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1 **Title:** The natural diversity and ecology of fission yeast.

2 **Running head:** Natural fission yeast diversity and ecology

3

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11

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13

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15

16 **Abstract**

17 While the fission yeast is a powerful model of eukaryote biology, there have been
18 few studies of quantitative genetics, phenotypic or genetic diversity. Here I survey
19 the small collection of fission yeast diversity research. I discuss what we can infer
20 about the ecology and origins of *Schizosaccharomyces pombe* from microbiology
21 field studies and the few strains that have been collected.

22

23 **Introduction**

24 *Schizosaccharomyces pombe* research began in the 1940s (Fantes and Hoffman
25 2016) and is now a potent model of eukaryote biology, with a well-annotated
26 curated genome (Wood *et al.* 2002; McDowall *et al.* 2015), an extensive battery
27 of technical methods and genome-scale tools (Hoffman, Wood and Fantes 2015;
28 Hagan *et al.* 2016) and regular international meetings devoted to its study. Part of
29 the important utility of fission yeast as a model is that it contains many vertebrate
30 orthologs that are not present in budding yeast (Hoffman, Wood and Fantes
31 2015), so it provides a complement for studies of cell biology.

32 The majority of fission yeast research has used the strains described by
33 Leupold with its three mating types (Leupold 1949), and mutants derived from
34 these strains. Studies of diversity or quantitative genetics have been few and far
35 between. By contrast there is an extensive literature describing diversity and
36 quantitative genetics in the budding yeast *Saccharomyces cerevisiae* and its wild
37 relative *Saccharomyces paradoxus*, and a range of related species (Peter and
38 Schacherer 2016). These include QTL studies

39 (Swinnen, Thevelein and Nevoigt 2012; Liti and Louis 2012; Fay 2013;
40 Bloom *et al.* 2013; Märtens *et al.* 2016), genome-scale analysis of diversity (Liti
41 *et al.* 2009; Schacherer *et al.* 2009) and analysis of diversity and evolution in the
42 natural environment (Robinson, Pinharanda and Bensasson 2016; Leducq *et al.*
43 2016). In this review, I survey fission yeast diversity research, and I discuss what
44 little is known about the origins and natural ecology of this species.

45

46 **Defining fission yeast species**

47 Collections of *Schizoaccharomyces* strains were classified into three groups based
48 on crossing and protoplast fusion (Sipiczki *et al.* 1982), phenotypic characters
49 (Bridge and May 1984), DNA optical reassociation and physiological characters

50 (Vaughan Martini 1991), simplifying the rather complex list of potential ‘species’
51 into three (*Schizosaccharomyces pombe*, *S. japonicus*, *S. octosporus*).
52 *Schizosaccharomyces cryophilus* was identified much later as a contaminant of a
53 *S. octosporus* strain (CBS7191) from Denmark, and the species description was
54 accompanied by a draft genome (Helston *et al.* 2010).

55 The genomes and transcriptomes of *S. japonicus*, *S. octosporus* and an
56 improved *S. cryophilus* genome were described in 2011, showing that the
57 *Schizosaccharomyces* genus is as divergent on the protein level as the human-
58 amphioxus divergence (~55% amino acid identity) (Rhind *et al.* 2011). This
59 analysis described the conservation of orthologous groups, conservation of
60 transcription, the evolution of mating type regions and transposons. It also
61 features the first sequencing of a non-reference strain of *S. pombe*, concluding that
62 the within-species diversity was < 1% (confirmed later with studies of more
63 strains (Fawcett *et al.* 2014; Jeffares *et al.* 2015)). The current clade of only four
64 highly divergent fission yeast species is a limitation for evolutionary studies, since
65 evolutionary constraints can be estimated only inaccurately, and non-coding sites
66 that are in general subject to weaker purifying selection tend to be saturated
67 (Rhind *et al.* 2011). None of the *Schizosaccharomyces* species is sufficiently
68 closely related to *S. pombe* to reliably determine ancestral nucleotide states.

69

70 **Early (pre-genome sequence) diversity studies**

71 An early field study of this species was conducted by Florenzano *et al.*, who
72 showed that *S. pombe* was frequently present on grapes in Sicilian vineyards
73 (Florenzano, Balloni and Materassi 1977). Phenotypic characterization began with
74 analysis of xerotolerance (resistance to high solute concentrations) in 27 *S. pombe*
75 strains (Ganthala, Marshall and May 1994). One the first genetic analysis of
76 diversity within *S. pombe* described the intron content of mitochondrial genomes
77 in 26 strains, showing presence/absence polymorphisms in group I and group II
78 introns (Zimmer *et al.* 1987). Interestingly, there appears to be no intron presence
79 polymorphisms in the nuclear genomes of sequenced strains (Mourier & Jeffares,
80 unpublished analyses), though on the longer scale fission yeasts have certainly
81 undergone intron gain and loss (Mourier and Jeffares 2003; Jeffares, Mourier and
82 Penny 2006; Rhind *et al.* 2011).

83 In a prelude to genome-scale analyses, three studies began to explore
84 genetic and phenotypic diversity on a larger scale. Gomes *et al.*, collected 27

85 strains from seven Brazilian cachaça distilleries, and characterised osmotolerance,
86 trehalose accumulation and ethanol tolerance, showing that these strains could
87 grow in 50% glucose and 10% ethanol (Gomes *et al.* 2002). They also explored
88 population structure using RAPD-PCR (random amplified polymorphic DNA
89 PCR), demonstrating local population structure in Brazilian cachaça strains.
90 RAPD-PCR was a useful method to characterise diversity prior to next generation
91 sequencing, but the development of 26 primers for microsatellite PCR now
92 provide a simple method to genotype strain collections (Patch and Aves 2007).
93 Brown *et al.* assembled 81 natural isolates of *S. pombe* including samples from all
94 continents (except Antarctica), and measured a large assembly of phenotypic
95 characters, including growth parameters in 42 liquid media and cell length (Brown
96 *et al.* 2011). This analysis also described diversity at three locations, and
97 estimated that the global effective population size of this species is 10^7 (a figure
98 that remained after genome-wide analysis (Farlow *et al.* 2015)). Most
99 interestingly, this work described extensive karyotype diversity within this
100 collection, including reciprocal translocations, duplications and inversions,
101 showing that the ribosomal repeats were located on different chromosome ends in
102 different strains.

103

104 **Genome-wide sequence analyses**

105 The creation and analysis of the only fission yeast recombinant strain library
106 was published in 2014 (Clément-Ziza *et al.* 2014). This study used a two-parent
107 segregant panel and described expression QTLs (eQTLs) from both protein-
108 coding and non-coding transcripts, during growth and stress conditions.
109 Interestingly this study discovered a larger proportion of associations between
110 genetic variants and non-coding transcripts than coding transcripts. The most
111 significant variant, that affected 44% of eQTL associations and growth rate, was a
112 frameshift in the *swc5* gene - part of a complex that affects histone deposition.
113 Detailed analysis showed that this frameshift caused increased antisense
114 transcription and decreased sense transcription, providing an example of the
115 molecular events that influenced a complex trait such as growth. Further analyses
116 of segregant panels are in progress, describing positive selection and the genetic
117 control of RNA and protein levels (Clément-Ziza, pers. comm.).

118 An analysis of segregant pool based mapping (bulk segregant analysis) from
119 a two-parent cross showed that this method was feasible in fission yeast (Hu, Suo

120 and Du 2015). Hu *et al.* localised the probable causal allele of maltose deficiency
121 by sequencing pools grown with and without maltose. The analysis was
122 complicated by an inversion in the reference strain, but few other wild strains
123 (Jeffares *et al.* 2017), which reduces the local recombination rate (Clément-Ziza *et*
124 *al.* 2014).

125 Two genome-wide analyses of genetic diversity in *S. pombe* were published
126 soon afterwards (Fawcett *et al.* 2014; Jeffares *et al.* 2015). Both analyses
127 described recombination rate and population structure, and showed that exons,
128 UTRs and introns were the main targets of purifying selection. Estimates of
129 diversity (π) were $\sim 3 \times 10^{-3}$ (pairwise comparison have an average of 3 SNPs/kb),
130 slightly higher than the budding yeast *Saccharomyces cerevisiae* (1×10^{-3}) (Liti *et*
131 *al.* 2009). From the genetic diversity and mutation rates, the effective population
132 size of *S. pombe* has been estimated to be 12 million, on a similar scale to budding
133 yeast (3 million) (Farlow *et al.* 2015).

134 The analysis of Fawcett *et al.* (32 strains) described some unusual patterns
135 of diversity that were likely due to soft selective sweeps, and either balancing
136 selection or introgression from some unknown fission yeast outgroup (Fawcett *et*
137 *al.* 2014). Jeffares *et al.* (161 strains) described transposon insertions and included
138 analysis of quantitative traits, their heritability and quantitative genetics using the
139 genome-wide association study (GWAS) approach (Jeffares *et al.* 2015). This
140 study located 1,400 variants that were significantly associated with traits despite
141 the very small sample size, showing that the combination of simple tractable
142 genetics with the capability to measure traits accurately with abundant repeat
143 measurements in well-controlled environments, is a powerful combination.
144 Further analysis with the same strain collection described structural variants
145 showing that they are both transient and contribute considerably to quantitative
146 traits and reproductive isolation (Jeffares *et al.* 2017). Interestingly the variance in
147 wine-making traits, such as malic acid accumulation and glucose/fructose
148 utilisation (Benito *et al.* 2016), appeared to be caused entirely by structural
149 variants.

150 Two genome-scale analyses of the mutation rate estimated the point
151 mutation rate to be 1.7×10^{-10} (or 2.0×10^{-10}) per base per generation (Farlow *et*
152 *al.* 2015; Behringer and Hall 2015), very similar to estimates for the budding
153 yeast *Saccharomyces cerevisiae* (estimates at 3 and 1.67×10^{-10}) (Lynch *et al.*
154 2008; Zhu *et al.* 2014). Both studies noted a strong bias towards small insertions,

155 over deletions, which occur primarily in the non-protein regions of the genome, a
156 pattern that is retained in natural genetic diversity (Jeffares *et al.* 2015).

157

158 **Reproductive isolation**

159 One topic that has received particular attention is the study of mating types
160 and reproductive isolation. Since the outset of fission yeast research, it was clear
161 homothallic strains could mutate to more or less stable heterothallic genotypes (h^+
162 or h^-) (Leupold 1949). Natural isolates also vary genetically at mating type
163 regions and in their mating behavior, with some strains mutating more frequently
164 from h^+ to h^- and vice versa (Schlake and Gutz 1993). In an interesting
165 demonstration that reproductive isolation could evolve via pre-zygotic
166 mechanisms, Sieke *et al.* created three novel reproductive groups with different
167 pheromone-receptor pairs (Seike, Nakamura and Shimoda 2015). Given these
168 changes it is likely that pre-zygotic reproductive isolation occurs within some
169 populations.

170 Several studies described the low spore viability that results from many
171 inter-strain matings (Kondrat'eva and Naumov 2001; Teresa Avelar *et al.* 2013;
172 Zanders *et al.* 2014; Naumov and Kondratieva 2015; Jeffares *et al.* 2015).
173 Viability ranges from pairs showing $< 1\%$ viable offspring to strains with 90%
174 viable, similar a range observed for *species* of budding yeast with that have much
175 higher genetic divergence than fission yeast strains (Liti, Barton and Louis 2006),
176 consistent with *S. pombe* strains being 'on the verge of speciation' (Naumov and
177 Kondratieva 2015) (**Figure 1A**). Some homothallic strains are also ineffective at
178 mating with their own genotype (Kondrat'eva and Naumov 2001; Jeffares *et al.*
179 2015).

180 Since most crosses do produce mating bodies and asci (Xavi Marsellach,
181 pers. comm.), the isolation is generally post-zygotic (intrinsic reproductive
182 isolation). The accumulation of genetic factors that reduce mating success
183 within these relatively closely related strains is probably due to the low
184 frequency of outbreeding in fission yeast. Based on the decay in linkage between
185 wild isolates Farlow *et al.* estimated that *S. pombe* mate with a genetically
186 dissimilar individual on average every 800,000 generations (Farlow *et al.* 2015),
187 far less frequently than the estimates 50,000 generation for *S. cerevisiae* (Ruderfer
188 *et al.* 2006). Given this frequency of, it is not surprising that the existing strains

189 have accumulated genetic factors that preclude interbreeding in the ~2300 years
190 since these strains have drifted apart (Jeffares *et al.* 2015).

191 There are at least three (non-exclusive) genetic causes for the reproductive
192 isolation of fission yeasts. Spore killing (meiotic drive), has been proposed to be a
193 mechanism (Kondrat'eva and Naumov 2001; Zanders *et al.* 2014; Naumov and
194 Kondratieva 2015). Many of the crosses analysed by Kondratieva *et al.* from
195 genetically divergent strains and produced strong deviations from expected
196 Mendelian ratios (Kondrat'eva and Naumov 2001; Naumov, Kondratieva and
197 Naumova 2015) (Figure 1B), while the analyses of Zanders *et al.* concluded that
198 there were meiotic drive elements on all three chromosomes (Zanders *et al.* 2014).

199 Two recent analyses have demonstrated that members of the *wtf* gene
200 family mediate drive with a spore killer-antidote system (Hu *et al.* 2017; Nuckolls
201 *et al.* 2017). Hu *et al.* demonstrate that *wtf9* and *wtf27* genes from the non-
202 reference strain (CBS5557/JB4) drive segregation distortion in when mated to the
203 reference strain, that this drive is independent of genomic location. Nuckolls *et al.*
204 show that *wtf4* promotes distortion in crosses between the reference strain and the
205 kombucha strain (SPK1820/YFS276/JB1180, as initially sequenced by the Broad
206 Institute (Rhind *et al.* 2011)). Other strains analysed by Kondratieva *et al.* also
207 show very biased segregation (Figure 1B).

208 Collectively, these analyses show that the spore killer (or poison) and
209 antidote functions can be separated by mutations. In the natural state, there are
210 two transcripts that mediate killer/antidote functions (Nuckolls *et al.* 2017). While
211 the killer protein variant is distributed in all four spores of the asci, the antidote
212 remains only within cells with the relevant *wtf* genotype. Since *wtf* genes encode
213 membrane-spanning domains they may travel between asci. The genetics of the
214 poison-antidote systems are complex, in that there are multiple *wtf* genes in
215 different strains that have degenerated to contain the poison and antidote
216 functions, antidote only, or no function. Both analyses show that *wtf* genes are
217 particularly genetically diverse (Figure 1C). However, they do not show an excess
218 of high Tajima's D values (Tajima 1989)(Figure 1C), a genetic diversity
219 parameter which is one of the expected signatures of balancing selection.

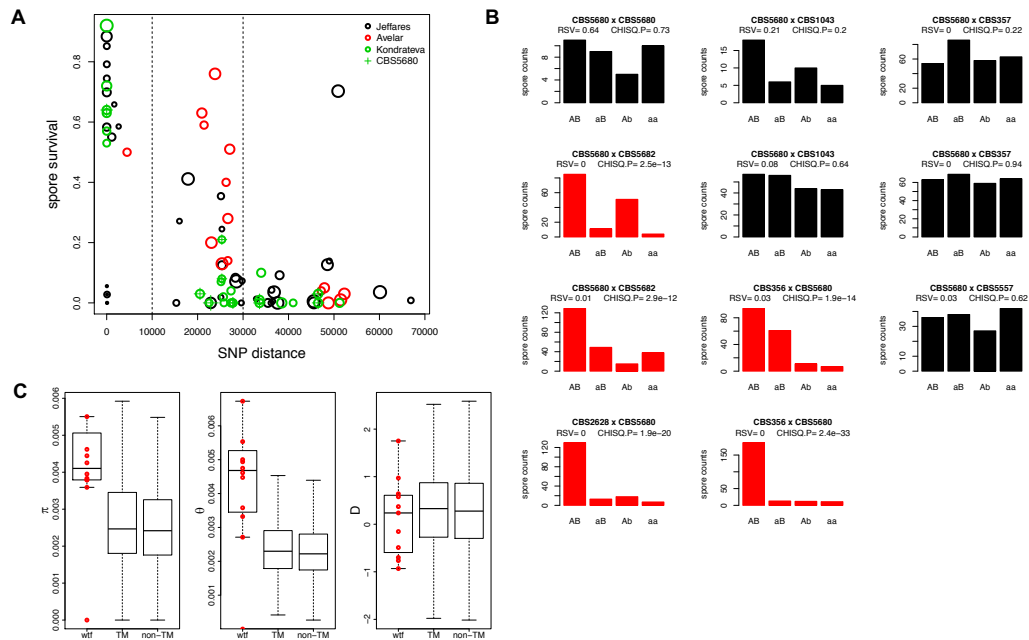
220 Reproductive isolation may also be the result of the aneuploidy that occurs
221 when parents differ in chromosomal inversions and translocations. For example,
222 engineered inversions and translocations reduce spore viability by ~40% (Teresa
223 Avelar *et al.* 2013). *S. pombe* strains do have extensive karyotype differences

224 (Brown *et al.* 2011; Naumov, Kondratieva and Naumova 2015; Jeffares *et al.*
225 2017), including a strain that maintains four (rather than the usual three)
226 chromosomes (Brown *et al.* 2014). There is a significant association between
227 viability and the SV-distance between parents (Jeffares *et al.* 2017), though
228 viability declines at less than 40% viability per variant. This is probably because
229 natural structural variants are biased to chromosome ends that do not contain
230 essential genes (Jeffares *et al.* 2015), due to selection for those that do not cause
231 lethal aneuploidies. Structural variants may also contribute to drive (Zanders *et al.*
232 2014).

233 Formally, reproductive isolation may also be due to Bateson-Dobzhansky-
234 Muller interactions (BDMIs) or any of the other genetic mechanisms of negative
235 epistasis (Nei and Nozawa 2011). However segregation data from random spores
236 (Kondrat'eva and Naumov 2001; Naumov and Kondratieva 2015) and dissected
237 tetrads is inconsistent with simple two-locus BDMIs, which are expected to
238 produce small deviations from expected segregation patterns (even when the
239 affected alleles were strongly linked to markers) (Hou and Schacherer 2016).
240 Ultimately meiotic drive, epistasis and structural variants may have interacting
241 effects on viability, since locally adapted haplotypes are predicted to develop
242 within areas of reduced recombination (Kirkpatrick and Barton 2006).

243 With all these studies of population genetics (reproductive isolation,
244 divergence dating, diversity measures, population size *etc.*) the analyses are based
245 on a small collection of strains that are a worldwide sample of mostly human
246 commensals (see below), so conclusions may not represent natural populations.

247



248

249

250

Figure 1. Intrinsic reproductive isolation in *S. pombe*.

251

A) Random spore viability from three studies shows a decline in spore survival

252

with genetic distance (SNP distance) between parents. The size of circles indicates

253

the lowest self-mating viability of parents. Data from (Kondrat'eva and Naumov

254

2001; Teresa Avelar *et al.* 2013; Jeffares *et al.* 2015). Crosses involving the strain

255

CBS5680 (as in part B) are indicated with cross hairs. The range of genetic

256

differences that have highly variable effects on viability (10,000 – 30,000 SNPs)

257

is indicated with vertical dashed lines. The outlier at top right is JB848/CBS10475

258

(Brazil) x JB870/CBS10499 (South Africa), which appears to be real (Xavier

259

Marsellach, pers. comm.). **B)** segregation of control markers in random spore

260

analysis show strong deviations from the expected 1:1:1:1 ratio, data from

261

(Kondrat'eva and Naumov 2001). For one strain (CBS5680/JB873, from Poland)

262

we show the counts of control markers (aB and Ab are parental types, AB, ab are

263

recombinants, see Kondrat'eva *et al.* for details). Segregation counts whose χ^2 test

264

P-values were < 0.05 are plotted with red bars. Plot text shows the parents of the

265

cross, the random spore viability (RSV) and the χ^2 test P-value (CHISQ.P).

266

C) *wtf* genes have high pairwise diversity within strains compared to all other

267

transmembrane domain containing and non-TM genes (π , left panel), high

268

numbers of segregating sites (θ , middle panel), but are not outliers for Tajima's D

269

(which is calculated from the ratio of the two, D, right panel). Plots show diversity

270

estimators from 57 strains, red circle indicate individual values for *wtf* genes.

271 Predicted transmembrane proteins were collected from a query of Pombase
272 (www.pombase.org), diversity data from (Jeffares *et al.* 2015).

273

274 **Genetics and the reference strain**

275 The fission yeast community has worked almost exclusively with one reference
276 strain, and spontaneous mutants generated from this strain (Fantes and Hoffman
277 2016). This laboratory strain is a natural isolate, and is not an unusual strain
278 phenotypically. It does not appear to be adapted to the standard rich or minimal
279 media, since it does not grow particularly rapidly in these media compared to wild
280 strains. There are several important discoveries that are relevant to the fission
281 yeast researcher. Firstly, Wild strains can differ from the reference by up to
282 68,000 SNPs and up to 24 structural variations, which contribute to phenotypic
283 variation between strains (Clément-Ziza *et al.* 2014; Jeffares *et al.* 2015; Hu, Suo
284 and Du 2015; Jeffares *et al.* 2017). I summarise the structural differences between
285 strains in Supplementary Figure 1. Secondly, the structural differences and
286 meiotic drive elements that wild strains contain complicate crosses between
287 strains, by reducing spore viability and skewing the proportions of alleles that are
288 produced in the offspring (Kondrat'eva and Naumov 2001; Kondrateva and
289 Naumov 2011; Clément-Ziza *et al.* 2014; Hu, Suo and Du 2015; Nuckolls *et al.*
290 2017; Hu *et al.* 2017).

291

292 **The ecology of fission yeast**

293 There have been few published attempts to systematically collect fission
294 yeast strains (Gomes *et al.* 2002; Benito *et al.* 2013; Hellberg 2013). However,
295 fission yeasts have been serendipitously discovered in a variety of microbiological
296 studies (Table 1, Figure 2). Sources have generally been traditional non-
297 industrialised fermentations, produced without any intentional inoculation from
298 substrates that contain high concentrations of sugars. When quantitative estimates
299 of species abundances are included *Schizosaccharomyces* yeasts were generally
300 minor components of these fermentations, with the exceptions of kombucha, some
301 cachaça fermentations and baijiu (from tea, sugar cane and sorghum respectively)
302 (Pataro, Guerra and Peixoto 2000; Teoh, Heard and Cox 2004; Wu, Xu and Chen
303 2012).

304 Perhaps more informative for fission yeast ecology, are the cases where

305 fission yeasts have been discovered in natural substrates such as palm wine (a
 306 fermentation of palm sap) (Theivendirarajah and Chrystopher 1987;
 307 Amanchukwu, Obafemi and Okpokwasili 1989; Ouoba *et al.* 2012). Fission yeast
 308 are also present in natural fermentations of fruits such as *Coffea arabica* and
 309 *Theobroma cacao* (from which coffee and cocoa beans are harvested respectively)
 310 (Silv *et al.* 2000; Schwan and Wheals 2004). Collectively, the field studies show
 311 that fission yeasts are a component of natural microbial communities that ferment
 312 botanical sugars in several geographic regions.

313 Including the strains present in stock collections and in field studies the
 314 most common substrates for fission yeast have been palm wine, grape wine, high-
 315 sugar substrates (molasses, cane sugar, honey) and fruits (Figure 2). Three
 316 selective media to have been described to enrich for fission yeast (Florenzano,
 317 Balloni and Materassi 1977; Hellberg 2013; Benito *et al.* 2013), so further
 318 systematic collections from similar locations and substrates should be possible in
 319 the future.

320

321 **Table 1. *Schizosaccharomyces* in field microbiology**

Substrate	Location	Reference
Grape must	Sicily	(Florenzano, Balloni and Materassi 1977)
Grapes	Ukraine	(Bayraktar 2014)
Palm wine	Sri Lanka	(Atputharajah, Widanapathirana and Samarajeewa 1986; Theivendirarajah and Chrystopher 1987)
Palm wine	Nigeria	(Sanni and Lönner 1993; Amanchukwu, Obafemi and Okpokwasili 2006)
Palm wine	Burkina Faso	(Ouoba <i>et al.</i> 2012)
Rum	Haiti	(Fahrasmane, Ganou-Parfait and Parfait 1988)
Molasses, raisin	Japan/Thailand/Taiwan	(Ishitane 1985)

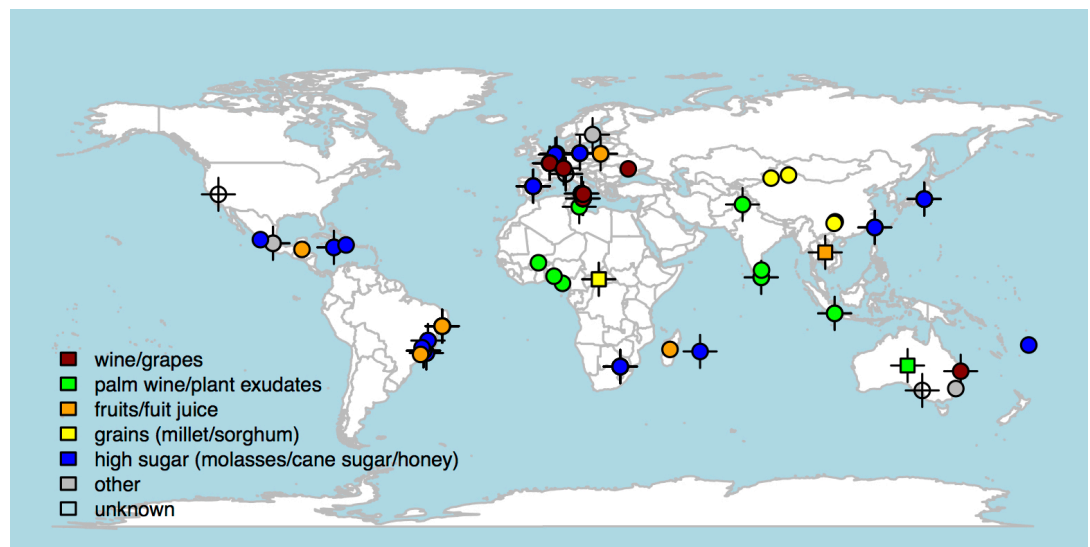
Tequila	Mexico	(Lachance 1995)
Coffee cherries	Brazil Madagascar	(Silv <i>et al.</i> 2000) (Ravelomanana <i>et al.</i> 1984)
Cachaça (from sugar cane)	Brazil	(Pataro, Guerra and Peixoto 2000; Gomes <i>et al.</i> 2002)
Kombucha (fermented tea)	Australia**	(Teoh, Heard and Cox 2004)
Cocoa pulp	Belize	(Schwan and Wheals 2004)
Baijiu (distillate of fermented sorghum)	China	(Wu, Xu and Chen 2012)
Traditional breweries	China	Fen-Yang Bai, pers. comm.
Honey	Fiji	(Ponici and Wimmer 1986)
Honey	Spain	(Benito <i>et al.</i> 2014)

322 * Not microbiological study itself, refers to earlier work.

323 ** From commercial kombucha brewers.

324

325



326

327

328 **Figure 2. Fission yeast locations and substrates.** The locations and
329 substrates where fission yeast have been discovered, including all strains that have
330 been sequenced from stock centers (Fawcett *et al.* 2014; Jeffares *et al.* 2015), and
331 reports from field studies (Table 1). Sequenced strains are marked with cross-
332 hairs, and strains isolated from uncertain locations are marked with a square.

333

334 **The origin of fission yeast**

335 *S. pombe* is now globally distributed (Figure 2), but we know little about its
336 origin and dispersal. We have estimated that these strains began to spread globally
337 in from ~340 BCE (95% confidence interval 1875 BCE–1088 CE), and that the
338 current collection of strains from Brazilian cachaça originated from the remainder
339 in about ~1620 CE (confidence interval 1422–1752 CE) (Jeffares *et al.* 2015), a
340 hint that like budding yeast and *C. elegans*, this model has probably been
341 dispersed as a commensal (most likely in fermented beverages).

342 The reference strain originated from French grapes (Osterwalder 1924). The
343 common belief is that *S. pombe* originated from Africa, perhaps because the initial
344 species description was from an African millet beer isolate (Lindner 1893;
345 Vorderman 1894). While genetic analysis is consistent with exchange between
346 African and European stocks (Jeffares *et al.* 2015), and some strains have been
347 collected from traditional African fermentations, there is no scientific evidence for
348 an African origin of this species. There are very few studies of the microbial
349 constituents of millet beer from Africa (I could find none than specifically
350 mentioned *S. pombe*, and one description of sorghum beer that did not mention *S.*
351 *pombe* (Kayode *et al.* 2011)). Since fission yeasts can be major components of
352 kombucha, which has been traditionally produced in China (Sreeramulu, Zhu and
353 Knol 2000; Teoh, Heard and Cox 2004), palm wine which is widely produced in
354 Asia (Table 1, Figure 2), and in traditional Chinese breweries (Fen-Yang Bai,
355 pers. comm.), China is an equally good candidate for the initial origin of *S.*
356 *pombe*.

357

358 **Why study diversity in fission yeast?**

359 The small genomes of budding yeasts enabled the early development of
360 population genomics methods (Liti *et al.* 2009; Schacherer *et al.* 2009), and now
361 large scale accurate quantitative genetics analyses (Bloom *et al.* 2013; Märtens *et*
362 *al.* 2016). The continuing advance of sequence throughput, analysis software and

363 laboratory methods (eg: RAD-seq) have now made population genomics
364 approaches available to any species. However, the abundance of genome-scale
365 data and technical tools and the small non-redundant genomes of yeasts make
366 them attractive models for systems biology, including approaches to
367 understanding genetic diversity and traits (Parts 2014). Fission yeast has the
368 benefit of being haploid (so that F1 generations need not be intercrossed). As with
369 budding yeast, fission yeast has abundant heritable phenotypic diversity in
370 growth, stress responses, cell morphology, and cellular biochemistry that is yet to
371 be explored with powerful quantitative genetics (Brown *et al.* 2011; Clément-Ziza
372 *et al.* 2014; Jeffares *et al.* 2015; 2017). Yeasts are also powerful tools for detailed
373 study of evolutionary processes using pooled time-series sequencing and other
374 high-throughput approaches that would be expensive or unfeasible in other
375 species (Cubillos *et al.* 2011; Hou *et al.* 2015). Finally, studies by Benito *et al.*
376 show that some non-reference *S. pombe* strains have potential in the winemaking
377 industry (Benito *et al.* 2014; 2016), so diverse strains could well have potential
378 elsewhere in biotechnology.

379

380 **Acknowledgements**

381 I thank Mathieu Clément-Ziza for commentary about unpublished work and
382 Xavier Marsellach for discussions.

383

384 **Supplementary data**

385 All used for plots is available at figshare at:

386 https://figshare.com/projects/The_natural_diversity_and_ecology_of_fission_yeast/21761

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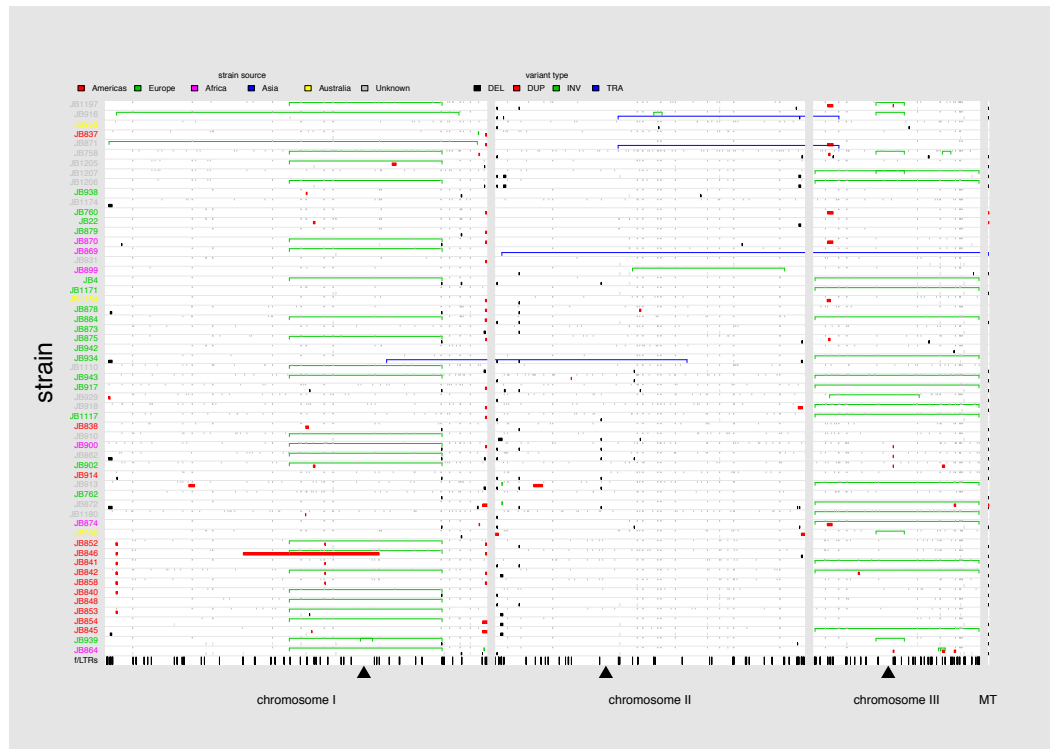
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401 **Supplementary Figure 1. Structural variants present in wild fission yeast**402 **strains.** Using predictions from short read data (Jeffares *et al.* 2017), I show the

403 genomic location of structural variants (SVs) in wild strains contain that differ

404 from the standard laboratory isolate (Leupold's 972) . I show deletions (black),

405 duplications (red), inversions (green) and translocations (blue). SVs present in

406 each of the 57 non-clonal strains are shown within the white horizontal bars, with

407 strain names coloured according to their continent of origin. Tfl-type

408 retrotransposon insertions that are present in some, but not all strains are shown at

409 grey ticks at the tops of bars. The positions of fixed Tfl-type retrotransposon

410 insertions are indicated on the last row (f/LTRs). Centromeres are indicated with

411 black triangles.

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413

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