Transposable elements as drivers of reproductive isolation: A framework for testing hybridization-induced escalation of genetic conflicts

Fritjof Lammers [1*] & Valentina Peona[1,2*], Reto Burri [1]

[1] Swiss Ornithological Institute, Sempach, Switzerland

[2] Department of Bioinformatics and Genetics, Swedish Natural History Museum, Stockholm, Sweden

* contributed equally

Corresponding Author: Reto Burri (reto.burri@vogelwarte.ch)

Short running title: Escalation of genetic conflict upon hybridization

Keywords: genetic conflict, transposable elements, hybridization, DNA methylation, reproductive isolation

Abstract

Contrary to long-held views, the exchange of genetic diversity between species by hybridization is now recognized as an important process contributing to the evolution of biodiversity. However, hybridization has molecular consequences beyond the exchange of genetic variation. The clash of divergent genomes upon hybridization can escalate genetic conflicts previously resolved in parental species – notably between transposable elements (TEs) and their repressors – and unleash a cascade of molecular events inducing the evolution of chromosomal rearrangements (structural variants, SVs). Novel TE insertions and other SVs are often associated with cancer, reduced longevity, and genomic disorders, and usually inflict high fitness costs, thus they likely incur major constraints to gene flow among species and may constitute an integral component of reproductive isolation. Furthermore, in the presence of gene flow, certain SVs (e.g., inversions) may help protect beneficial combinations of alleles across genes from breaking up. By mediating an increased input of SVs, TE reactivation may therefore act as an architect of SVs underlying reproductive isolation. While the hypothesis of such TE-related reproductive barriers is not new, tests of its predictions – especially in the wild – remain scarce. With this perspective we aim at synthesizing ideas and providing a roadmap to stimulate research targeted towards an increased understanding of hybridization-induced TE reactivation and its potential knock-on effects for the evolution of structural variation and reproductive isolation. We argue that technological advances now provide unprecedented opportunities to identify genomic, transcriptomic and epigenomic signatures of such re-escalating genetic conflict upon hybridization and present a practical roadmap to guide researcher to investigate such genetic conflict in wild populations. While our perspective is centred predominantly on vertebrates, with adaptations to the taxon-specific TE biology, our roadmap represents a generalizable framework for the study of hybridization-induced escalations of TE-related genomic conflicts.

Lay summary

Different components of the genome can conflict with each other. One of the most prominent examples of such a genetic conflict is the proliferation of transposable elements (TEs) despite genomes' defence mechanisms. TEs are repetitive sequences that can replicate themselves – often with a fitness cost for the host genome. Therefore, multiple silencing mechanisms evolved to counteract the uncontrolled activity of TEs. While conflicts between active TEs and their repressors are usually at least partially resolved within species, they may be unmasked by hybridization, because recombination between parental genomes can uncouple active TEs from their repressors.

Based on previous reports of hybridization-induced TE reactivation by genome, transcriptome and methylation sequencing we here synthesize an integrative framework for testing for hybridization-induced TE-mediated genetic conflict in natural populations. To this end, we describe the molecular processes that form the foundation of this framework and review the current state of research in the field. Finally, we provide a practical roadmap to sampling design, sequencing technologies and bioinformatic analyses.

Background

Hybridization was long seen as a process counteracting the evolution of biodiversity (Seehausen et al., 2008). However, a wealth of genomic studies now show that hybridization is an important contributor to the evolution of phenotypic and species diversity (Abbott et al., 2013; Arnold & Kunte, 2017; Taylor & Larson, 2019). Disease resistance and environmental adaptation in humans in part hails from exchange with Neanderthals and Denisovans (Huerta-Sánchez et al., 2014; Zhou et al., 2021); wing-colour phenotypes involved in mimicry of Heliconius butterflies are a result of adaptive introgression (The Heliconius Genome Consortium, 2012); and hybridization led to the evolution of new species in dog-relatives and birds (Gopalakrishnan et al., 2018; Runemark et al., 2018). Examples from butterflies, birds, and fish highlight that the exchange of genetic variation by hybridization can fuel entire species radiations (Berner & Salzburger, 2015; Edelman et al., 2019; Stryjewski & Sorenson, 2017).

However, hybridization has molecular consequences beyond the exchange of genetic variation that may contribute to the evolution of reproductive isolation and yet have received limited attention. Especially, insights into the hybridization-induced escalation of genetic conflicts (e.g. by TE reactivation) in the wild and its role in speciation remain limited (Fontdevila, 2019; Rebollo et al., 2010). Since successful identifications of genomic regions underlying reproductive isolation remain scarce (Maheshwari & Barbash, 2011; Wolf & Ellegren, 2017), a widening of focus is now advisable, specifically towards genetic conflicts between co-adapted components of the genome, such as transposable elements (TEs) and the mechanisms regulating their activity (Chuong et al., 2017; Dion-Côté & Barbash, 2017).

TEs as drivers of genome evolution and genetic conflict

TEs are DNA sequences abundantly present in virtually all eukaryotic genomes. TEs are able to increase in copy number by transposing or copying themselves to multiple new locations within host genomes thanks to the molecular machinery encoded within their sequence (Hayward & Gilbert, 2022). Thereby, TEs can increase their copy number in gametes and defy Mendelian inheritance, which is why they are considered selfish genetic elements. TEs encompass a large diversity of sequences, protein domains and replicative mechanisms. However, almost all TE sequences include a regulatory motif that ensures their expression and, in autonomous TEs, a sequence coding for proteins enabling their transposition (Chuong et al., 2017). Non-autonomous TEs, that is, elements that partially or completely lack their protein coding sequence, require their own promoter to get expressed, but they can still be transposed by proteins produced in *trans* by other TEs (Okada et al., 1997). TEs not able to be transcribed and translated are usually referred to as "TE-derived sequences". Therefore, the spreading of TEs within both genomes and populations depends on TEs' capability of self-replication, and no positive effect or function for the host is required for their maintenance and spread. While TEs can become 'domesticated' (exapted) by the host genome and thereby can serve

an important role in the evolution of regulatory network (Chuong et al., 2017), most TEs have no positive effect or function for the host, but, rather, may spread and be maintained neutrally or in spite of slightly deleterious effects (Lynch & Conery, 2003).

As selfish elements that often have deleterious fitness effects, TEs and their replicative nature are in direct conflict with the host genome. To control the deleterious effects of TE activity, the genomes evolved mechanisms to repress TE activity. However, these mechanisms can be a double-edged sword for host genomes causing additional conflicts by silencing regulatory motifs within domesticated TEs or nearby TE insertions (Sentmanat & Elgin, 2012). The importance and magnitude of host-TE conflicts are mirrored in a remarkable diversity of redundant repression mechanisms that evolved to counteract TE activity at pre- and post-transcriptional levels.

To understand the importance of TEs in the origin of genetic conflict, we need to understand the mutagenic effects of TE activity. Successful TE activity implies multiple levels of mutagenesis for the host genome: (i) active TEs cause new, identical insertions scattered across the genome, thereby causing structural mutations that can disrupt genic or regulatory sequences (Kazazian et al., 1988; Taşkesen et al., 2012; Wallace et al., 1991); (ii) TE insertions are a target for repressive methylation marks that can spread beyond the sequence of the TE itself. Such changes of the epigenetic state of the surrounding genomic regions, so-called "methylation spill overs", can cause changes in the regulation of nearby genes (Hollister & Gaut, 2009 but see Huang & Lee, 2024) and in the local mutation rates; the same process can happen to regulatory elements that originated from TEs (e.g., domesticated TEs) or share sequence similarity (Erwin et al., 2015). (iii) TEs often include regulatory motifs in their sequences that can serve as alternative promoters and contribute to changes in hosts' gene regulation (Goubert et al., 2020; Sun et al., 2021); and (iv) the spread of identical TE copies across the genome can trigger non-allelic homologous recombination (ectopic recombination) and mediate the evolution of new structural variants (SVs) (Delprat et al., 2009; K. Han et al., 2008; Kupiec & Petes, 1988). Ectopic recombination between TE copies can result in either the deletion (e.g., frequently observed between the terminal repeats of LTR retrotransposons) or inversion of the intervening sequence depending on the relative orientation of the TE copies involved (Balachandran et al., 2022; Mager & Goodchild, 1989). The propensity of a genome to undergo ectopic recombination coupled with the rate of TE activity (and population parameters like effective population size and selective pressures) largely shape the evolution of genome size and structure (Kapusta et al., 2017; Lynch & Conery, 2003) and influence genome stability (Carvalho & Lupski, 2016). Unchecked TE activity can therefore disrupt gene regulation and function (i-iii) or lead to structural rearrangements and genome size variation (iv; Kapusta et al., 2017).

Finally, TE insertions can also have direct fitness consequences for the host. They have been associated with (i) many diseases, including cancer (Payer & Burns, 2019); (ii) changes in longevity (Elsner et al., 2018; Nguyen & Bachtrog, 2021; Ricci et al., 2023); and (iii) the evolution of phenotypic traits, like the industrial melanism in pepper moths (Hof et al., 2016), tail loss in apes and humans (Xia et al., 2024), and the evolution of the mammalian placenta (Emera & Wagner, 2012). Given their often-detrimental

impact, TEs are largely selected against and silenced by multiple molecular mechanisms (see Mechanisms of TE repression section), but they usually persist in a species. This persistence can be attributed to several mechanisms. For example, selection is less efficient in removing insertions of active TEs from regions of low recombination or low gene density (e.g., sex-limited chromosomes; Peona et al., 2021b) than in frequently recombining proportions of the genome. Similarly, some TEs evolved mechanisms that minimise their deleterious effects on the host genome; for instance, they self-methylate or disguise themselves as introns (de Mendoza et al., 2018; Gozashti et al., 2022). This results in reduced selection against their presence in genomes. In addition, events of horizontal TE transfer can cause the invasion of host genomes by ever new types of TEs (Schaack et al., 2010). The general evolutionary success of TEs shows that the repression mechanisms of the host genomes are imperfect, and the conflict between TEs and their repressors is ongoing.

Mechanisms of TE repression

Given the negative effects of TE activity on genome stability and host fitness, genomes have evolved multiple repression systems to counteract TE activity. These repression systems regulate TE activity by reducing or blocking transcription of TE sequences (pre-transcriptional regulation) or by degrading TE transcripts and preventing their insertion into the genome (post-transcriptional regulation; Deniz et al., 2019).

Pre-transcriptional regulation of TE activity by DNA methylation and heterochromatinization

Pre-transcriptionally, TE activity is suppressed by epigenetic DNA modifications that inhibit TE transcription. Vertebrates achieve this primarily by cytosine methylation in CpG context within TEs (Figure 1A) and by histone modifications, such as trimethylation of H3K9 and H3K27 (Deniz et al., 2019). DNA methylation at CpG sites silences TE expression by reducing transcription factor binding capacity (Kaluscha et al., 2022), while histone modification primarily regulates the formation of heterochromatin and thereby renders TE sequences inaccessible for transcription (Slotkin & Martienssen, 2007). In addition, DNA methylation and histone modifications interact to maintain and stabilize TE repression. For example, DNA methylation attracts histone-modifying enzymes, and H3K9me3 and H3K27me3-mediated heterochromatin stabilizes DNA methylation. Thus, pre-transcriptional repression of TEs is an effective and partially redundant mechanism that protects the genome from uncontrolled activity (Guo et al., 2021). While epigenetic marks are known for their plasticity to environmental perturbations, which as such can also influence TE repression efficacy (Guo et al., 2021; Miousse et al., 2015; Pecinka et al., 2010), the epigenetic silencing of TEs is strongly genetically determined (e.g., Aravin & Bourc'his, 2008).

Post-transcriptional regulation of TE activity by the PIWI pathway

In animals, post-transcriptional TE regulation is primarily mediated by the PIWI (P-element induced wimpy testis) pathway (Czech et al., 2018; Deniz et al., 2019; Siomi et al., 2011). The PIWI pathway

involves the formation of complexes of PIWI proteins and piRNAs (piwi-interacting RNAs) that degrade TE transcripts in the cytoplasm (Tóth et al., 2016; Figure 1B). PIWI genes were originally discovered in *Drosophila* as regulatory proteins involved in stem and germ cell differentiation (Vagin et al., 2006) and have been subsequently found in many animal clades, including vertebrates. In contrast, plants, for instance, rely on siRNA-guided proteins (Liu & Zhao, 2023). piRNAs are small non-coding RNAs derived from transcribed TEs or from genomic clusters (Pritam & Signor, 2025). The formation of piRNA clusters involves the incorporation of TEs into piRNA clusters by a stochastic process that is driven by genome-wide insertion of TEs over evolutionary time. These clusters act as reservoirs and produce piRNAs that recognize and silence TEs. Based on their sequence homology, piRNAs help PIWI proteins to target and degrade TE transcripts. The PIWI-pathway can also epigenetically protect the genome from novel TE insertions by the deposition of repressive histone methylation marks, such as H3K9me3, that promote heterochromatin formation and transcriptional silencing pre-transcriptionally (Taskopru et al., 2024).

Epigenetic TE repression is genetically determined

Both, methylation and the PIWI pathway, depend on genetic components that recognize TE sequences and target them for repression or degradation. Consequently, the functionality of both mechanisms depends on the coevolution of the repression machinery's specificity with TE sequences. Therefore, the exchange of genetic material upon hybridization can disrupt the linkage of co-evolved repressors and TEs and consequently deregulate TE activity. In the following we outline how this may trigger a cascade of molecular events that by inducing an increased burden of structural mutation provides a path towards reproductive isolation.

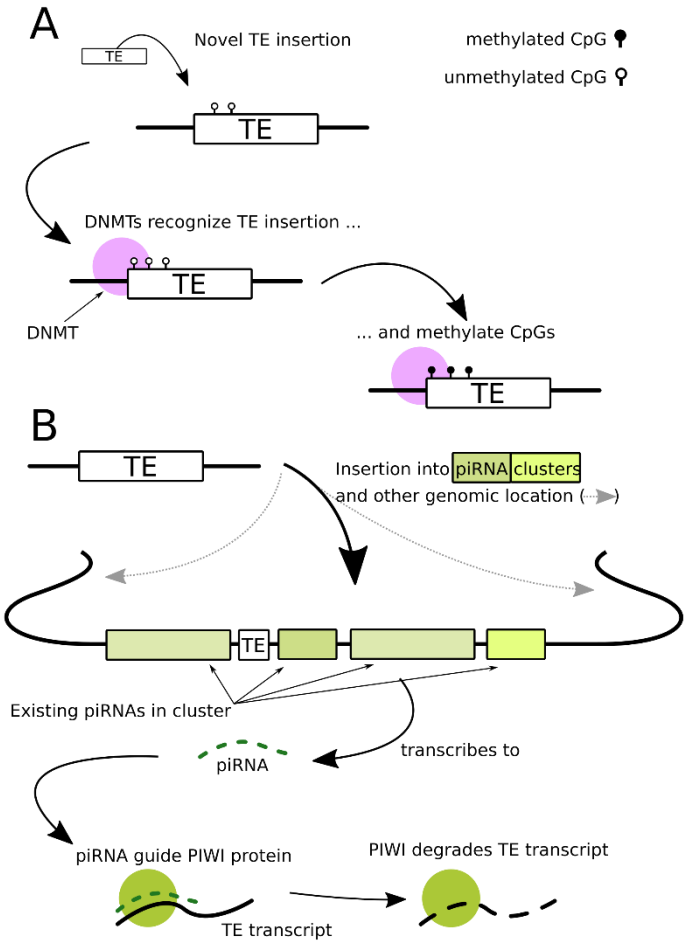

Figure 1 DNA methylation and piRNA-mediated transcript degradation regulate TE activity. **A)** DNA-methyltransferases (DNMTs) detect novel TEs as they insert into a new genomic locus (black line), where existing CpG sites in the TE sequence are methylated *de novo*. **B)** Post-transcriptional repression mediated by the PIWI pathway. When TE insertions insert into piRNA clusters (green rectangles), they can be stored as a reservoir from which TE-derived piRNAs are expressed and guide PIWI proteins to TE transcripts and mark them for degradation.

Hybridization-induced evolution of TE-related reproductive barriers

Hybridization-induced escalation of the genetic conflict between TEs and their repressors

Novel TE insertions can rapidly accumulate in lineages that receive limited gene flow. The fast co-evolution of the TE repression machinery results in an at least partial resolution of the genetic conflict between TEs and their repressors. Because also the methylation- and piRNA-mediated TE repressors can quickly diverge between lineages (Herrera & Bazaga, 2011; Liebl et al., 2013; Lira-Medeiros et al., 2010; Massicotte et al., 2011), the genetic conflicts between TEs and their repressors can be solved differently within each lineage (Dion-Côté, Renaut, Normandeau, et al., 2014; Lerat et al., 2019; Oppold et al., 2017). When such lineages hybridize, the clash of divergent genomes and divergently resolved genetic conflicts may result in the mis-regulation of TE repression (a phenomenon termed "genomic shock"; McClintock, 1984), and entail fitness costs (Maheshwari & Barbash, 2011; Rebollo et al., 2010). Hybridization-induced fitness costs have been most prominently studied in *Drosophila* crosses where the presence and absence of the P-element, a *Drosophila*-specific TE, across both parental species led to imminent hybrid sterility (Kidwell et al. 1977). While such "hybrid dysgenesis" (Kidwell et al. 1977) entails strong reproductive isolation due to a single TE, the fitness effects of TEs may often be much more subtle, reduce fitness only cumulatively across many TEs and at the scale of populations. Furthermore, these changes become most relevant in backcrossed individuals and not F1 hybrids where TEs and their repressors are still co-inherited (Figure 2A and B). However, in advanced-generation hybrids and backcrosses, recombination starts dissociating lineage-specific TEs and repressors from one another. This unmasks previously resolved genetic conflicts and may induce bursts of TE proliferation (Michalak, 2009; Oliver & Greene, 2012) in both germline and somatic cells. While TE insertions into the germline may contribute to long-term genome instability, somatic insertions may impact individuals by worsening their health and ultimately reducing fitness (Payer & Burns, 2019). Finally, physiological stress imposed by reduced fitness of hybrids may induce additional changes in DNA methylation that can further increase TE activity (Rey et al., 2016).

Several lines of evidence for hybridization-induced TE reactivation exist from experimental systems such as *Drosophila* (Kidwell et al., 1977) and yeast (Tusso et al., 2021), however examples from naturally hybridizing populations are limited. In *Drosophila* F1 crosses, despite initial TEs reactivation, they were then re-silenced during the individuals' lifetime (Khurana et al., 2011). Examples from wild populations that hybridize include mice (Brown et al., 2012), marsupials (Metcalfe et al., 2007; O'Neill et al., 2001) and fish. Insights from whitefish indicate that hybrids experience increased rates of TE transcription and insertion (Dion-Côté, Renaut, Normandeau, et al., 2014; Dion-Côté & Barbash, 2017; Laporte et al., 2019) and hybridization in sunflowers induced a burst of TE transcription (Kawakami et al., 2010; Ungerer & Kawakami, 2013).

Given that TE activity is primarily regulated by epigenetic modifications, hybridization-induced TE reactivation may be largely caused by the disruption of these repression mechanisms. Evidence for this comes for instance from wallabies, whose hybrid genomes are both hypomethylated and experienced TE reactivation (O'Neill et al., 2001).

In this context, there can be genomic regions that disproportionally contribute to the origin and escalation of host-TE conflicts, for example, sex chromosomes. The lower recombination rate and effective population size of sex chromosomes often leads to an asymmetric accumulation of active TEs in the non-recombining chromosome (i.e., Y and W chromosomes; Peona, et al., 2021b). If the non-recombining sex chromosome is much denser in active TEs than the rest of the chromosomes (e.g., "toxic Y" [Nguyen & Bachtrog, 2021], "W refugium" [Peona et al., 2021b]), it can represent a rich source of potential host-TE conflicts upon hybridization and backcrossing and by doing that representing a contributing factor to Haldane's rule (Peona et al. 2021b). However, if the non-recombining sex chromosome also features piRNA clusters, then it will represent a less impactful source of conflicts especially in the early hybrid generations. Therefore, depending on the TE composition of sex chromosomes, they could either exacerbate or defuse host-TE conflicts.

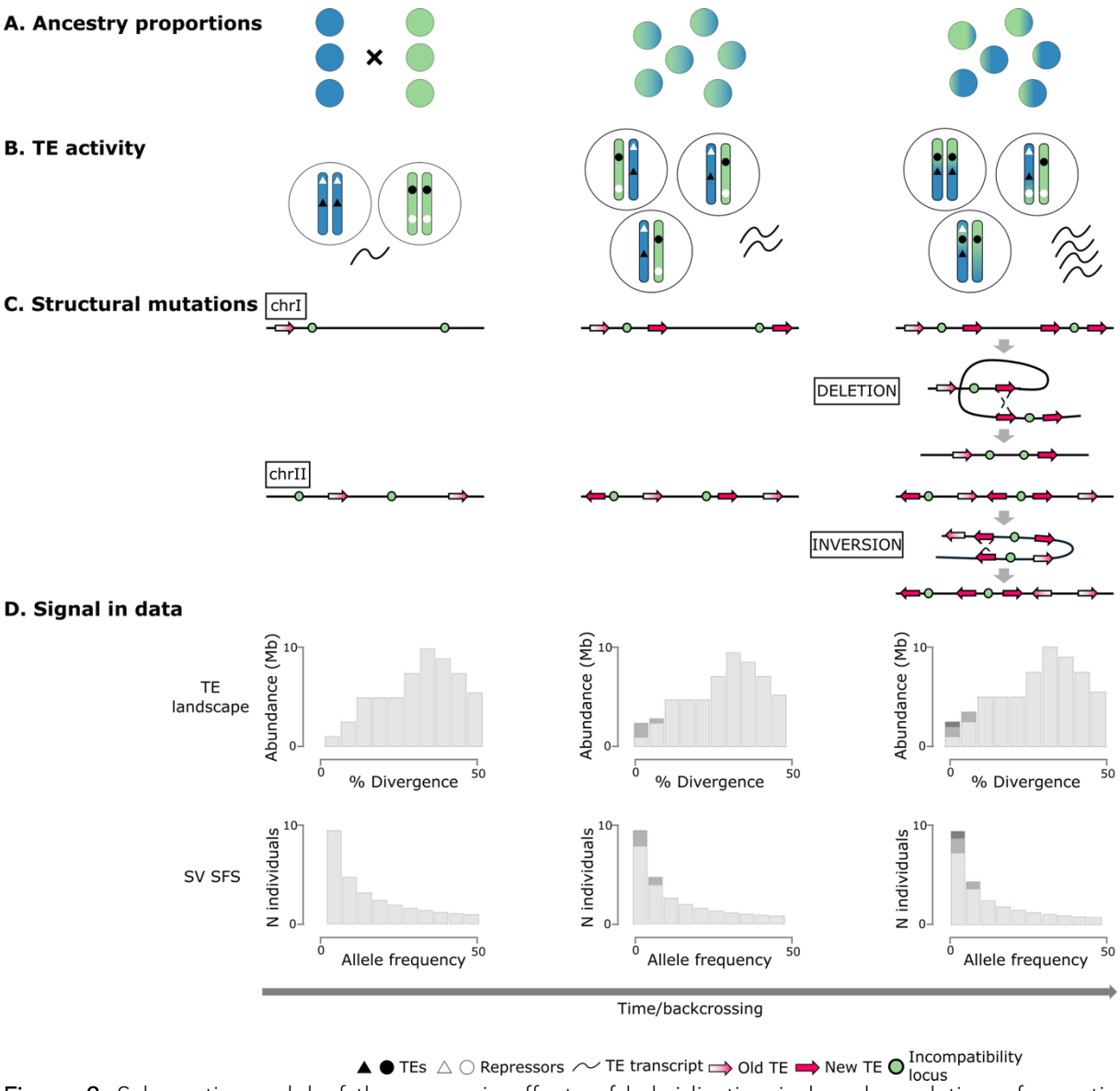

Figure 2. Schematic model of the genomic effects of hybridization-induced escalation of genetic conflicts. A) Two parental lineages (blue and green) cross and produce hybrids with different ancestry proportions as hybridization and backcrossing continues. B) TEs (black-filled shapes) are coupled with their repressors (white-filled shapes) in the parental lineages representing resolved genetic conflicts and only a little TE activity (represented as TE transcripts by wiggly black lines) is expected to be observed. As hybridization occurs, recombination between the parental genomes will uncouple TEs from their repressors and TE activity is expected to increase. C) An increased TE transcription leads to more new identical TE insertions that act as substrate for ectopic recombination and structural mutations. In the upper chromosome (chrI), an example of ectopic recombination between identical repeats oriented in the same direction resulting in a deletion of the intervening sequence is depicted. In chrII, an example of ectopic recombination between identical repeats oriented in the opposite direction resulting in the inversion of the intervening sequence is shown. In both cases, SVs change the linkage between hybrid incompatibility loci and resulting haplotypes can be stronger reproductive barriers. D) Expected genomic signatures of escalation of genetic conflicts shown as progression of TE divergence landscapes and SV site frequency spectra (SFS). As TE activity increases, an increased number of TE insertions in recent times (percentage of divergence of TEs from their consensus sequences) and of derived and low-frequency SV alleles should be observed in repeat landscape and SFS plots in comparison to the parental lineages and between categories of hybrids based on ancestry proportions. The recent gain of TEs and SVs is coloured in darker shades of grey.

Increased rates of chromosome rearrangements following hybridization-induced TE proliferation

Repetitive sequences of the genome, including TEs and sequences derived therefrom, can lead to increased rates of ectopic recombination (reviewed in Kent et al., 2017). These, in turn, can result in higher rates of structural mutation (Aasegg Araya et al., 2025; Balachandran et al., 2022; Munasinghe et al., 2023). The idea that evolutionary events that lead to bursts of repetitive sequence expansion – notably bursts of TE proliferation – may be followed by pulses of structural mutation and even lead to vast genome reorganization (Fontdevila, 2005, 2019) has received less attention to date, especially in the context of hybridization-related evolution. It therefore seems important to stress that the escalation of TE-related genetic conflict may have effects beyond increased TE activity. It may induce a cascade that, via a higher risk of ectopic recombination, causes chromosome rearrangements and ultimately leads to increased genome instability when happening in the germline (McClintock, 1984; Fontdevila, 2005; Rebollo et al., 2010; Rogers, 2015; Figure 2C).

Furthermore, the mis-regulation of methylation that may occur upon hybridization may not only entail TE reactivation but also on its own increase rates of chromosome rearrangements. This is because DNA methylation often suppresses recombination, and hypomethylation directly increases the risk of ectopic recombination (Zamudio et al., 2015). An example of such dynamics is represented by the genomes of hybrid wallabies, which on top of TE reactivation also experience increased rates of structural mutation due to hypomethylation (O'Neill et al., 2001).

The escalation of TE-related genetic conflicts upon hybridization can thus have a profound impact on the stability of genome architecture at both the level of individual lifespans (e.g., higher risk of cancer and faster ageing (Brown et al., 2020; Burns, 2017)), and the macroevolutionary level (e.g., chromosome mis-segregation (Ferree & Barbash, 2009)).

Fitness costs of escalating genetic conflicts: Constraints to gene flow between species?

In hybrids of divergent lineages, previously resolved genetic conflicts between TEs and their repressors may provide a substrate for hybrid incompatibilities (Kidwell et al., 1977; Maheshwari & Barbash, 2011) and for the evolution of intrinsic postzygotic reproductive barriers (Crespi & Nosil, 2013; Durand et al., 2012; Levin & Moran, 2011; Petrov et al., 1995; Rebollo et al., 2010). This contrasts hybrid dysgenesis in *Drosophila* that leads to sterility in the first generation of hybrids (Kidwell et al. 1977). Instead, in this framework TE reactivation contributes to a cumulative, less immediate build-up of fitness disadvantages in hybrids.

First, as outlined above, disease, reduced longevity, and other phenotypic effects can incur fitness costs directly related to (specific) TEs (Elsner et al., 2018; Hof et al., 2016; Nguyen & Bachtrog, 2021; Payer & Burns, 2019; Ricci et al., 2023). Second, and less directly associated with TEs themselves, similar fitness costs may establish through increased rates of structural mutation. Finally, the entire

cascade of hybridization-induced effects on TE repression mechanisms, TE activity, and structural mutation, may inflict less direct fitness costs in the form of, for instance, genome instability, metabolic costs related to the replication of more repeat-enriched genomes, or deregulated gene expression due to, for instance, epigenetic changes. In locations of the genome (and epigenome) where such fitness costs are anchored, hybridization is expected to have the strongest fitness effects and be selected against. Notably, compared to direct fitness costs on the individual level (i.e. disease, reduced longevity), the indirect effects of increased TE activity may show small effects on the individual, but only become apparent – and evolutionarily relevant – on the population level. The escalation of genetic conflicts upon hybridization may therefore inflict major constraints to genetic exchange between species (Crespi & Nosil, 2013; Maheshwari & Barbash, 2011; Rebollo et al., 2010; Seehausen et al., 2008).

A pathway towards molecular reinforcement of species barriers?

Finally, we suggest that by mediating an increased input of SVs (e.g., inversions), the escalation of TE-related genetic conflicts may constitute an avenue towards a molecular reinforcement of species barriers.

SVs, notably large chromosomal inversions, contribute to reproductive barriers in diverse organisms (e.g., reviewed in Kapun & Flatt, 2019; Knief et al., 2024), for example by impeding proper meiotic chromosome pairing in hybrids, or by protecting haplotypes composed of co-adapted allelic combinations from introgression and recombination (Hoffmann & Rieseberg, 2008). In known examples, alternative inversion haplotypes are usually fixed between reproductively isolated lineages, and their combination in hybrids has fitness costs that constitute a reproductive barrier. It may thus seem paradox to suggest that hybridization may be a source of inversions that act as reproductive barriers. However, correlations between the presence of inversions between species and the species' potential for hybridization in birds (Hooper & Price, 2017), for instance, may support a link between hybridization and the formation of inversions. A possible explanation for this paradox: the increased input of inversions in hybridizing species are the consequence of escalated TE-related conflict. If such inversions lock up allelic combinations that reinforce species barriers, they may segregate out of hybrid zones and be selected towards fixation in parental species.

In conclusion, because of escalated TE-related conflict and downstream knock-on effects on structural mutation, hybridization may itself fuel hybrid populations with the very same genetic variation that can contribute to stronger reproductive barriers – and ultimately reinforce reproductive barriers (**Figure 2C**).

Testing the escalation of TE-related genetic conflict upon hybridization in the wild: A roadmap

Several studies have shown that hybridization can induce an escalation of genetic conflicts between

TEs and their repressors. Still, insights into their importance and the magnitude of their effects in natural populations remain limited. Advances in genome sequencing technology now provide unprecedented opportunities to investigate and test the hypothesis of TE reactivation upon hybridization and the latter's downstream effects on chromosome rearrangements. Long-read sequencing helps overcoming previous limitations in resolving repetitive sequences in genome assemblies (Peona et al., 2021) and enables characterizing the landscapes of SVs and epigenetic modifications, notably DNA methylation. In the next section, after outlining the general requirements towards sampling design, we describe the predictions related to the hypothesis of the escalation of TE-related genetic conflict, and for each prediction provide specific requirements towards study design and the analyses approaches.

Sampling design

An adequate sampling design constitutes the foundation for testing all predictions. Such a sampling includes (i) samples from the parental species involved and (ii) a dense sampling of hybrid zones. As it is unclear, at which admixture proportions the predicted effects are most pronounced, it is crucial that this dense sampling includes hybrids from a broad range of ancestry classes. Finally, (iii) multiple individuals from adequate outgroups help distinguishing between ancestral and derived state of TE insertions and other SVs occurring in the lineage of interest.

An experimental setup that enables testing all the predictions below includes samples that provide access to transcriptomic, genomic, and epigenomic variation (Figure 3). Due to tissue- and potential age-specific effects on transcription (De Cecco et al., 2013; Warmuth et al., 2022) and epigenomic variation – such as methylation (Fraga et al., 2005; Loyfer et al., 2023) – the ideal sampling needs to consider the same tissue across individuals. Individual age and other factors may impact the molecular variation under investigation and should be accounted for by recording these data and be included in statistical inferences. Regarding environmental variation, we note that hybrid zones may provide natural common garden-like experiments (Buerkle & Lexer, 2008) in which divergent genomes and epigenomes experience a similar environment, and environmental effects on methylation are in part accounted for (Mueller et al., 2025).

The choice of tissue(s) may often need to balance between the ideal versus the realistic and feasible. Germline tissues have the great advantage of providing insights into molecular variation of evolutionary relevance. However, sampling germline tissues is often lethal and amongst other issues may imply limited sample sizes. On the other hand, molecular variation characterized from non-invasively sampled somatic tissues may include somatic mutations less relevant to evolution. Still, somatic mutations in hybrids may be caused by TE reactivation, have fitness consequence, and as such can be evidence for the presented hypothesis. In addition, somatic mutations, despite being ten-to hundredfold more abundant than germline mutation (Milholland et al., 2017). are distributed randomly in non-clonal cell populations and observed at low allele frequencies. Thereby somatic mutations often exhibit unique properties compared to germline mutations, that allow selective

filtering based on allele frequency. Typically, non-invasive sampling will involve blood, that is a robust approximator for epigenetic variation in the soma (Derks et al., 2016; Husby, 2022). Still, any effects in line with the hypothesis of hybridization-induced escalation of genetic conflict may be tissue specific. The ultimate study design would thus include a diversity of tissues. In addition to the limitations discussed in connection with germline tissues, such a design may usually be limited by the costs it still entails.

Testing the predictions

The following sections lay out analysis strategies to address five predictions made by the hypothesis of hybridization-induced escalation of genetic conflict between TEs and their repressors and its downstream effects. We treat the second and third predictions jointly, because the according analyses are largely identical.

1. Hybridization disrupts epigenetic silencing of transposable elements

The initial step of a hybridization-induced escalation of TE-related genetic conflicts is a failure in the epigenetic repression of TEs in an admixed genome that induces changes in epigenetic patterns. This disruptive pattern can be surveyed by testing many different epigenetic markers. However, DNA methylation is a main epigenetic modification of interest, because (i) methylation is the primary epigenetic modification that silences TEs (in vertebrates, but also plants) and (ii) technology is available to comprehensibly characterize genome-wide patterns of methylation at population scale. In comparison to the parental lineages, hybrids are predicted to show transgressive methylation patterns that are related to (de-)repression of TEs.

This prediction is based on assumption that methylation is determined genetically. As outlined in the section "TE repression", TE regulation depends on correct recognition of TE sequences by the molecular machinery responsible for *de novo* methylation of the genome during embryogenesis. At this developmental stage, the genome of most vertebrates is nearly completely demethylated and provides a window for TEs to become reactivated if not successfully silenced. Based on previous theoretical predictions (Fontdevila, 2005) and empirical observations that have linked deregulation of methylation and TE repression (O'Neill et al., 2001) we here lay out the data generation and analysis steps to study such effects in an evolutionary framework.

Measuring DNA methylation states related to TE-regulation requires the precise mapping of methylation signals in TE sequences and their promoters. This step is complicated by the repetitiveness of TEs that leads to multi-mapping of short reads and consequently inaccurate methylation calls. In non-model organisms, methylation signals can be obtained by whole-genome sequencing using short- or long-read technologies and methylation-specific analysis workflows. For short-reads, bisulfite-sequencing (BS-Seq) has long been the gold-standard for genome-wide or reduced representation mapping of 5mC modifications. Recently, it started being superseded by enzymatic conversion methods (EM-Seq) that are more reliable in deaminating unmethylated cytosines while causing less DNA degradation upon treatment (Y. Han et al., 2022). Compared to these short read-based technologies, long reads offer multiple advantages. Both Oxford Nanopore

(ONT) and PacBio HiFi long-read sequencing yield data on epigenetic DNA modifications together with genetic sequences (Ni et al., 2023). With average read length of >10 kilo base pairs (kb), long reads span complex repetitive elements, thereby enabling a reliable readout of methylation states within specific TE loci. Additionally, long reads improve assembling repetitive regions in genome assemblies and improve SV calling and genotyping in mapped genomes (Rech et al., 2022). While still being more expensive than short-read sequencing, prices for ONT and HiFi sequencing have decreased markedly with recent sequencing platforms. Thus, despite somewhat higher requirements for the DNA material, long-read sequencing becomes accessible for studying TE-based genetic conflict in hybridizing natural populations.

The computational analysis of methylation-sequencing data is guided by the expectations of observable patterns in methylation states across parental lineages and hybrid individuals. Given that DNA methylation is the primary repressor of TE activity, sequences of functional TEs or their regulatory regions may become hypomethylated in hybrids compared to the parental lineages. This prediction can be tested by assessing the methylation states of full-length TE sequences and their flanking regions in parental and hybrid populations. Due to limitations of mapping short-reads to repetitive regions, this analysis may best be based on long-reads. In addition, screening the genome for differentially methylated loci or regions (DMLs, DMRs) between parental populations and hybrids may give insights into methylation de-regulation with potential effects on the expression and insertion of TEs. For example, TE loci that are constitutively hypermethylated in parental populations may show transgressive hypomethylation in hybrids, indicating hybridization-induced deregulation of methylation patterns. However, potential statistical limitations of DML and DMR analyses are currently being discussed and call for careful interpretation of the results (Mueller et al., 2025). Additionally, statistical analyses should consider the potential of DNA methylation to respond to environmental stress (Makarevitch et al., 2015; Rey et al., 2016). Ideally this requires including environmental variables as covariates in the analyses to identify potential confounding signal in the data. In model-based DML/DMR analyses (Lea et al., 2015; Park & Wu, 2016) covariates can be included in the statistical model. More explicitly, distance-based redundancy analyses have been used to estimate the contribution of different variables to the epigenetic variation and identify sites driven by specific variables captured in those variables (Merondun & Wolf, 2025; Ruiz-Arenas & González, 2017).

2. TE transcription levels are deregulated in hybrids compared to parental populations

If hybridization deregulates the methylation of TEs, it can induce burst of TE expression (Dion-Côté, Renaut, & Normandeau Eric and Bernatchez, 2014; McClintock, 1984) . The identification of such bursts requires TE expression data that can be acquired by transcriptome sequencing. To assess the changes in transcription levels of TEs in hybrids with respect to the parental lineages, it is necessary to sequence their RNA and consider some key points regarding data production and analysis. Ideally, individuals' transcriptomes should be sequenced with short reads and long-read technologies like Iso-seq to ensure the detection of locus-specific differential expression given a reference genome and a TE annotation (Lee et al., 2025) . If only short-read RNA-seq is available, the differential expression of

specific TE loci is less precise (but see Schwarz et al., 2022). This reference-based approach can be replaced by mapping RNA-seq data to a TE library (Peona et al., 2021). In the latter case, it is possible to detect differential expression of TEs on the level of TE subfamilies, that is, if specific subfamilies show higher expression in hybrids than in parental lineages. In addition, it is important to carefully choose the type of RNA prior to sequencing. While polyadenylated mRNA selection is often the best choice, some TEs (e.g., TEs with a pol III promoter) do not present a poly-A tail upon transcription (Kramerov & Vassetzky, 2011). It is therefore necessary to know the overall TE composition of the genomes of interest before choosing the RNA extraction strategy.

Bioinformatics analysis of transgressive expression of TEs follow the same methodology used for transgressive expression of genes in hybrids (Papoli Yazdi et al., 2022) that require to be compared to both parental lineages. Therefore, to control for parental-specific differences in expression, transcriptome data is required for all the lineages involved.

3 & 4. Hybrid genomes feature elevated rates of TE insertions and SV variation compared to parental populations.

The increased expression of TEs may ultimately result in increased rates of TE insertion of structural mutation in hybrid lineages. Inferring the latter involves identifying TE and SV polymorphisms segregating in all involved lineages and comparing the levels of derived TEs and SVs between hybrid and parental lineages. The best approach to reliably and possibly completely call and genotype TEs and SVs relies on reference-free pangenome graphs estimated from long-read and other long-range information (such as Hi-C; Groza et al., 2024). Currently, this approach is still limited to a few study systems, mostly because of sampling requirements (e.g., high quality and quantity of DNA) and sequencing costs. An alternative to pangenomes is a reference-based approach that calls SVs from long reads from multiple individuals based on a high-quality chromosome-level assembly for one of the parental lineages (Weissensteiner et al., 2020). Additionally, to reduce sequencing costs, it is possible to use short reads from a higher number of individuals to genotype the characterised SVs (Chen et al., 2019; Peona et al., 2022). Lastly, SV calling and genotyping solely based on short reads can be implemented when using a high-quality reference genome assembly (David et al., 2024). However, short reads are inefficient in calling TE insertions, which are the most important type of SV to consider for testing the predictions presented here. Finally, any approach to identify TE polymorphisms relies on the availability of a high-quality TE library to be able to properly classify SVs as TE insertions.

Once TEs and SVs have been identified and quantified, we move to the comparison between hybrid and parent populations, which needs to account for multiple factors. In simple cases, hybrid populations may be expected to show overall higher rates of TE insertion and increased derived variation compared to parental lineages. However, realistically, we expect these patterns to be much subtler and intricate. First, recently evolved genetic variation usually segregates at low frequencies. Accordingly, the TE insertions and SVs we are interested in when testing the present predictions are expected to occur in hybrids at low frequencies or even as singletons. As a crucial implication, the

discovery of TEs and SVs (that is TE and SV calling) obligately needs to include a large enough sample size of hybrid individuals (singletons not included in the discovery set cannot be genotyped). Second, more complex situations may involve parental genomes that differ in TE and SV content. If the parental genomes differ in TE abundance, it is critical to establish a baseline for the expected TE abundance in hybrids based on admixture proportions. Only then it is possible to accurately assess any additional input of TE insertion caused by TE reactivation. Furthermore, also demographic effects such as changes in effective population size can affect integration rates of TEs on the population level and may produce false-positive signals of TE activity bursts (Horvath et al., 2022). To control for such demographic effects, SNPs need to be analysed in parallel to TEs and SVs. In the presence of bursts of TE insertion and structural mutation, we expect increased rates of the former that, however, are not paralleled in SNPs. To this end, site frequency spectrum (SFS) analysis can, for example, give an overview of the allele frequency dynamics of TEs and SNPs in the different lineages. A TE SFS that has the same shape as the SNP SFS, indicates that TE dynamics are mostly shaped by the same evolutionary and demographic processes as the ones that affect SNPs. Furthermore, it is important to account for allele's ages when comparing SFS between TEs and SNPs, because TE bursts are often temporarily limited. Such episodic bursts may be obscured in global analyses when alleles' frequencies do not account for age (Horvath et al., 2022). To date the variants, TEs and SVs are ideally phased and polarized, as this enables more precise comparisons of mutation rates at different age categories (Horvath et al., 2022; Figure 2D). Age estimates of TEs and SNPs can be obtained from various methods that depend on the variant type and the available data: While the age of TEs in general can be estimated as the genetic distance (e.g., using Kimura 2-p model) between an insertion and its consensus sequence, more precise age estimates can be obtained for specific groups of TEs. For instance, LTR retrotransposons harbour two terminal repeats that, because of their replicative mechanism, will be identical upon insertion. The independent mutations accumulated by the two identical repeats can therefore be used to estimate the upper bound of their insertion age (Weissensteiner et al., 2020). If phasing information is available for SVs as well as for SNPs, then coalescent-based methods like GEVA (Albers & McVean, 2020) can be used to estimate the age of all variants. If a reliable (that is, phased and polarised) SV dataset cannot be obtained, then comparing the TE landscapes of hybrids to the parental lineages can give first insights into the dynamics of TEs in different ancestry backgrounds (Figure 2D). However, repeat landscapes need to be interpreted with caution, since they do not take demographic dynamics and evolutionary processes into account.

5. Gene flow upon hybridization is restricted in genomic regions involved or deriving from TE-related genetic conflict

An increased input of TE insertions and SVs in hybrids may reduce hybrid fitness and, moreover, result in SVs that may strengthen reproductive barriers. Both of these outcomes may restrict gene flow in the genomic regions (i) that are involved in the escalating TE-related genetic conflict or (ii) the SVs resulting therefrom. Testing these predictions is notoriously challenging and, at the current stage of research, a comprehensive roadmap towards this goal remains difficult to formulate. Reaching the

goal may usually remain daunting.

In a scenario under which increased TE proliferation or the spread of SVs itself reduces hybrid fitness, selection is expected to act against allelic combinations contributing to TE proliferation and the spread of disadvantageous SVs. If TE proliferation is a result of disrupted methylation regulation due to heterospecific allelic combinations in cis- or trans-acting regulators of methylation, the targets of selection against introgression may be widely spread across the genome. In this case, barriers to gene flow would be spread genome-wide and detecting them will be a formidable challenge. In contrary, in simple cases, where incompatible heterospecific allele combinations causing TE reactivation are located close to TEs, such incompatibilities may show as patterns of increased genetic differentiation or decreased introgression concentrated around TEs (likewise for SVs resulting from TE reactivation). Yet, these patterns may be much more diffusely spread and difficult to associate with the processes described here, if relevant loci are less tightly bound to TEs and SVs but widespread, for instance in trans. Demonstrating a scenario where the reactivation of TEs upon hybridization leads to genome-wide reproductive barriers in our opinion will often first require advanced insights into basic aspects of how hybridization may deregulate TE repression, starting with a better understanding of how the variation repressing TE activity, notably methylation, is regulated and evolves under hybridization.

A scenario of molecular reinforcement, such as described above, makes predictions that may at least in part be more straightforward to test. At a macroevolutionary scale, a higher abundance of SVs in species pairs that have the opportunity to hybridize, such as is observed in birds (Hooper & Price, 2017), may provide indirect hints towards the importance of such a scenario. Direct evidence, however, would consist in (i) population genetic evidence for restricted gene flow around SVs, such as inversions, for which (ii) the origin can be pinpointed to hybridization. While addressing the first point may often be straightforward (Dobzhansky & Sturtevant, 1938; Kapun & Flatt, 2019; Kirkpatrick & Barton, 2006), demonstrating an SV's hybrid origin will in many instances be daunting. Analyses to this end may demonstrate a higher abundance of such SVs inside compared to outside hybrid zones, if the SV has not spread across parental species ranges or its fitness advantages are highest inside the hybrid zones. However, similar patterns may establish of SVs that evolved elsewhere have a higher fitness in hybrid zones than outside. Inferences of SVs' origins may thus often depend on geographic patterns of diversity linked to such an SV, with decreasing diversity linked to the derived variant away from the hybrid zone pinpointing the origin to the hybrid zone. In cases where the SV fixed in the parental lineages, this may be the only evidence for a hybrid-derived origin. However, in many instances, such patterns may never establish, for instance when positive selection rapidly fixes an SV and depletes it from all linked variation, or over time be masked other population genetic processes, such as genetic drift and recombination.

The demonstration of restricted gene flow in genomic regions related to the genetic conflicts caused by TEs would provide ultimate evidence for the latter's evolutionary importance. Promising vertebrate model systems from the wild that could be suitable to test this scenario are, for example, hybridising white killifish, swordtail fish, mice and birds (Langdon et al., 2024; Roussel et al., 2025; Trier et al.,

2014; Wang et al., 2015).

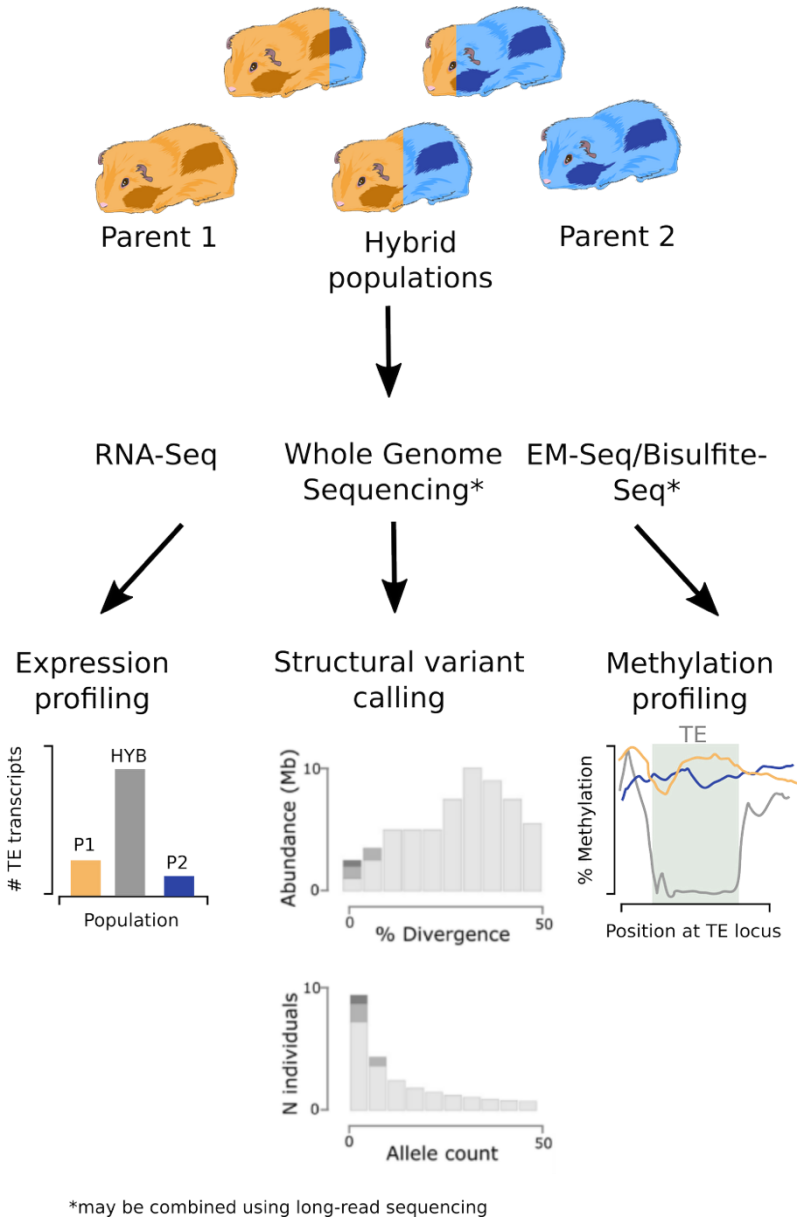

Figure 3. Roadmap to test for signatures of hybridization-induced TE related genetic conflict. From sampling two parental populations and their hybrid zones with different levels of ancestry proportions, multi-model OMICS-data is generated and tested for specific signatures in hybrids that indicate elevated TE expression, higher abundance of low divergent TE insertions and low-frequency TE alleles and hypomethylation of TE sequences.

Conclusions

Despite evidence arguing for an important contribution of TEs to reproductive isolation (Dion-Côté & Barbash, 2017; Fontdevila, 2005; Michalak, 2009; Rebollo et al., 2010), the relevance of the genetic conflict between TEs and their repressors for the evolution of species barriers has received limited testing. In this perspective, we described how the disruption of the tightly regulated interplay between

TEs and their repressors upon hybridization may unleash a cascade of molecular changes that may reinforce genetic incompatibilities and provide a quick path towards reproductive isolation. If valid, our hypothesis may be manifested in higher-than-expected rates of structural mutation driven by TE insertions as well as marked changes in DNA methylation in hybrid individuals. We predict that empirical studies of TE-related genetic conflict in hybrid zones may shed light into the molecular underpinnings of emerging reproductive isolation and contribute to our understanding of hybridization for the evolution of biodiversity.

References

Aasegg Araya, R., Reinar, W. B., Tørresen, O. K., Goubert, C., Daughton, T. J., Hoff, S. N. K., Baalsrud, H. T., Brieuc, M. S. O., Komisarczuk, A. Z., Jentoft, S., Cerca, J., & Jakobsen, K. S. (2025). Chromosomal Inversions Mediated by Tandem Insertions of Transposable Elements. *Genome Biology and Evolution*, *17*(8), evaf131. https://doi.org/10.1093/gbe/evaf131

Abbott, R., Albach, D., Ansell, S., Arntzen, J. W., Baird, S. J. E., Bierne, N., Boughman, J., Brelsford, A., Buerkle, C. A., Buggs, R., Butlin, R. K., Dieckmann, U., Eroukhmanoff, F., Grill, A., Cahan, S. H., Hermansen, J. S., Hewitt, G., Hudson, A. G., Jiggins, C., … Zinner, D. (2013). Hybridization and speciation. *Journal of Evolutionary Biology*, *26*(2), 229–246. https://doi.org/10.1111/j.1420-9101.2012.02599.x

Albers, P. K., & McVean, G. (2020). Dating genomic variants and shared ancestry in population-scale sequencing data. *PLOS Biology*, *18*(1), e3000586-. https://doi.org/10.1371/journal.pbio.3000586

Aravin, A. A., & Bourc'his, D. (2008). Small RNA guides for de novo DNA methylation in mammalian germ cells. *Genes & Development*, *22*(8), 970–975. https://doi.org/10.1101/gad.1669408

Arnold, M. L., & Kunte, K. (2017). Adaptive Genetic Exchange: A Tangled History of Admixture and Evolutionary Innovation. *Trends in Ecology & Evolution*, *32*(8), 601–611. https://doi.org/10.1016/j.tree.2017.05.007

Balachandran, P., Walawalkar, I. A., Flores, J. I., Dayton, J. N., Audano, P. A., & Beck, C. R. (2022). Transposable element-mediated rearrangements are prevalent in human genomes. *Nature Communications*, *13*(1), 7115. https://doi.org/10.1038/s41467-022-34810-8

Berner, D., & Salzburger, W. (2015). The genomics of organismal diversification illuminated by adaptive radiations. *Trends in Genetics*, *31*(9), 491–499. https://doi.org/10.1016/j.tig.2015.07.002

Brennecke, J., Malone, C. D., Aravin, A. A., Sachidanandam, R., Stark, A., & Hannon, G. J. (2008). An Epigenetic Role for Maternally Inherited piRNAs in Transposon Silencing. *Science*, *322*(5906), 1387–1392. https://doi.org/10.1126/science.1165171

Brown, E. J., Nguyen, A. H., & Bachtrog, D. (2020). The Y chromosome may contribute to sex-specific ageing in Drosophila. *Nature Ecology & Evolution*, *4*(6), 853–862. https://doi.org/10.1038/s41559-020-1179-5

Brown, J. D., Piccuillo, V., & O'Neill, R. J. (2012). Retroelement Demethylation Associated with Abnormal Placentation in Mus musculus × Mus caroli Hybrids1. *Biology of Reproduction*, *86*(3), 88, 1–10. https://doi.org/10.1095/biolreprod.111.095273

Buerkle, C. A., & Lexer, C. (2008). Admixture as the basis for genetic mapping. *Trends in Ecology & Evolution*, *23*(12), 686–694. https://doi.org/10.1016/j.tree.2008.07.008

Burns, K. H. (2017). Transposable elements in cancer. *Nature Reviews Cancer*, *17*(7), 415–424. https://doi.org/10.1038/nrc.2017.35

Carvalho, C. M. B., & Lupski, J. R. (2016). Mechanisms underlying structural variant formation in genomic disorders. *Nature Reviews Genetics*, *17*(4), 224–238. https://doi.org/10.1038/nrg.2015.25

Chen, S., Krusche, P., Dolzhenko, E., Sherman, R. M., Petrovski, R., Schlesinger, F., Kirsche, M., Bentley, D. R., Schatz, M. C., Sedlazeck, F. J., & Eberle, M. A. (2019). Paragraph: A graph-based structural variant genotyper for short-read sequence data. *Genome Biology*, *20*(1), 291. https://doi.org/10.1186/s13059-019-1909-7

Chuong, E. B., Elde, N. C., & Feschotte, C. (2017). Regulatory activities of transposable elements: from conflicts to benefits. *Nature Reviews Genetics*, *18*(2), 71–86. https://doi.org/10.1038/nrg.2016.139

Crespi, B., & Nosil, P. (2013). Conflictual speciation: species formation via genomic conflict. *Trends in Ecology & Evolution*, *28*(1), 48–57. https://doi.org/10.1016/j.tree.2012.08.015

Czech, B., Munafò, M., Ciabrelli, F., Eastwood, E. L., Fabry, M. H., Kneuss, E., & Hannon, G. J. (2018). PiRNA-guided genome defense: From biogenesis to silencing. *Annual Review of Genetics*, *52*(1), 131–157. https://doi.org/10.1146/annurev-genet-120417-031441

David, G., Bertolotti, A., Layer, R., Scofield, D., Hayward, A., Baril, T., Burnett, H. A., Gudmunds, E., Jensen, H., & Husby, A. (2024). Calling Structural Variants with Confidence from Short-Read Data in Wild Bird Populations. *Genome Biology and Evolution*, *16*(4), evae049. https://doi.org/10.1093/gbe/evae049

De Cecco, M., Criscione, S. W., Peterson, A. L., Neretti, N., Sedivy, J. M., & Kreiling, J. A. (2013). Transposable elements become active and mobile in the genomes of aging mammalian somatic tissues. *Aging*, *5*(12), 867–883. https://doi.org/10.18632/aging.100621

de Mendoza, A., Bonnet, A., Vargas-Landin, D. B., Ji, N., Li, H., Yang, F., Li, L., Hori, K., Pflueger, J., Buckberry, S., Ohta, H., Rosic, N., Lesage, P., Lin, S., & Lister, R. (2018). Recurrent acquisition of cytosine methyltransferases into eukaryotic retrotransposons. *Nature Communications*, *9*(1), 1341. https://doi.org/10.1038/s41467-018-03724-9

Delprat, A., Negre, B., Puig, M., & Ruiz, A. (2009). The Transposon Galileo Generates Natural Chromosomal Inversions in Drosophila by Ectopic Recombination. *PLOS ONE*, *4*(11), e7883-. https://doi.org/10.1371/journal.pone.0007883

Deniz, Ö., Frost, J. M., & Branco, M. R. (2019). Regulation of transposable elements by DNA modifications. *Nat. Rev. Genet.*, *20*(7), 417–431.

Derks, M. F. L., Schachtschneider, K. M., Madsen, O., Schijlen, E., Verhoeven, K. J. F., & van Oers, K. (2016). Gene and transposable element methylation in great tit (Parus major) brain and blood. *BMC Genomics*, *17*(1), 332. https://doi.org/10.1186/s12864-016-2653-y

Dion-Côté, A.-M., & Barbash, D. A. (2017). Beyond speciation genes: an overview of genome stability in evolution and speciation. *Current Opinion in Genetics & Development*, *47*, 17–23. https://doi.org/https://doi.org/10.1016/j.gde.2017.07.014

Dion-Côté, A.-M., Renaut, S., Normandeau, E., & Bernatchez, L. (2014). RNA-seq Reveals Transcriptomic Shock Involving Transposable Elements Reactivation in Hybrids of Young Lake Whitefish Species. *Molecular Biology and Evolution*, *31*(5), 1188–1199. https://doi.org/10.1093/molbev/msu069

Dion-Côté, A.-M., Renaut, S., & Normandeau Eric and Bernatchez, L. (2014). RNA-seq Reveals Transcriptomic

Shock Involving Transposable Elements Reactivation in Hybrids of Young Lake Whitefish Species. *Mol. Biol. Evol.*, *31*(5), 1188–1199.

Dobzhansky, T., & Sturtevant, A. H. (1938). INVERSIONS IN THE CHROMOSOMES OF DROSOPHILA PSEUDOOBSCURA. *Genetics*, *23*(1), 28–64. https://doi.org/10.1093/genetics/23.1.28

Durand, S., Bouché, N., Perez Strand, E., Loudet, O., & Camilleri, C. (2012). Rapid Establishment of Genetic Incompatibility through Natural Epigenetic Variation. *Current Biology*, *22*(4), 326–331. https://doi.org/10.1016/j.cub.2011.12.054

Edelman, N. B., Frandsen, P. B., Miyagi, M., Clavijo, B., Davey, J., Dikow, R. B., García-Accinelli, G., Van Belleghem, S. M., Patterson, N., Neafsey, D. E., Challis, R., Kumar, S., Moreira, G. R. P., Salazar, C., Chouteau, M., Counterman, B. A., Papa, R., Blaxter, M., Reed, R. D., … Mallet, J. (2019). Genomic architecture and introgression shape a butterfly radiation. *Science*, *366*(6465), 594–599. https://doi.org/10.1126/science.aaw2090

Elsner, D., Meusemann, K., & Korb, J. (2018). Longevity and transposon defense, the case of termite reproductives. *Proceedings of the National Academy of Sciences*, *115*(21), 5504–5509. https://doi.org/10.1073/pnas.1804046115

Emera, D., & Wagner, G. P. (2012). Transposable element recruitments in the mammalian placenta: Impacts and mechanisms. *Briefings in Functional Genomics*, *11*(4), 267–276. https://doi.org/10.1093/bfgp/els013

Erwin, A. A., Galdos, M. A., Wickersheim, M. L., Harrison, C. C., Marr, K. D., Colicchio, J. M., & Blumenstiel, J. P. (2015). piRNAs Are Associated with Diverse Transgenerational Effects on Gene and Transposon Expression in a Hybrid Dysgenic Syndrome of D. virilis. *PLOS Genetics*, *11*(8), e1005332-. https://doi.org/10.1371/journal.pgen.1005332

Ferree, P. M., & Barbash, D. A. (2009). Species-specific heterochromatin prevents mitotic chromosome segregation to cause hybrid lethality in Drosophila. *PLoS Biology*, *7*(10), e1000234. https://doi.org/10.1371/journal.pbio.1000234

Fontdevila, A. (2005). Hybrid genome evolution by transposition. *Cytogenet. Genome Res.*, *110*, 49–55.

Fontdevila, A. (2019). Hybrid Genome Evolution by Transposition: An Update. *Journal of Heredity*, *110*(1), 124–136. https://doi.org/10.1093/jhered/esy040

Fraga, M. F., Ballestar, E., Paz, M. F., Ropero, S., Setien, F., Ballestar, M. L., Heine-Suñer, D., Cigudosa, J. C., Urioste, M., Benitez, J., Boix-Chornet, M., Sanchez-Aguilera, A., Ling, C., Carlsson, E., Poulsen, P., Vaag, A., Stephan, Z., Spector, T. D., Wu, Y.-Z., … Esteller, M. (2005). Epigenetic differences arise during the lifetime of monozygotic twins. *Proceedings of the National Academy of Sciences*, *102*(30), 10604–10609. https://doi.org/10.1073/pnas.0500398102

Gopalakrishnan, S., Sinding, M.-H. S., Ramos-Madrigal, J., Niemann, J., Samaniego Castruita, J. A., Vieira, F. G., Carøe, C., Montero, M. de M., Kuderna, L., Serres, A., González-Basallote, V. M., Liu, Y.-H., Wang, G.-D., Marques-Bonet, T., Mirarab, S., Fernandes, C., Gaubert, P., Koepfli, K.-P., Budd, J., … Gilbert, M. T. P. (2018). Interspecific Gene Flow Shaped the Evolution of the Genus Canis. *Current Biology*, *28*(21), 3441-3449.e5. https://doi.org/10.1016/j.cub.2018.08.041

Goubert, C., Zevallos, N. A., & Feschotte, C. (2020). Contribution of unfixed transposable element insertions to human regulatory variation. *Philosophical Transactions of the Royal Society B: Biological Sciences*,

694 *375*(1795), 20190331. https://doi.org/10.1098/rstb.2019.0331
695 Gozashti, L., Roy, S. W., Thornlow, B., Kramer, A., Ares, M., & Corbett-Detig, R. (2022). Transposable elements
696 drive intron gain in diverse eukaryotes. *Proceedings of the National Academy of Sciences*, *119*(48),
697 e2209766119. https://doi.org/10.1073/pnas.2209766119
698 Groza, C., Schwendinger-Schreck, C., Cheung, W. A., Farrow, E. G., Thiffault, I., Lake, J., Rizzo, W. B., Evrony, G.,
699 Curran, T., Bourque, G., & Pastinen, T. (2024). Pangenome graphs improve the analysis of structural
700 variants in rare genetic diseases. *Nature Communications*, *15*(1), 657. https://doi.org/10.1038/s41467-
701 024-44980-2
702 Guo, W., Wang, D., & Lisch, D. (2021). RNA-directed DNA methylation prevents rapid and heritable reversal of
703 transposon silencing under heat stress in Zea mays. *PLOS Genetics*, *17*(6), e1009326-.
704 https://doi.org/10.1371/journal.pgen.1009326
705 Han, K., Lee, J., Meyer, T. J., Remedios, P., Goodwin, L., & Batzer, M. A. (2008). L1 recombination-associated
706 deletions generate human genomic variation. *Proceedings of the National Academy of Sciences*, *105*(49),
707 19366–19371. https://doi.org/10.1073/pnas.0807866105
708 Han, Y., Zheleznyakova, G. Y., Marincevic-Zuniga, Y., Kakhki, M. P., Raine, A., Needhamsen, M., & Jagodic, M.
709 (2022). Comparison of EM-seq and PBAT methylome library methods for low-input DNA. *Epigenetics*,
710 *17*(10), 1195–1204.
711 Hayward, A., & Gilbert, C. (2022). Transposable elements. *Current Biology*, *32*(17), R904–R909.
712 https://doi.org/10.1016/j.cub.2022.07.044
713 Herrera, C. M., & Bazaga, P. (2011). Untangling individual variation in natural populations: ecological, genetic and
714 epigenetic correlates of long-term inequality in herbivory. *Molecular Ecology*, *20*(8), 1675–1688.
715 https://doi.org/https://doi.org/10.1111/j.1365-294X.2011.05026.x
716 Hof, A. E. van't, Campagne, P., Rigden, D. J., Yung, C. J., Lingley, J., Quail, M. A., Hall, N., Darby, A. C., & Saccheri,
717 I. J. (2016). The industrial melanism mutation in British peppered moths is a transposable element. *Nature*,
718 *534*(7605), 102–105.
719 Hoffmann, A. A., & Rieseberg, L. H. (2008). Revisiting the Impact of Inversions in Evolution: From Population
720 Genetic Markers to Drivers of Adaptive Shifts and Speciation? *Annual Review of Ecology, Evolution, and*
721 *Systematics*, *39*(Volume 39, 2008), 21–42.
722 https://doi.org/https://doi.org/10.1146/annurev.ecolsys.39.110707.173532
723 Hollister, J. D., & Gaut, B. S. (2009). Epigenetic silencing of transposable elements: A trade-off between reduced
724 transposition and deleterious effects on neighboring gene expression. *Genome Research*, *19*(8), 1419–
725 1428. https://doi.org/10.1101/gr.091678.109
726 Hooper, D. M., & Price, T. D. (2017). Chromosomal inversion differences correlate with range overlap in passerine
727 birds. *Nature Ecology & Evolution*, *1*(10), 1526–1534. https://doi.org/10.1038/s41559-017-0284-6
728 Horvath, R., Menon, M., Stitzer, M., & Ross-Ibarra, J. (2022). Controlling for Variable Transposition Rate with an
729 Age-Adjusted Site Frequency Spectrum. *Genome Biology and Evolution*, *14*(2), evac016.
730 https://doi.org/10.1093/gbe/evac016
731 Huang, Y., & Lee, Y. C. G. (2024). Blessing or curse: how the epigenetic resolution of host-transposable element
732 conflicts shapes their evolutionary dynamics. *Proceedings of the Royal Society B: Biological Sciences*,

291(2020), 20232775. https://doi.org/10.1098/rspb.2023.2775

Huerta-Sánchez, E., Jin, X., Asan, Bianba, Z., Peter, B. M., Vinckenbosch, N., Liang, Y., Yi, X., He, M., Somel, M., Ni, P., Wang, B., Ou, X., Huasang, Luosang, J., Cuo, Z. X. P., Li, K., Gao, G., Yin, Y., … Nielsen, R. (2014). Altitude adaptation in Tibetans caused by introgression of Denisovan-like DNA. *Nature*, *512*(7513), 194–197. https://doi.org/10.1038/nature13408

Husby, A. (2022). Wild epigenetics: insights from epigenetic studies on natural populations. *Proceedings of the Royal Society B: Biological Sciences*, *289*(1968), 20211633. https://doi.org/10.1098/rspb.2021.1633

Kaluscha, S., Domcke, S., Wirbelauer, C., Stadler, M. B., Durdu, S., Burger, L., & Schübeler, D. (2022). Evidence that direct inhibition of transcription factor binding is the prevailing mode of gene and repeat repression by DNA methylation. *Nature Genetics*, *54*(12), 1895–1906. https://doi.org/10.1038/s41588-022-01241-6

Kapun, M., & Flatt, T. (2019). The adaptive significance of chromosomal inversion polymorphisms in Drosophila melanogaster. *Molecular Ecology*, *28*(6), 1263–1282. https://doi.org/https://doi.org/10.1111/mec.14871

Kapusta, A., Suh, A., & Feschotte, C. (2017). Dynamics of genome size evolution in birds and mammals. *Proceedings of the National Academy of Sciences*, *114*(8), E1460–E1469. https://doi.org/10.1073/pnas.1616702114

Kawakami, T., Strakosh, S. C., Zhen, Y., & Ungerer, M. C. (2010). Different scales of Ty1/copia-like retrotransposon proliferation in the genomes of three diploid hybrid sunflower species. *Heredity*, *104*(4), 341–350. https://doi.org/10.1038/hdy.2009.182

Kazazian, H. H., Wong, C., Youssoufian, H., Scott, A. F., Phillips, D. G., & Antonarakis, S. E. (1988). Haemophilia A resulting from de novo insertion of L1 sequences represents a novel mechanism for mutation in man. *Nature*, *332*(6160), 164–166. https://doi.org/10.1038/332164a0

Kent, T. V, Uzunović, J., & Wright, S. I. (2017). Coevolution between transposable elements and recombination. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *372*(1736), 20160458. https://doi.org/10.1098/rstb.2016.0458

Khurana, J. S., Wang, J., Xu, J., Koppetsch, B. S., Thomson, T. C., Nowosielska, A., Li, C., Zamore, P. D., Weng, Z., & Theurkauf, W. E. (2011). Adaptation to P Element Transposon Invasion in Drosophila melanogaster. *Cell*, *147*(7), 1551–1563. https://doi.org/10.1016/j.cell.2011.11.042

Kidwell, M. G., Kidwell, J. F., & Sved, J. A. (1977). Hybrid dysgenesis in Drosophila melanogaster: a syndrome of aberrant traits including mutation, sterility and male recombination. *Genetics*, *86*(4), 813–833. https://doi.org/10.1093/genetics/86.4.813

Kirkpatrick, M., & Barton, N. (2006). Chromosome Inversions, Local Adaptation and Speciation. *Genetics*, *173*(1), 419–434. https://doi.org/10.1534/genetics.105.047985

Knief, U., Müller, I. A., Stryjewski, K. F., Metzler, D., Sorenson, M. D., & Wolf, J. B. W. (2024). Evolution of Chromosomal Inversions across an Avian Radiation. *Molecular Biology and Evolution*, *41*(6), msae092. https://doi.org/10.1093/molbev/msae092

Kramerov, D. A., & Vassetzky, N. S. (2011). Origin and evolution of SINEs in eukaryotic genomes. *Heredity*, *107*(6), 487–495. https://doi.org/10.1038/hdy.2011.43

Kupiec, M., & Petes, T. D. (1988). Allelic and ectopic recombination between Ty elements in yeast. *Genetics*, *119*(3), 549–559. https://doi.org/10.1093/genetics/119.3.549

Langdon, Q. K., Groh, J. S., Aguillon, S. M., Powell, D. L., Gunn, T., Payne, C., Baczenas, J. J., Donny, A., Dodge, T. O., Du, K., Schartl, M., Ríos-Cárdenas, O., Gutiérrez-Rodríguez, C., Morris, M., & Schumer, M. (2024). Swordtail fish hybrids reveal that genome evolution is surprisingly predictable after initial hybridization. *PLOS Biology*, *22*(8), e3002742-. https://doi.org/10.1371/journal.pbio.3002742

Laporte, M., Le Luyer, J., Rougeux, C., Dion-Côté, A.-M., Krick, M., & Bernatchez, L. (2019). DNA methylation reprogramming, TE derepression, and postzygotic isolation of nascent animal species. *Sci. Adv.*, *5*(10), eaaw1644.

Lea, A. J., Tung, J., & Zhou, X. (2015). A Flexible, Efficient Binomial Mixed Model for Identifying Differential DNA Methylation in Bisulfite Sequencing Data. *PLOS Genetics*, *11*(11), e1005650-. https://doi.org/10.1371/journal.pgen.1005650

Lee, S., Barbour, J. A., Tam, Y. M., Yang, H., Huang, Y., & Wong, J. W. H. (2025). LocusMasterTE: integrating long-read RNA sequencing improves locus-specific quantification of transposable element expression. *Genome Biology*, *26*(1), 72. https://doi.org/10.1186/s13059-025-03522-9

Lerat, E., Goubert, C., Guirao-Rico, S., Merenciano, M., Dufour, A.-B., Vieira, C., & González, J. (2019). Population-specific dynamics and selection patterns of transposable element insertions in European natural populations. *Molecular Ecology*, *28*(6), 1506–1522. https://doi.org/https://doi.org/10.1111/mec.14963

Levin, H. L., & Moran, J. V. (2011). Dynamic interactions between transposable elements and their hosts. *Nature Reviews Genetics*, *12*(9), 615–627. https://doi.org/10.1038/nrg3030

Liebl, A. L., Schrey, A. W., Richards, C. L., & Martin, L. B. (2013). Patterns of DNA Methylation Throughout a Range Expansion of an Introduced Songbird. *Integrative and Comparative Biology*, *53*(2), 351–358. https://doi.org/10.1093/icb/ict007

Lira-Medeiros, C. F., Parisod, C., Fernandes, R. A., Mata, C. S., Cardoso, M. A., & Ferreira, P. C. G. (2010). Epigenetic Variation in Mangrove Plants Occurring in Contrasting Natural Environment. *PLOS ONE*, *5*(4), e10326-. https://doi.org/10.1371/journal.pone.0010326

Liu, B., & Zhao, M. (2023). How transposable elements are recognized and epigenetically silenced in plants? *Current Opinion in Plant Biology*, *75*, 102428. https://doi.org/https://doi.org/10.1016/j.pbi.2023.102428

Loyfer, N., Magenheim, J., Peretz, A., Cann, G., Bredno, J., Klochendler, A., Fox-Fisher, I., Shabi-Porat, S., Hecht, M., Pelet, T., Moss, J., Drawshy, Z., Amini, H., Moradi, P., Nagaraju, S., Bauman, D., Shveiky, D., Porat, S., Dior, U., … Kaplan, T. (2023). A DNA methylation atlas of normal human cell types. *Nature*, *613*(7943), 355–364. https://doi.org/10.1038/s41586-022-05580-6

Lynch, M., & Conery, J. S. (2003). The Origins of Genome Complexity. *Science*, *302*(5649), 1401–1404. https://doi.org/10.1126/science.1089370

Mager, D. L., & Goodchild, N. L. (1989). Homologous recombination between the LTRs of a human retrovirus-like element causes a 5-kb deletion in two siblings. *American Journal of Human Genetics*, *45*(6), 848.

Maheshwari, S., & Barbash, D. A. (2011). The genetics of hybrid incompatibilities. *Annu. Rev. Genet.*, *45*(1), 331–355.

Makarevitch, I., Waters, A. J., West, P. T., Stitzer, M., Hirsch, C. N., Ross-Ibarra, J., & Springer, N. M. (2015). Transposable Elements Contribute to Activation of Maize Genes in Response to Abiotic Stress. *PLOS Genetics*, *11*(1), e1004915-. https://doi.org/10.1371/journal.pgen.1004915

Massicotte, R., Whitelaw, E., & Angers, B. (2011). DNA methylation: A source of random variation in natural populations. *Epigenetics*, *6*(4), 421–427. https://doi.org/10.4161/epi.6.4.14532

McClintock, B. (1984). The Significance of Responses of the Genome to Challenge. *Science*, *226*(4676), 792–801. https://doi.org/10.1126/science.15739260

Merondun, J., & Wolf, J. B. W. (2025). DNA Methylation Reflects Cis-Genetic Differentiation Across the European Crow Hybrid Zone. *Molecular Ecology*, e70026. https://doi.org/https://doi.org/10.1111/mec.70026

Metcalfe, C. J., Bulazel, K. V, Ferreri, G. C., Schroeder-Reiter, E., Wanner, G., Rens, W., Obergfell, C., Eldridge, M. D. B., & O'Neill, R. J. (2007). Genomic Instability Within Centromeres of Interspecific Marsupial Hybrids. *Genetics*, *177*(4), 2507–2517. https://doi.org/10.1534/genetics.107.082313

Michalak, P. (2009). Epigenetic, transposon and small RNA determinants of hybrid dysfunctions. *Heredity (Edinb.)*, *102*(1), 45–50.

Milholland, B., Dong, X., Zhang, L., Hao, X., Suh, Y., & Vijg, J. (2017). Differences between germline and somatic mutation rates in humans and mice. *Nature Communications*, *8*(1), 15183. https://doi.org/10.1038/ncomms15183

Miousse, I. R., Chalbot, M.-C. G., Lumen, A., Ferguson, A., Kavouras, I. G., & Koturbash, I. (2015). Response of transposable elements to environmental stressors. *Mutation Research/Reviews in Mutation Research*, *765*, 19–39. https://doi.org/https://doi.org/10.1016/j.mrrev.2015.05.003

Mueller, S. A., Merondun, J., Lečić, S., & Wolf, J. B. W. (2025). Epigenetic variation in light of population genetic practice. *Nature Communications*, *16*(1), 1028. https://doi.org/10.1038/s41467-025-55989-6

Munasinghe, M., Read, A., Stitzer, M. C., Song, B., Menard, C. C., Ma, K. Y., Brandvain, Y., Hirsch, C. N., & Springer, N. (2023). Combined analysis of transposable elements and structural variation in maize genomes reveals genome contraction outpaces expansion. *PLOS Genetics*, *19*(12), e1011086-. https://doi.org/10.1371/journal.pgen.1011086

Nguyen, A. H., & Bachtrog, D. (2021). Toxic Y chromosome: Increased repeat expression and age-associated heterochromatin loss in male Drosophila with a young Y chromosome. *PLoS Genetics*, *17*(4 April 2021), 2020.07.21.214528. https://doi.org/10.1371/journal.pgen.1009438

Ni, P., Nie, F., Zhong, Z., Xu, J., Huang, N., Zhang, J., Zhao, H., Zou, Y., Huang, Y., Li, J., Xiao, C.-L., Luo, F., & Wang, J. (2023). DNA 5-methylcytosine detection and methylation phasing using PacBio circular consensus sequencing. *Nat. Commun.*, *14*(1), 4054.

Okada, N., Hamada, M., Ogiwara, I., & Ohshima, K. (1997). SINEs and LINEs share common 3' sequences: a review. *Gene*, *205*(1), 229–243. https://doi.org/https://doi.org/10.1016/S0378-1119(97)00409-5

Oliver, K. R., & Greene, W. K. (2012). Transposable elements and viruses as factors in adaptation and evolution: an expansion and strengthening of the TE-Thrust hypothesis. *Ecology and Evolution*, *2*(11), 2912–2933. https://doi.org/https://doi.org/10.1002/ece3.400

O'Neill, R. J. W., Eldridge, M. D. B., & Graves, J. A. M. (2001). Chromosome heterozygosity and de novo chromosome rearrangements in mammalian interspecies hybrids. *Mammalian Genome*, *12*(3), 256–259. https://doi.org/10.1007/s003350010270

Oppold, A.-M., Schmidt, H., Rose, M., Hellmann, S. L., Dolze, F., Ripp, F., Weich, B., Schmidt-Ott, U., Schmidt, E., Kofler, R., Hankeln, T., & Pfenninger, M. (2017). Chironomus riparius (Diptera) genome sequencing reveals

the impact of minisatellite transposable elements on population divergence. *Molecular Ecology*, *26*(12), 3256–3275. https://doi.org/https://doi.org/10.1111/mec.14111

Papoli Yazdi, H., Ravinet, M., Rowe, M., Sætre, G.-P., Guldvog, C. Ø., Eroukhmanoff, F., Marzal, A., Magallanes, S., & Runemark, A. (2022). Extensive transgressive gene expression in testis but not ovary in the homoploid hybrid Italian sparrow. *Molecular Ecology*, *31*(15), 4067–4077. https://doi.org/https://doi.org/10.1111/mec.16572

Park, Y., & Wu, H. (2016). Differential methylation analysis for BS-seq data under general experimental design. *Bioinformatics*, *32*(10), 1446–1453. https://doi.org/10.1093/bioinformatics/btw026

Payer, L. M., & Burns, K. H. (2019). Transposable elements in human genetic disease. *Nature Reviews Genetics*, *20*(12), 760–772. https://doi.org/10.1038/s41576-019-0165-8

Pecinka, A., Dinh, H. Q., Baubec, T., Rosa, M., Lettner, N., & Scheid, O. M. (2010). Epigenetic Regulation of Repetitive Elements Is Attenuated by Prolonged Heat Stress in Arabidopsis . *The Plant Cell*, *22*(9), 3118–3129. https://doi.org/10.1105/tpc.110.078493

Peona, V., Blom, M. P. K., Frankl-Vilches, C., Milá, B., Ashari, H., Thébaud, C., Benz, B. W., Christidis, L., Gahr, M., Irestedt, M., & Suh, A. (2022). The hidden structural variability in avian genomes. *BioRxiv*, 2021.12.31.473444. https://doi.org/10.1101/2021.12.31.473444

Peona, V., Blom, M. P. K., Xu, L., Burri, R., Sullivan, S., Bunikis, I., Liachko, I., Haryoko, T., Jønsson, K. A., Zhou, Q., Irestedt, M., & Suh, A. (2021). Identifying the causes and consequences of assembly gaps using a multiplatform genome assembly of a bird-of-paradise. *Molecular Ecology Resources*, *21*(1), 263–286. https://doi.org/10.1111/1755-0998.13252

Peona, V., Palacios-Gimenez, O. M., Blommaert, J., Liu, J., Haryoko, T., Jønsson, K. A., Irestedt, M., Zhou, Q., Jern, P., & Suh, A. (2021a). The avian W chromosome is a refugium for endogenous retroviruses with likely effects on female-biased mutational load and genetic incompatibilities. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *376*(1833), 20200186. https://doi.org/10.1098/rstb.2020.0186

Peona, V., Palacios-Gimenez, O. M., Blommaert, J., Liu, J., Haryoko, T., Jønsson, K. A., Irestedt, M., Zhou, Q., Jern, P., & Suh, A. (2021b). The avian W chromosome is a refugium for endogenous retroviruses with likely effects on female-biased mutational load and genetic incompatibilities. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *376*(1833), 20200186. https://doi.org/10.1098/rstb.2020.0186

Petrov, D. A., Schutzman, J. L., Hartl, D. L., & Lozovskaya, E. R. (1995). Diverse transposable elements are mobilized in hybrid dysgenesis in Drosophila virilis. *Proc. Natl. Acad. Sci. U. S. A.*, *92*(17), 8050–8054.

Pritam, S., & Signor, S. (2025). Evolution of piRNA-guided defense against transposable elements. *Trends in Genetics*, *41*(5), 390–401. https://doi.org/https://doi.org/10.1016/j.tig.2024.11.011

Rebollo, R., Horard, B., Hubert, B., & Vieira, C. (2010). Jumping genes and epigenetics: Towards new species. *Gene*, *454*(1–2), 1–7.

Rech, G. E., Radío, S., Guirao-Rico, S., Aguilera, L., Horvath, V., Green, L., Lindstadt, H., Jamilloux, V., Quesneville, H., & González, J. (2022). Population-scale long-read sequencing uncovers transposable elements associated with gene expression variation and adaptive signatures in Drosophila. *Nature Communications*, *13*(1), 1948. https://doi.org/10.1038/s41467-022-29518-8

Rey, O., Danchin, E., Mirouze, M., Loot, C., & Blanchet, S. (2016). Adaptation to Global Change: A Transposable

Element–Epigenetics Perspective. *Trends Ecol. Evol.*, *xx*, 1–13.

Ricci, M., Peona, V., Boattini, A., & Taccioli, C. (2023). Comparative analysis of bats and rodents' genomes suggests a relation between non-LTR retrotransposons, cancer incidence, and ageing. *Scientific Reports*, *13*(1), 9039. https://doi.org/10.1038/s41598-023-36006-6

Rogers, R. L. (2015). Chromosomal Rearrangements as Barriers to Genetic Homogenization between Archaic and Modern Humans. *Molecular Biology and Evolution*, *32*(12), 3064–3078. https://doi.org/10.1093/molbev/msv204

Roussel, A.-J., Suh, A., Ruiz-Ruano, F. J., & Dion-Côté, A.-M. (2025). Accumulation of a Biparentally Inherited Neptune Transposable Element in Natural Killifish Hybrids (Fundulus diaphanus × F. heteroclitus). *Molecular Biology and Evolution*, *42*(11), msaf266. https://doi.org/10.1093/molbev/msaf266

Ruiz-Arenas, C., & González, J. R. (2017). Redundancy analysis allows improved detection of methylation changes in large genomic regions. *BMC Bioinformatics*, *18*(1), 553. https://doi.org/10.1186/s12859-017-1986-0

Runemark, A., Fernández, L. P., Eroukhmanoff, F., & Sætre, G.-P. (2018). Genomic Contingencies and the Potential for Local Adaptation in a Hybrid Species. *The American Naturalist*, *192*(1), 10–22. https://doi.org/10.1086/697563

Schaack, S., Gilbert, C., & Feschotte, C. (2010). Promiscuous DNA: horizontal transfer of transposable elements and why it matters for eukaryotic evolution. *Trends in Ecology & Evolution*, *25*(9), 537–546. https://doi.org/10.1016/j.tree.2010.06.001

Schwarz, R., Koch, P., Wilbrandt, J., & Hoffmann, S. (2022). Locus-specific expression analysis of transposable elements. *Briefings in Bioinformatics*, *23*(1), bbab417. https://doi.org/10.1093/bib/bbab417

Seehausen, O. L. E., Takimoto, G., Roy, D., & Jokela, J. (2008). Speciation reversal and biodiversity dynamics with hybridization in changing environments. *Molecular Ecology*, *17*(1), 30–44. https://doi.org/https://doi.org/10.1111/j.1365-294X.2007.03529.x

Sentmanat, M. F., & Elgin, S. C. R. (2012). Ectopic assembly of heterochromatin in Drosophila melanogaster triggered by transposable elements. *Proceedings of the National Academy of Sciences*, *109*(35), 14104–14109. https://doi.org/10.1073/pnas.1207036109

Siomi, M. C., Sato, K., Pezic, D., & Aravin, A. A. (2011). PIWI-interacting small RNAs: the vanguard of genome defence. *Nat. Rev. Mol. Cell Biol.*, *12*(4), 246–258.

Slotkin, R. K., & Martienssen, R. (2007). Transposable elements and the epigenetic regulation of the genome. *Nat. Rev. Genet.*, *8*(4), 272–285.

Stryjewski, K. F., & Sorenson, M. D. (2017). Mosaic genome evolution in a recent and rapid avian radiation. *Nature Ecology & Evolution*, *1*(12), 1912–1922. https://doi.org/10.1038/s41559-017-0364-7

Sun, M., Wolf, G., Wang, Y., Senft, A. D., Ralls, S., Jin, J., Dunn-Fletcher, C. E., Muglia, L. J., & Macfarlan, T. S. (2021). Endogenous Retroviruses Drive Lineage-Specific Regulatory Evolution across Primate and Rodent Placentae. *Molecular Biology and Evolution*, *38*(11), 4992–5004. https://doi.org/10.1093/molbev/msab223

Taşkesen, M., Collin, G. B., Evsikov, A. V, Güzel, A., Özgül, R. K., Marshall, J. D., & Naggert, J. K. (2012). Novel Alu retrotransposon insertion leading to Alström syndrome. *Human Genetics*, *131*(3), 407–413. https://doi.org/10.1007/s00439-011-1083-9

Taskopru, E., Bacchi, M., Overheul, G. J., Linthout, C., Koenraadt, C. J. M., Qu, J., Miesen, P., Halbach, R., & van Rij, R. P. (2024). The piRNA pathway mediates transcriptional silencing of LTR retrotransposons in the soma and germline of Aedes mosquitoes. *BioRxiv*, 2024.11.28.623418.

Taylor, S. A., & Larson, E. L. (2019). Insights from genomes into the evolutionary importance and prevalence of hybridization in nature. *Nature Ecology & Evolution*, *3*(2), 170–177. https://doi.org/10.1038/s41559-018-0777-y

The Heliconius Genome Consortium. (2012). Butterfly genome reveals promiscuous exchange of mimicry adaptations among species. *Nature*, *487*(7405), 94–98. https://doi.org/10.1038/nature11041

Tóth, K. F., Pezic, D., Stuwe, E., & Webster, A. (2016). The piRNA pathway guards the germline genome against transposable elements. *Adv. Exp. Med. Biol.*, *886*, 51–77.

Trier, C. N., Hermansen, J. S., Sætre, G.-P., & Bailey, R. I. (2014). Evidence for Mito-Nuclear and Sex-Linked Reproductive Barriers between the Hybrid Italian Sparrow and Its Parent Species. *PLOS Genetics*, *10*(1), e1004075-. https://doi.org/10.1371/journal.pgen.1004075

Tusso, S., Suo, F., Liang, Y., Du, L.-L., & Wolf, J. B. W. (2021). *Reactivation of transposable elements following hybridization in fission yeast*.

Ungerer, M. C., & Kawakami, T. (2013). Transcriptional Dynamics of LTR Retrotransposons in Early Generation and Ancient Sunflower Hybrids. *Genome Biology and Evolution*, *5*(2), 329–337. https://doi.org/10.1093/gbe/evt006

Vagin, V. V., Sigova, A., Li, C., Seitz, H., Gvozdev, V., & Zamore, P. D. (2006). A distinct small RNA pathway silences selfish genetic elements in the germline. *Science*, *313*(5785), 320-324.

Wallace, M. R., Andersen, L. B., Saulino, A. M., Gregory, P. E., Glover, T. W., & Collins, F. S. (1991). A de novo Alu insertion results in neurofibromatosis type 1. *Nature*, *353*(6347), 864–866. https://doi.org/10.1038/353864a0

Wang, R. J., White, M. A., & Payseur, B. A. (2015). The Pace of Hybrid Incompatibility Evolution in House Mice. *Genetics*, *201*(1), 229–242. https://doi.org/10.1534/genetics.115.179499

Warmuth, V. M., Weissensteiner, M. H., & Wolf, J. B. W. (2022). Accumulation and ineffective silencing of transposable elements on an avian W Chromosome. *Genome Research*, *32*(4), 671–681. https://doi.org/10.1101/gr.275465.121

Weissensteiner, M. H., Bunikis, I., Catalán, A., Francoijs, K. J., Knief, U., Heim, W., Peona, V., Pophaly, S. D., Sedlazeck, F. J., Suh, A., Warmuth, V. M., & Wolf, J. B. W. (2020). Discovery and population genomics of structural variation in a songbird genus. *Nature Communications*, *11*(1), 3403. https://doi.org/10.1038/s41467-020-17195-4

Wolf, J. B. W., & Ellegren, H. (2017). Making sense of genomic islands of differentiation in light of speciation. *Nature Reviews Genetics*, *18*(2), 87–100. https://doi.org/10.1038/nrg.2016.133

Xia, B., Zhang, W., Zhao, G., Zhang, X., Bai, J., Brosh, R., Wudzinska, A., Huang, E., Ashe, H., Ellis, G., Pour, M., Zhao, Y., Coelho, C., Zhu, Y., Miller, A., Dasen, J. S., Maurano, M. T., Kim, S. Y., Boeke, J. D., & Yanai, I. (2024). On the genetic basis of tail-loss evolution in humans and apes. *Nature*, *626*(8001), 1042–1048. https://doi.org/10.1038/s41586-024-07095-8

Zamudio, N., Barau, J., Teissandier, A., Walter, M., Borsos, M., Servant, N., & Bourc'his, D. (2015). DNA

methylation restrains transposons from adopting a chromatin signature permissive for meiotic recombination. *Genes & Development*, *29*(12), 1256–1270. https://doi.org/10.1101/gad.257840.114

Zhou, S., Butler-Laporte, G., Nakanishi, T., Morrison, D. R., Afilalo, J., Afilalo, M., Laurent, L., Pietzner, M., Kerrison, N., Zhao, K., Brunet-Ratnasingham, E., Henry, D., Kimchi, N., Afrasiabi, Z., Rezk, N., Bouab, M., Petitjean, L., Guzman, C., Xue, X., … Richards, J. B. (2021). A Neanderthal OAS1 isoform protects individuals of European ancestry against COVID-19 susceptibility and severity. *Nature Medicine*, *27*(4), 659–667. https://doi.org/10.1038/s41591-021-01281-1