UPPSALA
UNIVERSITET

Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology 1851

Adaptive divergence in fission yeast

From experimental evolution to evolutionary genomics

SERGIO IVAN TUSSO GOMEZ

ACTA
UNIVERSITATIS
UPSALIENSIS
UPPSALA
2019

ISSN 1651-6214
ISBN 978-91-513-0743-5
urn:nbn:se:uu:diva-392422

Dissertation presented at Uppsala University to be publicly examined in Lindahlsalen, Evolutionsbiologiskt centrum, Norbyvägen 18A , Uppsala, Friday, 25 October 2019 at 13:00 for the degree of Doctor of Philosophy. The examination will be conducted in English. Faculty examiner: Professor Roger K. Butlin (Universities of Sheffield, UK).

Abstract
Tusso Gomez, S. I. 2019. Adaptive divergence in fission yeast. From experimental evolution to evolutionary genomics. *Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology* 1851. 68 pp. Uppsala: Acta Universitatis Upsaliensis. ISBN 978-91-513-0743-5.

How adaptation and population differentiation occur is fundamental to understand the origin of biodiversity. Work in speciation alongside the increased ease of generating genomic data have allowed the exploration of genomic changes relevant to adaptation. However, it remains challenging to infer the underlying mechanisms from genomic patterns of divergence governed by both genomic properties and external selective pressures. The chronological order of genomic changes, evolutionary history and selective forces can rarely be inferred from natural populations.

Currently, I see two promising ways to tackle the problem of the genomic underpinnings of divergence: (1) evolution experiments simulating adaptation and population divergence and measuring genomic changes as they occur through time; (2) empirical studies of closely related populations in which the extent of divergence varies, allowing us to infer the chronology of the genomic changes. In my Ph.D. research I applied these two approaches, using the fungus *Schizosaccharomyces pombe*. First, I experimentally tested the potential for ecological divergence with gene flow, and investigated genomic and phenotypic changes associated with this process. Next, I studied genomic data obtained from natural populations sampled worldwide. In both cases, the genetic inference relied on different sequencing technologies including the Illumina, Pacific Biosciences and Oxford Nanopore platforms.

The experiment explored the effect of gene flow on phenotype and fitness, and uncovered potential molecular mechanisms underlying adaptive divergence. In **paper I** we demonstrate the emergence of specialisation under low gene flow, but generalist strategies when gene flow was high. Evolved phenotypes were largely influenced by standing genetic variation subject to opposite antagonistic pleiotropy complemented by new mutations enriched in a subset of genes. In **paper II**, we show that the experimental selective regime also had an effect on mating strategies, result of temporal ecological heterogeneity and selection for mating efficiency. We found that the evolution of mating strategies was explained by a trade-off between mating efficiency and asexual growth rate dependent on environmental stability. **Papers III** and **IV** consider the role of gene flow in natural populations. In **paper III,** we provide evidence that gene flow also played a predominant role in adaptive divergence in nature. All strains resulted from recent hybridization between two ancestral groups manifested in large phenotypic variation and reproductive isolation.This demographic history of hybridization was confirmed in **paper IV** focusing on patterns of mitochondrial diversity, adding evidence for the geographic distribution of the ancestral populations and potential for horizontal gene transfer from a distant yeast clade.

Keywords: adaptation, divergence, selection, experimental evolution, genomics, genome evolution, population genetics, fission yeast, phenotypic variation

Sergio Ivan Tusso Gomez, Department of Ecology and Genetics, Evolutionary Biology, Norbyvägen 18D, Uppsala University, SE-75236 Uppsala, Sweden.

ISSN 1651-6214
ISBN 978-91-513-0743-5
urn:nbn:se:uu:diva-392422 (http://urn.kb.se/resolve?urn=urn:nbn:se:uu:diva-392422)

To my parents

List of Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals.

I **Tusso, S**., Nieuwenhuis, BPS., Immler, S., Wolf, JBW. (-) Parallelism in adaptive divergence and the modulating effect of gene flow. *Manuscript.*

II Nieuwenhuis, BPS.*, **Tusso, S.***, Bjerling, P., Stångberg, J., Wolf, JBW.**, Immler, S.** (2018) Repeated evolution of self-compatibility for reproductive assurance. *Nature Communications*, 9(1):1639

III **Tusso, S.**, Nieuwenhuis, BPS., Sedlaczeck, FJ., Jeffares, D., Wolf, JBW. (2019) Ancestral admixture is the main determinant of global biodiversity in fission yeast. *Molecular Biology and Evolution*, 36(9):1975-1989

IV Tao, YT., Suo, F., **Tusso, S.**, Wang, YK., Huang, S., Wolf, JBW., Du, Li-Lin. (2019) Intraspecific diversity of fission yeast mitochondrial genome. *Genome Biology and Evolution*, 11(8): 2312-2329

* These authors contributed equally to the study.

The following papers were published or submitted during the course of my doctoral studies, but are not part of this thesis.

Peart, CR.*, **Tusso, S.***, Pophaly, SD.*, Botero-Castro, F., Wu, C., Aurioles-Gamboa, D., Baird, A., Bickham, J., Forcada, J., Galimberti, F., Gelatt, TS., Gemmell, NJ., Hoffman, JI., Kovacs, KM., Kunnasranta, M., Lydersen, C., Nyman, T., Oliviera, LR., Orr, AJ., Sanvito, S., Trillmich, F., Valtonen, M., Shafer, ABA., Wolf, JBW. (-) Determinants of genetic variation across eco-evolutionary scales and their implications for the Anthropocene. *Submitted.*

Ekkers, DM., **Tusso, S.**, Moreno-Gomez, S., Rillo, MC., Kuipers, OP., Sander van Doorn, G. (-) Metabolic architecture gives rise to defines trade-offs that driveforce specialization and adaptive diversification in *Lactococcus lactis*. *Submitted*.

Amaya-Márquez M., **Tusso, S.**, Hernández, J., Jiménez, JD., Wells, H., Abramson, C. (-) Olfactory learning in the stingless bee *Melipona eburnea* Friese (Apidae: Meliponini). *Submitted*.

Shafer, ABA.*, Peart, CR.*, **Tusso, S.**, Maayan, I., Brelsford, A., Wheat, CW., Wolf, JBW. (2017) Bioinformatic processing of RAD-seq data dramatically impacts downstream population genetic inference. *Methods in Ecology and Evolution*, 8:907–917

Hoyos, M., **Tusso, S.**, Bedoya, TR., Gaviria, ASM., Bloor, P. (2017) A simple and cost-effective method for obtaining DNA from a wide range of animal wildlife samples. *Conservation Genetics Resources*, 9:513–521

Loire, E.*, **Tusso, S.***, Caminade, P., Severac, D., Boursot, P., Ganem, G., Smadja, CM. (2017) Do changes in gene expression contribute to sexual isolation and reinforcement in the house mouse? *Molecular Ecology*, 26:5189–5202

Promerová, M., Alavioon, G., **Tusso, S.**, Burri, R., Immler, S. (2017) No evidence for MHC class II-based non-random mating at the gametic haplotype in Atlantic salmon. *Heredity*, 118:563–567

Tusso, S., Morcinek, K., Vogler, C., Schupp, PJ., Caballes, CF., Vargas, S., Wörheide, G. (2016) Genetic structure of the crown-of-thorns seastar in the Pacific Ocean, with focus on Guam. *PeerJ*, 4:e1970.

* These authors contributed equally to the study.

Abbreviations

DNA	Deoxyribonucleic acid
SNP	Single nucleotide polymorphism
bp, kb, Mb	Base pair, Kilo base, Mega base
SV	Structural variation
Indel	Insertion / Deletion
TE	Transposable element
LTR	Long terminal region
R^2	Coefficient of determination
PCA	Principal component analyses
π	Nucleotide diversity
θ_w	Watterson theta
Taj's D	Tajima's D
μ	Mutation rate
N_e	Effective population size
D_{xy}	Average number of pairwise differences
LD	Linkage disequilibrium
DSB	DNA double-strand breaks
PCR	Polymerase chain reaction
SW	Switcher
NS	Non-Switcher

Contents

Introduction

There are millions of species in nature constituting the biodiversity on the planet. But, how did this biodiversity arise? And how does it continue to do so? What are the inherent features and characteristics of a population that can lead to differentiation and subsequently the origin of new species? Which environmental factors, if any, are driving this differentiation? Several decades after the merger of Charles Darwin's seminal work 'On the Origin of Species' with Mendel's insight into heritable principles into the Evolutionary Synthesis, the study of adaptation and speciation remain a central priority in evolutionary biology. While recent empirical work has begun to describe the genomics of early species diversification, the nature of underlying evolutionary processes remains a subject of debate.

Influenced by the rapid development in sequencing technology, genome-scale studies have become common place in evolutionary biology research. Genome-wide population surveys of natural populations have provided significant insight into the evolutionary processes and the genetic basis underlying adaptation and speciation (Savolainen, Lascoux, and Merilä 2013; Seehausen et al. 2014). By studying patterns of segregating genetic variation across the entire genomes of multiple individuals and populations sampled in their natural environment, evolutionary processes governing the accumulation of genetic differences between incipient evolutionary lineages can be inferred (Ravinet et al. 2017; Wolf and Ellegren 2017). However, this inference remains challenging, as several factors from both intrinsic genomic properties and external selective pressures can interact producing similar genomic signatures. In general, selective forces directing the evolutionary process and the chronological order of genomic changes can rarely be inferred in natural populations. In this thesis, I used two strategies to disentangle intrinsic and extrinsic evolutionary drivers: 1) evolutionary experiments to simulate the diversification process and documenting phenotypic and genomic changes as they occur through time and 2) empirical case studies of closely related taxa or populations with varying divergence levels, to infer the evolutionary history, the temporal order of the genomic changes and the influence of different selective pressures and genomic features in diversification.

In my research, I took both of these approaches using the fungus *Schizosaccharomyces pombe* (commonly known as fission yeast) as a model system. Through experimental evolution, I aimed to gather evidence for the possibility of ecological adaptation and divergence with gene flow and uncovered the potential molecular mechanisms underlying the process (Paper I). I found that the applied experimental selective regime had strong effects on the evolution of mating strategies resulting from temporal ecological heterogeneity and selection for mating efficiency (Paper II). I found that historic gene flow has also played a predominant role for adaptation and differentiation in natural populations. I demonstrated that all known strains within the species resulted from recent hybridization between two ancestral groups which manifested in large effects on phenotypic variation and reproductive isolation (Paper III). The resulting demographic history observed in the nuclear genome was corroborated by patterns of mitochondrial genetic diversity, adding evidence for the geographic distribution of the ancestral populations and potential for horizontal gene transfer from a distant yeast clade (Paper IV).

Local adaptation

Understanding the origin of genetic variation and how it is translated into phenotypic variation are central topics in speciation genomics. New mutations transmitted between generations are subjected to genetic drift and/or selection, resulting in changes in the frequency of genetic variants within populations. New mutations that confer an environmental advantage or generally an increase in fitness (beneficial mutations) will have higher chance of overcoming drift, being positively selected and increasing in frequency in the population. Conversely, deleterious mutations will tend to be reduced in frequency and eventually be lost in the population. Over time and in a stable environment, the genetic composition of the population (*genetic pool*) will tend to accumulate neutral variants, but also shift towards high frequency of genotypes that confer an adaptive advantage to the specific environmental conditions relative to the ancestral state.

Variation to fuel adaptation can thus not only arise from new emerging mutations (e.g. Linnen et al. 2013; Laurent et al. 2016), but also from standing genetic variants already present in the population from the past (*standing genetic variation*) (e.g. Feder et al. 2003; Jones et al. 2012; Monnahan, Colicchio, and Kelly 2015) or acquired by introgression from other populations (e.g. Hedrick 2013; Clarkson et al. 2014; Norris et al. 2015; Leducq et al. 2016). Selection on standing genetic variation presents several features relative to adaptation from new mutations, which have led to the hypothesis that standing variation may be the main source of variation during the initial steps of adaptation. For instance, 1) as alleles are already present, evolution

can occur faster; 2) under new environments, the higher allele frequencies of existing standing variation reduce the probability for variants to be lost by genetic drift; and 3) the adaptive value of these variants could have been tested previously in alternative temporal space contexts or other populations (Barrett and Schluter 2008; Tigano and Friesen 2016).

Genetic variants additionally differ in the effect size they exert on fitness. Variants with large effect are predicted to contribute more to adaptation and to be more resistant to loss by genetic drift (Yeaman and Otto 2011). However, simulation studies have shown that small effect variants could also substantially contribute to adaptation if there is high standing genetic variation (Yeaman 2015). In addition to effect size, the fixation or loss of genetic variants will depend of genetic architecture of the trait to which they contribute. For instance, quantitative traits can be defined by multiple loci, but each locus can additionally have multiple contributing alleles in the population. High genetic variation would thus result in a larger pool of allelic combinations from which selection could act.

Progress in understanding multilocus adaptation has paved the way for a thorough conceptual description of its role in evolution. Simulations by Franssen et al., (2017) addressing the role of genetic architecture in multilocus adaptation suggest three main phases during adaptation of quantitative traits: during the first phase, all adaptive alleles increase in frequency, but only until the mean population phenotype reaches a fitness optimum. As a phenotype can be reached by different combinations of multiple alleles and locus, individual genetic variants do not necessarily reach fixation. In the second phase, phenotypic variation is maximised, and no changes in allele frequency are observed. During the last phase, there is variation in allele frequency, but changes are stochastic since adaptive loci will go to fixation or extinction by drift, without changes in mean population fitness. Other models have shown additionally that: 1) the number of contributing variants decreases with an increase in recombination rate, while their effect tend to decrease; 2) small effect variants tend to cluster in linked groups with larger effect variants; 3) an increase in selection allows for larger physical dispersion of contributing linked clusters (Yeaman and Whitlock 2011); and 4) when the number of adaptive alleles is large, the expected allele frequency changes are small (Franssen, Kofler, and Schlötterer 2017).

Understanding multilocus adaptation also faces methodological challenges. Observed allele changes in natural populations are small for the most part, suggesting dominance of small effect variants, which limits the power to detect those adaptive variants from stochastic changes with existing tools (Yeaman 2015; Tigano and Friesen 2016). The most promising strategy to study quantitative adaptive traits is probably to combine multiple approach-

es. A synthetic view of quantitative traits is possible by combining evidence from gene enrichment analyses using annotation of biological pathways (Daub et al. 2013), information from changes in allele frequency under differential selection (Latta 1998; Le Corre and Kremer 2012; Bourret, Dionne, and Bernatchez 2014), genotype-phenotype association tests (Stephan, Stegle, and Beyer 2015), and genotype – environment and geography associations (Hancock et al. 2011; Günther and Coop 2013; Berg and Coop 2014). Regardless of the technical limitations, several empirical studies have provided insights into the genomics of adaptation. For example, the role of standing genetic variation in population differentiation has been tested in several systems (e.g. sticklebacks Feulner et al. 2013; cichlids Fan and Meyer 2014; pea aphids Duvaux et al. 2015; and monkey flowers Flagel, Willis, and Vision 2014). One important contribution of these studies is that in addition to single nucleotide polymorphisms (SNPs), other type of genetic variants were found to contribute to local adaptation, including structural variants (insertions, deletions, inversions and translocations) and copy number variation (described in more detail below).

Gene flow

The maintenance of genetic and phenotypic variation within a population largely depends on environmental heterogeneity, the effect size distribution of mutations and gene flow (Yeaman and Jarvis 2006). *Gene flow* refers to the movement of alleles and their establishment into a different gene pool (Endler 1977). The importance of gene flow in evolutionary biology has been studied since early 1930s, primarily with theoretical work (e.g. Haldane and Haldane 1930; Wright 1931; Slatkin 1987). With the emergence of high-throughput sequencing technologies, tools and methods for quantification, empirical studies of gene flow have lately gained momentum (Wolf, Lindell, and Backström 2010; Feder, Egan, and Nosil 2012).

Progress in characterizing gene flow on adaptation and speciation has brought unprecedented challenges. For instance, understanding the effect of gene flow in the context of adaptation requires knowledge on the genetic basis of adaptive traits, and the correlation between them and their fitness effects, which frequently are not clear (Barrett and Hoekstra 2011; Tigano and Friesen 2016).However, the progress in understanding the effect of gene flow on adaptation and speciation has brought unprecedented challenges. For instance, understanding the effect of gene flow in the context of adaptation requires characterization of the genetic basis of adaptive traits, and the correlation between them and their fitness effects, which frequently are not clear (Barrett and Hoekstra 2011; Tigano and Friesen 2016).

Gene flow can impact adaptation in three ways: first, interbreeding between individuals from a single panmictic population, differentiating by a polymorphic trait or genomic region reducing competition (adaptive divergence); second, when there is gene flow between separate, but interconnected, populations which are adapting locally (ecological divergence); or thirdly, when gene flow occurs after populations have substantially diverged (secondary contact or hybridization). In general, as described above, spatially heterogeneous selection regimes will tend to increase overall genetic divergence, but it is counteracted by the homogenising effect of gene flow. To complicate matters, temporal environmental variation also has shown to play a key role. Simulations suggest that intermediate levels of gene flow result in maximum local adaptation if there is temporal environmental variation (Blanquart and Gandon 2011).

Ecological divergence in the presence of gene flow is theoretically conditional to the nature of underlying fitness costs to local adaptation (Felsenstein 1981; Maclean 2005; Martin, Otto, and Lenormand 2006; Agrawal 2009; Smadja and Butlin 2011). For instance, under strong divergent selection and low migration rate, hybrids between ecologically differentiated populations are rapidly removed. The production of un-adapted hybrids incurs an evolutionary cost which will ultimately favour the selection for recombination suppressors and reproductive barriers. Inversely, an increase in gene-flow will homogenise populations, erode local adaption and reduce the efficiency of local adaptation by exchanging locally adapted alleles, and impede the coupling of variants needed for local adaptation (Balkau and Feldman 1973; Lenormand 2002; Blanquart and Gandon 2011). Ecological divergence in the presence of gene flow is theoretically conditional to the nature of underlying fitness costs to local adaptation (Felsenstein 1981; Maclean 2005; Martin, Otto, and Lenormand 2006; Agrawal 2009; Smadja and Butlin 2011). For instance, under strong divergent selection and low migration rate, hybrids between ecologically differentiated populations are rapidly removed. The production of un-adapted hybrids incurs an evolutionary cost which will ultimately favour the selection for recombination suppressors and reproductive barriers. Inversely, an increase in gene-flow will homogenise populations, eroding local adaption and reducing the efficiency of local adaptation by exchanging locally adapted alleles, and impeding the coupling of variants needed for local adaptation (Balkau and Feldman 1973; Lenormand 2002; Blanquart and Gandon 2011).

Different models have been studied as to how genetic variation of adapting populations is shaped as a function to gene flow and the underling genetic background (e.g. Bürger and Akerman 2011; Yeaman and Otto 2011; Feder et al. 2012). For example, simulations show that the probability for a beneficial mutation to establish in a population depends primarily on the strength

of selection (*s*) and the rate of gene flow (*m*). Without gene flow the probability of establishment depends solely on selection coefficient of the new mutation (s/2; Feder et al. 2012). With gene flow, a new beneficial mutation has a higher probability of fixation when *s* is higher than *m.* If this is not the case, the new variant will be swamped from the local genetic pool (Haldane and Haldane 1930; Wright 1931; Lenormand 2002; Yeaman and Otto 2011; Feder et al. 2012). In the presence of linkage, if selection on a new mutation is lower than gene flow, but linked to a differentiated locus under strong selection (s > 2m; Feder et al. 2012), the fixation probability of the first mutation is increased. Thus, in the presence of gene flow, adaptation from standing genetic variation is governed by the complex interaction of selection, migration and recombination.

Hybridization

Hybrids are organisms carrying genetic material from two differentiated, parental genomes, displaying a range of mixed traits present in the original parent lineages, as well as emerging new properties, including novel phenotypes. Events of natural hybridization between diverging lineages have been extensively recognised as an important evolutionary process and key element for diversification (Mallet 2005; Abbott et al. 2013; Gladieux et al. 2014; Mixão and Gabaldón 2018). Hybridization has been observed in a broad range of taxonomic systems including plants, animals and fungi with diverse geographical distributions (Mallet 2005; Abbott et al. 2013; Abbott 1992; Arnold 1992; Seehausen 2004; Grant, Grant, and Petren 2005; Schumer, Rosenthal, and Andolfatto 2014). A few intensively studied examples of hybridization have shown its role for adaptive radiation, transfer of adaptive loci within and between species, and the formation of new species (Rieseberg 2003; Fonseca et al. 2004; Seehausen 2004; Masuelli et al. 2009; Lee et al. 2013; Lunt et al. 2014). In fungi, hybridization has not been as extensively studied as in other groups, but recent genomic technologies have enabled the identification of several fungal hybrids, belonging to diverse clades (Morales and Dujon 2012; Pryszcz et al. 2014; 2015; Leducq et al. 2016).

Transgressive traits are a common feature of hybridization, corresponding to those traits with range variation beyond the observed range in either of the parental clades. A well-known example of this is the sunflower *Helianthus paradoxus*, a hybrid species between *H. annuus* and *H. petiolaris* (Welch and Rieseberg 2002a; 2002b). In this case, the hybrid species can survive under soil conditions with higher salt concentration relative to both parental species. This enables hybrids to thrive under high salinity environments unsuitable for either parental species. Transgressive phenotypes are common in

hybrids, feature that has been exploited during the domestication of several hybrid species, including several crops, and yeast strains for beer and wine (Pretorius and Bauer 2002; Warschefsky et al. 2014; Gibson and Liti 2015; Bevan et al. 2017; Mixão and Gabaldón 2018).

Even though an increasing number of examples for adaptive hybridization are emerging, it is also expected that early stages of hybridization are physiologically unfit, primarily due to the interaction between differentiated genomic components that have been evolving independently (known as *genomic shock*) (McClintock 1984). Even if new combinations could result in an adaptive advantage for hybrids, the persistence of hybrid forms will depend on purging existing deleterious interactions. As suggested by the Bateson–Dobzhansky–Muller model (Dobzhansky 1934; Muller 1942; Bateson 1909), the admixture between divergent genetic backgrounds can result in incompatibilities which can then influence the survival or fertility of the hybrid individual (*BDM-incompatibilities*). Incompatible allelic combinations that result in negative fitness effects, will be removed by selection, which prevent further mixing of those specific genomic regions. These regions are of particular interest, because theoretical and empirical studies suggest that the reduction in gene flow in those genomic regions could allow for the accumulation of additional mutations over time, causing further divergence and potentially eventual speciation (Wu 2001; Presgraves 2010). The identification of these incompatibilities could provide insights into the mechanisms driving isolation and adaptive divergence (Orr 1995; Wolf and Ellegren 2017). Selection acting against recombination between incompatible variants will change haplotype frequencies in the population such that patterns of linkage disequilibrium (LD) are distorted in a specific manner. Systematic analysis of LD distortion along the genome can therefore be used to identify candidate BDM-incompatibilities during hybridization (Colomé-Tatché and Johannes 2016).

Considering both adaptive and detrimental genomic variation being combined in hybrid genomes, we expect hybrids to show differential contribution of genes from parental linages in the long run (Grant, Grant, and Petren 2005; Bacilieri et al. 1996; McDonald et al. 2001; Thulin and Tegelström 2002). This process is known as *genome stabilisation* and can either be facilitated or impeded by mechanisms such as genetic recombination, gene conversion, or structural variation (Payseur and Rieseberg 2016). Note, however, that imbalance in the parental contribution is not only determined by selection, but also defined by asymmetry in introgression from the most common to the most rare lineage (Dowling, Smith, and Brown 1989; Taylor and Hebert 1993). Differential gene flow produces variation among hybrids and across the genome; variation upon which selection can act (Sankararaman et al. 2014; Schumer et al. 2016). This will have important consequences on

long-term hybrid persistence, adaptation and consequently speciation (Rieseberg 2003; Chiba 1993; Ortíz-Barrientos et al. 2002; Servedio and Saetre 2003; Gee 2004; Saetre et al. 2008).

Trade-Offs

Thus far, my focus has been on a single trait or a set of independent traits under selection. The interaction between traits in the context of selection is another important component for several evolutionary models for diversification, particularly those attempting to understand the evolution of life history traits. Trade-off refer to the increase in fitness given by the change in one trait, while decreasing fitness in a second trait (Roff and Fairbairn 2007). It is of general belief within evolutionary biology that trait evolution is commonly restricted by fitness trade-offs (Lynch and Gabriel 1987; Charnov 1989; Stearns 1992; Roff 1993; Whitlock 1996; Reznick, Nunney, and Tessier 2000; Roff 2002; Spichtig and Kawecki 2004; Roff and Fairbairn 2007; Bono et al. 2017). Fitness trade-offs between life history traits have been described in several studies, both in the laboratory and natural populations, commonly measured as the statistical correlation between traits (e.g. Bartlett, Wilfert, and Boots 2018; Lasne, Leblanc, and Gillet 2018; Mariette et al. 2018; Pinheiro-Costa et al. 2018; Støstad et al. 2019; Zhu et al. 2019). However, little is known on how trade-offs evolve (Roff and Fairbairn 2007; Bono et al. 2017).

At the genetic level, trade-offs can either be driven by selection on genetic variants that are neutral in alternative environments (conditional neutrality) (Reboud and Bell 1997; MacLean and Bell 2002; Ostrowski, Ofria, and Lenski 2007; Behe 2010; Anderson et al. 2013; Kvitek and Sherlock 2013; Samani and Bell 2016), or are detrimental in alternative environments (antagonistic pleiotropy) (Cooper and Lenski 2000; Lunzer et al. 2002; Nosil and Crespi 2004; Craig Maclean 2005; Gray and Goddard 2012; Hietpas et al. 2013; Samani and Bell 2016). Given the lack of fitness effects of variants under conditional neutrality in alternative environments, it is not expected these variants will contribute to adaptive polymorphism within populations, but rather to transient variation between populations in the presence of gene flow (Fry 1996; Kawecki 1997; Bono et al. 2017). Therefore, conditionally neutral variants are expected to be homogenised over time (Hall, Lowry, and Willis 2010; Fournier-Level et al. 2011). Under antagonistic pleiotropy, variants positively selected in one environment are selected against in alternative ecological conditions (Cooper and Lenski 2000; Lunzer et al. 2002; Nosil and Crespi 2004; Cooper and Lenski 2010; Hietpas et al. 2013; Samani and Bell 2016).

Although empirical data are still limited to be able to make general conclusions, contrary to expectations, experiments and natural populations more frequently show more evidence for conditional neutrality than antagonistic pleiotropy (e.g. Reboud and Bell 1997; MacLean and Bell 2002; Weinig et al. 2003; Gardner and Latta 2006; Verhoeven et al. 2008; Lowry et al. 2009; Latta, Gardner, and Staples 2010; Anderson et al. 2013; Leinonen et al. 2013; Remington et al. 2013; Soudi, Reinhold, and Engqvist 2015; Samani and Bell 2016; Bono et al. 2017). Two potential sources of bias have been suggested: 1) detection bias by statistical significance, since antagonistic pleiotropy is more difficult to detect than conditional neutrality, as the former requires significance in two environments, while the latter only requires change in one environment (Anderson et al. 2013) and 2) sampling bias because most empirical evidence are from studies performed in predominantly selfing organisms (Bono et al. 2017).

On the other hand, both theoretical and empirical results provide a potential scenario where the difference of prevalence is biologically plausible which is the presence of both antagonistic pleiotropy and conditionally neutral variants during the adaptation process (Fisher 1930; Ostrowski, Rozen, and Lenski 2005; Duffy, Turner, and Burch 2006; Schick, Bailey, and Kassen 2015; Dillon et al. 2016). If this is the case, the fixation of each type of variants are expected to depend on both ecological and evolutionary history (Bono et al. 2017). First, local adaptation in a homogeneous and stable environment is expected to involve antagonistic pleiotropic variants more frequently than in heterogeneous environments (Kassen 2002). The reason being adaptation in homogeneous environments does not need to incur associated fitness costs from alternative environments that are not experienced locally. Conversely, heterogeneous environments impose a higher number of environments locally, which results in selection against the cost of adaptation given by antagonistic pleiotropic variants. This is presumably strong in temporally heterogeneous environments as individuals experience the full range of environmental variation, as opposed to strict spatial heterogeneity where the population could be geographically structured (Kassen 2002; Bono et al. 2017).

There are other biological reasons to observe conditional neutrality more frequently than antagonistic pleiotropy. Variants involved in local adaptation can derive from standing genetic variation or from new mutations (evolutionary history). However, during colonization to new environments, antagonistic pleiotropic variants are expected to be in lower frequency, and therefore a higher contribution of conditionally neutral variants is predicted (Barrett and Schluter 2008; Matuszewski, Hermisson, and Kopp 2015; Dittmar et al. 2016). Additionally, antagonistic pleiotropic variants could have different contributions to adaptation depending on the degree of divergence between

populations from alternative environments. For instance, the pleiotropic cost of the alternative environment can be more pronounced with increasing divergence, making the adaptation process less constrained in early steps when antagonistic pleiotropy has not evolved. Early fixed antagonistic pleiotropic variants could also be compensated later by a new interacting loci (Lenski 1988; Kassen 2002; Jasmin and Zeyl 2013; Bono et al. 2015; Schick, Bailey, and Kassen 2015).

Methodologically, one way to measure the response to selection, and the presence of trade-offs is using genetic and phenotypic variance co-variance matrices, which define if some evolutionary trajectories might be constrained. Variance co-variance matrices between variables are symmetric, allowing the reduction of variance observed in traits to orthogonal axes (as many traits or genetic variants as applicable) into a principal component analysis (PCA). Each principal component explains a fraction of the total variance, given by the eigenvalue. In the PCA, restricted evolutionary trajectories can be identified when the eigenvalue is close to zero (Kirkpatrick and Lofsvold 1992; Blows and Hoffmann 2005; Roff and Fairbairn 2007).

In this framework, if a functional trade-off between fitness traits is present, the ability to detect it will depend on the variance and co-variance of the traits. This means that in the absence of phenotypic variation, the trade-off, even if present, may not be visible. This can be the case for populations evolving under strong selection in which phenotypic variation for specific traits is reduced or fixed. On the contrary, the presence of variation may allow observing a trade-off when present, but it also begs the question of whether this variation is given by genetic variation or phenotypic plasticity. Even under laboratory conditions, variation between individuals is expected as a result, in part, of environmental variation, and thus, this variation can be from phenotypic plasticity rather than heritable genetic variation. In the presence of genetic variation, it has been suggested this variation can be maintained by: mutation-selection equilibrium; antagonistic pleiotropy; correlated selection; and spatio-temporal heterogeneity balance (Lynch and Gabriel 1987; Houle 1991; 1998; Roff and Fairbairn 2007).

Structural variation

Adaptation at the population level has been studied extensively theoretically and empirically. Owing to technical limitations associated with the inference of several types of genetic variants, most studies in large populations explored genetic variation at the level of single nucleotides (SNPs) (Wolf and Ellegren 2017). However, increasing evidence highlights the importance of other types of genetic variation such as structural genomic variation (SV).

Structural variants (SVs) comprising chromosomal rearrangements, insertions, deletions and duplications, also significantly contribute to the overall genetic differentiation of populations with important evolutionary and functional consequences (Redon et al. 2006; Weischenfeldt et al. 2013). SVs impact complex phenotypic traits (Purcell et al. 2014; Schwander, Libbrecht, and Keller 2014; Küpper et al. 2016; Lamichhaney et al. 2016), can alter regional recombination rates and affect gene regulation (Rieseberg 2001), contribute to rapid evolution by gene duplications (Redon et al. 2006), allow the coupling of barrier loci for reproductive isolation (Rieseberg 2001; Hoffmann and Rieseberg 2008), and can confer fitness advantage (Avelar et al. 2013; Fisher et al. 2018). However, despite increasing attention on SVs, there is a gap in our knowledge regarding the extent of variation at the population level and the evolutionary dynamics of this variation within and between populations in the context of adaptation and speciation.

Study system

Within the kingdom fungi, the two best studied groups of fungi are the ascomycetes or the basidiomycetes group, which each contain many different yeasts. Yeast taxa are distinguished by their unicellular lifestyle (contrary to multicellular mycelia) and their ability for sugar fermentation. Ascomycetes are characterised by their sexual spores being packed in a specialised structures called the ascus. The fission yeast *Schizosaccharomyces pombe*, the model I used in these studies, belongs to the ascomycete yeast clade. Other well-known examples of ascomycetes are the common yeast (*Saccharomyces cerevisiae* – also known as brewer's yeast or baking yeast) and filamentous fungi used in the laboratory and industry such as *Aspergillus* and *Neurospora* (Hoffman, Wood, and Fantes 2015). The basidiomycetes contain a large variety of species, including smuts, rusts and the macroscopic mushroom fungi.

The genus *Schizosaccharomyces* has four species: *S. pombe*, *S. japonicas*, *S. octosporus* and *S. cryophilus* (Rhind et al. 2011). The divergence between *S. pombe* and its closest relative species has been estimated to around 120 million years (Rhind et al. 2011). The genus *Schizosaccharomyces* is part of the Taphrinomycetes, group that diverged early within the ascomycetes around 600 million years (Lutzoni et al. 2018). This early divergence places the fission yeast and common yeast phylogenetically distant, with a divergence time comparable to the divergence within all animal clades, or the emergence of embryophytes in plants (Sipiczki 2000; 2004; Dunn et al. 2014; Lutzoni et al. 2018).

Schizosaccharomyces pombe as a model species

Yeasts as model systems present several advantages over other systems, particularly for experimental work. *S. pombe* has traditionally been used as a model to study molecular and cell biology in fundamental processes defining the eukaryotic cell. Currently, *S. pombe* is used in more than 300 laboratories around the world producing more than 300 publications per year. The importance of *S. pombe* in cell biology is owing to cellular similarities with more complex multicellular eukaryotes; similarities which are not present or too highly diverged in other yeast species such as the common yeast *S. cerevisiae* (Hoffman, Wood, and Fantes 2015; Fantes and Hoffman 2016). These similarities include fundamental cell cycle control (Nurse 1990), cytokinesis (Goyal et al. 2011), mitosis and meiosis regulation (Harigaya and Yamamoto 2007), DNA repair and recombination mechanisms (Phadnis, Hyppa, and Smith 2011), centromere structure (Allshire and Karpen, 2008), epigenetic control (Goto and Nakayama 2012), and cell morphogenesis (Fawcett et al. 2014). Laboratory strains can be maintained either as haploid or diploid, allowing us to assess allele dominance. Yeasts also have the ability to retain plasmids, providing the potential to introduce genetic material to the nuclear genome. The use of these plasmids facilitate gene editing and gene cloning (Hoffman, Wood, and Fantes 2015). Compared with other model systems, yeasts have a high homologous recombination rate. For example, 1 cM in humans correspond to an average 1 Mbp (Kong et al. 2004), while it is only around 2500 bp in common yeast (Olson et al. 1986) and 6250 bp in fission yeast (Fowler et al. 2014; Hoffman, Wood, and Fantes 2015). In the context of this study, yeast has the major advantage that it can be maintained in large population sizes. This translates to a high efficiency of selection relative to genetic drift and allows for the identification of rare mutants in the gene(s) involved in traits of interest.

Genomic and wet lab resources

The haploid *S. pombe* reference strain was the sixth eukaryote to have its entire genome sequenced (Wood et al. 2002). The genome is 13.8 Mb in size, similar to the baker's yeast genome of 12.5 Mb. It is contained in only 3 chromosomes (5.7, 4.6, and 3.5 Mb each) compared to the 16 chromosomes found in baker's yeast. Similar to animal chromosomes, *S. pombe* has large and complex repeated centromeres of around 40, 69 and 110 kb for chromosomes I, II and III, composed of a central core flanked by repeats. Similar to baker's yeast, subtelomeric regions are transcriptionally silent (Aparicio, Billington, and Gottschling 1991; Nimmo et al. 1998), containing several genes mostly related to membrane transporters and response to stress and surface glycoproteins (Goffeau et al. 1996; Wood et al. 2002; Hansen et al. 2005; Rhind et al. 2011).

Despite its importance as a model organism and the existence of collections with samples from all over the world, most of the research has been based on a single isogenic strain (not containing genetic variants) isolated in 1921 by A. Osterwalder. This strain was isolated from grape juice in Montpellier, France and deposited at the Dutch culture collection. Genetic studies in *S. pombe* started during the 1940s when Urs Leupold used this strain from which three strains were isolated the differed only in the mating type system. These strains were 968 h^{90}, 972 h^{-}, and 975 h^{+}, which are now the ancestors for almost all genetic studies on *S. pombe* (Brown et al. 2011; Hoffman, Wood, and Fantes 2015; Fantes and Hoffman 2016). From these strains, a broad range of mutants has been isolated for multiple purposes. As most of the research in *S. pombe* has been performed in these unique strains, genetic diversity in natural populations is largely unexplored (Jeffares et al. 2015; Jeffares 2018).

Population structure

Until recently, genetic diversity in natural populations remained largely unexplored (Jeffares et al. 2015; Jeffares 2018). Over the last decade, there have been several attempts to infer parts of the evolutionary history and ecology of S. pombe. One recent study used a large, comprehensive sampling of mostly human-associated, globally-distributed samples, finding remarkably reduced genetic structure with moderate genetic diversity ($\pi = 3\times10^{-3}$ substitutions per site; Jeffares et al. 2015). They estimated that the divergence of all samples started only around 2300 years ago, reaching the Americas around 400 years ago, consistent with the beginning of European colonialism (Jeffares et al. 2015). This estimate of genetic diversity is similar to previous works (Brown et al. 2011; Rhind et al. 2011; Fawcett et al. 2014), albeit slightly higher than observed in the budding yeast S. cerevisiae ($\pi = 1\times10^{-3}$; Liti et al. 2009). Nonetheless, budding yeast shows clear differentiation between populations and more defined genetic structure. A detailed genetic interrogation suggests that the S. pombe genome is under selective constraints, with signatures of worldwide purifying selection or balancing selection (Fawcett et al. 2014; Jeffares et al. 2015).

Ecology and phenotypic variation

Although very little is known about the ecology, origin, and evolutionary history of *S. pombe* (Jeffares 2018), the remarkable lack of genetic structure despite a cosmopolitan distribution contrasts with observations of phenotypic and reproductive variation. Trait variation is usually not structured by geographical regions (Jeffares et al. 2015), yet *S. pombe* has abundant heritable phenotypic diversity in growth, stress responses, cell morphology, and cellular biochemistry (Brown et al. 2011; Clément-Ziza et al. 2014; Jeffares

et al. 2015; Jeffares 2018). *S. pombe* is generally found in traditional non-industrialized fermentations in substrates with high concentrations of sugars, including tea, kombucha, sugar cane, sorghum, palm sap, harvested coffee and cocoa beans, honey, and other fruits (Jeffares 2018). It is also used to produce distilled spirits such as rum, tequila or cachaça (Gomes et al. 2002), as well as to reduce acidity in wine given it ability to use malic acid (Volschenk, van Vuuren, and Viljoen–Bloom 2003).

Several studies have reported low spore viability in crosses between strains (Kondrat'eva and Naumov 2001; Avelar et al. 2013; Zanders et al. 2014; Jeffares et al. 2015; Naumov, Kondratieva, and Naumova 2015; Hu et al. 2017). This viability ranges from less than 1% to 90%, and even crosses within the same strain can present reduced mating efficiency (Kondrat'eva and Naumov 2001; Marsellach 2017). The incompatibility in general is considered to be post-zygotic as mating bodies and haploid spores are still produced (Kondrat'eva and Naumov 2001; Jeffares et al. 2015).

Combining these two sources of evidence, genetic and phenotypic results, is perplexing. On the one hand, there is a global pattern of reduced genetic structure and strong selection. On the other hand, there is large phenotypic diversity and strong reproductive isolation between strains. Functional work has been done in few strains, and at least three potential non-exclusive genetic causes for reproductive isolation have been identified in *S. pombe*. (1) Structural genomic variants (SV) including large chromosomal rearrangements have been implicated. Large SVs, mostly inversions and translocations, can result in aneuploidy during chromosome segregation after mating reducing spore viability. Around 12 of such large SVs have been observed in some strains including the reference strain (Rhind et al. 2011; Avelar et al. 2013; Zanders et al. 2014). (2) Presence of species-specific selfish elements or meiotic drives, inducing segregation distortion by spore killing in crosses between strains (Zanders et al. 2014; Hu et al. 2017; Nuckolls et al. 2017). (3) Bateson–Dobzhansky–Muller incompatibilities or related genetic mechanisms resulting in negative epistasis (Nei and Nozawa 2011). Understanding the relative importance of these variables in reproductive isolation and phenotypic differentiation requires comparative genomics and functional work in a more extensive sampling, which has previously been limited to around a dozen samples.

Life cycle and mating

The life cycle of *S. pombe* transitions between (1) growth by mitotic asexual divisions when there are adequate nutritional conditions, or (2) sexual propagation triggered by starvation (Leupold 1949; Beach and Klar 1984; Leupold 1993), a process that is regulated by internal and external cues

(**Figure 1a**). During asexual growth in nutrient-rich medium, the cells are cylindrical with hemispherical ends and are about 3.5 µm in diameter. After cell division, cells are around 8 µm long and growth occurs mostly longitudinally, where length constitutes an adequate measurement for cell age (Hoffman, Wood, and Fantes 2015). During cell division, a transverse septum appears in the middle of the cell, producing two nearly identical daughter cells (Hoffman, Wood, and Fantes 2015). This type of cell division differentiates *S. pombe* from other yeasts, including the common yeast, which produces asymmetrical cell during division. Right after mitosis, the G1 phase in *S. pombe* is very short, leading to a rapid replication of DNA (S phase). Consequently, at the end of the S phase, there are two nuclei in one cell, each of them containing two copies of chromosomes (sister chromatids). The G2 phase starts after cytokinesis, and cells remain in this phase for about three-quarters of the cell cycle, containing fully replicated chromosomes. This means haploid cells in a growing population contain two copies of identical sister chromatids (Hoffman, Wood, and Fantes 2015; Hagan et al. 2016). Alternatively, cells change from asexual growth to sexual reproduction when there is a mating partner and nutrient starvation. First, cells with alternative mating types (described below) conjugate. The conjugation consists of an initial cell wall degradation to produce a single cell. Then, the nuclei fuse producing a diploid zygote. The nucleus starts meiosis, thus producing four haploid nuclei. A wall is then formed around each nucleus, producing four individual spores, albeit encased in the original zygote (ascus). When nutrients are no longer limiting, the ascus wall will be lysed, exposing the four spores to the environment. Spores remain dormant until they are exposed to favourable growth conditions, where they can germinate and grow again asexually (Egel 1973; Schmidt and Gutz 1994). A fraction of the diploid cells can also propagate asexually rather than continuing with the sexual cycle.

Different strains of *S. pombe* have different mating systems, divided into homothallic and heterothallic, first studied by Urs Leupold (Leupold 1949; Fantes and Hoffman 2016) (**Figure 1b**). In general, homothallism (in paper II, this is referred to as the *switcher*) refers to possession within a single individual of the mating components to reproduce sexually. For example, an individual with both "male" and "female" reproductive structures, allowing it to potentially self-reproduce, is classified as homothallic. In many unicellular organisms like fission yeast, homothallism describes the phenomenon of switching of mating type (a change of the genes at the mating type locus from plus to minus or vice versa), within the offspring of a single cell during asexual growth (Hanson and Wolfe 2017). Conversely, heterothallic strains (in paper II, this is referred as the *non-switcher*) only express one of the two mating types (either plus or minus). Thus, during starvation, a single homothallic cell has the potential for mating-type switching after cell division

followed by sexual reproduction, while a heterothallic strains cannot undergo mating-type switching and therefore can only undergo the sexual cycle in the presence of cells of the opposite mating type.

The mating type in fission yeast is defined by the mating locus *mat1,* which can have the alleles Plus (h+) or Minus (h-). However, in the wild homothallic strain h^{90}, the active *mat1* locus is adjacent to two silent copies *mat2-P* (with allele Plus) and *mat3-M* (with allele Minus). During asexual growth, one of the two silent copies can be copied into the active locus *mat1* by non-homologous recombination, changing the allele between mating types in the active locus. As a result, the daughter cell will have the opposite active mating type compared to the mother cell, and the two cells are now able to sexually conjugate (Hanson and Wolfe 2017).

Spontaneous mutations in the wild h^{90} configuration can result in heterothallic strains (non-switchers) (Beach and Klar 1984). Although the mutation occurs in low frequency (between 1 in 10,000 and 1 in 1 million cell divisions), an extensive sampling of natural strains around the world found that around 10% were heterothallic (Jeffares et al. 2015). Some known heterothallic genotypes are h^{-S}, h^{+S} or h^{+N} (Beach and Klar 1984; Heim 1990). In the h^{-S} and h^{+S} genotypes, there is a deletion including one of the silent copies, leaving only the active locus and one silent copy (in this case called *mat1* and *mat2:3* respectively). The new variant can still undergo non-homologous recombination between the two copies. However, after one switch, the daughter cell will have both copies (*mat1* and *mat2:3*) with the same mating type allele, as subsequently, there are not father allele switching. In the h^{+N} genotype, there is an insertion of the fragment from *mat2-P* to *mat3-M,* replacing *mat1* (inserted *mat* copies now called *mat1:2* and *mat3:1*). The first copy (*mat1:2*) is now the active copy, and the second one (*mat3:1*) is silent. The switching occurs between the silent *mat3:1* and the original two silent copies (*mat2* and *mat3*). As a result, the active locus *mat1:2* does not switch, maintaining the same mating type allele (Nielsen and Egel 2007; Hanson and Wolfe 2017).

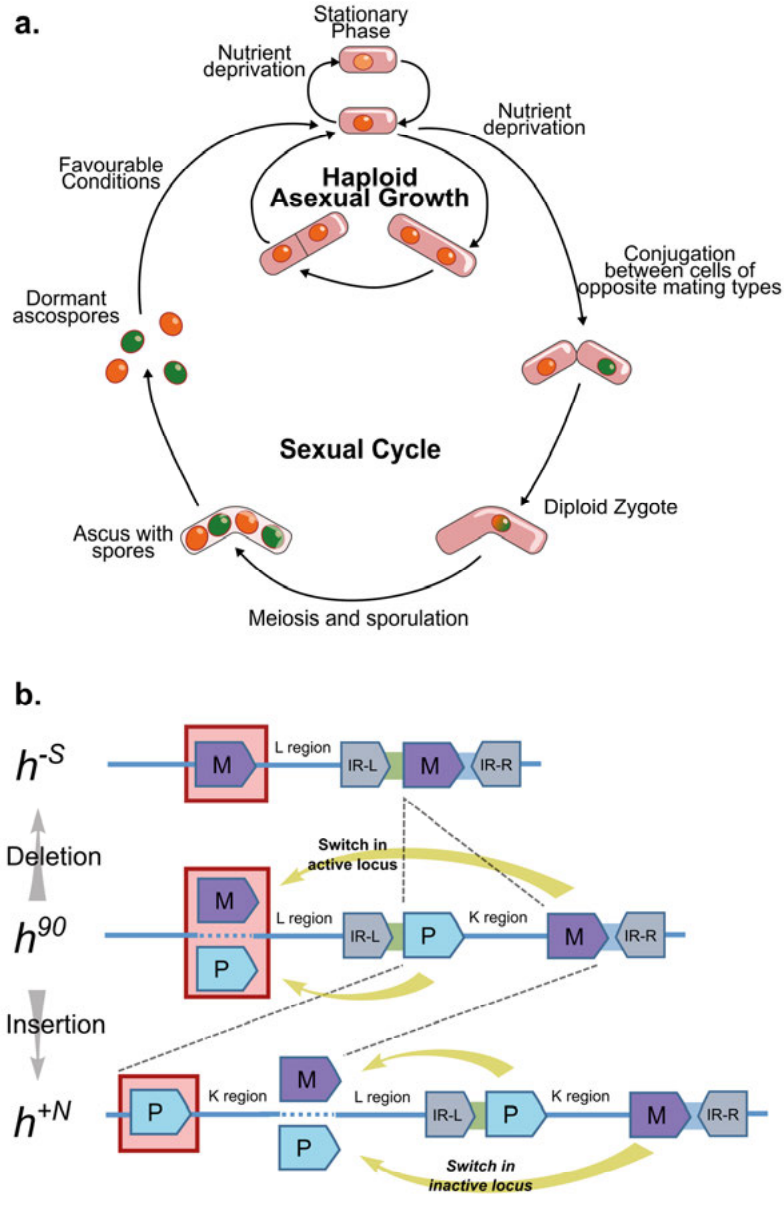

Figure 1. a. The life cycle of *S. pombe* (Figure edited from Nielsen and Egel 2007). **b.** Genetic organization of the mating region. Schematic of the genetic arrangement for homothallics (h^{90}; middle) and the two most common heterothallics (h^{-S} and h^{+N}; top and bottom, respectively). Regions are not to scale. In homothallic strain, the active mating-type locus *mat1* (red box) switches between *Plus* (P) and *Minus* (M), using the silent cassettes as templates during cell division. The heterothallic strain h^{-S} contains only the minus cassette, due to deletion of part of the silent region. The heterothallic strain h^{+N} is the result of a duplication of almost the entire silent mating type and only expresses the P cassette at *mat1* (Figure edited from Nieuwenhuis et al. 2018).

Methods

This section briefly describes some of the most relevant methods used in the four papers of this thesis.

Genome sequencing data

Information derived from whole genome sequencing is becoming one of the most prevalently used data type in evolutionary biology. The general workflow is as follows: DNA is extracted from whole organisms or parts of organisms, the identity and order of nucleotides in a small section of the genome is determined by sequencing, and the larger scale order along the genome of these pieces can be inferred by algorithmic processing. The traditional method begins with the polymerase chain reaction (PCR; Saiki et al. 1985), to amplify a locus of interest. The order of the nucleotides along this locus is then determined by chain-termination DNA sequencing (known as S*anger sequencing*; Sanger, Nicklen, and Coulson 1977). Sanger sequencing can only produce one fragment with roughly 700 bp of high quality sequence before the quality decreases. The sequencing of larger fragments, therefore, requires the amplification of multiple fragments, which are then analytically 'stitched' together. The limitation in sequencing output of Sanger sequencing, was alleviated by high-throughput sequencing data, implemented with next generation sequencing technologies. The high throughput and significantly lower price per sequenced nucleotide allowed the production of whole genomic data from multiple individuals or even populations. Early high-throughput sequencing methods were still limited by the length of any given fragment but this was changed with the introduction of third-generation, long read sequencing technology (Eid et al. 2009). Using a combination of these high-throughput data types provide a more complete view of the genome. In this thesis, three main types of sequencing technologies were used: *Illumina sequencing-by-synthesis* (Bentley et al. 2008), single-molecule real-time sequencing (Pacific BioSciences technology, or *PacBio*; Eid et al. 2009) and nanopore sequencing (Oxford Nanopore technology, or *MinIon*). Different sequencing technologies have different advantages and disadvantages, including data-specific biases, error rate, sequencing cost, and general data handling.

Existing sequencing technologies are still unable to produce a single contiguous sequence corresponding to a whole chromosome. Whole genome sequencing consists of data from millions of shorter, individually sequenced reads from a single sample. Those reads are derived from a sample in which DNA has been fragmented, and fragments are used as a template for sequencing. For *Illumina* sequencing, a single template DNA can be sequenced from both ends (from the 5' or 3' strand), producing pair-reads. Each read is most commonly either 100 or 150 bp long. Using a template DNA normally of around 700 bp, would result in a pair of reads with an expected distance around 350 - 400 bp left un-sequenced between them. The main limitation of collecting small reads, rather than whole chromosome sequences, is that those reads need to be compared to an already existing whole-genome assembly (*reference genome*). Attempting to assemble short reads into larger contigs, although possible, would still result in a highly fragmented chromosome. Reads are usually compared to the reference genome by comparing the sequence and finding regions in the reference genome that correspond to the sequence found in individual reads (*mapping*). The mapping process then compiles individual reads into corresponding genomic regions, which subsequently will allow comparison between multiple reads falling in the same region. If the sequenced sample has genetic variation relative to the reference genome, those differences will be observed on multiple reads mapping to those genomic regions. In all papers of this thesis, the primary focus are individual base variation, known as single nucleotide polymorphisms (SNPs), but structural variation (insertions, deletions, inversions and translocations) are likewise explored in paper II, III and IV.

Genotyping

The accurate identification of genetic variants requires that those variants are supported by multiple reads. In principle, for a given variant, reads mapping to a genomic region should show the alternative base for all reads (in homozygous or haploid individuals) or in half of the reads (in heterozygous individuals). However, in practice this is not necessarily the case. Frequently reads mapping to the same region can consist of more than two alleles or the ratio of reads consisting alternative alleles deviate from the 50:50 expectation in heterozygous diploids. This can occur as a result of DNA contamination, sequencing errors, incorrect mapping of reads or mapping of the same read in multiple genomic regions. Therefore, the correct identification of genetic variants (*genotyping*) improves with increasing *sequence coverage* (the number of reads mapping to the same genome region – or *read depth*), thus the number of reads with the "correct" genotype is higher relative to those with false alternative alleles or the result of sequencing error.

The need for increased sequencing depth to improve genotyping means that whole-genome sequencing for multiple individuals and populations can easily become very expensive. A given sequencing run produces a maximum specific sequence output. The resulting coverage per individual is therefore a function of the size of the genomic target and the number of individuals. In *S. pombe* this is not a major obstacle as the genome size is small (~13 Mb) and high coverage can be achieved from a single sequencing run (e.g. a single *lane* for Illumina sequencing or *SMRT cell* for long read PacBio sequencing). In order to have a more efficient sequencing strategy, in this thesis individual samples were first labelled with unique sequence tag identifiers (i.e. barcodes). This allows multiple samples to be sequenced in a single lane and then later be assigned to the correct individual. Additionally, for paper I, each sample constitutes a whole population with potentially multiple genotypes, rather than an individual genotype. In this case, the strategy was to sequence the whole population without barcodes (*pool sequencing*). Pooled sequencing cannot provide individual whole genome genotypes. Instead, each read pair is derived from a single DNA molecule, which itself comes from an individual cell. The total sequencing coverage is used as an approximation for sampling individual chromosomes from the original population given enough starting individuals. Sequencing depth/coverage for each genetic variant is then used to estimate allele frequency in the original population (Schlötterer et al. 2014). The estimate of allele frequency derives from counting the number of reads supporting each allele out of the total number of reads mapping to the same genomic location. This approach is widely used and the robustness has been supported empirically (Zhu et al. 2012), but there is concern regarding the accuracy to identify low frequency variants as the signal is mixed with sequencing errors (Cutler and Jensen 2010).

Genome *de novo* assembly

The use of reference genomes has several methodological implications. A main one being that the power to identify genetic variation is limited to genomic regions present in the reference genome. If a sample has a genomic region that is not present in the reference genome, reads corresponding to those regions will not map, and will be omitted. As an alternative approach one can assemble the genome from the reads of each individual separately instead of mapping them onto a reference. Individual reads can be compared with each other, merging reads when they have overlapping sequence, and extending the sequences into larger fragments (*contigs*). Contigs associated with one another can then be linked into even larger fragments (s*caffolds*). This genome-wide extension of fragments will result in an assembly of a new genome without the use of reference genome (*de-novo assembly*; Ekblom and Wolf 2014; Nimmy and Kamal 2015). The quality of the as-

sembly, based on the completeness of chromosome sequences, will depend on a combination of methodological and intrinsic biological variables. Examples of methodological factors include the quantity and quality of the original sampled DNA, degree of fragmentation, fragment size selection before sequencing, read length, and total number and quality of obtained reads. Similarly, biological factors that affect the quality of genome assembly include: genome size, ploidy level, GC content, population genetic diversity, and repeat content or in general sequence similarity.

In order to improve the *de novo* assemblies that we generated in this thesis we used more recent long-read sequencing technologies (*PacBio* and *MinIon*). In this case, template DNA is sequenced in a single direction, but the resulting reads are substantially longer (~ 5 to 100 kb). Long reads improve the quality of assembled genomes and can result in chromosome level *de novo* assemblies for several species, especially those with small genome size (Chin et al. 2013). As a disadvantage, long read sequencing technologies are substantially more expensive and error rate during sequencing is higher. Therefore, the use of long read data has been primarily directed to improve *de novo* genomes and the identification of large structural variation, not possible using short *Illumina* reads (Weissensteiner et al. 2017; Peona, Weissensteiner, and Suh 2018). On the other hand, a combination of short and long reads can complement the disadvantages from either technology. For instance, *de novo* assemblies from long read data can leverage the higher quality of short read data through polishing (error correction) (Walker et al. 2014). The resulting genome assembly can be used then similar to mapping methods. Assembled genomes can be used either to map reads from other samples on these new genomes, or assembled genomes can be directly compared to identify genetic variation.

Population genetic diversity

Once there is a set of genetic variants genotyped from multiple individuals and populations, these data can be used to summarise variation within population and differentiation between populations. There are several summary statistics to describe genetic variation, but in this thesis the primary focus is the use of estimators of the theoretical parameter θ, representing the genetic diversity expected in a hypothetical population, in which all variants are neutral and at mutation-drift equilibrium (populations in which the rate of new neutral mutations is balanced by loss or fixation by genetic drift). The main benefit of using estimators of θ is that theoretical expectations can be compared to empirical observations, and the deviation from expectation allows us to make inferences about demographic and selective processes. In general, at mutation-drift equilibrium the amount of genetic diversity that a

population can sustain (θ) is proportional to the effective population size (N_e) and the mutation rate (μ):

$$\theta = 4N_e\mu$$

Considering a single biallelic nucleotide polymorphism, each allele will be in the population in a frequency of p_i, such that the sum of both alleles is 1. One way to measure the variation in a single site is the *heterozygosity* (*H*), defined as:

$$H = \frac{n}{n-1}(1 - \sum p_i^2)$$

Where *n* is the number of sequences sampled. The heterozygosity represents the expected probability of sampling the two alternative alleles when two sequences are sample randomly. The heterozygosity from each variant (H_j) can be summed to estimate genetic diversity in the whole sequence or genome (π) as such:

$$\pi = \sum_{j=1}^{S} H_j$$

Where *S* is the number of polymorphic sites and H_j is the heterozygosity in the *j*th polymorphic site (M Nei and Li 1979; Masatoshi Nei and Tajima 1981; Fumio Tajima 1983). This value is normally reported as a normalised value according to the total sequence length. Alternatively, π can be calculated as the mean number of pairwise nucleotide differences between any two sequences: where *S* is the number of polymorphic sites and H_j is the heterozygosity in the *j*th segregating site (Nei and Li 1979; Nei and Tajima 1981; Tajima 1983). This value is normally reported as a normalised value according to the total sequence length. Alternatively, π can be calculated as the mean number of pairwise nucleotide differences between any two sequences:

$$\pi = \frac{\sum_{i<j} k_{ij}}{n(n-1)/2}$$

Where k_{ij} is the total number of nucleotide difference between the *i*th and *j*th sequences, from a sample of *n* sequences.

An alternative measurement of genetic diversity is *Watterson's theta* (θ_W) (Ewens 1974; Watterson 1975), defined as:

$$\theta_W = \frac{S}{\sum_{i=1}^{n-1} \frac{1}{i}}$$

Both θ_W and π are expected to be equal in a neutrally evolving population of constant size. However, when assumptions to this neutral expectation are violated, the two estimators behave differently. These violations can happen if the population has had a demographic change (expansion or contraction) or is under selection. Thus, these demographic or selective processes can be quantified by looking at the normalised difference between θ_W and π (*Tajima's D*; Tajima 1989):

$$D = \frac{\pi - \theta_W}{\sqrt{Var(\pi - \theta_W)}}$$

If a population is behaving as expected under a neutral model, *D* has a value around 0. In contrast, negative *D* will be generally interpreted as either evidence for selective sweeps removing genetic variation or population expansion, and positive *D* is interpreted as evidence of either balancing selection or population contraction.

Population structure

Inferring consistent differences between individuals allows us to identify *population structure* and the amount of time for the divergence between populations to occur. Similar to genetic diversity, several statistics have been proposed. One commonly used statistic to measure differentiation between two populations is F_{st} (Wright 1931; 1943; 1951; Weir 1996), defined as:

$$F_{st} = \frac{\sum_i \frac{(p_i - \bar{p})^2}{K - 1} \frac{n_i}{\bar{n}}}{\bar{p}\bar{q}}$$

Where K is the number of populations, $\bar{p}$ and $\bar{q}$ is the average frequency of alleles *A* and *a* respectively among populations. Each population *i* has n_i samples and a sample allele frequency p_i. $\bar{n}$ is the average sample size. However, although *Fst* is commonly used, it is strongly influenced by levels of variation within a population (Charlesworth 1998; Jakobsson, Edge, and Rosenberg 2013). For this reason F_{st} is considered a *relative* measurement of differentiation ranging from 0 to 1 (Cruickshank and Hahn 2014). As an alternative, other statistics of differentiation are used as *absolute* measurements. One example is D_{xy} (Nei 1973; Nei and Li 1979; Nei 1987), which is the average number of pair-wise differences between sequences from two populations. D_{xy} is an absolute measurement of differentiation, which is not

influenced by levels of variation within populations (although it depends on mutation rate). It is calculated as:

$$D_{xy} = \sum_{ij} x_i \, y_j k_{ij}$$

Where x_i and y_j are the frequencies of the ith haplotype from population X and the jth haplotype from population Y respectively. k_{ij} is the average total number of nucleotide differences between the pair of haplotypes from each population.

Experimental evolution

Here *experimental evolution* will be defined as the study of genetic and phenotypic changes in populations under controlled and reproducible environmental conditions over multiple generations (Garland and Rose 2009). Environmental conditions can be in the laboratory (artificial environments) or in nature (i.e. droughts, fires, invasions, or epidemics). Environmental conditions can involve various aspects of abiotic, biotic or demographic variables. Experimental populations are then compared with an un-manipulated control population or, in some cases they are compared to the ancestral population prior to experimental manipulation. Thus, the primary advantage of experimental evolution is the ability to have multiple replicates and controls for studying the evolutionary process. Exposing multiple populations to the same novel environment increases our ability to distinguish predictable responses to the novel environment from stochastic differences.

In experimental evolution, as opposed to *"artificial selection"* or *"selective breeding"*, selection on individuals within a population across generations is not based on a single phenotypic trait in particular or a combination of traits. Instead, selection is imposed by the environment (local conditions). The population can respond to those novel conditions and increase their fitness through subsequent generations relative to the ancestral population. Thus, differential phenotypic adaptive responses can be observed between different replicate populations (Garland and Rose 2009). Experiments of artificial selection primarily focus on understanding the organismal properties (i.e. the physiological or genetic bases of a specific trait). On the contrary, evolutionary experiments tend to focus more on testing general evolutionary theories, and the organism itself is used as the conduit for such tests (Garland and Rose 2009; Kawecki et al. 2012).

In contrast to comparative studies in natural systems, evolutionary experiments present several other advantages. For instance, in evolutionary exper-

iments there is no uncertainty about ancestral conditions, which is determined *a priori* in the experimental design. Additionally, environmental conditions can be adjusted, such as restricting variation in single variables or a combination of variables, allowing us to identify the response of a population to those specific conditions (Garland and Rose 2009). Using experimental evolution, populations can respond to selection by enhancing changes in phenotypic traits, which can subsequently be used for functional studies (i.e. identifying the genetic bases of phenotypic traits) (Bennett 2003). Despite these advantages, evolutionary experiments have several drawbacks. Although evolutionary experiments have been conducted in natural ecosystems (Reznick, Bryga, and Endler 1990), most studies tend to be restricted to laboratory conditions. Consequently, the imposed environmental conditions are simplified versions of the multidimensionality encountered in natural environments. Additionally, as the experimental design generally requires multiple populations of large size and short generation times, experiments are restricted to organisms easily maintained in the laboratory. However, the range of available systems is phylogenetically broad, including bacteria, algae, protists, fungi, animal and plants. It is important to note that experimental evolution does not aim to rigorously reproduce nature. Instead, its utility is the ability to test general evolutionary theories under simplified conditions (Bennett and Lenski 1999; Garland and Rose 2009).

In addition to tracking the change in phenotype or certain traits through generations, experimental evolution can also screen for potential causal effects of genetic variants and their relationships with fitness (Kawecki et al. 2012). In this way, genetic variants can come from a focal set of previously identified loci of interest and the ancestral population of the experiment will be polymorphic for those variants. Throughout the experiment, conditions will be manipulated and the response of the population will be measured via changes in allele frequencies (e.g. Murray and Cutter 2011). Experiments can also be done without previous knowledge of the genetic variants involved. For instance, the ancestral population can come from a pool of individuals from one or several natural populations, resulting in a genetic pool upon which selection can directly act during the experiment. The ancestral population for the experiment can also contain no genetic variation (ie. isogenic), in which case evolutionary changes rely on *de novo* mutations occurring randomly over generations. Populations are then subject to selection, and resulting populations can be sequenced to identify genetic variants with divergence in allele frequency (e.g. Kosheleva and Desai 2018). These changes can be the result of adaptation to the experimental conditions, but they can also be due to drift, physical linkage to the causal variant or statistical false-positives. These type of experiments have been used to explore a broad range of topics including testing new technologies, identifying new genes, testing potential genetic interactions (epigenetics), identifying emer-

gence of new functions, estimating mutation rate, identifying size effect distribution, and understanding convergent evolution and patterns of molecular evolution (Kawecki et al. 2012).

Research aims

The main aim of this thesis was to integrate multiple approaches to identify potential evolutionary processes and factors governing adaptation in the fission yeast *Schizosaccharomyces pombe*. I primarily tested the role of gene flow and genetic admixture on the resulting distribution of phenotypic variation and adaptive divergence between populations. Specific aims for each of the presented papers as part of this thesis are listed below:

Paper I – assess the role of gene flow in adaptive divergence by means of experimental evolution, and to identify genetic changes underlying the process and potential intrinsic genetic and phenotypic properties constraining evolutionary trajectories.

Paper II – explore the molecular, ecological and evolutionary drivers of the emergence and persistence of switching and non-switching mating strategies in *Schizosaccharomyces pombe*.

Paper III – elucidate the evolutionary history of *Schizosaccharomyces pombe*, and identify the importance of genome-wide admixture, structural mutations and negative epistatic selection in explaining phenotypic variation and reproductive isolation.

Paper IV – provide new genetic resources and give insights into the evolutionary history in *Schizosaccharomyces pombe* based on diversity patterns of the mitochondrial genome.

Summary of papers

Paper I

Parallelism in adaptive divergence and the modulating effect of gene flow

It has been a central topic in evolutionary research how adaptation to different environments may contribute to the origin of genetic and phenotypic divergence. In isolated populations, directional, habitat-specific selection is predicted to foster local specialisation with the potential to promote reproductive isolation, and ultimately speciation (Shafer and Wolf 2013). In connected populations this process is counteracted by gene flow. Here, the interplay between the strength of selection, the degree of gene flow and the genetic architecture of adaptive traits will determine the evolutionary outcome ranging from full admixture, reduced population structure and the emergence of generalist phenotypes (Ronce and Kirkpatrick 2001) to local specialisation followed by the emergence of reproductive isolation (Smadja and Butlin 2011);

Although the adaptation process has been studied in isolated populations, experimental insights into the role of gene flow between populations adapting to contrasting environments are essentially lacking (Gray and Goddard 2012). Here, we performed an evolutionary experiment aiming to identify the role of gene flow on local adaptation, the quality and strength of fitness trade-offs, the importance of standing genetic variation, and the potential genetic architecture driving adaptation with gene flow. 132 populations of the fission yeast *Schizosaccharomyces pombe* were subjected to disruptive selection on settling speed as an environmental factor (named top and bottom selection). Different migration rates were allowed between contrasting environmental conditions, dividing populations along a gradient of gene flow (allopatry, parapatry, local mating and sympatry).

After 53 sexual cycles, fitness components for asexual growth, reproductive efficiency and performance after top and bottom selection were quantified. The degree of adaptation and the strength and direction of the underlying fitness trade-offs showed strong dependency on gene flow. Isolated popula-

tions adapting to either of the two habitats (allopatry) showed consistent convergent adaptation, resulting in population specialisation for local conditions. In the presence of gene flow divergence between top and bottom populations was lost. However, populations with the highest level of gene flow (sympatry) evolved two phenotypically differentiated subpopulations similar to the allopatric populations. Yet, one of the subpopulations was characterized by a generalist strategy with improved performance after both top and bottom selection. Populations experienced a consistent intrinsic trade-off between asexual growth and reproductive efficiency across all levels of gene flow. Other correlations between ecological fitness components, however, depended on gene flow and genetic background. For instance, under high levels of gene flow (local mating, sympatry) growth and performance under both ecological selection were negatively correlated, while there was no relationship in allopatry and parapatry.

Whole-genome re-sequencing of all 132 populations revealed that in contrast to the variation and high complexity of traits under selection, variance in fitness components was mostly attributed to few standing genetic variants in the absence of gene flow. Under conditions of high gene flow, however, adaptation was mostly driven by novel, population specific mutations which were enriched in a subset of genes. We found that this difference may be attributed to the presence of multiple standing genetic variants, each under antagonistic pleiotropy, favoured in one environment but deleterious under alternative ecological conditions. As the pleiotropic cost was only present under conditions of gene flow, adaptation with gene flow relied on the emergence of new mutations subject to conditional neutrality or beneficial in both ecological conditions. Additionally, in the experiment we identified pairs of loci in close proximity showing opposite antagonistic pleiotropy, i.e. the derived mutations were beneficial in opposite environments. Coupling of loci subject to divergent selection is expected promote reproductive isolation.

We presented empirical evidence for convergence shaping life history trait evolution and the genetic composition of populations. Gene flow modulated life history correlations and introduced a shift in strategy from specialisation to one environment to generalist genotypes. Depending on the level of gene flow, fitness variation was strongly driven by both standing and new genetic variation under opposite antagonistic pleiotropy.

Paper II

Repeated evolution of self-compatibility for reproductive assurance

Sexual reproduction is a wide spread characteristic for the majority of eukaryotic organisms. Yet, while sexual reproduction presents potential evolutionary advantages such as increased adaptive rates (Muller 1932; Maynard Smith 1971) it requires the costly fusion of two compatible gametes of opposite sexes or mating types. In fungi, mate compatibility is regulated by a mating-types locus, such that only individuals with different alleles in this locus (e.g. *Plus* and *Minus* alleles) can mate. This presents a potential challenge, as the probability of encountering a suitable partner can be reduced by up to 50% when two sexes or mating types exist (Fisher 1930). This could have significant effects on individual and population fitness, especially in structured, lowly dense populations with an obligatory sexual phase in their lifecycle (Nieuwenhuis and James 2016).

The evolutionary responses to the selective pressures for mating can be self-compatibility, hermaphroditism, or in fungi, homothallism. In the later, an individual is compatible with all others, including itself and thus can perform intra-clonal mating, while maintaining the potential to outcross. One common form of homothallism is mating-type switching, also present in *Schizosaccharomyces pombe*, in which individuals have genetic information for the two mating types, but only one is active. During asexual growth, daughter cells render the active locus, switching between mating type alleles.

While the molecular mechanisms of mating-type switching are known in much detail, little is known about its evolutionary origin and the forces that drive the selection for self-compatibility at the population level. Different species of yeast have the switching or the non-switching phenotype, and even populations within species can be polymorphic for the trait. In this study, we explored potential molecular, ecological and evolutionary drivers for the emergence and persistence of switching and non-switching reproductive strategies.

Using phylogenetic analyses we inferred the evolutionary history of mating-type region in 57 differentiated worldwide clades. We found substantial phenotypic variation in switching ability accompanied by structural genetic diversity suggesting multiple inferred transitions from the ancestral state of switching to a non-switching strategy. Next, we tested the emergence of the switching strategy in the laboratory, conducting an evolutionary experiment, in which selection for mating was applied under low population densities of non-switching parental strains. We found that after 25 cycles of selection,

nine of the 20 evolved replicate populations contained the switching phenotype in frequencies from 0.15 to 1.0. The presence of both mating alleles in individual strains and the switching mechanism was confirmed by PCR and whole genome long-read SMRT sequencing.

The remarkable ease with which the novel mating-type switching genotypes arose and could be selected for under experimental conditions suggests that this trait can, in principle, readily emerge and persist in nature. Using fitness measurements and individual based simulations we explored the benefits and costs of switching during sexual and asexual reproduction. As predicted, we found that the switching phenotype had a strong selective advantage during sexual reproduction, intensified under low population densities. However, we also demonstrated a fitness cost for switching during asexual growth with switching strains incurring the cost of lower growth rates.

Our findings illuminate a trade-off between the benefits of reproductive assurance and its fitness costs during prolonged asexual growth. This trade-off may explain the polymorphic condition of mating strategies in natural populations. Our data predict that non-switcher strains would be more prevalent in stable and rich environmental conditions, or in locations where resources are regularly replenished. Conversely, switcher strains would have an adaptive advantage under temporally unstable environments where cycles of sexual reproduction are selectively favoured. These results emphasize the importance of ecological factors driving the evolution of mating strategies.

Paper III

Ancestral admixture is the main determinant of global biodiversity in fission yeast

Events of natural hybridization between diverging linages have been extensively recognised as an important evolutionary process and key element for diversification (Abbott et al. 2013). A few extensively studied examples of hybridization have shown its role in adaptive radiation, transfer of adaptive loci within and between species, and the formation of novel, ecologically differentiated species. Upon hybridization, new combinations of genetic variants from divergent parental linages emerge, consequently expanding phenotypic variation. When combined along the genome, adaptive variants from the parental linages can be compatible, enhance adaptation and by the interaction of recombination and selection are expected to be brought into linkage. Novel combinations may however also be incompatible and reduce fitness in hybrids. Selection and heterogeneous recombination thus will lead

to differential admixture of the two parental backgrounds along the genome of hybrids.

In this study, we integrated whole-genome sequencing data from a global sample collection of the fission yeast *Schizosaccharomyces pombe* to explore the role of hybridization during adaptation. We found that the evolutionary history of the species involves recent hybridization between two divergent ancestral populations. Hybridization occurred within the last 20-60 sexual outcrossing generations, a timeframe consistent with recent human-induced migration at the onset of intensified transcontinental trade. Comparing the distribution of ancestral blocks along the genome between all samples identified 8 discrete clusters sharing similarly in admixture histories. These clusters did not group geographically suggesting recent global dispersion after hybridization followed by local differentiation.

Next, we assessed the consequences of hybridization on phenotypic variation making use of a large data set of quantitative traits. Phenotypic variation in over 200 measured phenotypes was strongly influenced by hybridization between ancestral populations. Generally, phenotypes in hybrids resembled the parental distribution (dominant phenotypes), or were intermediate between parental values. In several cases, hybrid phenotypes exceeded the distribution of both parental populations (transgressive phenotypes), a feature considered important for the colonization of new environments. Heritable phenotypic variation was explained near-exclusively by haplotype blocks distribution and shared ancestry. Likewise, the degree of postzygotic reproductive isolation quantified as spore viability was very well predicted by the degree of shared ancestry.

Haplotype block distribution along the genome showed large variation in heterogeneity in ancestry exceeding neutral expectations. We tested if this heterogeneity could be explained by incompatibilities of large effect structural mutants, including insertions, deletions, inversion and translocations. We predicted SVs from *de novo* genomes and found little effect on haplotype distribution. SVs were found to be in low frequency with little divergence between ancestral populations. This suggests that the observed SVs with phenotypic and fitness effect were recent, and more generally, that SV were under strong purifying selection. Conversely, we found strongly increased statistical linkage between ancestral populations along the genome, even over large genomic regions. This is consistent with negative epistatic interactions of incompatible genetic backgrounds contributing to the heterogeneity in the distribution and frequency of ancestry along the genome.

This study provides detailed insight into the evolutionary forces and phenotypic consequences of admixture in natural populations. It highlights the

importance of hybridization in generating biodiversity and promoting novel evolutionary trajectories.

Paper IV

Intraspecific diversity of fission yeast mitochondrial genome

The mitochondrion is a vital organelle within eukaryotic cells that catalyses an exothermal process providing chemical energy accessible to the cell. Mitochondria contain their own genetic material separated from the nuclear genome. This property can been exploited to obtain an independent assessment an organism's evolutionary history. Surprisingly in *Schizosaccharomyces pombe*, despite its importance as a model system in cell biology, little is known about its ecology, origin, and evolutionary history (Jeffares 2018).

In this study, we provide new insights into mitochondrial diversity and evolutionary history of natural *S. pombe* population samples. We performed *de-novo* mitochondrial genome assembly and annotation from 192 globally distributed samples. All strains contained a total of 69 non-clonal haplotypes substantially varying in length from 17 to 26 kb. Differences in mitochondrial size were due to variation in presence–absence polymorphisms of intronic sequence. Additional to the 7 known introns in *S. pombe* (*cox1-I1a, cox1-I1b, cox1-I2a, cox1-I2b, cox1-I3, cob-I1,* and *cox2-I1*), 3 of which are present in the reference strain (*cox1-I1b, cox1-I2b,* and *cob-I1*), we identified 2 additional introns (*cox1-I1b0* and *cox1-I4*). *Cox1-I1b0* showed orthology to *cox1-I1b* when present, but sequence similarity between them was below 71% for most of the sequence. *Cox1-I4* did not share close relationship with any known intron, and instead showed higher sequence similarity with the highly divergent *cox1-ai1* and *cox1-ai2* introns in *S. cerevisiae.* This observation suggests that *Cox1-I4* may have been inserted by horizontal gene transfer from a species in the Saccharomycetaceae clade.

Based on patterns of mitochondrial diversity, we infer that all haplotypes originated from two ancestral populations which recently hybridized; a result consistent with previous work focussing on the nuclear genome (Tusso et al. 2019). Despite high levels of inter-lineage admixture, for the large majority of strains we found no evidence of mitochondrial recombination. We calculated that the divergence time between the two ancestral populations was around 31 million generations ago. Assuming the generation time in the wild for *S. pombe* is similar the common yeast (*Saccharomyces cerevisiae*), the estimated number of generations corresponds to around 78000 years. Although we emphasize the high uncertainty of this estimate in years, diver-

gence time between ancestral populations would fall within the most recent glacial period between 110000 and 12000 years ago. We speculate that the expansions of ice sheets resulted in fragmentation of the ancestral population into two allopatric subpopulations, one of them restricted to southern Europe, and the second one predominating in Asia. These subpopulations may have recently hybridized upon secondary contact, a process believed to be associated with human migration (see paper III).

These results contribute to our understanding of the evolutionary history of *S. pombe* and contribute genomic resources for future studies.

Conclusions and future prospects

What is the origin of biodiversity? Insights on evolutionary processes resulting in population differentiation and speciation rely on clear understanding on the origin of genetic variation, its translation into phenotypic variation, and how phenotypes within the population are environmentally selected. Advances in theoretical work with a constant increase in our ability to generate genomic data, has allowed interpreting the evolutionary history of natural populations in a suite of organisms. However, theoretical predictions remain challenging since interactions between intrinsic genomic properties, external environmental pressures and demographic processes can result in similar genomic patterns.

In this thesis, I present a series of studies in which multiple approaches were integrated to unravel the evolutionary processes governing adaptive divergence. These include comparative genomics, population genetic simulations and analyses, as well as experimental evolution. This work is centred on the fission yeast *Schizosaccharomyces pombe*, a cell biological model species that has so far gained little attention in evolutionary research. This thesis is a clear example of its potential to tackle fundamental questions in evolutionary biology.

All four chapters of the thesis provide strong evidence of the pervasive impact of gene flow and genetic admixture on the resulting distribution of phenotypic variation and adaptive divergence between populations. The effect of gene flow was tested in two scenarios: first, under experimental evolution phenotypic variation and local adaptation emerging under differentiated environments was modulated by different levels of gene flow; second, variation in phenotypic traits and reproductive isolation resulted after secondary contact of natural already divergent populations (hybridization). We demonstrated that the evolutionary trajectory is affected by gene flow, shifting from local specialisation in the absence of gene flow, to reduced adaptive divergence and emergence of a generalist strategist under high levels of gene flow. This work not only adds to the evidence that hybridization is an important evolutionary process. It dissects the effect of gene flow relative to other variables and their interaction like population evolutionary history, environmental stability and selection regime it experiences, available stand-

ing genetic variation, genetic architecture of the traits under selection and intrinsic organismal and ecologically induced phenotypic trade-offs.

In addition to selection acting on independent genetic variants, an important result from this thesis is the apparent persistence of genic "adaptation hotspots", gene interaction and linkage between genetic variants. These interactions showed to have a profound effect in the resulting genetic and phenotypic variation observed in both experimental and natural populations. For instance, genetic variants in experimental populations rarely reached fixation. Instead, evolutionary convergence was observed at the gene level, with multiple alternative variants in the same set of genes. Several of these variants responded to disruptive selection with antagonistic pleiotropy effect, acting in both synergistic and opposite directions. Similarly, genetic composition after hybridisation in natural populations showed to be non-random showing large variation in the heterogeneity of introgression from both ancestral populations suggesting selective constrains for admixture between genetic backgrounds.

Furthermore, this thesis presents insights into the effect of structural variation on phenotypic divergence. Structural variation in *S. pombe* has previously been shown to have large effect on phenotypic variation and reproductive isolation. Consistent with this notion, SVs induced by non-homologous recombination in the mating type locus played an important role in the reestablishment of self-compatibility during experimental evolution and for naturally occurring polymorphism in mating system. However, genome-wide genetic variation of SV showed a different picture. Here, the evolution of structural variants was heavily restricted by selective constrains, with little divergence driven by selective sweeps from structural mutations nor by neutral processes like genetic drift. Although we identified a large set of structural variants, they showed to be under strong purifying selection, resulting in little long term divergence between populations. Observed structural variants were rather recent, in low frequency and population specific.

The high complexity of different factors interacting during evolution with gene flow makes it difficult to make general conclusions for other systems. To date only a limited number of studies have directly tested the role of gene flow on adaptive divergence experimentally. This thesis thus contributes valuable results, but at the same time, it exposes the potential for significantly different results under alternative experimental conditions or systems. Experimental evolution is a promising approach, which combined with comparative genomics can shed light on the evolutionary history of species and factors driving it. Additional empirical work both in *S. pombe* and beyond is needed, testing alternative conditions and phylogenetic groups in order to identify consistent general patterns.

Svenks Sammanfattning

Kunskapen om hur differentiering och adaptation sker i naturen är fundamental för att förstå biodiversitetens ursprung. Nya teoretiska resultat inom artbildning, tillsammans med den ökande kapaciteten att på ett enkelt sätt generera sekvensdata från hela genom, har främjat en utforskning av de genomiska förändringar som är relevanta för adaptationsprocesser. Dock är det fortsatt utmanande att uttyda evolutionära processer från genomiska divergensmönster, som styrs både av intrinsiska genomegenskaper och extrinsiska selektionsstryck. Den kronologiska ordningen av genomförändringar samt de evolutionära krafter som driver divergensprocesser kan sällan uttydas direkt från naturliga populationer av sexuellt reproducerande organismer.

För tillfället ser jag två lovande väger att hantera frågan om divergensens genomiska ursprung: (1) evolutionära experiment med syfte att simulera divergensprocesser och mäta genomiska förändringar medan de sker; (2) empiriska fallstudier av närbesläktade arter och populationer i vilka divergensens omfattning varierar, vilket tillåter oss att inferera den kronologiska ordningen av genomförändringar och inflytandet av andra faktorer. I min doktorandforskning applicerade jag dessa två tillvägagångssätt genom att använda svampen *Schizosaccharomyces pombe* som modellsystem. Först testade jag experimentellt potentialen för ekologisk divergens med genflöde (d.v.s. utan geografiska barriärer mellan populationer), och undersökte genomiska och fenotypiska förändringar associerade med denna process. Sedan kompletterade jag de experimentella resultaten genom att studera genomdata tillförskansade från naturliga populationer världen över. I både de experimentella och empiriska tillvägagångssätten baserades den genetiska inferensen på en varietet av sekvenseringsteknologier såsom Illumina-, Pacbio- och MinIonsekvensering.

De evolutionära experimenten utforskade effekten av genflöde på fenotyp- och fitnessfördelning samt påvisade potentiella molekylära mekanismer underliggandes den adaptiva divergensen (Artikel I). Vi påvisade uppkomsten av lokal specialisering under en låg nivå av genflöde och generalism, när genflödesnivån tilläts vara högre. Utvecklade fenotyper var främst influerade av den redan tillgängliga genetiska variationen under motsatt antagonistisk pleiotropi. Även nya mutationer berikade vissa gener. Det experimentella selektionsskicket visade sig också ha en stark effekt på parningsstrategi som

ett resultat av temporär ekologisk heterogenitet och selektion för parningseffektivitet (Artikel II). Evolutionen av parningsstrategier kan förklaras av en "trade-off" mellan parningseffektivitet och asexuell tillväxt, beroende av stabilitet i miljön. I naturliga populationer påvisade vi att genflöde har en essentiell roll i adaptiv divergens. Vi visade att alla kända *S. pombe* släktlinjer var och en har uppstått ur nyligen skedda hybridiseringar mellan två ursprungliga grupper med stora effekter på fenotypisk variation och reproduktiv isolering (Artikel III). Den demografiska historien som uttyds ur kärngenomet bekräftades med mitokondrisk genetisk diversitet, vilket är ytterligare bevis för de ursprungliga populationernas geografiska hemvist och potential för horisontell genöverföring från en avlägsen grupp av jäst (Artikel IV).

Translated to Swedish by Jesper Boman

Acknowledgements

First and foremost, I would like to thanks my supervisor, Jochen Wolf, for his boundless patience, guidance and support throughout this large journal. Your fascination and enthusiasm for science is admirable, and it was a constant motivation to persuade through the multiple challenges. It has been already some years since the moment I came for the first time to your office. Then, close to finish my master, I think talking to you for the first time, I clearly did not have idea what I was getting into. That was our first two to three hours meeting, out of many more coming…. By then, I had been working in this project probably for almost a year, and contrary to the confused faces I normally would get from people when hearing about my master thesis, I was surprised to find the opposite. What I found was a massive excitement. You were very quickly standing, drawing matrices and plots on the board, and discussing all the endless possibilities of studies with the experiment. Such a good energy and love for science. I cannot think of a better way to start a PhD. A couple of months later I was starting my PhD in the group. Thanks a lot for having always the doors open, for your constant and valuable input, and especially for your confidence in my abilities, always finding the way to promote my ideas (no so good for the crow folks who could not sequence as many samples thanks to me). The head of the department never seems to be busy enough for a short two or three hour's discussion on a single plot, which I always appreciated. It has been a pleasure to work with you and I look forward to more collaboration in the future.

I am also infinitely graceful to my co-supervisor Simone Immler. This project would have not been possible but with the support from the person who first believed it may be a good idea. Thanks a lot for putting your hand and trust on an idea that back then was just that, *an idea*. Who would image this idea would grow into a whole PhD thesis. I would like to express my gratitude for welcome me to the group and for you constant support and critical comments. Thanks a lot for the positive attitude and for being such a friendly person.

Many thanks to current and past members of the Department of Ecology and Genetics for creating such as amazing working environment. First, it was fantastic to always have someone to ask and an expert for almost any topic. But most importantly, thanks a lot for being so friendly and great people.

First, I would like to deeply thank Bart Nieuwenhuis. My pombe buddy. I do not think neither of us would have imaging that we will end up working together for so much time. Such a combination, ah?! I want to thank you for many things, for taking the time to teach me all the basics to manipulate pombe. Thanks you for supervising my Master project and for the constant scientific input since then. I have learnt a lot of things from you. Not only restricted to science, for instance, now I read my emails twice before sending them and my pronunciation of the *v* and *b* has improved a lot. Thanks a lot for consider my opinion in all decision from the beginning and for the infinite patience to discuss for hours over and over the same point. Five years later and I am still not sure who is the most stubborn between us, but I am sure the combination definitely led to better results and more consistent conclusions. Thanks a lot for all the helping with the reading, writing, scripting, in the lab during holidays, and more. I especially would like to thank you for all the interesting chats. I do not think there can be awkward silences with you, you have an incredible ability to bring interesting (and random) topics to talk about, and I really enjoyed it.

Many thanks to Sylvain Glémin and Martin Lascoux. I own you most of my understanding to population genetics and coalescent theory. Thanks a lot for helping me out with my basic questions in the most patient way. Thanks to Douglas Scofield. There was always an excuse with a technical bioinformatics error or an *Uppmax* problem to have a nice conversation with you. Conversations with you were always very intellectually stimulating and chill so I enjoy them a lot. Plus, you solved my problems like in less than one minute. Thanks to Alex Suh and Valentina Peona. I would be still trying to figure out what a TE is if not for your help. Thanks for taking the time to listen to my results and my many questions on repeats, and for being always very friendly and nice. I would like to thank Annette Berger and Frida Svedberg. I do not know how many times I asked exactly the same question, or requested exactly the same certificates or information. Thanks a lot for replying every single time equally patient. You saved me more than once under my administrative ineptitude. Thanks a lot to Jesper Boman who translated the summary to Swedish. I would like to thank a very especial group of nerds with whom I shared a lot of time trying to understand the mysteries of the sometimes indecipherable equations contained in the Gillespie's book. That was a lot of fun! Willian Silva, Lore Ament, Paulina Bolibar, Homa Papoli, and Matthias Weissensteiner.

My life in Uppsala was full of exciting, fun and special moments thanks to several people and friends. Now I am full of very nice memories thanks to you all. Willian, although it may see as we are constantly discussing without sense (mostly you), I really thank you for being around. You are such a nice friend to share some personal thoughts and random ideas. Paulina, *mi chavi-*

ta, *no mames*, you are the best. I really liked to chat with you, mostly while eating and gossiping. You constantly brought the Mexican happiness just by talking. Just thinking on you makes me smile and I thank you for that. Sarahi, it is difficult to find people with similar sense of humour. I thank you for all those histories you used to tell me which would make me laugh for hours. Lore, thanks for being always there, for all your scientific input, for helping me out with scripts (quite handy), for reading three chapters of my thesis!, for helping me with the whole preparation of my defence, but especially for being such a honest and reliable friend. I really enjoyed all the fantastic adventures with you. Many nice memories. Thanks to my favourite Ozzie and dancing partner, Alex C. It was a lot of fun to be around you. I really enjoyed our time dancing salsa and bachata or just talking about random stuff. You are such a lovely and special person. It is very easy to get in a good mood while talking to you. Homa, thanks a lot for taking the time to organize material in order to understand the variable basic concepts in calculus, trigonometry, geometry and others. I always found your discipline highly remarkable and an example to follow. I really appreciate the interest and dedication you put to everything. You are a fantastic scientist and great friend. Berrit, thanks a lot for saving me when I was lost and without money in Turkey. I always remember that, you were my hero. Ghazal, although you want to look like disinterested, dry and in your own business, you are just the sweetest person ever. Thanks for the good times. Roy, the grumpy cat, such a cool guy. The true is, you are always super nice and in a good mood. TJ!, one of the nicest persons I have ever met. You are the definition of goodness and appreciate the great moments with your fantastic sarcastic sense of humour. We need to practice more our secret handshake. Venkat, my friend, you introduced me to the world of the chicken biryani and I will never forget that. Mercè, although we never managed to understand each other in Spanish, I really enjoy to chat to you and it was a lot of fun to keep trying. Foteini, tons of great memories. Thanks a lot for the great tour in Greece, for being such a great and lovely person and for teaching me *stiff salsa*. Who would have though *φιλάμε μανιχα* would be so effective?.... Mooscito Blom, thanks a lot for making every single trip/dinner/gathering so much fun. Thanks a lot to Nina for all the fun playing complicated board games, without pushing me to play fast.

After leaving Uppsala, coming to Munich by myself was made less daunting with the fantastic energy of many people I met there. I would like to especially thank Ana, Josh, Claire, Matthias, Bernadette, Sebis, Ricardo, Uli, Bart, Fidel, Steffi, Benedict, Vera, Richard, Justin, Saurabh, James, Marlen, Elisa, Catarina, Clara and Arka with whom I spent most of my time in Munich. Thanks a lot to my extraordinary office mates Claire and Matthias. That was a very interesting and fun combination. The mix of peculiarities of each of us made this a unique working experience. Odd as it sounds I really

appreciated the mix of eccentricities, the singing, the drumming, the awkward staring "at the window", the regular changes of angle for the window and the door, as well as the discussions about the right moment to close the blinds or the disturbance of the lights. Added to the friendly and always positive attitude, I thank you for the constant input on figures, writing, politics, science, politics in science, family, life, etc. I would like to thank Josh for being such a great friend, for the positive energy, for reading the kappa, for bringing me gossip from other countries, and especially for all those cakes almost every week! Thanks a lot to my favourite *guatemalteca* friend, Ana, I loved all the time we spent together, it was a lot of fun, thanks a lot for bringing a little piece of the Latino American energy. Ah, and thanks for teaching me about the *Guacamol*, the *Enchilad,* and the *Tacs,* always with good corn *tortills*. I would like to thank Uli and Ricardo, I really admire your attitude towards science and it was always very rewarding and exciting to talk about it with you. I also thank Ricardo for helping me finding a new apartment. Sebis, always willing to help and to make others happy, thanks a lot for being a great friend.

I am also infinitely grateful to Ingrid, Hilde, Gay, Ana, and especially to Bernadette. Mit all der Hilfe, die Sie mir gaben, habe ich tatsächlich die Zeit, an meiner Diplomarbeit zu arbeiten. Vielen Dank, dass du immer so freundlich und geduldig mit meinem Mangel an Deutsch bist.

I thank my MEME buddies Marina, Miya, Lore, and Bere. It is great to have you close all these years. Thanks a lot for making my life in Europe a lot happier. Especial thanks to my friends Sandri, Deivis, Anilla, and David A. I would like to especially thank two people, first Juan Sebastian Lopez Fernandez, a fantastic friend who always accompanied me when I needed a bit of the Colombian air far from my family, and K, who has been constantly there. Listening, motivating, talking, and inspiring. Thanks a lot for all the crazy and cool trips together. Life in academia can be very demanding and sometimes stressful. And I do not think this work would have been possible without having the constant save space I have with Fer. You are my rock and my peace through thick and thin. It is difficult to take a break, isolated from science and you were that, the person that makes me feel safe and gives me comfort. I really thank you for the patients listening and for bringing a lot of new experiences and excitement to my life. No enough words to thank you all.

Finalmente, probablemente los protagonistas de todo y quienes realmente fueron los fundadores de este trabajo. No hay palabras suficiente para agradecer mi familia. Mis padres, quienes a pesar de la distancia, sigues justo ahí, a mi lado, siempre muy atentos a todo lo que pase conmigo. Les dedico este trabajo, no solo por buscar siempre lo mejor para mí y porque

busque siempre mis sueños, sino también por ser un gran ejemplo a seguir. Un agradecimiento muy especial para mis hermanos, y mis sobrinos Aleja y Samuel. El amor por ustedes es mi motos principal de vida. Finalmente quiero enviar un agradecimiento especial a mis tías Marina, Hilda y Miriam. Ya perdí la cuenta del número de veces que ustedes me han dado la mano y una voz de aliento cuando más lo necesitaba. Los amo a todos infinitamente.

References

Abbott, R., D. Albach, S. Ansell, J. W. Arntzen, S. J. E. Baird, N. Bierne, J. Boughman, et al. 2013. Hybridization and Speciation. *Journal of Evolutionary Biology* 26 (2): 229–46.

Abbott, Richard J. 1992. Plant Invasions, Interspecific Hybridization and the Evolution of New Plant Taxa. *Trends in Ecology & Evolution* 7 (12): 401–5.

Agrawal, Aneil F. 2009. Spatial Heterogeneity and the Evolution of Sex in Diploids. *The American Naturalist* 174 (S1): S54–70.

Anderson, Jill T., Cheng-Ruei Lee, Catherine A. Rushworth, Robert I. Colautti, and Thomas Mitchell-Olds. 2013. Genetic Trade-Offs and Conditional Neutrality Contribute to Local Adaptation: Genetic Basis of Local Adaptation. *Molecular Ecology* 22 (3): 699–708.

Aparicio, Oscar M., Barbara L. Billington, and Daniel E. Gottschling. 1991. Modifiers of Position Effect Are Shared between Telomeric and Silent Mating-Type Loci in *S. Cerevisiae*. *Cell* 66 (6): 1279–87.

Arnold, Michael L. 1992. Natural Hybridization as an Evolutionary Process. *Annual Review of Ecology and Systematics* 23 (1): 237–61.

Bacilieri, Roberto, Alexis Ducousso, Rémy J. Petit, and Antoine Kremer. 1996. Mating System and Asymmetric Hybridization in a Mixed Stand of European Oaks. *Evolution* 50 (2): 900–908.

Balkau, Beverley J., and Marcus W. Feldman. 1973. Selection for Migration Modification. *Genetics* 74 (1): 171–74.

Barrett, Rowan D. H., and Hopi E. Hoekstra. 2011. Molecular Spandrels: Tests of Adaptation at the Genetic Level. *Nature Reviews Genetics* 12 (11): 767–80.

Barrett, Rowan D. H., and Dolph Schluter. 2008. Adaptation from Standing Genetic Variation. *Trends in Ecology & Evolution* 23 (1): 38–44.

Bartlett, Lewis J., Lena Wilfert, and Michael Boots. 2018. A Genotypic Trade-off between Constitutive Resistance to Viral Infection and Host Growth Rate. *Evolution* 72 (12): 2749–57.

Bateson, W. 1909. *Heredity and Variation in Modern Lights. In Darwin and Modern Science*. Cambridge: Seward AC (ed). Cambridge University Press.

Beach, D.H., and A.J. Klar. 1984. Rearrangements of the Transposable Mating-Type Cassettes of Fission Yeast. *The EMBO Journal* 3 (3): 603–10.

Behe, Michael J. 2010. Experimental Evolution, Loss-of-Function Mutations, and "the First Rule of Adaptive Evolution." *The Quarterly Review of Biology* 85 (4): 419–45.

Bennett, Albert F., and Richard E. Lenski. 1999. Experimental Evolution and Its Role in Evolutionary Physiology. *Integrative and Comparative Biology* 39 (2): 346–62.

Bennett, Albert F. 2003. Experimental Evolution and the Krogh Principle: Generating Biological Novelty for Functional and Genetic Analyses. *Physiological and Biochemical Zoology* 76 (1): 1–11.

Bentley, David R., Shankar Balasubramanian, Harold P. Swerdlow, Geoffrey P. Smith, John Milton, Clive G. Brown, Kevin P. Hall, et al. 2008. Accurate Whole Human Genome Sequencing Using Reversible Terminator Chemistry. *Nature* 456 (7218): 53–59.

Berg, Jeremy J., and Graham Coop. 2014. A Population Genetic Signal of Polygenic Adaptation. *PLOS Genetics* 10 (8): e1004412.

Bevan, Michael W., Cristobal Uauy, Brande B. H. Wulff, Ji Zhou, Ksenia Krasileva, and Matthew D. Clark. 2017. Genomic Innovation for Crop Improvement. *Nature* 543 (7645): 346–54.

Blanquart, F., and S. Gandon. 2011. Evolution of Migration in a Periodically Changing Environment. *The American Naturalist* 177 (2): 188–201.

Blows, Mark W., and Ary A. Hoffmann. 2005. A Reassessment of Genetic Limits to Evolutionary Change. *Ecology* 86 (6): 1371–84.

Bono Lisa M., Gensel Catharine L., Pfennig David W., and Burch Christina L. 2015. Evolutionary Rescue and the Coexistence of Generalist and Specialist Competitors: An Experimental Test. *Proceedings of the Royal Society B: Biological Sciences* 282 (1821): 20151932.

Bono, Lisa M., Leno B. Smith, David W. Pfennig, and Christina L. Burch. 2017. The Emergence of Performance Trade-Offs during Local Adaptation: Insights from Experimental Evolution. *Molecular Ecology* 26 (7): 1720–33.

Bourret, Vincent, Mélanie Dionne, and Louis Bernatchez. 2014. Detecting Genotypic Changes Associated with Selective Mortality at Sea in Atlantic Salmon: Polygenic Multilocus Analysis Surpasses Genome Scan. *Molecular Ecology* 23 (18): 4444–57.

Brown, William R. A., Gianni Liti, Carlos Rosa, Steve James, Ian Roberts, Vincent Robert, Neil Jolly, et al. 2011. A Geographically Diverse Collection of Schizosaccharomyces Pombe Isolates Shows Limited Phenotypic Variation but Extensive Karyotypic Diversity. *G3: Genes|Genomes|Genetics* 1 (7): 615–26.

Bürger, Reinhard, and Ada Akerman. 2011. The Effects of Linkage and Gene Flow on Local Adaptation: A Two-Locus Continent–Island Model. *Theoretical Population Biology* 80 (4): 272–88.

Charlesworth, B. 1998. Measures of Divergence between Populations and the Effect of Forces That Reduce Variability. *Molecular Biology and Evolution* 15 (5): 538–43.

Charnov, Eric L. 1989. Phenotypic Evolution under Fisher's Fundamental Theorem of Natural Selection. *Heredity* 62 (1): 113–16.

Chiba, Satoshi. 1993. Modern and Historical Evidence for Natural Hybridization between Sympatric Species in Mandarina (Pulmonata: Camaenidae). *Evolution* 47 (5): 1539–56.

Chin, Chen-Shan, David H. Alexander, Patrick Marks, Aaron A. Klammer, James Drake, Cheryl Heiner, Alicia Clum, et al. 2013. Nonhybrid, Finished Microbial Genome Assemblies from Long-Read SMRT Sequencing Data. *Nature Methods* 10 (6): 563–69.

Clarkson, Chris S., David Weetman, John Essandoh, Alexander E. Yawson, Gareth Maslen, Magnus Manske, Stuart G. Field, et al. 2014. Adaptive Introgression between *Anopheles* Sibling Species Eliminates a Major Genomic Island but Not Reproductive Isolation. *Nature Communications* 5 (June): 4248.

Clément-Ziza, Mathieu, Francesc X Marsellach, Sandra Codlin, Manos A Papadakis, Susanne Reinhardt, María Rodríguez-López, Stuart Martin, et al. 2014. Natural Genetic Variation Impacts Expression Levels of Coding, Non-Coding, and Antisense Transcripts in Fission Yeast. *Molecular Systems Biology* 10 (11): 764.

Colomé-Tatché, Maria, and Frank Johannes. 2016. Signatures of Dobzhansky–Muller Incompatibilities in the Genomes of Recombinant Inbred Lines. *Genetics* 202 (2): 825–41.

Cooper, Tim F, and Richard E Lenski. 2010. Experimental Evolution with *E. Coli* in Diverse Resource Environments. I. Fluctuating Environments Promote Divergence of Replicate Populations. *BMC Evolutionary Biology* 10 (1): 11.

Cooper, Vaughn S., and Richard E. Lenski. 2000. The Population Genetics of Ecological Specialization in Evolving *Escherichia Coli* Populations. *Nature* 407 (6805): 736–39.

Cruickshank, Tami E., and Matthew W. Hahn. 2014. Reanalysis Suggests That Genomic Islands of Speciation Are Due to Reduced Diversity, Not Reduced Gene Flow. *Molecular Ecology* 23 (13): 3133–57.

Cutler, David J., and Jeffrey D. Jensen. 2010. To Pool, or Not to Pool? *Genetics* 186 (1): 41–43.

Daub, Josephine T., Tamara Hofer, Emilie Cutivet, Isabelle Dupanloup, Lluis Quintana-Murci, Marc Robinson-Rechavi, and Laurent Excoffier. 2013. Evidence for Polygenic Adaptation to Pathogens in the Human Genome. *Molecular Biology and Evolution* 30 (7): 1544–58.

Dillon, Marcus M., Nicholas P. Rouillard, Brian Van Dam, Romain Gallet, and Vaughn S. Cooper. 2016. Diverse Phenotypic and Genetic Responses to Short-Term Selection in Evolving *Escherichia Coli* Populations. *Evolution* 70 (3): 586–99.

Dittmar, Emily L., Christopher G. Oakley, Jeffrey K. Conner, Billie A. Gould, and Douglas W. Schemske. 2016. Factors Influencing the Effect Size Distribution of Adaptive Substitutions. *Proceedings of the Royal Society B: Biological Sciences* 283 (1828): 20153065.

Dobzhansky, Th. 1934. Studies on Hybrid Sterility. *Zeitschrift Für Zellforschung Und Mikroskopische Anatomie* 21 (2): 169–223.

Dowling, Thomas E., Gerald R. Smith, and Wesley M. Brown. 1989. Reproductive Isolation and Introgression between *Notropis Cornutus* and *Notropis Chrysocephalus* (Family Cyprinidae): Comparison of Morphology, Allozymes, and Mitochondrial DNA. *Evolution* 43 (3): 620–34.

Duffy, Siobain, Paul E. Turner, and Christina L. Burch. 2006. Pleiotropic Costs of Niche Expansion in the RNA Bacteriophage Φ6. *Genetics* 172 (2): 751–57.

Dunn, Casey W., Gonzalo Giribet, Gregory D. Edgecombe, and Andreas Hejnol. 2014. Animal Phylogeny and Its Evolutionary Implications. *Annual Review of Ecology, Evolution, and Systematics* 45 (1): 371–95.

Duvaux, Ludovic, Quentin Geissmann, Karim Gharbi, Jing-Jiang Zhou, Julia Ferrari, Carole M. Smadja, and Roger K. Butlin. 2015. Dynamics of Copy Number Variation in Host Races of the Pea Aphid. *Molecular Biology and Evolution* 32 (1): 63–80.

Egel, Richard. 1973. Commitment to Meiosis in Fission Yeast. *Molecular and General Genetics MGG* 121 (3): 277–84.

Eid, John, Adrian Fehr, Jeremy Gray, Khai Luong, John Lyle, Geoff Otto, Paul Peluso, et al. 2009. Real-Time DNA Sequencing from Single Polymerase Molecules. *Science* 323 (5910): 133–38.

Ekblom, Robert, and Jochen B. W. Wolf. 2014. A Field Guide to Whole-Genome Sequencing, Assembly and Annotation. *Evolutionary Applications* 7 (9): 1026–42.

Endler, John A. 1977. *Geographic Variation, Speciation, and Clines*. Princeton University Press.

Ewens, W. J. 1974. A Note on the Sampling Theory for Infinite Alleles and Infinite Sites Models. *Theoretical Population Biology* 6 (2): 143–48.
Fan, Shaohua, and Axel Meyer. 2014. Evolution of Genomic Structural Variation and Genomic Architecture in the Adaptive Radiations of African Cichlid Fishes. *Frontiers in Genetics* 5.
Fantes, Peter A., and Charles S. Hoffman. 2016. A Brief History of Schizosaccharomyces Pombe Research: A Perspective Over the Past 70 Years. *Genetics* 203 (2): 621–29.
Fawcett, Jeffrey A., Tetsushi Iida, Shohei Takuno, Ryuichi P. Sugino, Tomoyuki Kado, Kazuto Kugou, Sachiko Mura, et al. 2014. Population Genomics of the Fission Yeast Schizosaccharomyces Pombe. *PLOS ONE* 9 (8): e104241.
Feder, Jeffrey L., Stewart H. Berlocher, Joseph B. Roethele, Hattie Dambroski, James J. Smith, William L. Perry, Vesna Gavrilovic, Kenneth E. Filchak, Juan Rull, and Martin Aluja. 2003. Allopatric Genetic Origins for Sympatric Host-Plant Shifts and Race Formation in Rhagoletis. *Proceedings of the National Academy of Sciences* 100 (18): 10314–19.
Feder, Jeffrey L., Scott P. Egan, and Patrik Nosil. 2012. The Genomics of Speciation-with-Gene-Flow. *Trends in Genetics* 28 (7): 342–50.
Feder, Jeffrey L., Richard Gejji, Sam Yeaman, and Patrik Nosil. 2012. Establishment of New Mutations under Divergence and Genome Hitchhiking. *Philosophical Transactions of the Royal Society B: Biological Sciences* 367 (1587): 461–74.
Felsenstein, Joseph. 1981. Skepticism towards Santa Rosalia, or Why Are There so Few Kinds of Animals? *Evolution* 35 (1): 124–38.
Feulner, Philine G. D., Frédéric J. J. Chain, Mahesh Panchal, Christophe Eizaguirre, Martin Kalbe, Tobias L. Lenz, Marvin Mundry, et al. 2013. Genome-Wide Patterns of Standing Genetic Variation in a Marine Population of Three-Spined Sticklebacks. *Molecular Ecology* 22 (3): 635–49.
Fisher, Kaitlin J., Sean W. Buskirk, Ryan C. Vignogna, Daniel A. Marad, and Gregory I. Lang. 2018. Adaptive Genome Duplication Affects Patterns of Molecular Evolution in *Saccharomyces Cerevisiae*. Edited by Sarah Perin Otto. *PLOS Genetics* 14 (5): e1007396.
Fisher, Ronald Aylmer. 1930. *The Genetical Theory of Natural Selection: A Complete Variorum Edition*. Oxford University Press.
Flagel, Lex E., John H. Willis, and Todd J. Vision. 2014. The Standing Pool of Genomic Structural Variation in a Natural Population of *Mimulus Guttatus*. *Genome Biology and Evolution* 6 (1): 53–64.
Fonseca, Dina M., Nusha Keyghobadi, Colin A. Malcolm, Ceylan Mehmet, Francis Schaffner, Motoyoshi Mogi, Robert C. Fleischer, and Richard C. Wilkerson. 2004. Emerging Vectors in the Culex Pipiens Complex. *Science* 303 (5663): 1535–38.
Fournier-Level, A., A. Korte, M. D. Cooper, M. Nordborg, J. Schmitt, and A. M. Wilczek. 2011. A Map of Local Adaptation in *Arabidopsis Thaliana*. *Science* 334 (6052): 86–89.
Fowler, Kyle R., Mariko Sasaki, Neta Milman, Scott Keeney, and Gerald R. Smith. 2014. Evolutionarily Diverse Determinants of Meiotic DNA Break and Recombination Landscapes across the Genome. *Genome Research* 24 (10): 1650–64.
Franssen, S U, R Kofler, and C Schlötterer. 2017. Uncovering the Genetic Signature of Quantitative Trait Evolution with Replicated Time Series Data. *Heredity* 118 (1): 42–51.
Fry, James D. 1996. The Evolution of Host Specialization: Are Trade-Offs Overrated? *The American Naturalist* 148 (November): S84–107.

Gardner, Kyle M., and Robert G. Latta. 2006. Identifying Loci under Selection across Contrasting Environments in *Avena Barbata* Using Quantitative Trait Locus Mapping. *Molecular Ecology* 15 (5): 1321–33.
Garland, Theodore, and Michael Robertson Rose. 2009. *Experimental Evolution: Concepts, Methods, and Applications of Selection Experiments*. University of California Press Berkeley, CA.
Gee, Jennifer M. 2004. Gene Flow across a Climatic Barrier between Hybridizing Avian Species, California and Gambel's Quail (*Callipepla Californica* and *C. Gambelii*). *Evolution* 58 (5): 1108–21.
Gibson, Brian, and Gianni Liti. 2015. *Saccharomyces Pastorianus*: Genomic Insights Inspiring Innovation for Industry. *Yeast* 32 (1): 17–27.
Gladieux, Pierre, Jeanne Ropars, Hélène Badouin, Antoine Branca, Gabriela Aguileta, Damien M. de Vienne, Ricardo C. Rodríguez de la Vega, Sara Branco, and Tatiana Giraud. 2014. Fungal Evolutionary Genomics Provides Insight into the Mechanisms of Adaptive Divergence in Eukaryotes. *Molecular Ecology* 23 (4): 753–73.
Goffeau, A., B. G. Barrell, H. Bussey, R. W. Davis, B. Dujon, H. Feldmann, F. Galibert, et al. 1996. Life with 6000 Genes. *Science* 274 (5287): 546–67.
Gomes, Fátima C.O, Carla Pataro, Juliana B Guerra, Maria J Neves, Soraya R Corrêa, Elizabeth S.A Moreira, and Carlos A Rosa. 2002. Physiological Diversity and Trehalose Accumulation in*Schizosaccharomyces Pombe* Strains Isolated from Spontaneous Fermentations during the Production of the Artisanal Brazilian *Cachaça*. *Canadian Journal of Microbiology* 48 (5): 399–406.
Goto, Derek B., and Jun-ichi Nakayama. 2012. RNA and Epigenetic Silencing: Insight from Fission Yeast: Epigenetic Silencing in Fission Yeast. *Development, Growth & Differentiation* 54 (1): 129–41.
Goyal, Anupama, Masak Takaine, Viesturs Simanis, and Kentaro Nakano. 2011. Dividing the Spoils of Growth and the Cell Cycle: The Fission Yeast as a Model for the Study of Cytokinesis. *Cytoskeleton* 68 (2): 69–88.
Grant, Peter R., B. Rosemary Grant, and K. Petren. 2005. Hybridization in the Recent Past. *The American Naturalist* 166 (1): 56–67.
Gray, Jeremy C., and Matthew R. Goddard. 2012. Gene-Flow between Niches Facilitates Local Adaptation in Sexual Populations. Edited by Michael Bonsall. *Ecology Letters* 15 (9): 955–62.
Günther, Torsten, and Graham Coop. 2013. Robust Identification of Local Adaptation from Allele Frequencies. *Genetics* 195 (1): 205–20.
Hagan, Iain M, Antony M Carr, Agnes Grallert, and Paul Nurse. 2016. *Fission Yeast: A Laboratory Manual*. Cold Spring Harbor Laboratory Press.
Haldane, JBS, and JBS Haldane. 1930. Theoretical Genetics of Autopolyploids. *Journal of Genetics* 22 (03).
Hall, M. C., D. B. Lowry, and J. H. Willis. 2010. Is Local Adaptation in Mimulus Guttatus Caused by Trade-Offs at Individual Loci? *Molecular Ecology* 19 (13): 2739–53.
Hancock, Angela M., David B. Witonsky, Gorka Alkorta-Aranburu, Cynthia M. Beall, Amha Gebremedhin, Rem Sukernik, Gerd Utermann, Jonathan K. Pritchard, Graham Coop, and Anna Di Rienzo. 2011. Adaptations to Climate-Mediated Selective Pressures in Humans. *PLOS Genetics* 7 (4): e1001375.

Hansen, Klavs R., Gavin Burns, Juan Mata, Thomas A. Volpe, Robert A. Martienssen, Jürg Bähler, and Geneviève Thon. 2005. Global Effects on Gene Expression in Fission Yeast by Silencing and RNA Interference Machineries. *Molecular and Cellular Biology* 25 (2): 590–601.

Hanson, Sara J., and Kenneth H. Wolfe. 2017. An Evolutionary Perspective on Yeast Mating-Type Switching. *Genetics* 206 (1): 9–32.

Harigaya, Yuriko, and Masayuki Yamamoto. 2007. Molecular Mechanisms Underlying the Mitosis–Meiosis Decision. *Chromosome Research* 15 (5): 523–37.

Hedrick, Philip W. 2013. Adaptive Introgression in Animals: Examples and Comparison to New Mutation and Standing Variation as Sources of Adaptive Variation. *Molecular Ecology* 22 (18): 4606–18.

Heim, Lutz. 1990. Construction of an h +Sstrain of Schizosaccharomyces Pombe. *Current Genetics* 17 (1): 13–19.

Hietpas, Ryan T., Claudia Bank, Jeffrey D. Jensen, and Daniel N. A. Bolon. 2013. Shifting Fitness Landscapes in Response to Altered Environments: Fitness Landscapes in Altered Environments. *Evolution* 67 (12): 3512–22.

Hoffman, Charles S., Valerie Wood, and Peter A. Fantes. 2015. An Ancient Yeast for Young Geneticists: A Primer on the *Schizosaccharomyces Pombe* Model System. *Genetics* 201 (2): 403–23.

Hoffmann, Ary A., and Loren H. Rieseberg. 2008. Revisiting the Impact of Inversions in Evolution: From Population Genetic Markers to Drivers of Adaptive Shifts and Speciation? *Annual Review of Ecology, Evolution, and Systematics* 39 (1): 21–42.

Houle, David. 1991. Genetic Covariance of Fitness Correlates: What Genetic Correlations Are Made of and Why It Matters. *Evolution* 45 (3): 630–48.

———. 1998. How Should We Explain Variation in the Genetic Variance of Traits? *Genetica* 102 (0): 241.

Hu, Wen, Zhao-Di Jiang, Fang Suo, Jin-Xin Zheng, Wan-Zhong He, and Li-Lin Du. 2017. A Large Gene Family in Fission Yeast Encodes Spore Killers That Subvert Mendel's Law. *ELife* 6 (June).

Jakobsson, Mattias, Michael D Edge, and Noah A Rosenberg. 2013. The Relationship between FST and the Frequency of the Most Frequent Allele. *Genetics* 193 (2): 515–28.

Jasmin, J.-N., and C. Zeyl. 2013. Evolution of Pleiotropic Costs in Experimental Populations. *Journal of Evolutionary Biology* 26 (6): 1363–69.

Jeffares, Daniel C. 2018. The Natural Diversity and Ecology of Fission Yeast. *Yeast* 35 (3): 253–60.

Jeffares, Daniel C., Charalampos Rallis, Adrien Rieux, Doug Speed, Martin Převorovský, Tobias Mourier, Francesc X. Marsellach, et al. 2015. The Genomic and Phenotypic Diversity of Schizosaccharomyces Pombe. *Nature Genetics* 47 (3): 235–41.

Jones, Felicity C., Manfred G. Grabherr, Yingguang Frank Chan, Pamela Russell, Evan Mauceli, Jeremy Johnson, Ross Swofford, et al. 2012. The Genomic Basis of Adaptive Evolution in Threespine Sticklebacks. *Nature* 484 (7392): 55–61.

Kassen, R. 2002. The Experimental Evolution of Specialists, Generalists, and the Maintenance of Diversity. *Journal of Evolutionary Biology* 15 (2): 173–90.

Kawecki, Tadeusz J. 1997. Sympatric Speciation Via Habitat Specialization Driven by Deleterious Mutations. *Evolution* 51 (6): 1751–63.

Kawecki, Tadeusz J., Richard E. Lenski, Dieter Ebert, Brian Hollis, Isabelle Olivieri, and Michael C. Whitlock. 2012. Experimental Evolution. *Trends in Ecology & Evolution* 27 (10): 547–60.

Kirkpatrick, Mark, and David Lofsvold. 1992. Measuring Selection and Constraint in the Evolution of Growth. *Evolution* 46 (4): 954–71.
Kondrat'eva, V. I., and G. I. Naumov. 2001. The Phenomenon of Spore Killing in *Schizosaccharomyces Pombe* Hybrids. *Doklady Biological Sciences* 379 (1): 385–88.
Kong, X., K. Murphy, T. Raj, C. He, P. S. White, and T. C. Matise. 2004. A Combined Linkage-Physical Map of the Human Genome. *The American Journal of Human Genetics* 75 (6): 1143–48.
Kosheleva, Katya, and Michael M Desai. 2018. Recombination Alters the Dynamics of Adaptation on Standing Variation in Laboratory Yeast Populations. *Molecular Biology and Evolution* 35 (1): 180–201.
Küpper, Clemens, Michael Stocks, Judith E. Risse, Natalie dos Remedios, Lindsay L. Farrell, Susan B. McRae, Tawna C. Morgan, et al. 2016. A Supergene Determines Highly Divergent Male Reproductive Morphs in the Ruff. *Nature Genetics* 48 (1): 79–83.
Kvitek, Daniel J., and Gavin Sherlock. 2013. Whole Genome, Whole Population Sequencing Reveals That Loss of Signaling Networks Is the Major Adaptive Strategy in a Constant Environment. Edited by Jianzhi Zhang. *PLoS Genetics* 9 (11): e1003972.
Lamichhaney, Sangeet, Guangyi Fan, Fredrik Widemo, Ulrika Gunnarsson, Doreen Schwochow Thalmann, Marc P. Hoeppner, Susanne Kerje, et al. 2016. Structural Genomic Changes Underlie Alternative Reproductive Strategies in the Ruff *(Philomachus Pugnax)*. *Nature Genetics* 48 (1): 84–88.
Lasne, Emilien, Camille Anne-Lise Leblanc, and Christian Gillet. 2018. Egg Size Versus Number of Offspring Trade-Off: Female Age Rather Than Size Matters in a Domesticated Arctic Charr Population. *Evolutionary Biology* 45 (1): 105–12.
Latta, Robert G. 1998. Differentiation of Allelic Frequencies at Quantitative Trait Loci Affecting Locally Adaptive Traits. *The American Naturalist* 151 (3): 283–92.
Latta, Robert G., Kyle M. Gardner, and David A. Staples. 2010. Quantitative Trait Locus Mapping of Genes Under Selection Across Multiple Years and Sites in *Avena Barbata*: Epistasis, Pleiotropy, and Genotype-by-Environment Interactions. *Genetics* 185 (1): 375–85.
Laurent, Stefan, Susanne P. Pfeifer, Matthew L. Settles, Samuel S. Hunter, Kayla M. Hardwick, Louise Ormond, Vitor C. Sousa, Jeffrey D. Jensen, and Erica Bree Rosenblum. 2016. The Population Genomics of Rapid Adaptation: Disentangling Signatures of Selection and Demography in White Sands Lizards. *Molecular Ecology* 25 (1): 306–23.
Le Corre, Valérie, and Antoine Kremer. 2012. The Genetic Differentiation at Quantitative Trait Loci under Local Adaptation. *Molecular Ecology* 21 (7): 1548–66.
Leducq, Jean-Baptiste, Lou Nielly-Thibault, Guillaume Charron, Chris Eberlein, Jukka-Pekka Verta, Pedram Samani, Kayla Sylvester, Chris Todd Hittinger, Graham Bell, and Christian R. Landry. 2016. Speciation Driven by Hybridization and Chromosomal Plasticity in a Wild Yeast. *Nature Microbiology* 1 (1): 15003.
Lee, Yoosook, Clare D. Marsden, Laura C. Norris, Travis C. Collier, Bradley J. Main, Abdrahamane Fofana, Anthony J. Cornel, and Gregory C. Lanzaro. 2013. Spatiotemporal Dynamics of Gene Flow and Hybrid Fitness between the M and S Forms of the Malaria Mosquito, Anopheles Gambiae. *Proceedings of the National Academy of Sciences* 110 (49): 19854–59.

Leinonen, Päivi H., David L. Remington, Johanna Leppälä, and Outi Savolainen. 2013. Genetic Basis of Local Adaptation and Flowering Time Variation in *Arabidopsis Lyrata*. *Molecular Ecology* 22 (3): 709–23.
Lenormand, Thomas. 2002. Gene Flow and the Limits to Natural Selection. *Trends in Ecology & Evolution* 17 (4): 183–89.
Lenski, Richard E. 1988. Experimental Studies of Pleiotropy and Epistasis in *Escherichia Coli*. Ii. Compensation for Maladaptive Effects Associated with Resistance to Virus T4. *Evolution* 42 (3): 433–40.
Leupold, Urs. 1949. *Die Vererbung von Homothallie und Heterothallie bei Schizosaccharomyces Pombe.* Zürich.
———. 1993. The Origin of *Schizosaccharomyces Pombe* Genetics. *The Early Days of Yeast Genetics*, 125–28.
Linnen, Catherine R., Yu-Ping Poh, Brant K. Peterson, Rowan D. H. Barrett, Joanna G. Larson, Jeffrey D. Jensen, and Hopi E. Hoekstra. 2013. Adaptive Evolution of Multiple Traits Through Multiple Mutations at a Single Gene. *Science* 339 (6125): 1312–16.
Liti, Gianni, David M. Carter, Alan M. Moses, Jonas Warringer, Leopold Parts, Stephen A. James, Robert P. Davey, et al. 2009. Population Genomics of Domestic and Wild Yeasts. *Nature* 458 (7236): 337–41.
Lowry, David B., Megan C. Hall, David E. Salt, and John H. Willis. 2009. Genetic and Physiological Basis of Adaptive Salt Tolerance Divergence between Coastal and Inland *Mimulus Guttatus*. *New Phytologist*, 776–88.
Lunt, David H., Sujai Kumar, Georgios Koutsovoulos, and Mark L. Blaxter. 2014. The Complex Hybrid Origins of the Root Knot Nematodes Revealed through Comparative Genomics. *PeerJ* 2 (May): e356.
Lunzer, Mark, Arvind Natarajan, Daniel E. Dykhuizen, and Antony M. Dean. 2002. Enzyme Kinetics, Substitutable Resources and Competition: From Biochemistry to Frequency-Dependent Selection in *Lac*. *Genetics* 162 (1): 485.
Lutzoni, François, Michael D. Nowak, Michael E. Alfaro, Valérie Reeb, Jolanta Miadlikowska, Michael Krug, A. Elizabeth Arnold, et al. 2018. Contemporaneous Radiations of Fungi and Plants Linked to Symbiosis. *Nature Communications* 9 (1): 1–11.
Lynch, Michael, and Wilfried Gabriel. 1987. Environmental Tolerance. *The American Naturalist* 129 (2): 283–303.
Maclean, R. Craig. 2005. Adaptive Radiation in Microbial Microcosms: Microbial Diversification. *Journal of Evolutionary Biology* 18 (6): 1376–86.
MacLean, R. Craig, and Graham Bell. 2002. Experimental Adaptive Radiation in *Pseudomonas*. *The American Naturalist* 160 (5): 569–81.
Mallet, James. 2005. Hybridization as an Invasion of the Genome. *Trends in Ecology & Evolution* 20 (5): 229–37.
Mariette, Nicolas, Alexander Kröner, Romain Mabon, Josselin Montarry, Bruno Marquer, Roselyne Corbière, Annabelle Androdias, and Didier Andrivon. 2018. A Trade-off between Sporangia Size and Number Exists in the Potato Late Blight Pathogen *Phytophthora Infestans*, and Is Not Altered by Biotic and Abiotic Factors. *Frontiers in Plant Science* 9 (December).
Marsellach, Xavi. 2017. A Non-Genetic Meiotic Repair Program Inferred from Spore Survival Values in Fission Yeast Wild Isolates: A Clue for an Epigenetic Ratchet-like Model of Ageing?, November.
Martin, Guillaume, Sarah P. Otto, and Thomas Lenormand. 2006. Selection for Recombination in Structured Populations. *Genetics* 172 (1): 593–609.

Masuelli, Ricardo W., Elsa L. Camadro, Luis E. Erazzú, María C. Bedogni, and Carlos F. Marfil. 2009. Homoploid Hybridization in the Origin and Evolution of Wild Diploid Potato Species. *Plant Systematics and Evolution* 277 (3): 143–51.
Matuszewski, Sebastian, Joachim Hermisson, and Michael Kopp. 2015. Catch Me If You Can: Adaptation from Standing Genetic Variation to a Moving Phenotypic Optimum. *Genetics* 200 (4): 1255–74.
Maynard Smith, J. 1971. What Use Is Sex? *Journal of Theoretical Biology* 30 (2): 319–35.
McClintock, B. 1984. The Significance of Responses of the Genome to Challenge. *Science* 226 (4676): 792–801.
McDonald, David B., Robert P. Clay, Robb T. Brumfield, and Michael J. Braun. 2001. Sexual Selection on Plumage and Behavior in an Avian Hybrid Zone: Experimental Tests of Male-Male Interactions. *Evolution* 55 (7): 1443–51.
Mixão, Verónica, and Toni Gabaldón. 2018. Hybridization and Emergence of Virulence in Opportunistic Human Yeast Pathogens. *Yeast* 35 (1): 5–20.
Monnahan, Patrick J., Jack Colicchio, and John K. Kelly. 2015. A Genomic Selection Component Analysis Characterizes Migration-Selection Balance. *Evolution* 69 (7): 1713–27.
Morales, Lucia, and Bernard Dujon. 2012. Evolutionary Role of Interspecies Hybridization and Genetic Exchanges in Yeasts. *Microbiology and Molecular Biology Reviews* 76 (4): 721–39.
Muller, H. 1942. Isolating Mechanisms, Evolution, and Temperature. In *Biol. Symp.*, 6:71–125.
Muller, Hermann Joseph. 1932. Some Genetic Aspects of Sex. *The American Naturalist* 66 (703): 118–38.
Murray, Rosalind L., and Asher D. Cutter. 2011. Experimental Evolution of Sperm Count in Protandrous Self-Fertilizing Hermaphrodites. *Journal of Experimental Biology* 214 (10): 1740–47.
Naumov, G. I., V. I. Kondratieva, and E. S. Naumova. 2015. Hybrid Sterility of the Yeast *Schizosaccharomyces Pombe*: Genetic Genus and Many Species in Statu Nascendi? *Microbiology* 84 (2): 159–69.
Nei, M, and W H Li. 1979. Mathematical Model for Studying Genetic Variation in Terms of Restriction Endonucleases. *Proceedings of the National Academy of Sciences of the United States of America* 76 (10): 5269–73.
Nei, Masatoshi. 1973. Analysis of Gene Diversity in Subdivided Populations. *Proceedings of the National Academy of Sciences* 70 (12): 3321–23.
———. 1987. *Molecular Evolutionary Genetics*. New York: Columbia University Press.
Nei, Masatoshi, and Masafumi Nozawa. 2011. Roles of Mutation and Selection in Speciation: From Hugo de Vries to the Modern Genomic Era. *Genome Biology and Evolution* 3 (January): 812–29.
Nei, Masatoshi, and Fumio Tajima. 1981. DNA Polymorphism Detectable by Restriction Endonucleases. *Genetics* 97 (1): 145–63.
Nielsen, Olaf, and Richard Egel. 2007. The Mat Genes of *Schizosaccharomyces Pombe*: Expression, Homothallic Switch, and Silencing. In *Sex in Fungi*, edited by John W. Taylor, James W. Kronstad, Joseph Heitman, and Lorna A. Casselton, 143–57. American Society of Microbiology.
Nieuwenhuis Bart P. S., and James Timothy Y. 2016. The Frequency of Sex in Fungi. *Philosophical Transactions of the Royal Society B: Biological Sciences* 371 (1706): 20150540.

Nieuwenhuis, Bart P. S., Sergio Tusso, Pernilla Bjerling, Josefine Stångberg, Jochen B. W. Wolf, and Simone Immler. 2018. Repeated Evolution of Self-Compatibility for Reproductive Assurance. *Nature Communications* 9 (1).

Nimmo, Elaine R., Alison L. Pidoux, Paul E. Perry, and Robin C. Allshire. 1998. Defective Meiosis in Telomere-Silencing Mutants of *Schizosaccharomyces Pombe*. *Nature* 392 (6678): 825–28.

Nimmy, Sonia Farhana, and M. S. Kamal. 2015. Next Generation Sequencing under de Novo Genome Assembly. *International Journal of Biomathematics* 08 (05): 1530001.

Norris, Laura C., Bradley J. Main, Yoosook Lee, Travis C. Collier, Abdrahamane Fofana, Anthony J. Cornel, and Gregory C. Lanzaro. 2015. Adaptive Introgression in an African Malaria Mosquito Coincident with the Increased Usage of Insecticide-Treated Bed Nets. *Proceedings of the National Academy of Sciences* 112 (3): 815–20.

Nosil, P., and B. J. Crespi. 2004. Does Gene Flow Constrain Adaptive Divergence or Vice Versa? A Test Using Ecomorphology and Sexual Isolation in *Timema Cristinae* Walking-Sticks. *Evolution* 58 (1): 102–12.

Nuckolls, Nicole L, María Angélica Bravo Núñez, Michael T Eickbush, Janet M Young, Jeffrey J Lange, Jonathan S Yu, Gerald R Smith, Sue L Jaspersen, Harmit S Malik, and Sarah E Zanders. 2017. Wtf Genes Are Prolific Dual Poison-Antidote Meiotic Drivers. *ELife* 6 (June).

Nurse, Paul. 1990. Universal Control Mechanism Regulating Onset of M-Phase. *Nature* 344 (6266): 503–8.

Olson, M. V., J. E. Dutchik, M. Y. Graham, G. M. Brodeur, C. Helms, M. Frank, M. MacCollin, R. Scheinman, and T. Frank. 1986. Random-Clone Strategy for Genomic Restriction Mapping in Yeast. *Proceedings of the National Academy of Sciences* 83 (20): 7826–30.

Orr, H. A. 1995. The Population Genetics of Speciation: The Evolution of Hybrid Incompatibilities. *Genetics* 139 (4): 1805–13.

Ortíz-Barrientos, Daniel, Jane Reiland, Jody Hey, and Mohamed A. F. Noor. 2002. Recombination and the Divergence of Hybridizing Species. In *Genetics of Mate Choice: From Sexual Selection to Sexual Isolation*, edited by W. J. Etges and M. A. F. Noor, 9:167–78. Dordrecht: Springer Netherlands.

Ostrowski, Ekizabeth A., Daniel E. Rozen, and Richard E. Lenski. 2005. Pleiotropic Effects of Beneficial Mutations in *Escherichia Coli*. *Evolution* 59 (11): 2343–52.

Ostrowski, Elizabeth A., Charles Ofria, and Richard E. Lenski. 2007. Ecological Specialization and Adaptive Decay in Digital Organisms. *The American Naturalist* 169 (1): E1–20.

Payseur, Bret A., and Loren H. Rieseberg. 2016. A Genomic Perspective on Hybridization and Speciation. *Molecular Ecology* 25 (11): 2337–60.

Peona, Valentina, Matthias H. Weissensteiner, and Alexander Suh. 2018. How Complete Are "Complete" Genome Assemblies?—An Avian Perspective. *Molecular Ecology Resources* 18 (6): 1188–95.

Phadnis, Naina, Randy W. Hyppa, and Gerald R. Smith. 2011. New and Old Ways to Control Meiotic Recombination. *Trends in Genetics* 27 (10): 411–21.

Pinheiro-Costa, Bruna Karen, José Neiva Mesquita-Neto, Juliana Ordones Rego, Clemens Schlindwein, Bruna Karen Pinheiro-Costa, José Neiva Mesquita-Neto, Juliana Ordones Rego, and Clemens Schlindwein. 2018. Trade off between Quantity and Size of Pollen Grains in the Heterandrous Flowers of *Senna Pendula* (Fabaceae). *Acta Botanica Brasilica* 32 (3): 446–53.

Presgraves, Daven C. 2010. The Molecular Evolutionary Basis of Species Formation. *Nature Reviews Genetics* 11 (3): 175–80.
Pretorius, Isak S, and Florian F Bauer. 2002. Meeting the Consumer Challenge through Genetically Customized Wine-Yeast Strains. *Trends in Biotechnology* 20 (10): 426–32.
Pryszcz, Leszek P., Tibor Németh, Attila Gácser, and Toni Gabaldón. 2014. Genome Comparison of *Candida Orthopsilosis* Clinical Strains Reveals the Existence of Hybrids between Two Distinct Subspecies. *Genome Biology and Evolution* 6 (5): 1069–78.
Pryszcz, Leszek P., Tibor Németh, Ester Saus, Ewa Ksiezopolska, Eva Hegedűsová, Jozef Nosek, Kenneth H. Wolfe, Attila Gacser, and Toni Gabaldón. 2015. The Genomic Aftermath of Hybridization in the Opportunistic Pathogen *Candida Metapsilosis*. *PLOS Genetics* 11 (10): e1005626.
Purcell, Jessica, Alan Brelsford, Yannick Wurm, Nicolas Perrin, and Michel Chapuisat. 2014. Convergent Genetic Architecture Underlies Social Organization in Ants. *Current Biology* 24 (22): 2728–32.
Ravinet, M., R. Faria, R. K. Butlin, J. Galindo, N. Bierne, M. Rafajlović, M. a. F. Noor, B. Mehlig, and A. M. Westram. 2017. Interpreting the Genomic Landscape of Speciation: A Road Map for Finding Barriers to Gene Flow. *Journal of Evolutionary Biology* 30 (8): 1450–77.
Reboud, Xavier, and Graham Bell. 1997. Experimental Evolution in Chlamydomonas. III. Evolution of Specialist and Generalist Types in Environments That Vary in Space and Time. *Heredity* 78 (5): 507–14.
Redon, Richard, Shumpei Ishikawa, Karen R. Fitch, Lars Feuk, George H. Perry, T. Daniel Andrews, Heike Fiegler, et al. 2006. Global Variation in Copy Number in the Human Genome. *Nature* 444 (7118): 444–54.
Remington, David L., Päivi H. Leinonen, Johanna Leppälä, and Outi Savolainen. 2013. Complex Genetic Effects on Early Vegetative Development Shape Resource Allocation Differences between *Arabidopsis Lyrata* Populations. *Genetics* 195 (3): 1087–1102.
Reznick, David A., Heather Bryga, and John A. Endler. 1990. Experimentally Induced Life-History Evolution in a Natural Population. *Nature* 346 (6282): 357–59.
Reznick, David, Leonard Nunney, and Alan Tessier. 2000. Big Houses, Big Cars, Superfleas and the Costs of Reproduction. *Trends in Ecology & Evolution* 15 (10): 421–25.
Rhind, Nicholas, Zehua Chen, Moran Yassour, Dawn A. Thompson, Brian J. Haas, Naomi Habib, Ilan Wapinski, et al. 2011. Comparative Functional Genomics of the Fission Yeasts. *Science* 332 (6032): 930–36.
Rieseberg, L. H. 2003. Major Ecological Transitions in Wild Sunflowers Facilitated by Hybridization. *Science* 301 (5637): 1211–16.
Rieseberg, Loren H. 2001. Chromosomal Rearrangements and Speciation. *Trends in Ecology & Evolution* 16 (7): 351–58.
Roff, Derek A. 1993. *Evolution of Life Histories: Theory and Analysis*. Springer Science & Business Media.
———. 2002. *Life History Evolution*.
Roff, Derek A., and D.J. Fairbairn. 2007. The Evolution of Trade-Offs: Where Are We? *Journal of Evolutionary Biology* 20 (2): 433–47.
Ronce, Ophélie, and Mark Kirkpatrick. 2001. When Sources Become Sinks: Migrational Meltdown in Heterogeneous Habitats. *Evolution* 55 (8): 1520–31.

Saetre, Glenn-Peter, Thomas Borge, Johan Lindell, Truls Moum, Craig R. Primmer, Ben C. Sheldon, Jon Haavie, Arild Johnsen, and Hans Ellegren. 2008. Speciation, Introgressive Hybridization and Nonlinear Rate of Molecular Evolution in Flycatchers: Molecular Evolution in Flycatchers. *Molecular Ecology* 10 (3): 737–49.

Saiki, R. K., S. Scharf, F. Faloona, K. B. Mullis, G. T. Horn, H. A. Erlich, and N. Arnheim. 1985. Enzymatic Amplification of Beta-Globin Genomic Sequences and Restriction Site Analysis for Diagnosis of Sickle Cell Anemia. *Science* 230 (4732): 1350–54.

Samani, Pedram, and Graham Bell. 2016. Experimental Evolution of the Grain of Metabolic Specialization in Yeast. *Ecology and Evolution* 6 (12): 3912–22.

Sanger, F., S. Nicklen, and A. R. Coulson. 1977. DNA Sequencing with Chain-Terminating Inhibitors. *Proceedings of the National Academy of Sciences* 74 (12): 5463–67.

Sankararaman, Sriram, Swapan Mallick, Michael Dannemann, Kay Prüfer, Janet Kelso, Svante Pääbo, Nick Patterson, and David Reich. 2014. The Genomic Landscape of Neanderthal Ancestry in Present-Day Humans. *Nature* 507 (7492): 354–57.

Savolainen, Outi, Martin Lascoux, and Juha Merilä. 2013. Ecological Genomics of Local Adaptation. *Nature Reviews Genetics* 14 (11): 807–20.

Schick, Alana, Susan F Bailey, and Rees Kassen. 2015. Evolution of Fitness Trade-Offs in Locally Adapted Populations of *Pseudomonas Fluorescens*. *The American Naturalist* 186 (S1): S48–59.

Schlötterer, Christian, Raymond Tobler, Robert Kofler, and Viola Nolte. 2014. Sequencing Pools of Individuals — Mining Genome-Wide Polymorphism Data without Big Funding. *Nature Reviews Genetics* 15 (11): 749–63.

Schmidt, H., and H. Gutz. 1994. The Mating-Type Switch in Yeasts. In *Growth, Differentiation and Sexuality*, edited by Joseph G. H. Wessels and Friedhelm Meinhardt, 283–94. The Mycota. Berlin, Heidelberg: Springer Berlin Heidelberg.

Schumer, Molly, Rongfeng Cui, Daniel L. Powell, Gil G. Rosenthal, and Peter Andolfatto. 2016. Ancient Hybridization and Genomic Stabilization in a Swordtail Fish. *Molecular Ecology* 25 (11): 2661–79.

Schumer, Molly, Gil G. Rosenthal, and Peter Andolfatto. 2014. How Common Is Homoploid Hybrid Speciation?: Perspective. *Evolution* 68 (6): 1553–60.

Schwander, Tanja, Romain Libbrecht, and Laurent Keller. 2014. Supergenes and Complex Phenotypes. *Current Biology* 24 (7): R288–94.

Seehausen, Ole. 2004. Hybridization and Adaptive Radiation. *Trends in Ecology & Evolution* 19 (4): 198–207.

Seehausen, Ole, Roger K. Butlin, Irene Keller, Catherine E. Wagner, Janette W. Boughman, Paul A. Hohenlohe, Catherine L. Peichel, et al. 2014. Genomics and the Origin of Species. *Nature Reviews Genetics* 15 (3): 176–92.

Servedio, M. R., and G.-P. Saetre. 2003. Speciation as a Positive Feedback Loop between Postzygotic and Prezygotic Barriers to Gene Flow. *Proceedings of the Royal Society B: Biological Sciences* 270 (1523): 1473–79.

Shafer, Aaron B. A., and Jochen B. W. Wolf. 2013. Widespread Evidence for Incipient Ecological Speciation: A Meta-Analysis of Isolation-by-Ecology. *Ecology Letters* 16 (7): 940–50.

Sipiczki, Matthias. 2000. Where Does Fission Yeast Sit on the Tree of Life? *Genome Biology* 1 (2): reviews1011.1.

———. 2004. Fission Yeast Phylogenesis and Evolution. In *The Molecular Biology of Schizosaccharomyces Pombe: Genetics, Genomics and Beyond*, edited by Richard Egel, 431–43. Berlin, Heidelberg: Springer Berlin Heidelberg.

Slatkin, M. 1987. Gene Flow and the Geographic Structure of Natural Populations. *Science* 236 (4803): 787–92.

Smadja, Carole M., and Roger K. Butlin. 2011. A Framework for Comparing Processes of Speciation in the Presence of Gene Flow. *Molecular Ecology* 20 (24): 5123–40.

Soudi, Shaghayegh, Klaus Reinhold, and Leif Engqvist. 2015. Host-Associated Divergence in Sympatric Host Races of the Leaf Beetle Lochmaea Capreae: Implications for Local Adaptation and Reproductive Isolation. *Biological Journal of the Linnean Society* 116 (1): 169–82.

Spichtig, Mathias, and Tadeusz J. Kawecki. 2004. The Maintenance (or Not) of Polygenic Variation by Soft Selection in Heterogeneous Environments. *The American Naturalist* 164 (1): 70–84.

Stearns, Stephen C. 1992. *The Evolution of Life Histories*.

Stephan, Johannes, Oliver Stegle, and Andreas Beyer. 2015. A Random Forest Approach to Capture Genetic Effects in the Presence of Population Structure. *Nature Communications* 6 (June): 7432.

Støstad, Hanna N., Melissah Rowe, Arild Johnsen, and Jan T. Lifjeld. 2019. Sperm Head Abnormalities Are More Frequent in Songbirds with More Helical Sperm: A Possible Trade-off in Sperm Evolution. *Journal of Evolutionary Biology* 32 (7): 666–74.

Tajima, F. 1989. Statistical Method for Testing the Neutral Mutation Hypothesis by DNA Polymorphism. *Genetics* 123 (3): 585–95.

Tajima, Fumio. 1983. Evolutionary Relationship of DNA Sequences in Finite Populations. *Genetics* 105 (2): 437–60.

Tao, Yu-Tian, Fang Suo, Sergio Tusso, Yan-Kai Wang, Song Huang, Jochen B. W. Wolf, and Li-Lin Du. 2019. Intraspecific Diversity of Fission Yeast Mitochondrial Genomes. *Genome Biology and Evolution* 11 (8): 2312–29.

Taylor, D. J., and P. D. Hebert. 1993. Habitat-Dependent Hybrid Parentage and Differential Introgression between Neighboringly Sympatric Daphnia Species. *Proceedings of the National Academy of Sciences* 90 (15): 7079–83.

Teresa Avelar, Ana, Lília Perfeito, Isabel Gordo, and Miguel Godinho Ferreira. 2013. Genome Architecture Is a Selectable Trait That Can Be Maintained by Antagonistic Pleiotropy. *Nature Communications* 4 (August): 2235.

Thulin, Carl-Gustaf, and Håkan Tegelström. 2002. Biased Geographical Distribution of Mitochondrial DNA That Passed the Species Barrier from Mountain Hares to Brown Hares (Genus *Lepus*): An Effect of Genetic Incompatibility and Mating Behaviour? *Journal of Zoology* 258 (3): 299–306.

Tigano, Anna, and Vicki L. Friesen. 2016. Genomics of Local Adaptation with Gene Flow. *Molecular Ecology* 25 (10): 2144–64.

Tusso, Sergio, Bart P. S. Nieuwenhuis, Fritz J. Sedlazeck, John W. Davey, Daniel C. Jeffares, and Jochen B. W. Wolf. 2019. Ancestral Admixture Is the Main Determinant of Global Biodiversity in Fission Yeast. *Molecular Biology and Evolution* 36 (9): 1975–89.

Verhoeven, K. J. F., H. Poorter, E. Nevo, and A. Biere. 2008. Habitat-Specific Natural Selection at a Flowering-Time QTL Is a Main Driver of Local Adaptation in Two Wild Barley Populations. *Molecular Ecology* 17 (14): 3416–24.

Volschenk, H., H. J. J. van Vuuren, and M. Viljoen–Bloom. 2003. Malo-Ethanolic Fermentation in *Saccharomyces* and *Schizosaccharomyces*. *Current Genetics* 43 (6): 379–91.

Walker, Bruce J., Thomas Abeel, Terrance Shea, Margaret Priest, Amr Abouelliel, Sharadha Sakthikumar, Christina A. Cuomo, et al. 2014. Pilon: An Integrated Tool for Comprehensive Microbial Variant Detection and Genome Assembly Improvement. Edited by Junwen Wang. *PLoS ONE* 9 (11): e112963.

Warschefsky, Emily, R. Varma Penmetsa, Douglas R. Cook, and Eric J. B. von Wettberg. 2014. Back to the Wilds: Tapping Evolutionary Adaptations for Resilient Crops through Systematic Hybridization with Crop Wild Relatives. *American Journal of Botany* 101 (10): 1791–1800.

Watterson, G.A. 1975. On the Number of Segregating Sites in Genetical Models without Recombination. *Theoretical Population Biology* 7 (2): 256–76.

Weinig, Cynthia, Lisa A. Dorn, Nolan C. Kane, Zachary M. German, Solveig S. Halldorsdottir, Mark C. Ungerer, Yuko Toyonaga, Trudy F. C. Mackay, Michael D. Purugganan, and Johanna Schmitt. 2003. Heterogeneous Selection at Specific Loci in Natural Environments in *Arabidopsis Thaliana*. *Genetics* 165 (1): 321–29.

Weir, B.S. 1996. *Genetic Data Analysis II: Methods for Discrete Population Genetic Data*. Genetic Data Analysis. Sunderland, MA: Sinauer Associates.

Weischenfeldt, Joachim, Orsolya Symmons, François Spitz, and Jan O. Korbel. 2013. Phenotypic Impact of Genomic Structural Variation: Insights from and for Human Disease. *Nature Reviews Genetics* 14 (2): 125–38.

Weissensteiner, Matthias H., Andy W. C. Pang, Ignas Bunikis, Ida Höijer, Olga Vinnere-Petterson, Alexander Suh, and Jochen B. W. Wolf. 2017. Combination of Short-Read, Long-Read, and Optical Mapping Assemblies Reveals Large-Scale Tandem Repeat Arrays with Population Genetic Implications. *Genome Research* 27 (5): 697–708.

Welch, Mark E., and Loren H. Rieseberg. 2002a. Habitat Divergence between a Homoploid Hybrid Sunflower Species, *Helianthus Paradoxus* (Asteraceae), and Its Progenitors. *American Journal of Botany* 89 (3): 472–78.

———. 2002b. Patterns of Genetic Variation Suggest a Single, Ancient Origin for the Diploid Hybrid Species *Helianthus Paradoxus*. *Evolution* 56 (11): 2126–37.

Whitlock, Michael C. 1996. The Red Queen Beats the Jack-Of-All-Trades: The Limitations on the Evolution of Phenotypic Plasticity and Niche Breadth. *The American Naturalist* 148 (November): S65–77.

Wolf, Jochen B. W., and Hans Ellegren. 2017. Making Sense of Genomic Islands of Differentiation in Light of Speciation. *Nature Reviews Genetics* 18 (2): 87–100.

Wolf, Jochen B. W., Johan Lindell, and Niclas Backström. 2010. Speciation Genetics: Current Status and Evolving Approaches. *Philosophical Transactions of the Royal Society B: Biological Sciences* 365 (1547): 1717–33.

Wood, V., R. Gwilliam, M.-A. Rajandream, M. Lyne, R. Lyne, A. Stewart, J. Sgouros, et al. 2002. The Genome Sequence of Schizosaccharomyces Pombe. *Nature* 415 (6874): 871–80.

Wright, Sewall. 1931. Evolution in Mendelian Populations. *Genetics* 16 (2): 97–159.

———. 1943. Isolation by Distance. *Genetics* 28 (2): 114–38.

———. 1951. The Genetical Structure of Populations 15: 323–54.

Wu, Chung-I. 2001. The Genic View of the Process of Speciation. *Journal of Evolutionary Biology* 14 (6): 851–65.

Yeaman, Sam. 2015. Local Adaptation by Alleles of Small Effect. *The American Naturalist* 186 (S1): S74–89.

Yeaman, Sam, and Andy Jarvis. 2006. Regional Heterogeneity and Gene Flow Maintain Variance in a Quantitative Trait within Populations of Lodgepole Pine. *Proceedings of the Royal Society B: Biological Sciences* 273 (1594): 1587–93.

Yeaman, Sam, and Sarah P. Otto. 2011. Establishment and Maintenance of Adaptive Genetic Divergence Under Migration, Selection, and Drift. *Evolution* 65 (7): 2123–29.

Yeaman, Sam, and Michael C. Whitlock. 2011. The Genetic Architecture of Adaptation Under Migration–Selection Balance. *Evolution* 65 (7): 1897–1911.

Zanders, Sarah E., Michael T. Eickbush, Jonathan S. Yu, Ji-Won Kang, Kyle R. Fowler, Gerald R. Smith, and Harmit Singh Malik. 2014. Genome Rearrangements and Pervasive Meiotic Drive Cause Hybrid Infertility in Fission Yeast. *ELife* 3 (June): e02630.

Zhu, Jinlei, Minghu Liu, Zhiming Xin, Zhimin Liu, and Frank M. Schurr. 2019. A Trade-off between Primary and Secondary Seed Dispersal by Wind. *Plant Ecology* 220 (4): 541–52.

Zhu, Yuan, Alan O. Bergland, Josefa González, and Dmitri A. Petrov. 2012. Empirical Validation of Pooled Whole Genome Population Re-Sequencing in Drosophila Melanogaster. *PLOS ONE* 7 (7): e41901.

Acta Universitatis Upsaliensis

Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology 1851

Editor: The Dean of the Faculty of Science and Technology

A doctoral dissertation from the Faculty of Science and Technology, Uppsala University, is usually a summary of a number of papers. A few copies of the complete dissertation are kept at major Swedish research libraries, while the summary alone is distributed internationally through the series Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology. (Prior to January, 2005, the series was published under the title "Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology".)

Distribution: publications.uu.se
urn:nbn:se:uu:diva-392422

ACTA
UNIVERSITATIS
UPSALIENSIS
UPPSALA
2019