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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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14

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 al.*  
124 *et 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 ( $\pi$ ) were  $\sim 3 \times 10^{-3}$  (pairwise comparison have an average of 3 SNPs/kb),  
130 slightly higher than the budding yeast *Saccharomyces cerevisiae* ( $1 \times 10^{-3}$ ) (Liti *et al.*  
131 *et 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 al.*  
137 *et 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  $1.7 \times 10^{-10}$  (or  $2.0 \times 10^{-10}$ ) per base per generation (Farlow *et al.*  
152 *et al.* 2015; Behringer and Hall 2015), very similar to estimates for the budding  
153 yeast *Saccharomyces cerevisiae* (estimates at 3 and  $1.67 \times 10^{-10}$ ) (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.

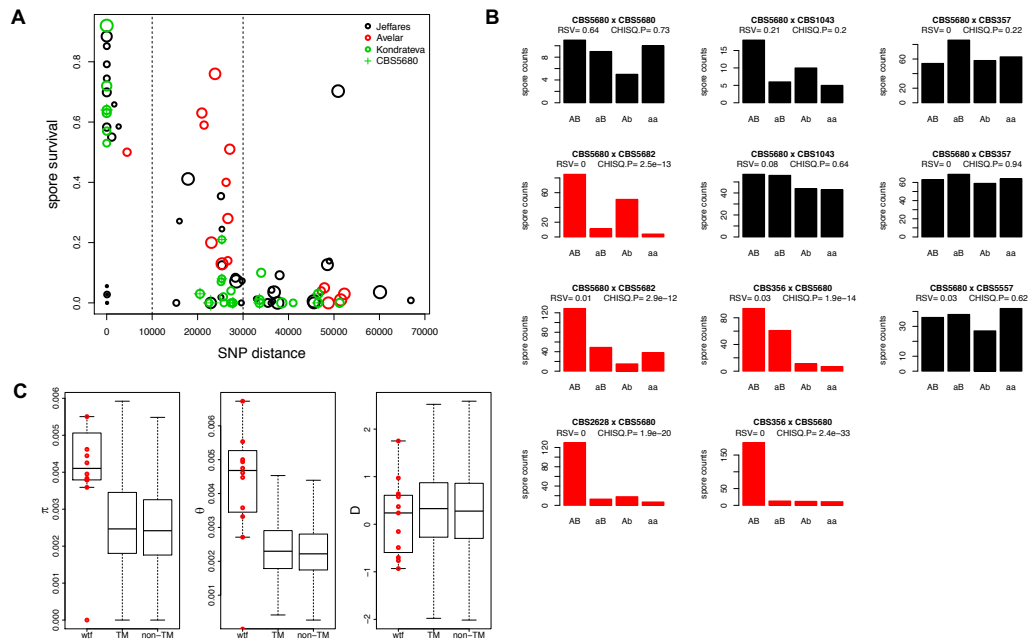
220       Reproductive isolation may also be the result of the aneuploidy that occurs  
221 when parents differ in chromosomal inversions and translocations. For example,  
222 engineered inversions and translocations reduce spore viability by ~40% (Teresa  
223 Avelar *et al.* 2013). *S. pombe* strains 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  $\chi^2$  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  $\chi^2$  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 ( $\pi$ , left panel), high

268

numbers of segregating sites ( $\theta$ , 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)

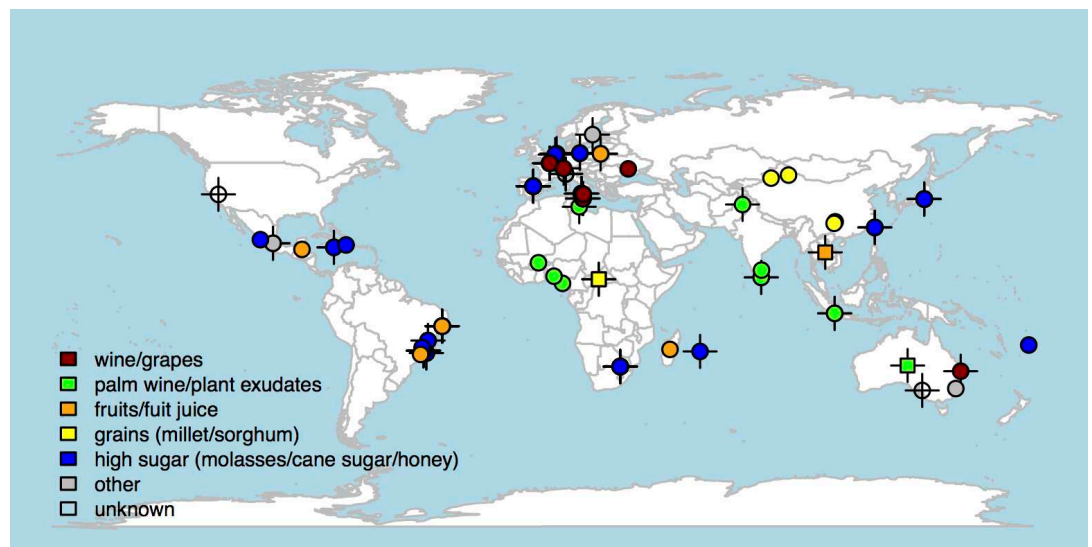
Tequila	Mexico	(Lachance 1995)
Coffee cherries	Brazil Madagascar	(Silv <i>et al.</i> 2000) (Ravelomanana <i>et al.</i> 1984)
Cachaça (from sugar cane)	Brazil	(Pataro, Guerra and Peixoto 2000; Gomes <i>et al.</i> 2002)
Kombucha (fermented tea)	Australia**	(Teoh, Heard and Cox 2004)
Cocoa pulp	Belize	(Schwan 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

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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\\_yeas](https://figshare.com/projects/The_natural_diversity_and_ecology_of_fission_yeast/21761)  
387 [t /21761](https://figshare.com/projects/The_natural_diversity_and_ecology_of_fission_yeast/21761)

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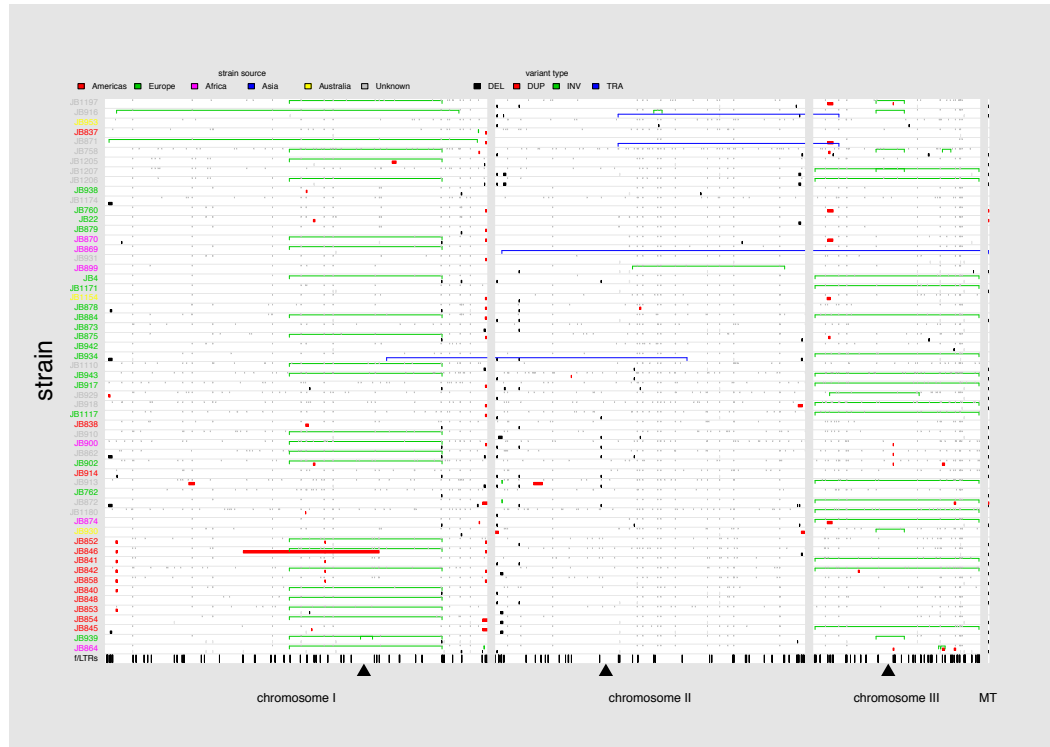
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401 **Supplementary Figure 1. Structural variants present in wild fission yeast**402 **strains.** Using predictions from short read data (Jeffares *et al.* 2017), I show the

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

414

415 **References**

416

417 Amanchukwu SC, Obafemi A, Okpokwasili GC. Single-cell-protein production

418 by *Schizosaccharomyces pombe* isolated from palmwine using hydrocarbon419 feedstocks. *Folia Microbiol* 1989;**34**:112–9.



- 420 Amanchukwu SC, Obafemi A, Okpokwasili GC. Hydrocarbon degradation and  
421 utilization by a palm-wine yeast isolate. *FEMS microbiology letters*  
422 2006;**57**:151–4.
- 423 Atputharajah JD, Widanapathirana S, Samarajeewa U. Microbiology and  
424 biochemistry of natural fermentation of coconut palm sap - ScienceDirect.  
425 *Food Microbiol* 1986.
- 426 Bayraktar VN. Biodiversity of yeast cultures isolated from littoral areas of the  
427 Regional Landscape Park «Tiligulskiy». *J. Optimization and Protection of*  
428 *Ecosystems* 2014.
- 429 Behringer MG, Hall DW. Genome-Wide Estimates of Mutation Rates and  
430 Spectrum in *Schizosaccharomyces pombe* Indicate CpG Sites are Highly  
431 Mutagenic Despite the Absence of DNA Methylation. *G3* 2015;**6**:149–60.
- 432 Benito Á, Jeffares DC, Palomero F *et al.* Selected *Schizosaccharomyces pombe*  
433 Strains Have Characteristics That Are Beneficial for Winemaking. *PLoS ONE*  
434 2016;**11**:e0151102.
- 435 Benito S, Gálvez L, Palmero F *et al.* *Schizosaccharomyces* selective differential  
436 media. *African Journal of Microbiology Research* 2013;**7**:3026–36.
- 437 Benito S, Palomero F, Calderón F *et al.* Selection of appropriate  
438 *Schizosaccharomyces* strains for winemaking. *Food Microbiol* 2014;**42**:218–  
439 24.
- 440 Bloom JS, Ehrenreich IM, Loo WT *et al.* Finding the sources of missing  
441 heritability in a yeast cross. *Nature* 2013;**494**:234–7.
- 442 Bridge PD, May JW. A numerical classification of fission yeasts of the genus  
443 *Schizosaccharomyces* Lindner. *Microbiology* 1984;**130**:1921–32.
- 444 Brown WRA, Liti G, Rosa C *et al.* A geographically diverse collection of  
445 *Schizosaccharomyces pombe* isolates shows limited phenotypic variation but  
446 extensive karyotypic diversity. Fay JC (ed.). *G3* 2011;**1**:615–26.
- 447 Brown WRA, Thomas G, Lee NCO *et al.* Kinetochore assembly and  
448 heterochromatin formation occur autonomously in *Schizosaccharomyces*

- 449 pombe. *Proc Natl Acad Sci USA* 2014;**111**:1903–8.
- 450 Clément-Ziza M, Marsellach FX, Codlin S *et al.* Natural genetic variation impacts  
451 expression levels of coding, non-coding, and antisense transcripts in fission  
452 yeast. *Mol Syst Biol* 2014;**10**:764–4.
- 453 Cubillos FA, Billi E, Zörgö E *et al.* Assessing the complex architecture of  
454 polygenic traits in diverged yeast populations. *Mol Ecol* 2011;**20**:1401–13.
- 455 Fahasmane L, Ganou-Parfait B, Parfait A. Research note: Yeast flora of Haitian  
456 rum distilleries. *Mircen Journal* 1988;**4**:239–41.
- 457 Fantes PA, Hoffman CS. A Brief History of *Schizosaccharomyces pombe*  
458 Research: A Perspective Over the Past 70 Years. *Genetics* 2016;**203**:621–9.
- 459 Farlow A, Long H, Arnoux S *et al.* The Spontaneous Mutation Rate in the Fission  
460 Yeast *Schizosaccharomyces pombe*. *Genetics* 2015;**201**:737–44.
- 461 Fawcett JA, Iida T, Takuno S *et al.* Population Genomics of the Fission Yeast  
462 *Schizosaccharomyces pombe*. *PLoS ONE* 2014;**9**:e104241.
- 463 Fay JC. The molecular basis of phenotypic variation in yeast. *Current Opinion in*  
464 *Genetics and Development* 2013;**23**:672–7.
- 465 Florenzano G, Balloni W, Materassi R. Contributo alla ecologia dei lieviti  
466 *Schizosaccharomyces* sulle uve. *Vitis* 1977;**16**:38–44.
- 467 Ganthala BP, Marshall JH, May JW. Xerotolerance in fission yeasts and the role  
468 of glycerol as compatible solute. *Archives of microbiology* 1994;**162**:108–13.
- 469 Gomes FCO, Pataro C, Guerra JB *et al.* Physiological diversity and trehalose  
470 accumulation in *Schizosaccharomyces pombe* strains isolated from  
471 spontaneous fermentations during the production of the artisanal Brazilian  
472 cachaça. *Can J Microbiol* 2002;**48**:399–406.
- 473 Hagan IM, Grallert A, Carr AM *et al.* eds. *Fission Yeast: a Laboratory Manual*.  
474 Cold Spring Harbour Laboratory Press, 2016.
- 475 Hellberg JEEU. Finding wild fission yeast: Where are they? *Genetics Society*  
476 *News* 2013;**68**:59–60.

- 477 Helston RM, Box JA, Tang W *et al.* Schizosaccharomyces cryophilus sp nov., a  
478 new species of fission yeast. *Fems Yeast Res* 2010;**10**:779–86.
- 479 Hoffman CS, Wood V, Fantes PA. An Ancient Yeast for Young Geneticists: A  
480 Primer on the Schizosaccharomyces pombe Model System. *Genetics*  
481 2015;**201**:403–23.
- 482 Hou J, Friedrich A, Gounot J-S *et al.* Comprehensive survey of condition-specific  
483 reproductive isolation reveals genetic incompatibility in yeast. *Nat Commun*  
484 2015;**6**:7214.
- 485 Hou J, Schacherer J. On the Mapping of Epistatic Genetic Interactions in Natural  
486 Isolates: Combining Classical Genetics and Genomics. *Methods Mol Biol*  
487 2016;**1361**:345–60.
- 488 Hu W, Jiang Z-D, Suo F *et al.* A large gene family in fission yeast encodes spore  
489 killers that subvert Mendel's law. *eLife* 2017;**6**:3025.
- 490 Hu W, Suo F, Du L-L. Bulk segregant analysis reveals the genetic basis of a  
491 natural trait variation in fission yeast. *Genome Biol Evol* 2015;**7**:3496–510.
- 492 Ishitane. Identification of yeasts isolated from high-sugar foods. Tokuoka K,  
493 Ishitane T, Goto S, et al. (eds.). *J Gen Appl Microbiol* 1985;**31**:411–27.
- 494 Jeffares DC, Jolly C, Hoti M *et al.* Transient structural variations have strong  
495 effects on quantitative traits and reproductive isolation in fission yeast. *Nat*  
496 *Commun* 2017;**8**:14061.
- 497 Jeffares DC, Jolly C, Hoti M *et al.* Transient structural variations have strong  
498 effects on quantitative traits and reproductive isolation in fission yeast.  
499 *Multiple values selected* **8**:14061EP–.
- 500 Jeffares DC, Mourier T, Penny D. The biology of intron gain and loss. *Trends*  
501 *Genet* 2006;**22**:16–22.
- 502 Jeffares DC, Rallis C, Rieux A *et al.* The genomic and phenotypic diversity of  
503 Schizosaccharomyces pombe. *Nature Genetics* 2015;**47**:235–41.
- 504 Kayode APP, Vieira-Dalode G, Linnemann AR *et al.* Diversity of yeasts involved  
505 in the fermentation of tchoukoutou, an opaque sorghum beer from Benin.

- 506 *African Journal of Microbiology Research* 2011;**5**:2737–42.
- 507 Kirkpatrick M, Barton N. Chromosome inversions, local adaptation and  
508 speciation. *Genetics* 2006;**173**:419–34.
- 509 Kondrat'eva VI, Naumov GI. The Phenomenon of Spore Killing in  
510 *Schizosaccharomyces pombe* Hybrids. *Doklady Biological Sciences* 2001.
- 511 Kondrateva VI, Naumov GI. POPULATION ANTAGONISM IN THE YEASTS  
512 *SCHIZOSACCHAROMYCES POMBE*. *Ecological genetics* 2011;**9**:21–6.
- 513 Lachance MA. Yeast communities in a natural tequila fermentation. *Antonie Van*  
514 *Leeuwenhoek* 1995;**68**:151–60.
- 515 Leducq J-B, Nielly-Thibault L, Charron G *et al.* Speciation driven by  
516 hybridization and chromosomal plasticity in a wild yeast. *Nat Microbiol*  
517 2016;**1**:15003.
- 518 Leupold U. Die Vererbung von Homothallie und Heterothallie bei  
519 *Schizosaccharomyces pombe*. *Compt Rend Trav Lab Carlsberg Ser*  
520 1949;**24**:381–480.
- 521 Lindner P. *Schizosaccharomyces pombe* n. sp., ein neuer Gärungserreger.  
522 *Wochenschrift für Brauerei* 1893;**10**:1298–300.
- 523 Liti G, Barton DBH, Louis EJ. Sequence diversity, reproductive isolation and  
524 species concepts in *Saccharomyces*. *Genetics* 2006;**174**:839–50.
- 525 Liti G, Carter DM, Moses AM *et al.* Population genomics of domestic and wild  
526 yeasts. *Nature* 2009;**458**:337–41.
- 527 Liti G, Louis EJ. Advances in quantitative trait analysis in yeast. Fay JC (ed.).  
528 *PLoS Genet* 2012;**8**:e1002912.
- 529 Lynch M, Sung W, Morris K *et al.* A genome-wide view of the spectrum of  
530 spontaneous mutations in yeast. *Proc Natl Acad Sci USA* 2008;**105**:9272–7.
- 531 Märtens K, Hallin J, Warringer J *et al.* Predicting quantitative traits from genome  
532 and phenome with near perfect accuracy. *Nat Commun* 2016;**7**:11512.

- 533 McDowall MD, Harris MA, Lock A *et al.* PomBase 2015: updates to the fission  
534 yeast database. *Nucleic Acids Res* 2015;**43**:D656–61.
- 535 Mourier T, Jeffares DC. Eukaryotic intron loss. *Science (New York, NY)*  
536 2003;**300**:1393–3.
- 537 Naumov G, Kondratieva V. Hybrid sterility of the yeast *Schizosaccharomyces*  
538 *pombe*: Genetic genus and many species in statu nascendi? *Microbiology*  
539 (00262617) 2015.
- 540 Naumov GI, Kondratieva VI, Naumova ES. [Hybrid Sterility of the Yeast  
541 *Schizosaccharomyces pombe*: Genetic Genus and Many Species in statu  
542 nascendi?]. *Mikrobiologiya* 2015;**84**:192–203.
- 543 Nei M, Nozawa M. Roles of mutation and selection in speciation: from Hugo de  
544 Vries to the modern genomic era. *Genome Biol Evol* 2011;**3**:812–29.
- 545 Nuckolls NL, Bravo Núñez MA, Eickbush MT *et al.* wtf genes are prolific dual  
546 poison-antidote meiotic drivers. *eLife* 2017;**6**:2235.
- 547 Osterwalder A. *Schizosaccharomyces liquefaciens* n.sp., eine gegen freie  
548 schweflige Säure widerstandsfähige Gärhefe. *Mitt Geb Lebensmittelunters*  
549 *Hyg* 1924;**15**:5–28.
- 550 Ouoba LII, Kando C, Parkouda C *et al.* The microbiology of Bandji, palm wine of  
551 *Borassus akeassii* from Burkina Faso: identification and genotypic diversity  
552 of yeasts, lactic acid and acetic acid bacteria. *J Appl Microbiol* 2012, DOI:  
553 10.1111/jam.12014.
- 554 Parts L. Genome-wide mapping of cellular traits using yeast. *Yeast* 2014;**31**:197–  
555 205.
- 556 Pataro C, Guerra JB, Peixoto MP. Yeast communities and genetic polymorphism  
557 of *Saccharomyces cerevisiae* strains associated with artisanal fermentation in  
558 Brazil. *Journal of Applied Microbiology* 2000;**89**:24–31.
- 559 Patch A-M, Aves SJ. Fingerprinting fission yeast: polymorphic markers for  
560 molecular genetic analysis of *Schizosaccharomyces pombe* strains.  
561 *Microbiology (Reading, Engl)* 2007;**153**:887–97.

- 562 Peter J, Schacherer J. Population genomics of yeasts: towards a comprehensive  
563 view across a broad evolutionary scale. *Yeast* 2016;**33**:73–81.
- 564 Ponici L, Wimmer FL. Characterization of the yeasts (Blastomycetes) in some  
565 Fijian honeys. *Acta alimentaria polonica* 1986.
- 566 Ravelomanana R, Guiraud JP, Vincent JC *et al.* Study of the yeast flora from the  
567 traditional cacao fermentation in Madagascar. *Revue des Fermentations et des*  
568 *Industries Alimentaires (Belgium)* 1984.
- 569 Rhind N, Chen Z, Yassour M *et al.* Comparative functional genomics of the  
570 fission yeasts. *Science (New York, NY)* 2011;**332**:930–6.
- 571 Robinson HA, Pinharanda A, Bensasson D. Summer temperature can predict the  
572 distribution of wild yeast populations. *Ecol Evol* 2016;**6**:1236–50.
- 573 Sanni AI, Lönner C. Identification of yeasts isolated from Nigerian traditional  
574 alcoholic beverages. *Food Microbiol* 1993;**10**:517–23.
- 575 Schacherer J, Shapiro JA, Ruderfer DM *et al.* Comprehensive polymorphism  
576 survey elucidates population structure of *Saccharomyces cerevisiae*. *Nature*  
577 2009;**458**:342–5.
- 578 Schlake T, Gutz H. Mating configurations in *Schizosaccharomyces pombe* strains  
579 of different geographical origins. *Curr Genet* 1993.
- 580 Schwan RF, Wheals AE. The microbiology of cocoa fermentation and its role in  
581 chocolate quality. *Crit Rev Food Sci Nutr* 2004;**44**:205–21.
- 582 Seike T, Nakamura T, Shimoda C. Molecular coevolution of a sex pheromone and  
583 its receptor triggers reproductive isolation in *Schizosaccharomyces pombe*.  
584 *Proc Natl Acad Sci USA* 2015;**112**:4405–10.
- 585 Silv CF, Schwan RF, Sousa Dias ES *et al.* Microbial diversity during maturation  
586 and natural processing of coffee cherries of *Coffea arabica* in Brazil. *Int J*  
587 *Food Microbiol* 2000;**60**:251–60.
- 588 Sipiczki M, Kucsera J, Ulaszewski S *et al.* Hybridization Studies by Crossing and  
589 Protoplast Fusion Within the Genus *Schizosaccharomyces* Lindner. *J Gen*  
590 *Microbiol* 1982;**128**:1989–2000.

- 591 Sreeramulu G, Zhu Y, Knol W. Kombucha fermentation and its antimicrobial  
592 activity. *J Agric Food Chem* 2000;**48**:2589–94.
- 593 Swinnen S, Thevelein JM, Nevoigt E. Genetic mapping of quantitative phenotypic  
594 traits in *Saccharomyces cerevisiae*. *Fems Yeast Res* 2012;**12**:215–27.
- 595 Tajima F. Statistical method for testing the neutral mutation hypothesis by DNA  
596 polymorphism. *Genetics* 1989;**123**:585–95.
- 597 Teoh AL, Heard G, Cox J. Yeast ecology of Kombucha fermentation. *Int J Food*  
598 *Microbiol* 2004;**95**:119–26.
- 599 Teresa Avelar A, Perfeito L, Gordo I *et al*. Genome architecture is a selectable  
600 trait that can be maintained by antagonistic pleiotropy. *Nat Commun*  
601 2013;**4**:2235.
- 602 Theivendirarajah K, Chrystopher RK. Microflora and microbial activity in  
603 palmyrah (*Borassus flabellifer*) palm wine in Sri Lanka. *Mircen Journal*  
604 1987;**3**:23–31.
- 605 Vaughan Martini A. Evaluation of phylogenetic relationships among fission yeast  
606 by nDNA/nDNA reassociation and conventional taxonomic criteria. *Yeast*  
607 1991;**7**:73–8.
- 608 Vorderman AG. *Analecta Op Bromatologisch Gebied: I II*. Geneeskundig  
609 Tijdschrift voor Nederlandsch-Indië, 1894:343–97.
- 610 Wood V, Gwilliam R, Rajandream M-A *et al*. The genome sequence of  
611 *Schizosaccharomyces pombe*. *Nature* 2002;**415**:871–80.
- 612 Wu Q, Xu Y, Chen L. Diversity of yeast species during fermentative process  
613 contributing to Chinese Maotai-flavour liquor making. *Letters in Applied*  
614 *Microbiology* 2012;**55**:301–7.
- 615 Zanders SE, Eickbush MT, Yu JS *et al*. Genome rearrangements and pervasive  
616 meiotic drive cause hybrid infertility in fission yeast. *eLife*  
617 2014;**2014**:e02630.
- 618 Zhu YO, Siegal ML, Hall DW *et al*. Precise estimates of mutation rate and  
619 spectrum in yeast. *Proc Natl Acad Sci USA* 2014;**111**:E2310–8.

620 Zimmer M, Welser F, Orlaer G *et al.* Distribution of mitochondrial introns in the  
621 species *Schizosaccharomyces pombe* and the origin of the group II intron in  
622 the gene encoding apocytochrome b. *Curr Genet* 1987;**12**:329–36.

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624 The author declares that there is no conflict of interest.