded that although the retrotransposons' ability to invade and spread provides their *raison d'être*, they can be exploited by the host and adapt to the host's needs (11,17,145). We call this the "lemonade-from-lemons" hypothesis. The lemonade-from-lemons hypothesis recognizes that retrotransposons can be both genomic trash and organismal treasure. The initial impairment to the host could provide the drive for co-evolution of the two entitites to their mutual benefit. Others have named retrotransposons "handy junk" (145). These authors argue that handy junk can evolve into "necessary junk," namely, elements that have become essential for organismal development.

What is the impact, negative or positive, of the heavy retrotransposon load in our genomes? We will confine our examples to mammalian LINEs and SINEs and initially consider how these elements can undermine their host. Although there are about a half-million L1s in the human genome, only about 6,000 are intact, and of these ~100 are functional, with varying degrees of activity (28). The active elements have the potential to wreak havoc on a genome in myriad ways: by inducing DSBs that can lead to misrepair or cell arrest and apoptosis (11, 17,72); by stimulating genome rearrangements via homologous recombination between non-

allelic repeats (17,59,83); and finally, by L1 integration and L1-mediated insertion of non-autonomous elements (eg. Alus) (61). The literature is replete with examples of the pathologies caused by these elements, including genetic diseases that result from retrotransposition events in the germ-line where L1s are most active, as well as cancers resulting from rare somatic events (5,11,103). The human disease phenotypes, of which there are already >50, range from blood disorders through cystic fibrosis to cancers.

The positive impact L1s and Alu sequences can have on their host goes beyond providing a source of genetic diversity and generating for evolving novel exons (208). Indeed, the exploitation of transposons as regulatory elements has become so commonplace as to have prompted the borrowing of a word from evolutionists describing the coopting phenomenon, “exaptation” (78). Exaptation of retrotransposons can be divided broadly into regulation of gene expression, developmental programming and telomere generation: First, it has been suggested that Alu sequences, which are present in >5% of human 3′-UTRs, may be a target for miRNA silencing, thereby providing a global orchestration of gene expression (145,180). Conversely, Alu elements contain many binding sites for transcription factors, and may therefore play a role in positive regulation and development (154). At the level of individual genes, Alu elements, with their internal polymerase III promoters, can regulate expression of their neighboring genes (85). Alus may play a role in translation as well (86,161). For even greater variety, Alu RNAs have recently been shown to act in trans as transcriptional represssors during the heat shock response in human cells (127).

Second, a role for L1s in epigenetic gene regulation and development may be emerging (145). Engineered L1s have been shown to have a preference to retrotranspose vicinal to neuronal genes to generate somatic mosaicism in rats and mice in neuronal precursor cells during differentiation (144). Such events influence neuronal cell fate in vitro. The transcription factor Sox2, which binds the L1 promoter, likely mediates this effect. Sox2 is plentiful and represses L1 transcription in neural stem cells, but is downregulated in differentiating cells allowing the L1 promoter to be turned on (33). Furthermore, SINEs have been recently implicated as enhancers of gene expression of the developing forebrain of the mouse (167), suggesting that both LINEs and SINEs may have critical roles in development of the mammalian neuronal network.

Finally, evidence is mounting in favor of modern telomerases, the RNP enzymes that synthesize telomere repeats at the ends of chromosomes, having evolved from retroelements (reviewed by 53). The integrity of chromosome ends is arguably the ultimate in utility, and the relationship between retrotransposons and telomerase is described below.

The end(s): retrotransposons and telomeres

Eukaryotic chromosomes terminate in telomeres, short head-to-tail arrays of repeat sequences, which compensate for the inability of DNA polymerase to completely replicate the ends of DNA. Telomeres are usually maintained by telomerase, an RNP that includes an RT and an RNA molecule which is reverse transcribed on the end of the chromosome (19,80). A broad range of data, from fruit-flies to mammals, suggest that telomeres may have their origins in TP retrotransposons.

The first evidence of the retrotransposon-telomere relationship is from the *HeT-A* and *TART* TP retrotransposons of *Drosophila*, which are dedicated to the maintenance of telomeres by repeated transposition to the chromosome ends. *TART* encodes two proteins, Gag and RT, while *HeT-A*, like Alu, does not encode its own RT (149). *Drosophila* chromosomes terminate in a mixed head-to-tail array of *HeT-A* and *TART* elements. After RNA is transcribed from *HeT-A* and *TART* arrays it moves to the cytoplasm where it is translated, forms an RNP that

moves back to the nucleus where it associates with chromosome ends and acts as a template for reverse transcription and subsequent telomere elongation (149).

With such an intercompartmental life cycle, it is not surprising that *HeT-A* and *TART*, like its TP retrotransposon relatives, have interactions with host factors. Of particular interest here is that the proteins that prevent telomere fusion and promote terminal gene conversion are repair proteins, some of which are analogous to those that are involved in L1 retrotransposition in fish and mammalian cells. Thus, ATM kinase, Mre11 3′−5′ exonuclease, Rad50, which processes DSBs, and the Ku complex, which promotes terminal gene conversion, affect the frequency of *HeT-A/TART* addition to telomeres (135,155). Interestingly, some proteins in the RNAi pathway also regulate *HeT-A/TART* in telomere elongation in the female germline (169). Specifically, *aubergine*, a member of the Argonaute protein family necessary for RISC assembly enhances telomeric retrotransposition.

The classical telomerase RNP of eukaryotes consists of an RT and an RNA encoded by a different gene. Not only are the RTs of telomerase related to retrotranposon RTs by sequence (3,123), but also a nuclease-deficient subset of PLEs, a phylogenetically diverse family of eukaryotic retrotransposons, were discovered in bdelloid rotifers and other eukaryotes at the tips of chromosomes (75). The connection to L1s is two-fold. First, in addition to the RT ORF, the terminal PLEs have a second ORF which resembles the chaperone-encoding ORF1 of L1. Second, it was recently demonstrated in mutant hamster cell lines, in which the protective nucleoprotein cap at telomeres was dysfunctional, and NHEJ was disabled, that nuclease-deficient L1s retrotranspose adjacent to telomeric repeats (142). These findings link nuclease-independent retrotransposition to telomeres, suggesting that TP retrotransposons helped prevent chromosome-end erosion at the very dawn of eukaryotic evolution.

Retro- and prospectives

From the foregoing sweep of TP and EP retrotransposons in hosts as diverse as microbes and man, it is clear that what these elements have in common is that by and large they encode enzymes that promote the initial steps of their movement, and that for later steps they borrow host machinery. The machinery that they commandeer is the very survival apparatus of the host: equipment for transcription, translation, replication, recombination and repair. What better way to ensure their own survival? The variety of ways in which the retrotransposons exploit these processes is startling, and we have only just begun looking!

The host retaliates to the retrotransposon onslaught with an armamentarium of silencing weaponry, to suppress retrotransposon transcription by promoter methylation, to chop up the mRNA by the RNAi pathway, and to sequester the transcript with APOBEC3. Whereas group II introns are self-silencing via splicing, the EP and other TP transposons use a mix of all three abovementioned silencing mechanisms, suggesting that the surveillance strategies may have developed against one element, evolved, and then were re-deployed against another. Interestingly, some organisms have shied away from specific forms of silencing: *S. cerevisiae* has no RNAi to silence Ty elements, and there has not yet been any description of histone deacetylases silencing LINEs and SINEs, despite the recruitment of these enzymes against retrotransposon proliferation in yeasts and plants.

A striking observation in several systems is the “host effect”, whereby the same element can have altered properties and can be subsumed in alternate pathways in different hosts. For example, the very same group II intron from *L. lactis* not only uses different retrotransposition pathways in its native host than in *E. coli*, but it does so orders of magnitude more efficiently. Another difference is that retrotransposition events in *L. lactis* are scattered around the chromosome, wherease, in *E. coli*, there is an integration bias for the origin (Ori) and terminal (Ter) macrodomains of the chromosome (47). Additionally, completion of the retrohoming

pathways in bacteria is recombinase independent, whereas a major pathway in yeast mitochondria is characterized by homologous recombination between donor and recipient (52,64,131,137). Likewise, L1s have a different fate in humans from chimpanzees, with there being a much higher fraction of active elements in humans, in which there is considerable individual diversity (173). Furthermore, although Alus inundate primate genomes, they are absent from mice, and their distribution in related primates is different, suggesting that exposure of the host to different environmental influences helps determine these patterns. Finally, among *Sacharomyces sensu stricto* species, the variation in copy number of Ty elements generally correlates with the geographic distribution of the species (124). These are but a few examples that illustrate how the hosts and environment fashion the fate of their invaders.

We have proposed the lemonade-from-lemons hypothesis for how these invasive, selfish retrotransposons coevolve with the host to develop useful functions: a range of up- and down-regulation of gene expression, developmental programming and telomere evolution. Perhaps the sweetest lemonade is provided by the CENP-B protein, a centromere protein evolved from a transposase to silence Tf2, a different transposable element!

Where might future studies of retrotransposons lead us? Almost certainly to more lemons and more lemondade: more examples of debilitating retrotransposon-mediated diseases, and more examples of spectacular retrotransposon-associated exaptations. But our mechanistic insights are still in their infancy: we need more functional genomic screens, particularly with the mammalian elements, and better in vitro retrotransposition assays, to determine precisely where and how host factors interact with the molecular machinery, to facilitate or block the retrotransposon life cycle.

Retrotransposons may eventually provide a handy toolbox for genetic manipulation. Group II introns have already been useful for gene targeting and making gene knockouts in bacterial cells, and they hold promise for targeted gene therapy in vertebrates, including mammals (114). Retroviral integrases too are being developed as gene-targeting agents (31), so one can easily imagine similar utility for the integrases of EP retrotransposons. Also, given the universal response of these elements to stress, might sensors for environmental and genotoxic agents be developed in vertebrate cells, including human cell lines? Whatever the future might hold, we need to better understand the hate-love relationship between retrotransposons and their hosts.

Sidebar 1

Retrotransposon integration preferences and the host

An important mechanism by which the host minimizes the deleterious consequences and capitalizes on the adaptive potential of retrotransposons is by directing integration into nonessential domains of the genome. In general, retrotransposons tend to integrate into specific chromatin domains, or macrodomains, of chromosomes where their presence is less harmful. For example, Ty1 integrates preferentially into a ~750-bp window upstream of tRNA genes, which are gene-poor regions of the *S. cerevisiae* genome. Although the mechanism of tethering integrase to this domain is not known, chromatin plays a role. As another example of integration preference, Ty3 is targeted to a site 1 to 2 bp upstream of tRNA genes, where it does not interfere with the internal RNA Pol III promoter (206), through an interaction of Ty3 with the RNA Pol III transcription factor TFIIIB. Although Tf1 preferentially inserts into RNA Pol II promoters via an interaction with the transcriptional activator, Atf1, disrupting the native promoter, Tf1 provides an alternative promoter ensuring the maintenance of gene expression (121).

Sidebar 2

Targeting and molecular mimicry

Ty5 is targeted to heterochromatic regions where expression is silenced. The targeting of Ty5 to heterochromatin is mediated by an interaction between the targeting domain (TD) of the retrotransposon-encoded integrase and the silent information regulator protein, Sir4, which interacts with a multitude of proteins and provides a molecular scaffold at the inner nuclear periphery (73). The retrotransposon targets heterochromatin by emulating the interaction between Sir4 and the inner nuclear membrane associated protein, Esc1 (25), which is important for assembly of the nuclear pore complex and tethers telomeres to the nuclear periphery. A functionally equivalent motif in TD and Esc1 was identified as being responsible for targeting integration on one hand and partitioning DNA on the other. Thus, retrotransposons exploit molecular mimicry to interweave themselves into cellular functions.

Acknowledgements

We thank Irina Arkhipova, Gary Dunny, Kenji Ichiyanagi and John Moran for ideas and critical reading of the manuscript, and the many authors who provided their manuscripts before publication. We are grateful to Maryellen Carl and John Dansereau for help with the manuscript and figures, respectively. Work in our laboratories is supported by NIH grants GM39422, GM44844, GM52072 and AI65301.

MINI-GLOSSARY

Exaptation, exploitation of transposons for use as regulatory elements of the host; Maturase, protein that stabilizes catalytically active RNA structure; Retrohoming, movement of group II intron to an intronless allele; Retrotransposition, movement of retrotransposon to chromosomal site, usually ectopic; Retrotransposon, mobile genetic element that is transferred via an RNA intermediate; Reverse transcriptase (RT), enzyme that transcribes complementary strand DNA (cDNA) using an RNA template; Reverse transcription, synthesis of cDNA from RNA template; Ribozyme, catalytic RNA; Target-primed reverse transcription (TPRT), integration mechanism that utilizes the 3′ hydroxyl group exposed during cleavage of a DNA strand as a primer for reverse transcription.

ABBREVIATIONS LIST

cAMP, cyclic adenosine monophosphate
DSB, double-strand break
EP retrotransposon, extrachromosomally-primed retrotransposon (also LTR retrotransposon)
LINE, long-interspersed nuclear element
LTR, long-terminal repeat
NHEJ, non-homologous end joining
ORF, open reading frame
ppGpp, guanosine tetraphosphate
RT, reverse transcriptase
SINE, short-interspersed nuclear element
TP retrotransposon, target-primed retrotransposon (also non-LTR retrotransposon)
TPRT, target-primed reverse transcription

Literature Cited

1. Aravin AA, Sachidanandam R, Girard A, Fejes-Toth K, Hannon GJ. Developmentally regulated piRNA clusters implicate MILI in transposon control. Science 2007;316:744–7. [PubMed: 17446352]
2. Arkhipova I, Meselson M. Transposable elements in sexual and ancient asexual taxa. Proc Natl Acad Sci U S A 2000;97:14473–7. [PubMed: 11121049]

3. Arkhipova IR, Pyatkov KI, Meselson M, Evgen'ev MB. Retroelements containing introns in diverse invertebrate taxa. Nat Genet 2003;33:123–4. [PubMed: 12524543]
4. Aye M, Irwin B, Beliakove-Bethell N, Chen E, Garrus J, Sandmeyer S. Host factors that affect Ty3 retrotransposition in Saccharomyces cerevisiae. Genetics 2004;168:1159–76. [PubMed: 15579677]
5. Babushok DV, Kazazian HH Jr. Progress in understanding the biology of the human mutagen LINE-1. Hum Mutat 2007;28:527–39. [PubMed: 17309057]
6. Bartlett MS, Gaal T, Ross W, Gourse RL. RNA polymerase mutants that destabilize RNA polymerase-promoter complexes alter NTP-sensing by rrn P1 promoters. J Mol Biol 1998;279:331–45. [PubMed: 9642041]
7. Batzer MA, Deininger PL. Alu repeats and human genomic diversity. Nat Rev Genet 2002;3:370–9. [PubMed: 11988762]
8. Beauregard A, Chalamcharla VR, Piazza CL, Belfort M, Coros CJ. Bipolar localization of the group II intron Ll.LtrB is maintained in Escherichia coli deficient in nucleoid condensation, chromosome partitioning and DNA replication. Mol Microbiol 2006;62:709–22. [PubMed: 17005014]
9. Becker KG, Swergold GD, Ozato K, Thayer RE. Binding of the ubiquitous nuclear transcription factor YY1 to a cis regulatory sequence in the human LINE-1 transposable element. Hum Mol Genet 1993;2:1697–702. [PubMed: 8268924]
10. Belancio VP, Hedges DJ, Deininger P. LINE-1 RNA splicing and influences on mammalian gene expression. Nucleic Acids Res 2006;34:1512–21. [PubMed: 16554555]
11. Belancio VP, Hedges DJ, Deininger P. Mammalian non-LTR retrotransposons: for better or worse, in sickness and in health. Genome Res 2008;18:343–58. [PubMed: 18256243]
12. Belancio VP, Roy-Engel AM, Deininger P. The impact of multiple splice sites in human L1 elements. Gene 2008;411:38–45. [PubMed: 18261861]
13. Belfort, M.; Derbyshire, V.; Parker, MM.; Cousineau, B.; Lambowitz, AM. Mobile introns: pathways and proteins.. In: Craig, N.; Craigie, R.; Gellert, M.; Lambowitz, A., editors. Mobile DNA II. ASM Press; 2002. p. 761-83.
14. Belhocine K, Yam KK, Cousineau B. Conjugative transfer of the Lactococcus lactis chromosomal sex factor promotes dissemination of the Ll.LtrB group II intron. J Bacteriol 2005;187:930–9. [PubMed: 15659671]
15. Beliakova-Bethell N, Beckham C, Giddings TH Jr. Winey M, Parker R, Sandmeyer S. Virus-like particles of the Ty3 retrotransposon assemble in association with P-body components. RNA 2006;12:94–101. [PubMed: 16373495]
16. Berretta J, Pinskaya M, Morillon A. A cryptic unstable transcript mediates transcriptional trans-silencing of the Ty1 retrotransposon in S. cerevisiae. Genes Dev 2008;22:615–26. [PubMed: 18316478]
17. Bestor TH. Cytosine methylation mediates sexual conflict. Trends Genet 2003;19:185–90. [PubMed: 12683970]
18. Bishop KN, holmes RK, Sheehy AM, Davidson NO, Cho S-J, Malim MH. Cytidine deamination of retroviral DNA by diverse APOBEC proteins. Curr Biol 2004;14:1392–6. [PubMed: 15296758]
19. Blackburn EH. Telomerases. Annu Rev Biochem 1992;61:113–29. [PubMed: 1497307]
20. Bogerd HP, Wiegand HL, Doehle BP, Lueders KK, Cullen BR. APOBEC3A and APOBEC3B are potent inhibitors of LTR-retrotransposon function in human cells. Nucleic Acids Res 2006;34:89–95. [PubMed: 16407327]
21. Bogerd HP, Wiegand HL, Hulme AE, Garcia-Perez JL, O'Shea KS, et al. Cellular inhibitors of long interspersed element 1 and Alu retrotransposition. Proc Natl Acad Sci U S A 2006;103:8780–5. [PubMed: 16728505]
22. Bonen L. Cis- and trans-splicing of group II introns in plant mitochondria. Mitochondrion 2008;8:26–34. [PubMed: 18006386]
23. Bonen L, Vogel J. The ins and outs of group II introns. Trends Genet 2001;17:322–31. [PubMed: 11377794]
24. Bourc'his D, Bestor TH. Meiotic catastrophe and retrotransposon reactivation in male germ cells lacking Dnmt3L. Nature 2004;431:96–9. [PubMed: 15318244]

25. Brady TL, Fuerst PG, Dick RA, Schmidt C, Voytas DF. Retrotransposon target site selection by imitation of a cellular protein. Mol Cell Biol 2008;28:1230–9. [PubMed: 18086891]
26. Branciforte D, Martin SL. Developmental and cell type specificity of LINE-1 expression in mouse testis: implications for transposition. Mol Cell Biol 1994;14:2584–92. [PubMed: 8139560]
27. Brass AL, Dykxhoorn DM, Benita Y, Yan N, Engelman A, et al. Identification of host proteins required for HIV infection through a functional genomic screen. Science 2008;319:921–6. [PubMed: 18187620]
28. Brouha B, Schustak J, Badge RM, Lutz-Prigge S, Farley AH, et al. Hot L1s account for the bulk of retrotransposition in the human population. Proc Natl Acad Sci U S A 2003;100:5280–5. [PubMed: 12682288]
29. Bryk M, Banerjee M, Conte D Jr. Curcio MJ. The Sgs1 helicase of Saccharomyces cerevisiae inhibits retrotransposition of Ty1 multimeric arrays. Mol Cell Biol 2001;21:5374–88. [PubMed: 11463820]
30. Buchon N, Vaury C. RNAi: a defensive RNA-silencing against viruses and transposable elements. Heredity 2006;96:195–202. [PubMed: 16369574]
31. Bushman FD. Integration site selection by lentiviruses: biology and possible control. Curr Top Microbiol Immunol 2002;261:165–77. [PubMed: 11892246]
32. Cam HP, Noma K, Ebina H, Levin HL, Grewal SI. Host genome surveillance for retrotransposons by transposon-derived proteins. Nature 2008;451:431–6. [PubMed: 18094683]
33. Cao X, Yeo G, Muotri AR, Kuwabara T, Gage FH. Noncoding RNAs in the mammalian central nervous system. Annu Rev Neurosci 2006;29:77–103. [PubMed: 16776580]
34. Carmell MA, Girard A, van de Kant HJ, Bourc'his D, Bestor TH, et al. MIWI2 is essential for spermatogenesis and repression of transposons in the mouse male germline. Dev Cell 2007;12:503–14. [PubMed: 17395546]
35. Carmell MA, Hannon GJ. RNase III enzymes and the initiation of gene silencing. Nat Struct Mol Biol 2004;11:214–8. [PubMed: 14983173]
36. Carpousis AJ. The RNA degradosome of Escherichia coli: an mRNA-degrading machine assembled on RNase E. Annu Rev Microbiol 2007;61:71–87. [PubMed: 17447862]
37. Cavalier-Smith T. Intron phylogeny: a new hypothesis. Trends Genet 1991;7:145–8. [PubMed: 2068786]
38. Chapman KB, Boeke JD. Isolation and characterization of the gene encoding yeast debranching enzyme. Cell 1991;65:483–92. [PubMed: 1850323]
39. Chen H, Lilley CE, Yu Q, Lee DV, Chou J, et al. APOBEC3A is a potent inhibitor of adeno-associated virus and retrotransposons. Curr Biol 2006;16:480–5. [PubMed: 16527742]
40. Cheng Z, Menees TM. RNA branching and debranching in the yeast retrovirus-like element Ty1. Science 2004;303:240–3. [PubMed: 14716018]
41. Chiu YL, Greene WC. The APOBEC3 Cytidine Deaminases: An innate defensive network opposing exogenous retroviruses and endogenous retroelements. Annu Rev Immunol 2007;26:317–53. [PubMed: 18304004]
42. Chiu YL, Witkowska HE, Hall SC, Santiago M, Soros VB, et al. High-molecular-mass APOBEC3G complexes restrict Alu retrotransposition. Proc Natl Acad Sci U S A 2006;103:15588–93. [PubMed: 17030807]
43. Coller J, Parker R. Eukaryotic mRNA decapping. Annu Rev Biochem 2004;73:861–90. [PubMed: 15189161]
44. Conlan LH, Stanger MJ, Ichiyanagi K, Belfort M. Localization, mobility and fidelity of retrotransposed group II introns in rRNA genes. Nucleic Acids Res 2005;33:5262–70. [PubMed: 16170154]
45. Coombes CE, Boeke JD. An evaluation of detection methods for large lariat RNAs. RNA 2005;11:323–31. [PubMed: 15661842]
46. Copertino DW, Hallick RB. Group II and group III introns of twintrons: potential relationships to nuclear pre-mRNA introns. Trends Biochem Sci 1993;18:467–71. [PubMed: 8108859]
47. Coros CJ, Landthaler M, Piazza CL, Beauregard A, Esposito D, et al. Retrotransposition strategies of the Lactococcus lactis Ll.LtrB group II intron are dictated by host identity and cellular environment. Mol Microbiol 2005;56:509–24. [PubMed: 15813740]

48. Coros CJ, Piazza CL, Smith D, Chalamcharla VR, Belfort M. A mutant screen implicates global regulators as modulators of group II intron retrohoming into the Escherichia coli chromosome. Submitted
49. Cost GJ, Boeke JD. Targeting of human retrotransposon integration is directed by the specificity of the L1 endonuclease for regions of unusual DNA structure. Biochemistry 1998;37:18081–93. [PubMed: 9922177]
50. Cost GJ, Feng Q, Jacquier A, Boeke JD. Human L1 element target-primed reverse transcription in vitro. EMBO J 2002;21:5899–910. [PubMed: 12411507]
51. Cousineau B, Lawrence S, Smith D, Belfort M. Retrotransposition of a bacterial group II intron. Nature 2000;404:1018–21. [PubMed: 10801134][see comment][erratum appears in *Nature* 2001 Nov 414(6859):84]Correction (2001) *Nature* 414, 84
52. Cousineau B, Smith D, Lawrence-Cavanagh S, Mueller JE, Yang J, et al. Retrohoming of a bacterial group II intron: mobility via complete reverse splicing, independent of homologous DNA recombination. Cell 1998;94:451–62. [PubMed: 9727488]
53. Curcio MJ, Belfort M. The beginning of the end: Links between ancient retroelements and modern telomerases. Proc Nat Acad Sci U S A 2007;104:9107–8.
54. Curcio MJ, Derbyshire KM. The outs and ins of transposition: from mu to kangaroo. Nat Rev Mol Cell Biol 2003;4:865–77.
55. Curcio MJ, Kenny AE, Moore S, Garfinkel DJ, Weintraub M, et al. S-phase checkpoint pathways stimulate the mobility of the retrovirus-like transposon Ty1. Mol Cell Biol 2007;27:8874–85. [PubMed: 17923678]
56. Dai J, Xie W, Brady TL, Gao J, Voytas DF. Phosphorylation regulates integration of the yeast Ty5 retrotransposon into heterochromatin. Mol Cell 2007;27:289–99. [PubMed: 17643377]
57. Dai L, Toor N, Olson R, Keeping A, Zimmerly S. Database for mobile group II introns. Nucleic Acids Res 2003;31:424–6. [PubMed: 12520040]
58. Dai L, Zimmerly S. ORF-less and reverse-transcriptase-encoding group II introns in archaebacteria, with a pattern of homing into related group II intron ORFs. RNA 2003;9:14–9. [PubMed: 12554871]
59. Deininger PL, Moran JV, Batzer MA, Kazazian HH Jr. Mobile elements and mammalian genome evolution. Curr Opin Genetics Dev 2003;13:651–8.
60. Del Campo M, Tijerina P, Bhaskaran H, Mohr S, Yang Q, et al. Do DEAD-box proteins promote group II intron splicing without unwinding RNA? Mol Cell 2007;28:159–66. [PubMed: 17936712]
61. Dewannieux M, Esnault C, Heidmann T. LINE-mediated retrotransposition of marked Alu sequences. Nat Genet 2003;35:41–8. [PubMed: 12897783]
62. Eickbush, TH. R2 and related site-specific non-long terminal repeat retrotransposons.. In: Craig, NL.; Craigie, R.; Gellert, M.; Lambowitz, AM., editors. Mobile DNA II. ASM Press; Washington, DC: 2002. p. 813-35.
63. Eickbush TH, Jamburuthugoda VK. The diversity of retrotransposons and the properties of their reverse transcriptases. Virus Res 2008;134:221–34. [PubMed: 18261821]
64. Eskes R, Liu L, Ma H, Chao MY, Dickson L, et al. Multiple homing pathways used by yeast mitochondrial group II introns. Mol Cell Biol 2000;20:8432–46. [PubMed: 11046140]
65. Evgen'ev MB, Arkhipova IR. Penelope-like elements--a new class of retroelements: distribution, function and possible evolutionary significance. Cytogenet Genome Res 2005;110:510–21. [PubMed: 16093704]
66. Feng Q, Moran JV, Kazazian HH Jr. Boeke JD. Human L1 retrotransposon encodes a conserved endonuclease required for retrotransposition. Cell 1996;87:905–16. [PubMed: 8945517]
67. Foster PL. Stress-induced mutagenesis in bacteria. Crit Rev Biochem Mol Biol 2007;42:373–97. [PubMed: 17917873]
68. Fuerst PG, Voytas DF. CEN plasmid segregation is destabilized by tethered determinants of Ty 5 integration specificity: a role for double-strand breaks in CEN antagonism. Chromosoma 2003;112:58–65. [PubMed: 12883945]
69. Gao J, Lee K, Zhao M, Qiu J, Zhan X, et al. Differential modulation of E. coli mRNA abundance by inhibitory proteins that alter the composition of the degradosome. Mol Microbiol 2006;61:394–406. [PubMed: 16771842]

70. Garfinkel DJ, Stefanisko KM, Nyswaner KM, Moore SP, Oh J, Hughes SH. Retrotransposon suicide: formation of Ty1 circles and autointegration via a central DNA flap. J Virol 2006;80:11920–34. [PubMed: 17005648]
71. Gasior SL, Roy-Engel AM, Deininger PL. ERCC1/XPF limits L1 retrotransposition. DNA Repair (Amst). 2008In press
72. Gasior SL, Wakeman TP, Xu B, Deininger PL. The human LINE-1 retrotransposon creates DNA double-strand breaks. J Mol Biol 2006;357:1383–93. [PubMed: 16490214]
73. Gasser SM, Cockell MM. The molecular biology of the SIR proteins. Gene 2001;279:1–16. [PubMed: 11722841]
74. Gilbert N, Lutz S, Morrish TA, Moran JV. Multiple fates of L1 retrotransposition intermediates in cultured human cells. Mol Cell Biol 2005;25:7780–95. [PubMed: 16107723]
75. Gladyshev EA, Arkhipova IR. Telomere-associated, endonuclese-deficient Penelope-like retroelements in diverse eukaryotes. Proc Natl Acad Sci U S A 2007;104:9352–7. [PubMed: 17483479]
76. Goodier JL, Zhang L, Vetter MR, Kazazian HH Jr. LINE-1 ORF1 protein localizes in stress granules with other RNA-binding proteins, including components of RNA interference RNA-induced silencing complex. Mol Cell Biol 2007;27:6469–83. [PubMed: 17562864]
77. Goodwin TJ, Poulter RT. The DIRS1 group of retrotransposons. Mol Biol Evol 2001;18:2067–82. [PubMed: 11606703]
78. Gould SJ, Vrba ES. Exaptation - a missing term in the science of form. Paleobiology 1982;8:4–15.
79. Gregan J, Kolisek M, Schweyen RJ. Mitochondrial Mg(2+) homeostasis is critical for group II intron splicing in vivo. Genes Dev 2001;15:2229–37. [PubMed: 11544180]
80. Greider CW. Telomere length regulation. Annu Rev Biochem 1996;65:337–65. [PubMed: 8811183]
81. Griffith JL, Coleman LE, Raymond AS, Goodson SG, Pittard WS, et al. Functional genomics reveals relationships between the retrovirus-like Ty1 element and its host Saccharomyces cerevisiae. Genetics 2003;164:867–79. [PubMed: 12871900]
82. Grivell LA. Transposition: mobile introns get into line. Curr Biol 1996;6:48–51. [PubMed: 8805219]
83. Han JS, Boeke JD. LINE-1 retrotransposons: modulators of quantity and quality of mammalian gene expression? Bioessays 2005;27:775–84. [PubMed: 16015595]
84. Han JS, Szak ST, Boeke JD. Transcriptional disruption by the L1 retrotransposon and implications for mammalian transcriptomes. Nature 2004;429:268–74. [PubMed: 15152245]
85. Hasler J, Strub K. Alu elements as regulators of gene expression. Nucleic Acids Res 2006;34:5491–7. [PubMed: 17020921]
86. Hasler J, Strub K. Alu RNP and Alu RNA regulate translation initiation in vitro. Nucleic Acids Res 2006;34:2374–85. [PubMed: 16682445]
87. Heidmann O, Heidmann T. Retrotranscription of a mouse IAP sequence tagged with an indicator gene. Cell 1991;64:159–70. [PubMed: 1846087]
88. Heyman T, Agoutin B, Friant S, Wilhelm FX, Wilhelm ML. Plus-strand DNA synthesis of the yeast retrotransposon Ty1 is initiated at two sites, PPT1 next to the 3' LTR and PPT2 within the pol gene. PPT1 is sufficient for Ty1 transposition. J Mol Biol 1995;253:291–303. [PubMed: 7563090]
89. Heyman T, Wilhelm M, Wilhelm FX. The central PPT of the yeast retrotransposon Ty1 is not essential for transposition. J Mol Biol 2003;331:315–20. [PubMed: 12888340]
90. Hirochika H, Okamoto H, Kakutani T. Silencing of retrotransposons in arabidopsis and reactivation by the ddm1 mutation. Plant Cell 2000;12:357–69. [PubMed: 10715322]
91. Huang HR, Rowe CE, Mohr S, Jiang Y, Lambowitz AM, Perlman PS. The splicing of yeast mitochondrial group I and group II introns requires a DEAD-box protein with RNA chaperone function. Proc Natl Acad Sci U S A 2005;102:163–8. [PubMed: 15618406]
92. Hulme AE, Bogerd HP, Cullen BR, Moran JV. Selective inhibition of Alu retrotransposition by APOBEC3G. Gene 2007;390:199–205. [PubMed: 17079095]
93. Hutvagner G, Simard MJ. Argonaute proteins: key players in RNA silencing. Nat Rev Mol Cell Biol 2008;9:22–32. [PubMed: 18073770]
94. Ichiyanagi K, Beauregard A, Belfort M. A bacterial group II intron favors retrotransposition into plasmid targets. Proc Natl Acad Sci U S A 2003;100:15742–7. [PubMed: 14673083]

95. Ichiyanagi K, Beauregard A, Lawrence S, Smith D, Cousineau B, Belfort M. Multiple pathways for the Ll.LtrB group II intron include reverse splicing into DNA targets. Mol Microbiol 2002;46:1259–71. [PubMed: 12453213]
96. Irelan JT, Gutkin GI, Clarke L. Functional redundancies, distinct localizations and interactions among three fission yeast homologs of centromere protein-B. Genetics 2001;157:1191–203. [PubMed: 11238404]
97. Irwin B, Aye M, Baldi P, Beliakova-Bethell N, Cheng H, et al. Retroviruses and yeast retrotransposons use overlapping sets of host genes. Genome Res 2005;15:641–54. [PubMed: 15837808]
98. Irwin EF, Ho JE, Kane SR, Healy KE. Analysis of interpenetrating polymer networks via quartz crystal microbalance with dissipation monitoring. Langmuir 2005;21:5529–36. [PubMed: 15924485]
99. Jacquier A. Self-splicing Group II and nuclear pre-mRNA introns: how similar are they? Trends Biochem Sci 1990;15:351–4. [PubMed: 2238045]
100. Jurka J, Kapitonov VV, Kohany O, Jurka MV. Repetitive sequences in complex genomes: structure and evolution. Annu Rev Genomics Hum Genet 2007;8:241–59. [PubMed: 17506661]
101. Karst SM, Rutz ML, Menees TM. The yeast retrotransposons Ty1 and Ty3 require the RNA Lariat debranching enzyme, Dbr1p, for efficient accumulation of reverse transcripts. Biochem Biophys Res Commun 2000;268:112–7. [PubMed: 10652222]
102. Kato Y, Kaneda M, Hata K, Kumaki K, Hisano M, et al. Role of the Dnmt3 family in de novo methylation of imprinted and repetitive sequences during male germ cell development in the mouse. Hum Mol Genet 2007;16:2272–80. [PubMed: 17616512]
103. Kazazian HH Jr. Mobile elements: drivers of genome evolution. Science 2004;303:1626–32. [PubMed: 15016989]
104. Kazazian HH, Moran JV. The impact of L1 retrotransposons on the human genome. Nat Genet 1998;19:19–24. [PubMed: 9590283]
105. Kim FJ, Battini JL, Manel N, Sitbon M. Emergence of vertebrate retroviruses and envelope capture. Virology 2004;318:183–91. [PubMed: 14972546]
106. Kipling D, Warburton PE. Centromeres, CENP-B and Tigger too. Trends Genet 1997;13:141–5. [PubMed: 9097724]
107. Klein JR, Dunny GM. Bacterial group II introns and their association with mobile genetic elements. Front Biosci 2002;7:d1843–56. [PubMed: 12133822]
108. Kornberg, A.; Baker, TA. DNA Replication. W.H. Freeman and Company; New York: 1992. p. 931
109. Kubo S, Seleme MC, Soifer HS, Perez JL, Moran JV, et al. L1 retrotransposition in nondividing and primary human somatic cells. Proc Natl Acad Sci U S A 2006;103:8036–41. [PubMed: 16698926]
110. Kulaev I, Kulakovskaya T. Polyphosphate and phosphate pump. Annu Rev Microbiol 2000;54:709–34. [PubMed: 11018142]
111. Kuramochi-Miyagawa S, Watanabe T, Gotoh K, Totoki Y, Toyoda A, et al. DNA methylation of retrotransposon genes is regulated by Piwi family members MILI and MIWI2 in murine fetal testes. Genes Dev 2008;22:908–17. [PubMed: 18381894]
112. Kurzynska-Kokorniak A, Jamburuthugoda VK, Bibillo A, Eickbush TH. DNA-directed DNA polymerase and strand displacement activity of the reverse transcriptase encoded by the R2 retrotransposon. J Mol Biol 2007;374:322–33. [PubMed: 17936300]
113. Lambowitz, AM.; Caprara, MG.; Zimmerly, S.; Perlman, PS. The RNA World, Second Edition. Cold Spring Harbor Laboratory Press; Cold Spring Harbor, NY: 1999. Group I and group II ribozymes as RNPs: clues to the past and guides to the future.; p. 451-85.
114. Lambowitz, AM.; Mohr, G.; Zimmerely, S. Group II intron homing endonucleases: ribonucleoprotein complexes with programmable target specificity.. In: Belfort, M.; Derbyshire, V.; Stoddard, BL.; Wood, DW., editors. Homing Endonucleases and Inteins. Springer-Verlag; 2005. p. 121-45.
115. Lambowitz AM, Zimmerly S. Mobile group II introns. Ann Rev Genet 2004;38:1–35. [PubMed: 15568970]
116. Lander ES, Linton LM, Birren B, Nusbaum C, Zody MC, et al. Initial sequencing and analysis of the human genome. Nature 2001;409:860–921. [PubMed: 11237011]

117. Larsen LS, Beliakova-Bethell N, Bilanchone V, Zhang M, Lamsa A, et al. Ty3 nucleocapsid controls localization of particle assembly. J Virol 2008;82:2501–14. [PubMed: 18094177]
118. Lee BS, Bi L, Garfinkel DJ, Bailis AM. Nucleotide excision repair/TFIIH helicases RAD3 and SSL2 inhibit short-sequence recombination and Ty1 retrotransposition by similar mechanisms. Mol Cell Biol 2000;20:2436–45. [PubMed: 10713167]
119. Lee BS, Lichtenstein CP, Faiola B, Rinckel LA, Wysock W, et al. Posttranslational inhibition of Ty1 retrotransposition by nucleotide excision repair/transcription factor TFIIH subunits Ssl2p and Rad3p. Genetics 1998;148:1743–61. [PubMed: 9560391]
120. Lee K, Zhan X, Gao J, Qiu J, Feng Y, et al. RraA. a protein inhibitor of RNase E activity that globally modulates RNA abundance in E. coli. Cell 2003;114:623–34. [PubMed: 13678585]
121. Leem YE, Ripmaster TL, Kelly FD, Ebina H, Heincelman ME, et al. Retrotransposon Tf1 is targeted to Pol II promoters by transcription activators. Mol Cell 2008;30:98–107. [PubMed: 18406330]
122. Lesage P, Todeschini AL. Happy together: the life and times of Ty retrotransposons and their hosts. Cytogenet Genome Res 2005;110:70–90. [PubMed: 16093660]
123. Lingner J, Hughes TR, Shevchenko A, Mann M, Lundblad V, Cech TR. Reverse transcriptase motifs in the catalytic subunit of telomerase. Science 1997;276:561–7. [PubMed: 9110970]
124. Liti G, Peruffo A, James SA, Roberts IN, Louis EJ. Inferences of evolutionary relationships from a population survey of LTR-retrotransposons and telomeric-associated sequences in the Saccharomyces sensu stricto complex. Yeast 2005;22:177–92. [PubMed: 15704235]
125. Luan DD, Korman MH, Jakubczak JL, Eickbush TH. Reverse transcription of R2Bm RNA is primed by a nick at the chromosomal target site: a mechanism for non-LTR retrotransposition. Cell 1993;72:595–605. [PubMed: 7679954]
126. Malik HS, Eickbush TH. Phylogenetic analysis of ribonuclease H domains suggest a late, chimeric origin of LTR retrotransposable elements and retroviruses. Genome Res 2001;11:1187–97. [PubMed: 11435400]
127. Mariner PD, Walters RD, Espinoza CA, Drullinger LF, Wagner SD, et al. Human Alu RNA is a modular transacting repressor of mRNA transcription during heat shock. Mol Cell 2008;29:499–509. [PubMed: 18313387]
128. Martin SL. The ORF1 protein encoded by LINE-1: structure and function during L1 retrotransposition. J Biomed Biotech 2006;2006:1–6.
129. Martinez-Abarca F, Barrientos-Duran A, Fernandez-Lopez M, Toro N. The RmInt1 group II intron has two different retrohoming pathways for mobility using predominantly the nascent lagging strand at DNA replication forks for priming. Nucleic Acids Res 2004;32:2880–8. [PubMed: 15155857]
130. Martínez-Abarca F, Garcia-Rodriguez FM, Toro N. Homing of a bacterial group II intron with an intron-encoded protein lacking a recognizable endonuclease domain. Mol Microbiol 2000;35:1405–12. [PubMed: 10760141]
131. Martínez-Abarca F, Toro N. RecA-independent ectopic transposition *in vivo* of a bacterial group II intron. Nucleic Acids Res 2000;28:4397–402. [PubMed: 11058141]
132. Martinez-Abarca F, Zekri S, Toro N. Characterization and splicing in vivo of a Sinorhizobium meliloti group II intron associated with particular insertion sequences of the IS630-Tc1/IS3 retroposon superfamily. Mol Microbiol 1998;28:1295–306. [PubMed: 9680217]
133. Mathias SL, Scott AF, Kazazian HH Jr. Boeke JD, Gabriel A. Reverse transcriptase encoded by a human transposable element. Science 1991;254:1808–10. [PubMed: 1722352]
134. Maxwell PH, Curcio MJ. Host factors that control long terminal repeat retrotransposons in Saccharomyces cerevisiae: implications for regulation of mammalian retroviruses. Eukaryot Cell 2007;6:1069–80. [PubMed: 17496126]
135. Melnikova L, Biessmann H, Georgiev P. The Ku protein complex is involved in length regulation of Drosophila telomeres. Genetics 2005;170:221–35. [PubMed: 15781709]
136. Michel F, Ferat J-L. Structure and activities of group II introns. Ann Rev Biochem 1995;64:435–61. [PubMed: 7574489]
137. Mills DA, Manias DA, McKay LL, Dunny GM. Homing of a group II intron from Lactococcus lactis subsp. *lactis* ML3. J Bacteriol 1997;179:6107–11. [PubMed: 9324259]
138. Mills DA, McKay LL, Dunny GM. Splicing of a group II intron involved in the conjugative transfer of pRS01 in lactococci. J Bacteriol 1996;178:3531–8. [PubMed: 8655550]

139. Mohr G, Del Campo M, Mohr S, Yang Q, Jia H, et al. Function of the C-terminal domain of the DEAD-box protein Mss116p analyzed in vivo and in vitro. J Mol Biol 2008;375:1344–64. [PubMed: 18096186]
140. Moran, JV.; Gilbert, N. Mammalian LINE-1 retrotransposons and related elements.. In: Craig, NL.; Craigie, R.; Gellert, M.; Lambowitz, AM., editors. Mobile DNA II. ASM Press; Washington, DC: 2002. p. 836-69.
141. Moran JV, Holmes SE, Naas TP, DeBerardinis RJ, Boeke JD, Kazazian HH Jr. High frequency retrotransposition in cultured mammalian cells. Cell 1996;87:917–27. [PubMed: 8945518]
142. Morrish TA, Garcia-Perez JL, Stamato TD, Taccioli GE, Sekiguchi J, Moran JV. Endonuclease-independent LINE-1 retrotransposition at mammalian telomeres. Nature 2007;446:208–12. [PubMed: 17344853]
143. Muckenfuss H, Hamdorf M, Held U, Perkovic M, Lower J, et al. APOBEC3 proteins inhibit human LINE-1 retrotransposition. J Biol Chem 2006;281:22161–72. [PubMed: 16735504]
144. Muotri AR, Chu VT, Marchetto MC, Deng W, Moran JV, Gage FH. Somatic mosaicism in neuronal precursor cells mediated by L1 retrotransposition. Nature 2005;435:903–10. [PubMed: 15959507]
145. Muotri AR, Marchetto MC, Coufal NG, Gage FH. The necessary junk: new functions for transposable elements. Hum Mol Genet 2007;16:R159–67. [PubMed: 17911158]Spec No. 2
146. Nakagawa H, Lee JK, Hurwitz J, Allshire RC, Nakayama J, et al. Fission yeast CENP-B homologs nucleate centromeric heterochromatin by promoting heterochromatin-specific histone tail modifications. Genes Dev 2002;16:1766–78. [PubMed: 12130537]
147. Nyswaner KM, Checkley MA, Yi M, Stephens RM, Garfinkel DJ. Chromatin-associated genes protect the yeast genome from Ty1 insertional mutagenesis. Genetics 2008;178:197–214. [PubMed: 18202368]
148. Ohshima K, Okada N. SINEs and LINEs: symbionts of eukaryotic genomes with a common tail. Cytogenet Genome Res 2005;110:475–90. [PubMed: 16093701]
149. Pardue ML, DeBaryshe PG. Retrotransposons provide an evolutionarily robust non-telomerase mechanism to maintain telomeres. Annu Rev Genet 2003;37:485–511. [PubMed: 14616071]
150. Perepelitsa-Belancio V, Deininger P. RNA truncation by premature polyadenylation attenuates human mobile element activity. Nat Genet 2003;35:363–6. [PubMed: 14625551]
151. Piskareva O, Schmatchenko V. DNA polymerization by the reverse transcriptase of the human L1 retrotransposon on its own template in vitro. FEBS Lett 2006;580:661–8. [PubMed: 16412437]
152. Plosky BS, Woodgate R. Switching from high-fidelity replicases to low-fidelity lesion-bypass polymerases. Curr Opin Genet Devel 2004;14:113–9. [PubMed: 15196456]
153. Pochart P, Agoutin B, Rousset S, Chanet R, Doroszkiewicz V, Heyman T. Biochemical and electron microscope analyses of the DNA reverse transcripts present in the virus-like particles of the yeast transposon Ty1. Identification of a second origin of Ty1DNA plus strand synthesis. Nucleic Acids Res 1993;21:3513–20. [PubMed: 8393990]
154. Polak P, Domany E. Alu elements contain many binding sites for transcription factors and may play a role in regulation of developmental processes. BMC Genomics 2006;7:133. [PubMed: 16740159]
155. Purdy A, Su TT. Telomeres: not all breaks are equal. Curr Biol 2004;14:R613–4. [PubMed: 15296775]
156. Rao NN, Liu S, Kornberg A. Inorganic polyphosphate in Escherichia coli: the phosphate regulon and the stringent response. J Bacteriol 1998;180:2186–93. [PubMed: 9555903]
157. Rattray AJ, Shafer BK, Garfinkel DJ. The Saccharomyces cerevisiae DNA recombination and repair functions of the RAD52 epistasis group inhibit Ty1 transposition. Genetics 2000;154:543–56. [PubMed: 10655210]
158. Ribet D, Harper F, Dupressoir A, Dewannieux M, Pierron G, Heidmann T. An infectious progenitor for the murine IAP retrotransposon: emergence of an intracellular genetic parasite from an ancient retrovirus. Genome Res 2008;18:597–609. [PubMed: 18256233]
159. Robinow C, Kellenberger E. The bacterial nucleoid revisited. Microbiol Reviews 1994;58:211–32.
160. Roman-Gomez J, Jimenez-Velasco A, Agirre X, Cervantes F, Sanchez J, et al. Promoter hypomethylation of the LINE-1 retrotransposable elements activates sense/antisense transcription and marks the progression of chronic myeloid leukemia. Oncogene 2005;24:7213–23. [PubMed: 16170379]

161. Rubin CM, Kimura RH, Schmid CW. Selective stimulation of translational expression by Alu RNA. Nucleic Acids Res 2002;30:3253–61. [PubMed: 12136107]
162. Rudin CM, Thompson CB. Transcriptional activation of short interspersed elements by DNA-damaging agents. Genes Chromosomes Cancer 2001;30:64–71. [PubMed: 11107177]
163. Saint-Ruf C, Pesut J, Sopta M, Matic I. Causes and consequences of DNA repair activity modulation during stationary phase in Escherichia coli. Crit Rev Biochem Mol Biol 2007;42:259–70. [PubMed: 17687668]
164. Sak BD, Eisenstark A, Touati D. Exonuclease III and the catalase hydroperoxidase II in Escherichia coli are both regulated by the katF gene product. Proc Natl Acad Sci U S A 1989;86:3271–5. [PubMed: 2541439]
165. Saldanha R, Chen B, Wank H, Matsuura M, Edwards J, Lambowitz AM. RNA and protein catalysis in group II intron splicing and mobility reactions using purified components. Biochemistry 1999;38:9069–83. [PubMed: 10413481]
166. Salem LA, Boucher CL, Menees TM. Relationship between RNA lariat debranching and Ty1 element retrotransposition. J Virol 2003;77:12795–806. [PubMed: 14610201]
167. Sasaki T, Nishihara H, Hirakawa M, Fujimura K, Tanaka M, et al. Possible involvement of SINEs in mammalian-specific brain formation. Proc Natl Acad Sci U S A 2008;105:4220–5. [PubMed: 18334644]
168. Sassaman DM, Dombroski BA, Moran JV, Kimberland ML, Naas TP, et al. Many human L1 elements are capable of retrotransposition. Nat Genet 1997;16:37–43. [PubMed: 9140393]
169. Savitsky M, Kwon D, Georgiev P, Kalmykova A, Gvozdev V. Telomere elongation is under the control of the RNAi-based mechanism in the Drosophila germline. Genes Dev 2006;20:345–54. [PubMed: 16452506]
170. Scholes DT, Banerjee M, Bowen B, Curcio MJ. Multiple regulators of Ty1 transposition in Saccharomyces cerevisiae have conserved roles in genome maintenance. Genetics 2001;159:1449–65. [PubMed: 11779788]
171. Scholes DT, Kenny AE, Gamache ER, Mou Z, Curcio MJ. Activation of a LTR-retrotransposon by telomere erosion. Proc Natl Acad Sci U S A 2003;100:15736–41. [PubMed: 14673098]
172. Sehgal A, Lee CY, Espenshade PJ. SREBP controls oxygen-dependent mobilization of retrotransposons in fission yeast. PLoS Genet 2007;3:e131. [PubMed: 17696611]
173. Seleme MC, Vetter MR, Cordaux R, Bastone L, Batzer MA, Kazazian HH Jr. Extensive individual variation in L1 retrotransposition capability contributes to human genetic diversity. Proc Natl Acad Sci U S A 2006;103:6611–6. [PubMed: 16618923]
174. Seraphin B, Simon M, Boulet A, Faye G. Mitochondrial splicing requires a protein from a novel helicase family. Nature 1989;337:84–7. [PubMed: 2535893]
175. Servomaa K, Rytomaa T. UV light and ionizing radiations cause programmed death of rat chloroleukaemia cells by inducing retropositions of a mobile DNA element (L1Rn). Int J Radiat Biol 1990;57:331–43. [PubMed: 1968498]
176. Sharon G, Burkett TJ, Garfinkel DJ. Efficient homologous recombination of Ty1 element cDNA when integration is blocked. Mol Cell Biol 1994;14:6540–51. [PubMed: 7523854]
177. Sharp PA. Five easy pieces. Science 1991;254:663. [PubMed: 1948046]
178. Shearman C, Godon J-J, Gasson M. Splicing of a group II intron in a functional transfer gene of Lactococcus lactis. Mol Microbiol 1996;21:45–53. [PubMed: 8843433]
179. Shiloh Y. ATM and related protein kinases: safeguarding genome integrity. Nat Rev Cancer 2003;3:155–68. [PubMed: 12612651]
180. Smalheiser NR, Torvik VI. Alu elements within human mRNAs are probable microRNA targets. Trends Genet 2006;22:532–6. [PubMed: 16914224]
181. Smith D, Zhong J, Matsuura M, Lambowitz AM, Belfort M. Recruitment of host functions suggests a repair pathway for late steps in group II intron retrohoming. Genes Dev 2005;19:2477–87. [PubMed: 16230535]
182. Soifer HS, Zaragoza A, Peyvan M, Behlke MA, Rossi JJ. A potential role for RNA interference in controlling the activity of the human LINE-1 retrotransposon. Nucleic Acids Res 2005;33:846–56. [PubMed: 15701756]

183. Solem A, Zingler N, Pyle AM. A DEAD protein that activates intron self-splicing without unwinding RNA. Mol Cell 2006;24:611–7. [PubMed: 17188036]

184. Speek M. Antisense promoter of human L1 retrotransposon drives transcription of adjacent cellular genes. Mol Cell Biol 2001;21:1973–85. [PubMed: 11238933]

185. Stenglein MD, Harris RS. APOBEC3B and APOBEC3F inhibit L1 retrotransposition by a DNA deamination-independent mechanism. J Biol Chem 2006;281:16837–41. [PubMed: 16648136]

186. Stribinskis V, Ramos KS. Activation of human long interspersed nuclear element 1 retrotransposition by benzo(a)pyrene, an ubiquitous environmental carcinogen. Cancer Res 2006;66:2616–20. [PubMed: 16510580]

187. Sundararajan A, Lee BS, Garfinkel DJ. The Rad27 (Fen-1) nuclease inhibits Ty1 mobility in Saccharomyces cerevisiae. Genetics 2003;163:55–67. [PubMed: 12586696]

188. Suzuki J, Kajikawa M, Yamaguchi K, Ichiyanagi K, Adachi N, et al. Genetic evidence that LINE retrotransposition depends on the nonhomologous end-joining repair pathway. submitted

189. Swergold GD. Identification, characterization, and cell specificity of a human LINE-1 promoter. Mol Cell Biol 1990;10:6718–29. [PubMed: 1701022]

190. Tchenio T, Casella JF, Heidmann T. Members of the SRY family regulate the human LINE retrotransposons. Nucleic Acids Res 2000;28:411–5. [PubMed: 10606637]

191. Tijerina P, Bhaskaran H, Russell R. Nonspecific binding to structured RNA and preferential unwinding of an exposed helix by the CYT-19 protein, a DEAD-box RNA chaperone. Proc Natl Acad Sci U S A 2006;103:16698–703. [PubMed: 17075070]

192. Tippin B, Pham P, Goodman MF. Error-prone replication for better or worse. Trends Microbiol 2004;12:288–95. [PubMed: 15165607]

193. Todd BL, Stewart EV, Burg JS, Hughes AL, Espenshade PJ. Sterol regulatory element binding protein is a principal regulator of anaerobic gene expression in fission yeast. Mol Cell Biol 2006;26:2817–31. [PubMed: 16537923]

194. Trelogan SA, Martin SL. Tightly regulated, developmentally specific expression of the first open reading frame from LINE-1 during mouse embryogenesis. Proc Natl Acad Sci U S A 1995;92:1520–4. [PubMed: 7878012]

195. Umezu K, Hiraoka M, Mori M, Maki H. Structural analysis of aberrant chromosomes that occur spontaneously in diploid Saccharomyces cerevisiae: retrotransposon Ty1 plays a crucial role in chromosomal rearrangements. Genetics 2002;160:97–110. [PubMed: 11805048]

196. Voytas DF. Genomics: fighting fire with fire. Nature 2008;451:412–3. [PubMed: 18216844]

197. Voytas, DF.; Boeke, JD. Ty1 and Ty5 of Saccharomyces cerevisiae.. In: Craig, NL.; Craigie, R.; Gellert, M.; Lambowitz, A., editors. Mobile DNA II. ASM Press; Washington, DC: 2002. p. 631-62.

198. Walsh CP, Chaillet JR, Bestor TH. Transcription of IAP endogenous retroviruses is constrained by cytosine methylation. Nat Genet 1998;20:116–7. [PubMed: 9771701]

199. Weiner AM. SINEs and LINEs: the art of biting the hand that feeds you. Curr Opin Cell Biol 2002;14:343–50. [PubMed: 12067657]

200. Weinstock KG, Mastrangelo MF, Burkett TJ, Garfinkel DJ, Strathern JN. Multimeric arrays of the yeast retrotransposon Ty. Mol Cell Biol 1990;10:2882–92. [PubMed: 2160587]

201. Wicker T, Sabot F, Hua-Van A, Bennetzen JL, Capy P, et al. A unified classification system for eukaryotic transposable elements. Nat Rev Genet 2007;8:973–82. [PubMed: 17984973]

202. Wiesenberger G, Waldherr M, Schweyen RJ. The Nuclear Gene MRS2 is Essential for the Excision of Group II Introns from Yeast Mitochondrial Transcripts *in vivo*. J Biol Chem 1992;267:6963–9. [PubMed: 1551905]

203. Yang N, Kazazian HH Jr. L1 retrotransposition is suppressed by endogenously encoded small interfering RNAs in human cultured cells. Nat Struct Mol Biol 2006;13:763–71. [PubMed: 16936727]

204. Yang N, Zhang L, Zhang Y, Kazazian HH Jr. An important role for RUNX3 in human L1 transcription and retrotransposition. Nucleic Acids Res 2003;31:4929–40. [PubMed: 12907736]

205. Ye Y, De Leon J, Yokoyama N, Naidu Y, Camerini D. DBR1 siRNA inhibition of HIV-1 replication. Retrovirology 2005;2:63. [PubMed: 16232320]

206. Yieh L, Kassavetis G, Geiduschek EP, Sandmeyer SB. The Brf and TATA-binding protein subunits of the RNA polymerase III transcription factor IIIB mediate position-specific integration of the gypsy-like element, Ty3. J Biol Chem 2000;275:29800–7. [PubMed: 10882723]

207. Yu F, Zingler N, Schumann G, Stratling WH. Methyl-CpG-binding protein 2 represses LINE-1 expression and retrotransposition but not Alu transcription. Nucleic Acids Res 2001;29:4493–501. [PubMed: 11691937]

208. Zhang XH, Chasin LA. Comparison of multiple vertebrate genomes reveals the birth and evolution of human exons. Proc Natl Acad Sci U S A 2006;103:13427–32. [PubMed: 16938881]

209. Zhao J, Lambowitz AM. Inaugural Article: A bacterial group II intron-encoded reverse transcriptase localizes to cellular poles. Proc Natl Acad Sci U S A 2005;102:16133–40. [PubMed: 16186487]

210. Zhao J, Niu W, Marcotte E, Lambowitz A. Group II intron protein localization and insertion sites are affected by polyphosphate. PLoS Biology. 2008In press

211. Zhong J, Karberg M, Lambowitz AM. Targeted and random bacterial gene disruption using a group II intron (targetron) vector containing a retrotransposition-activated selectable marker. Nucleic Acids Res 2003;31:1656–64. [PubMed: 12626707]

212. Zhong J, Lambowitz AM. Group II intron mobility using nascent strands at DNA replication forks to prime reverse transcription. EMBO J 2003;22:4555–65. [PubMed: 12941706]

213. Zhou L, Manias DA, Dunny GM. Regulation of intron function: efficient splicing in vivo of a bacterial group II intron requires a functional promoter within the intron. Mol Microbiol 2000;37:639–51. [PubMed: 10931357]

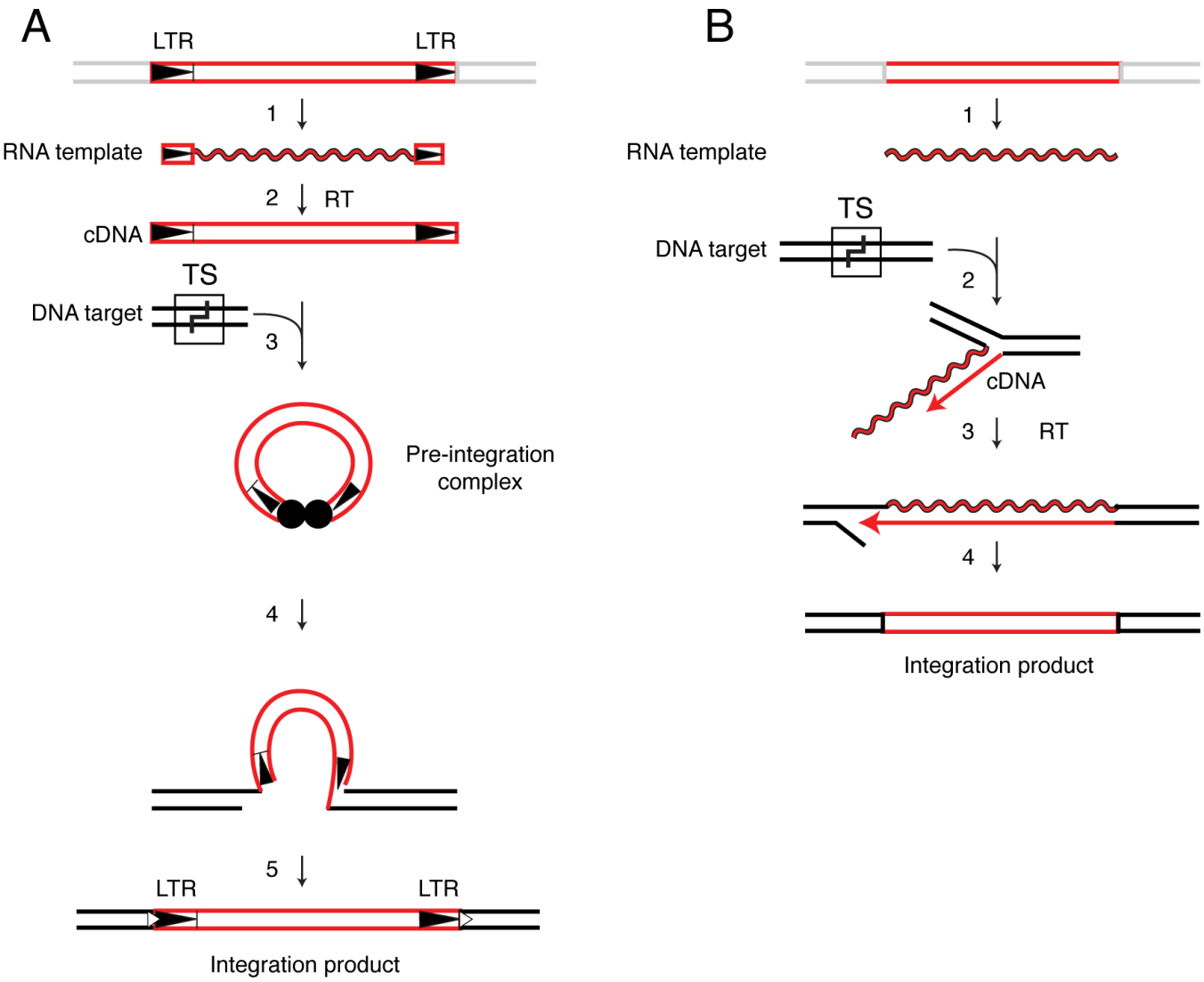

Figure 1.

Comparison of EP and TP retrotransposition pathways. The retrotransposon is colored red throughout; LTRs are represented by boxed, black arrowheads; gray lines represent donor chromosomes. The retrotransposon transcripts (A & B, step 1) are represented by red wavy lines, that are subsequently translated and reverse transcribed into cDNA. (A) Retrotransposition of EP retrotransposons. After cDNA synthesis (A, step 2) integrase or recombinase, represented by two black dots, allows *t*arget *s*ite (TS) access (A, step 3), in generating a TS duplication of uniform length (A, steps 4 and 5). (B) Retrotranspositon of TP retrotransposons. The first step of TPRT usually involves endonuclease cleavage of the first strand of the chromosomal DNA TS (B, step 2), exposing the 3′ hydroxyl that serves as the primer for reverse transcription (B, step 3). Late steps are conducted by host repair functions (B, step 4).

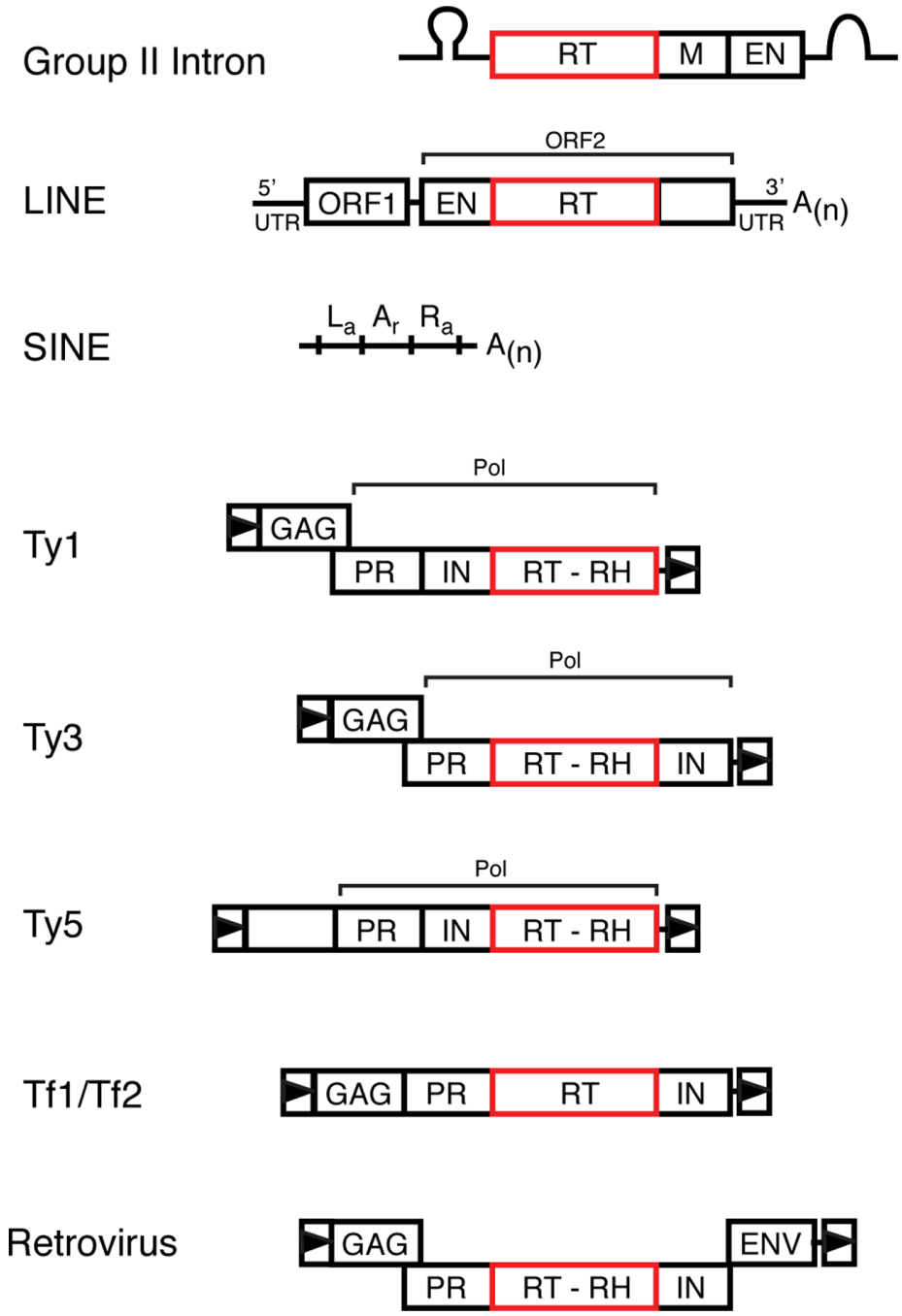

Figure 2.

Retrotransposon architecture. RNA maps of the retrotransposons described in this review are shown, with the reverse transcriptase (RT) sequence in red (not to scale). Rectangles represent protein-coding sequences. The stem-loops flanking the group II intron ORF represent the catalytic RNA. Coding sequences are as follows: M = maturase, EN = endonuclease, GAG = gag protein , PR = protease, IN = integrase, RH = ribonuclease H domain, Pol = polymerase domain, ENV = envelope protein. UTR = untranslated region, $A_{(n)}$ = poly(A) tail, L_a = left-arm region, A_r = adenosine-rich region, R_a = right-arm region, boxed triangles = LTRs. Some group II introns lact the EN domain (see text).

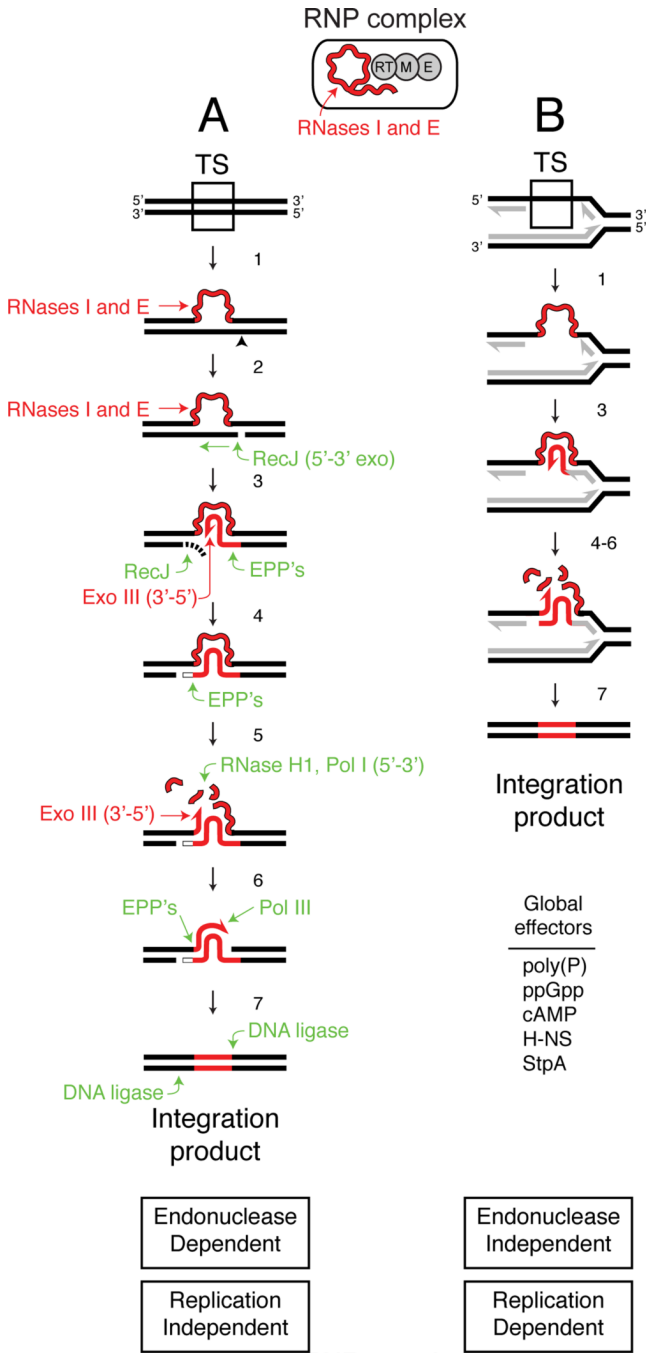

Figure 3.

Retromobility pathways of bacterial group II introns. (A) Endonuclease-dependent pathway via dsDNA. Reaction steps are as follows: (1) reverse splicing into DNA target, (2) bottom strand cleavage by IEP endonuclease, (3 and 4) cDNA synthesis by IEP RT, represented by solid red line (5) removal of intron RNA (6) second strand cDNA synthesis, and (7) ligation. Stimulatory (green) and inhibitory (red) host functions are superimposed on the schematic, with an arrow pointing to their putative site of action (adapted from 181). (B) Endonuclease-independent pathway via ssDNA at the replication fork. Newly replicated DNA is represented by a gray line. Reaction step numbering corresponds to (A). These pathways are for

retrohoming or retrotransposition in different intron/bacterial combinations as shown in the **sidebar table**. Global effectors that influence retromobility are listed in Table 1.

Sidebar to Figure 3 legend

Pathway	Intron	Host	Process
A	Ll.LtrB	*L. lactis*	Retrohoming
A	Ll.LtrB	*E. coli*	Retrotransposition
B	Ll.LtrB	*L. lactis*	Retrotransposition
B	RmInt1	*S. meliloti*	Retrohoming

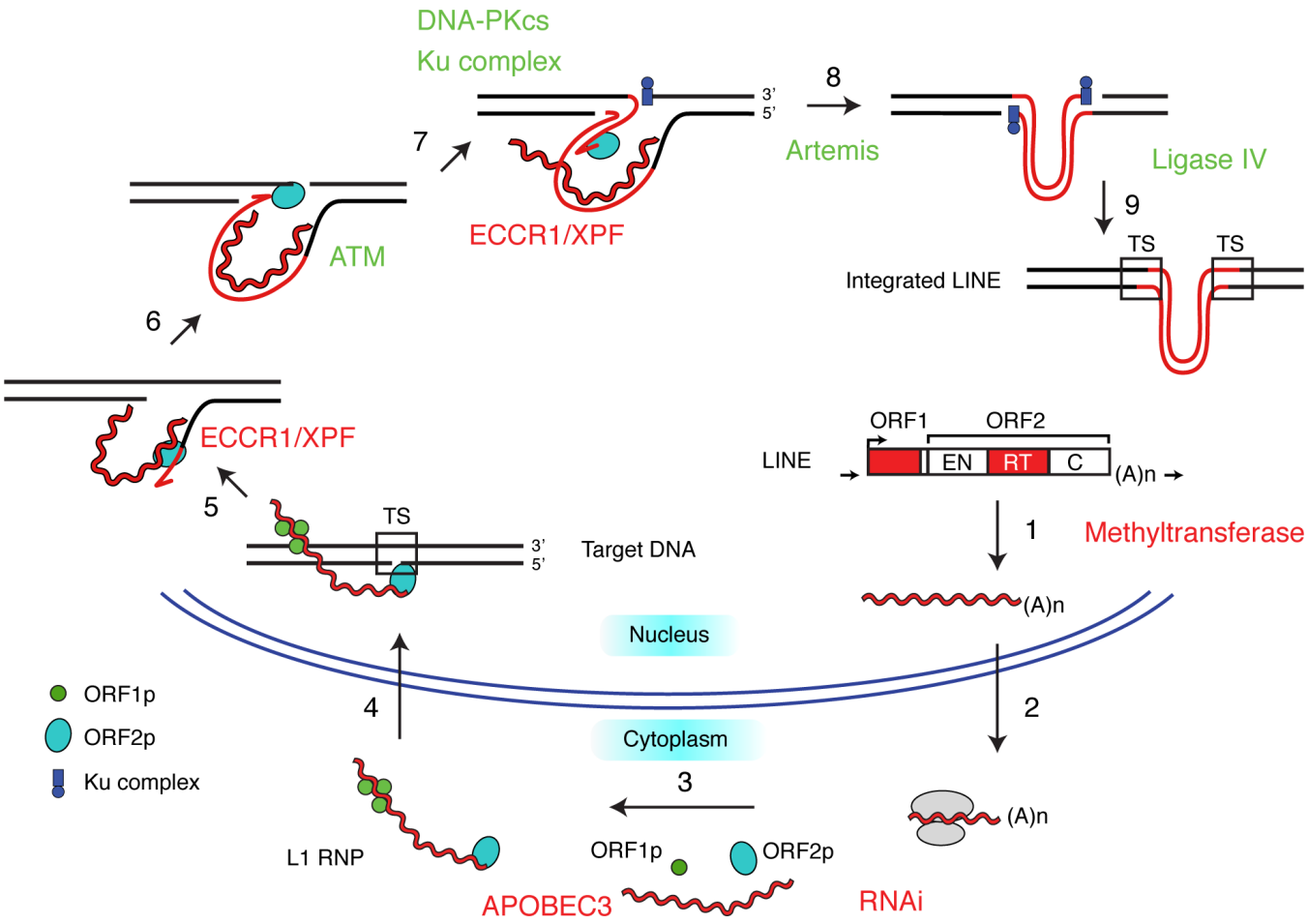

Figure 4.

L1 retrotransposition pathway. The L1 retrotransposon is transcribed in the nucleus from an internal Pol II promoter and the resulting full-length RNA is exported to the cytoplasm. ORF1 and ORF2 are translated and subsequently form an RNP which may form a higher order structure. The resulting RNP is transported into the nucleus where retrotransposition takes place by TPRT. Steps are as follows: (1) transcription, (2) export to cytoplasm and translation, (3) RNP formation, (4) first strand cleavage, (5) cDNA synthesis, (6) second-strand cleavage, (7) second-strand cDNA synthesis, (8) repair, (9) ligation. Host factors involved are indicated in green (stimulatory) or red (inhibitory) at their putative site of action.

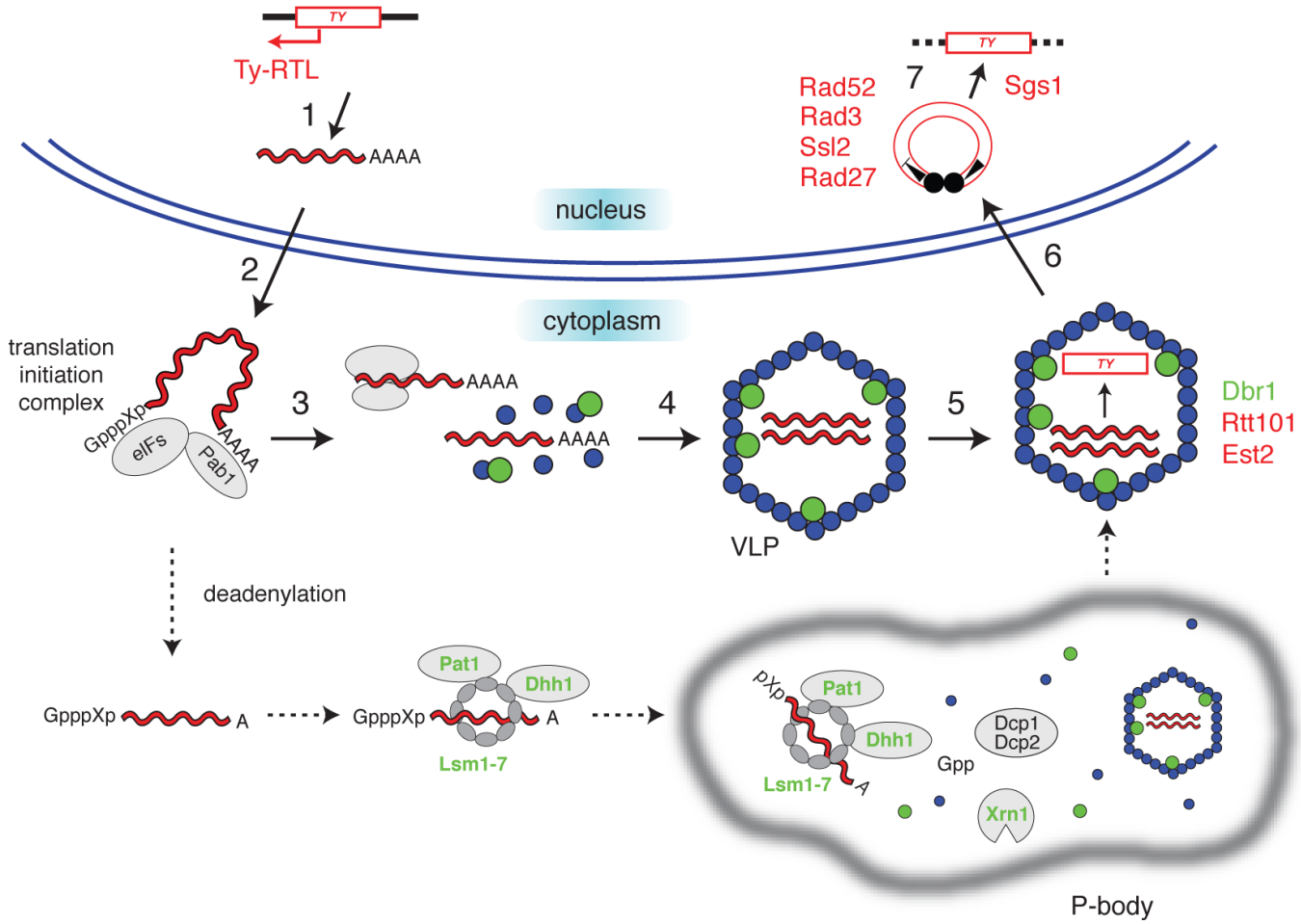

Figure 5.

Ty retrotransposition cycle. Wavy red lines represent retrotransposon RNA; the red rectangle is the DNA copy of the retrotransposon. Blue dots represent the Gag structural protein; blue and green shapes represent the Gag-Pol polyprotein. Hexagons represent the *v*irus-*l*ike *p*article (VLP). The stages in the pathway are as follows: (1) transcription of Ty element by RNA polymerase II, (2) transport into the cytoplasm (3) translation of retrotransposon mRNA, (4) VLP assembly and retrotransposon mRNA packaging, (5) reverse transcription, (6) import into the nucleus, and (7) integration of cDNA into the genome. Stimulatory (green) and inhibitory (red) host functions are represented near their putative sites of action. An enlargement of stages 3 and 4 is provided to illustrate the localization of Ty RNA and proteins in mRNA processing bodies (P body, grey enclosure). An active translation complex is indicated by binding of the GpppX cap of the mRNA to translation initiation factors (eIFs) and binding of the poly(A) tail to poly(A) binding protein, Pab1. Shortening of the poly(A) tail (deadenylation, dotted arrows) is the major mechanism of disassociation of mRNA from the translation initiation complex and binding by the decapping co-activators, Pat1, Dhh1 and Lsm1−7 (chain of grey ovals). These factors promote the localization of mRNA in P bodies, where mRNA decapping by Dcp1/Dcp2 and 5′−3′ degradation by Xrn1 occurs. VLP assembly may occur in P bodies.

Table 1

Host factor that affect group II intron retromobility in *E. coli*

Host Factor	Affect[a]	Identified Function	Putative effect on group II intron	Ref.
cAMP	S*	Global small-molecular regulator	Promotes retromobility frequency	(48)
Exo III	I	3′−5′ exonuclease	Degrade nascent cDNA	(181)
H-NS	S*	Nucleoid component, transcription regulator	Promotes retromobility frequency - global	(8)
Ligase	S	DNA ligase	Sealing in DNA nicks	(181)
MutD	S	3′−5′ exonuclease ε subunit of Pol III (*dnaQ*)	Repair second-strand cDNA synthesis	(181)
Pol I	S	5′−3′ exonuclease; removal of RNA primer from Okazaki fragments	Remove intron RNA template	(181)
Pol II	S	Repair polymerase (*polB*)	Repair polymerization across DNA-RNA junctions	(181)
Pol III	S	Replicative polymerase	Second-strand cDNA synthesis	(181)
Pol IV	S	Repair polymerase (*dinB*)	Repair polymerization	(181)
Pol V	S	Repair polymerase (*umuDC*)	Repair polymerization	(181)
poly(P)	S*	Global small-molecular regulator	Alters IEP localization and intron integration bias	(210)
ppGpp	S*	Global small-molecular regulator	Can promote retromobility frequency	(48)
RecJ	S	5′−3′ exonuclease	5′−3′ resection of DNA	(181)
RNase E	I	Ribonuclease; part of RNA degradosome	Reduce half-life of intron RNA	(181)
RNase H1	S	Ribonuclease; cleaves RNA strand in RNA/DNA hybrid	Remove intron RNA template	(181)
RNase I	I	Ribonuclease	Reduce half-life of intron RNA	(181)
StpA	S*	Nucleoid component, RNA chaperone	Promotes retromobility frequency - global	(8)

[a] S, Stimulates retromobility; I, Inhibits retromobility; S*, Stimulates retromobility into the chromosome only.

Table 2

Host factors that affect yeast retrotransposon mobility

Host factor	Affect[a]	Identified Function	Ty retrotransposon effect	Ref.
Abp1, Cbh1, Cbh2	I	Centromeric heterochromatin formation and chromosome segregation	Recruits Clr3 and Clr6 to silence Tf2	(32)
Atf1p	S	Transcription activator	Binds to Tf1 intergrase to mediate integration	(121)
Clr3, Clr6	I	Histone deacetylase	Silence Tf2	(32)
Dbr1	S	RNA debranching enzyme	Promotes Ty cDNA synthesis	(38,40)
Dhh1	S	Translational repressor	Post-transcriptional regulation	(15,98)
Est2	I	Catalytic subunit of telomerase	Telomere erosion leads to increased Ty1 mobility frequency	(171)
Lsm1−7	S	Decapping co-activator complex	Post-transcriptional regulation	(81)
Pat1	S	Translational repressor	Post-transcriptional regulation	(81,98)
Rad27	I	Fen1 homolog with 5′−3′ exo and 5′-flap endo activities	Prevents accumulation of cDNA and multimer formation	(187)
Rad3	I	Component of RNA pol II transcription factor, TFIIH	Prevents accumulation of cDNA	(118,119)
Rad52	I	Required for homologous recombination	Post-translational regulation of cDNA accumulation	(157)
Rtt101	I	Cullin component of E3-ubiquitin ligase	Inhibits cDNA accumulation	(55,98,170)
Sgs1	I	RecQ family helicase	Inhibits formation of multimeric Ty1 integration events	(29)
Sir4	S	Silent information regulator protein, provides molecular scaffold at the periphery	Interacts with the targeting domain of Ty5	(68)
Ssl2	I	Component of RNA pol II transcription factor, TFIIH	Prevents accumulation of cDNA and stability	(118,119)
TFIIIB	S	Transcription factor for RNA Pol III	Major determinants of Ty3 integration	(4,206)
XrnI	S	5'−3' exoribonuclease	Transcriptional and post-transcriptional regulation	(15,16,98)

[a] General affect on Ty retrotransposon mobility; S, Stimulatory; I, Inhibitory.