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A Comparison of Yeast Communities Found in Necrotic Tissue of Cladodes and Fruits of *Opuntia stricta* on Islands in the Caribbean Sea and Where Introduced into Australia

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Abstract. Yeast communities growing in the decaying tissues (cladodes and fruits) of *Opuntia stricta* (prickly pear cactus) and associated yeast vectors (*Drosophila* species) were compared in two geographic regions (Caribbean and eastern Australia). The Australian yeast community provides an interesting comparison to the Caribbean community, because the host plant *O. stricta* was introduced to Australia over 100 years ago. Many of the yeasts found in the Australian system also were introduced during a period of biological control (1926–1935) when they accompanied rotting prickly pear cladodes and insects shipped to Australia from the Americas. The yeast community composition (proportion of each species) is compared at several levels of organization: (1) within and between regions, (2) across seasons and years, and (3) within and between tissue types. The yeast species composition of the cladode communities are similar from locality to locality, season to season, and year to year, with the region-to-region similarity slightly less. The composition of the fruit-yeast communities are distinct from region to region and only show some overlap with the cladodes within regions when collected simultaneously in the same locality. It is suggested that the cladode-microorganism-*Drosophila* system is relatively closed (little extrinsic influence) whereas the fruit-microorganism-*Drosophila* system is open (large extrinsic influence).

Introduction

The composition of communities, in terms of species present and their proportional representation, is interesting from two standpoints. One is an evolutionary view of how and why a community arrives at a certain state (membership and relative numbers of species) over many generations. Thus, stasis and change are under the influences of evolutionary forces such as natural selection and chance. Another is an ecological view of how and why a community is found at a certain state at a particular time (e.g., within a generation where time is short on the evolutionary scale). Thus, environmental constraints, species-species interactions (e.g., competition or facilitation), predation, succes-

sional factors, and chance may play a role in shaping the community. The objective of this study was to try to evaluate the role of the extrinsic environment on the composition of yeast communities. We compared the yeast communities found in the decaying tissues of *Opuntia stricta* (prickly pear cactus) in their endemic environments (Pan-Caribbean) to communities found in an introduced environment (Australia). Some of the Australian localities are climatically different from those of the endemic populations, being at higher elevations and higher latitudes [12]. In addition, the indigenous yeast species in the local environment may influence the new communities in the introduced habitat.

Decaying cladodes or pads of *O. stricta* are utilized for feeding and breeding by a number of insects including drosophilids. In the Caribbean, *Drosophila mulleri, D. mayaguana,* undescribed species *S. D. stalkeri,* and *D. richardsoni* are all found to various degrees in decaying cladodes (W. B. Heed, unpublished data). In Australia, both *D. buzzatii* and *D. aldrichi* are known to use the cladodes but *D. buzzatii* is more common and widespread [1, 2]. In both regions fresh, healthy *Opuntia stricta* cladode tissue is attacked by the phycitid moth *Cactoblastis cactorum,* and rot initiation is thought to be principally due to this primary herbivore. The resulting damaged tissue is then utilized by a community of bacteria, yeasts, and molds on which the drosophilids and sometimes other insects feed, breed, and subsequently vector the microorganisms to new rots.

The fruits of *O. stricta* are seasonal, they mature in autumn or winter [6]. The fruits are deciduous, but rotting often begins on the plant after lesions are made by birds foraging on the succulent tissue. The holes left by birds are then occupied by insects, including ants, beetles, and dipterans, most notably drosophilids. In the Caribbean, a number of *Drosophila* species feed on the open fruit and some breed there. However, some species that breed elsewhere will also feed on this sugar-rich substrate and may be responsible for introducing microorganisms from other sources. The situation in Australia is similar in that birds may be primarily responsible for fruit damage, and several drosophilids (endemic and introduced), including the pad-breeding *D. buzzatii,* will feed on fruits.

The original introduction of *O. stricta* into Australia is not certain, but it is presumed to have come from the vicinity of trade ports in Texas, Florida and Cuba [8]. The first record of *Opuntia stricta* (originally referred to as *O. inermis,* a synonym) appears to be that of a plant brought from Sydney to Scone, New South Wales in 1839. However, this species is believed to have been growing in cultivation at Parramatta (near Sydney) prior to that date. Sydney was also the source for introduction of the species to Queensland in 1843.

Introduction of various insects and the establishment of biological control took place primarily in the years 1926 to 1930 when large shipments of material, including cut portions of prickly pear (*Opuntia*) with accompanying insects that feed on the cladodes (eggs, larvae and adults), were sent from the Americas. The last consignment arrived in December 1935. A total of 67 separate shipments (59 from the United States and 8 from Argentina) included 1,230 cases of material. The microbiota contained in these shipments, along with the accidental *Drosophila,* were probably the major source of the present day com-

munities of yeasts. We therefore presume the yeast communities of *O. stricta* in Australia to be a composite of species from both American continents. Furthermore, the communities found in the Caribbean are tentatively considered to be endemic to that region. It is of interest that *Cactoblastis cactorum* (indigenous to South America) found in the Caribbean was subsequently introduced to that region from Australia [9, 11–13] and that populations of *O. stricta* in the Caribbean are now under this similar selective pressure. Introduction of *Cactoblastis cactorum* from Australia to the Caribbean in 1957 was via egg sticks (which may not carry yeasts) and not by shipment of rotting materials [9].

Materials and Methods

Samples of necrotic *O. stricta* tissue (cladodes and fruits) were collected from 20 distinct localities (14 from various Caribbean islands, 6 from eastern Australia). These collection localities are listed in Table 1 along with the date and number of samples. The collection localities in the Caribbean region were pooled according to proximity (e.g., same island) to form 8 Caribbean localities. The Caribbean collections were made in May 1982 during cruise CF-8205 and in November 1983 during cruise CF-8314 of the research vessel Cape Florida in that region.

Samples were taken by removing several grams of decaying tissue from the infected cladodes or fruits of individual plants. No more than one sample per plant was taken. Each tissue sample was placed in a sterile Whirl-pak bag and stored in a cool box (4–10°C) until return to the laboratory for plating. Plating took place from 2–8 hours after collection. Homogenates were prepared by placing 1 g of the decaying tissue into 9 ml of sterile water and vortexing for 1 min. Dilutions of each homogenate were made in steps of 1/10 in sterile water followed by plating 0.1 ml of various dilutions as previously described [15]. In some cases one loopful (0.01–0.02 ml) of the original homogenate was directly streaked on the isolation plate. Plating was conducted on acidified (to pH 3.8 with 1N HCl) yeast extract-malt extract agar (YM, Difco), and incubation was at room temperature (25°C). A representative of each colony type from each sample was restreaked once or twice and the resulting culture was identified by standard recommended procedures [16]. Frequency of isolation was determined on a per plant basis by dividing the number of plants containing a particular yeast by the number of plants sampled. Some information is lost by this procedure but we have employed it so that earlier work on the Australian microbiota [3, 4] could be used in direct comparison. In addition, several complicating factors such as differences in rot moisture, variable adherence of yeast cells, and inability to equally homogenize all rot samples made within-sample estimates of yeast numbers less reliable. We have therefore focused our attention on sampling yeast communities within a locality. Similarity of two yeast communities was calculated as Pearson's product moment correlation coefficient (r). Those localities that yielded less than 15 yeast isolates from a particular substrate were not included in the similarity analysis (i.e., pad localities 3, 11; fruit localities 4, 6, 7). The physiological structure of yeast communities was analyzed by the method of Lachance and Starmer [10], summarized as follows. The species frequency matrix is multiplied by a matrix of physiological responses of the component yeast species, and each community is then described as a vector of mean physiological responses. The responses are compared with the expected mean responses of all yeast species known (data from Barnett et al. [5]) and expressed as standard normal deviates of their expected mean using a binomial model for smaller communities and a normal approximation for larger ones. A mean physiological response may then be viewed as not significant, or as significantly larger or smaller than expectation for a particular critical probability (e.g., 0.05). The proportion of community responses that appear significant by this analysis is given as S. In the present case, *Prototheca* entries were deleted from the data because its characteristics were not included in the physiological data by Barnett et al. [5]. The entry for fruit from Navassa Island is deleted also because it included only one yeast species.

Table 1. Collection localities, dates, substrates, and numbers for *Opuntia stricta* cladodes and fruits from two regions

Locality	Date	No. of samples	
		Cladodes	Fruits
(Caribbean Islands)			
Haiti			
Fond Parisienne	5/6/82	13	0
Jacmel	5/7/82	5	0
Total		18	0
Spanish Pt., Montserrat	5/15/82	17	0
British Virgin Islands			
Beef Island	5/17/82	1	0
Virgin Gorda	5/18/82	3	0
Total		4	0
Little Conception Island, Bahamas	11/18/83	16	3
Great Inagua, Bahamas	11/19/83	28	27
Navassa Island, U.S.A.	11/21/83	30	3
Jamaica			
Discovery Bay	11/23/83	15	10
Sandy Bay	11/23/83	11	0
Palisadoes, Kingston	11/23/83	2	0
Total		28	10
Cayman Islands			
Cayman Brac	11/25/83	16	15
Little Cayman Island	11/26/83	5	1
Grand Cayman Island	11/27/83	12	0
Total		33	16
(Australia)			
O'Hara (Denman), N.S.W.	7/27/84	18	9
Breeza, N.S.W.	8/5/84	19	0
Metz Gorge, N.S.W.	8/9/84	8	12
Trinkey, N.S.W.	8/13/84	5	7
Hemmant, Queensland	8/19/84	18	0
Lightning Ridge, N.S.W.	10/8/84	13	0

To produce a structured table of significantly deviating responses, the deviates were subjected to bidimensional clustering by equally weighted pair agglomeration of a cosine matrix. A dendrogram shows the cluster structure (Fig. 1), and partitions show groups of physiological responses differing from one another by cosine values of 0.8 or less.

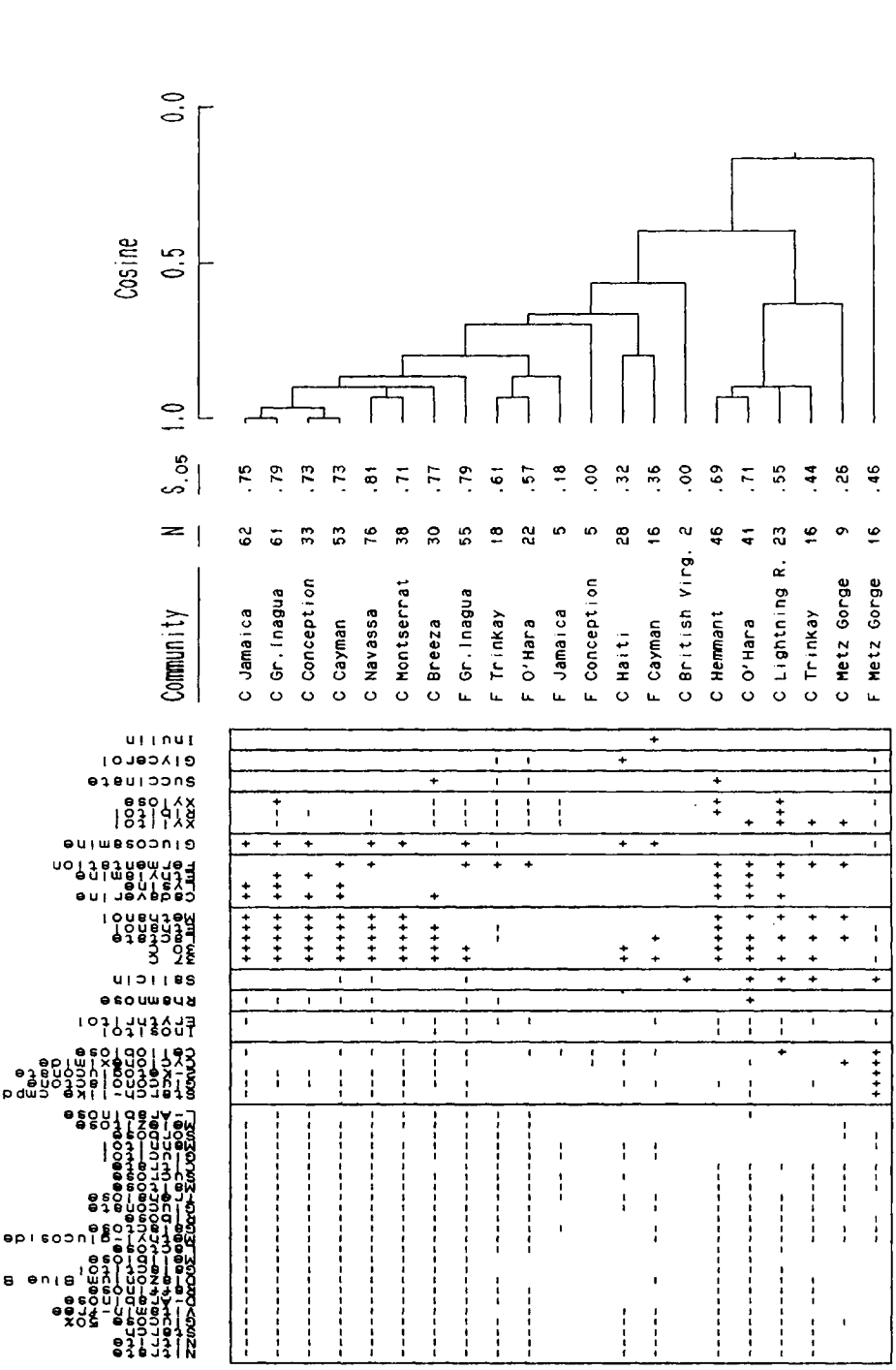

Fig. 1. Dendrogram showing cluster structure of yeast communities according to the physiological abilities of the constituent species [5]. Significant physiological deviations of each community from a standard community is shown in the diagram.

Results

Tables 2 and 3 list the proportions of the yeast species found in decaying cladodes and fruits, respectively. It is apparent from Table 2 that certain common yeasts in the Caribbean region are restricted to that area (i.e., *Pichia norvegensis, P. amethionina* var. "f", and the less common *P. mexicana*) as compared to the Australian collection. It should be noted that *P.* sp. "B" and strains belonging to the *Cryptococcus cereanus* complex have been found in Australia in previous studies of *O. stricta* yeasts [3, 4]. The reason for referring in Table 2 to the *Cr. cereanus* complex rather than to *Cr. cereanus* (or its teleomorph *Sporopachydermia cereana*) is that the G + C contents of many isolates were found to be much lower than that of *Cr. cereanus,* suggesting that we are dealing with related but different species. Strains of *Clavispora* sp. reported in Table 2 were mainly *Clavispora opuntiae* and a minor number of the related species *Clavispora lusitaniae.*

Those common yeasts found only in the Australian survey but not in the Caribbean are *Pichia opuntiae,* and to some extent *Hansenula californica, Candida boidinii,* and *Kloeckera apiculata.* The last species was found primarily in fruits and appeared in the cladodes, in low numbers, when fruits were present (compare Tables 2 and 3). Locality #14 also had fruits present but they were not sampled. Both *H. californica* (a common soil yeast) and *C. boidinii* (occasionally found in cactus rots) were recovered from rotting cladodes in Metz Gorge, N.S.W. (locality #11). This locality did not have *Drosophila buzzatii* nor *Cactoblastis cactorum* present and should be considered an exception since in all other Australian localities *D. buzzatii* and *C. cactorum* were actively feeding on and vectoring yeasts from rot to rot. With this in mind, *P. opuntiae* appears to be the only common cladode yeast not found in the Caribbean communities. The origin of *P. opuntiae* is uncertain as it has not been recovered from sources in the Caribbean, Mexico, or the United States. It possibly originated in Argentina where *D. buzzatii* was presumed to originate when biological control was instituted in Australia. Yeasts that were common to both the Caribbean islands and Australia include *Pichia cactophila, Candida sonorensis,* and *Clavispora* sp., and to a lesser extent *P.* sp. "B" and *Cr. cereanus* complex reported by Barker et al. [3] in their earlier survey.

The yeast species found in fruit overlap to a limited degree with the yeasts of cladode communities. In Australia, *P. opuntiae,* which is common in cladodes, was absent from fruit, whereas *P. membranaefaciens* and *P. nakasei* were recovered from fruit but not from cladode rots. Furthermore, *Kloeckera apiculata,* a very common fruit yeast, appeared infrequently in cladodes and occurred only when fruit was present (fruits were present at locality #14 but were not sampled). The Caribbean results for fruit are less clear for two reasons: (1) some localities (#4, 6, 7) were not adequately sampled, and (2) fruit from locality #8 were green (unripe) fruits. This leaves one collection (#5) for comparison. Excluding those species which were less than 5% of the community, *P. norvegensis* is common in both substrates. Four species are either exclusive to cladodes (*Cr. cereanus* complex), or more common in cladodes (*P. cactophila, C. sonorensis,* and *Prototheca*). Three species are either exclusive to or are more frequent in fruits (*Pichia* sp. "B," *Candida krusei,* and *Kloeckera apis*). Two

Table 2. Relative frequency of yeast species isolated from cladodes of *Opuntia stricta* collected at localities (see Table 1) in the Caribbean and Australia

Yeast species / Locality #	Caribbean Islands								Australia					
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Pichia cactophila	.214	.316	.5	.273	.213	.329	.274	.264	.171	.2	0	0	.152	.087
P. norvegensis	0	.026	0	.152	.197	.013	.065	.019	0	0	0	0	0	0
P. opuntiae	0	0	0	0	0	0	0	0	.098	.467	0	.313	.065	0
P. sp. "B"[a]	0	0	0	0	0	.039	.048	.038	0	0	0	0	0	0
P. kluyveri	0	0	0	.061	0	0	0	.057	.073	0	.091	.063	.043	0
P. membranaefaciens	0	0	0	.030	0	0	0	0	0	0	0	0	0	0
P. amethionina var. "f"	0	.053	0	.030	.016	.105	.032	.038	0	0	0	0	0	0
P. mexicana	.143	0	0	0	0	.026	.016	0	0	0	0	0	0	0
Hansenula polymorpha	0	0	0	0	.016	0	0	0	0	0	0	0	0	0
H. californica	0	0	0	0	0	0	0	0	0	0	.182	0	0	0
Candida sonorensis	.036	.184	0	.212	.23	.263	.258	.264	.220	.133	.273	.313	.304	.391
C. boidinii	0	0	0	0	0	0	0	0	0	0	.364	0	.022	0
C. mucilagina	0	0	0	0	.016	0	0	0	0	0	0	0	.087	.043
C. guilliermondii	0	0	0	0	0	0	.016	0	0	0	0	0	0	0
C. catenulata	0	0	0	0	0	0	0	.019	0	0	0	0	0	0
C. krusei	0	0	0	0	.016	0	0	0	0	0	0	0	0	0
Clavispora sp.	.179	.211	.5	0	.033	.039	.081	.019	.390	.167	0	.25	.283	.435
Kloeckera apiculata	0	0	0	0	0	0	0	0	.049	0	0	.063	0	.043
K. apis	0	0	0	0	.016	0	0	0	0	0	0	0	0	0
Cryptococcus cereanus complex[a]	.107	.079	0	.242	.164	.092	.129	.151	0	0	0	0	0	0
Cr. luteolus	.036	0	0	0	.016	0	0	0	0	0	0	0	0	0
Cr. albidus	.036	0	0	0	0	0	0	.019	0	0	0	0	0	0
Cr. laurentii	.036	0	0	0	0	0	0	0	0	0	0	0	.022	0
Cr. infirmo-miniatus	0	0	0	0	0	0	0	0	0	0	.091	0	0	0
Rhodotorula graminis	0	0	0	0	0	0	0	.057	0	0	0	0	0	0
Prototheca zopfii	.214	.132	0	0	.066	.092	.081	.057	0	.033	0	0	.022	0
Total no. of isolates	28	38	2	33	61	76	62	53	41	30	11	16	46	23

[a] Even though these species were not recovered from Australian localities in this survey, they were isolated by Barker et al. [3, 4] in Australia in earlier investigations of *O. stricta* and other *Opuntia* cladodes

Table 3. Relative frequency of yeast species isolated from fruits of *Opuntia stricta* collected at localities (see Table 1) in the Caribbean and Australia

	Caribbean Islands					Australia		
Yeast species Locality #	4	5	6	7	8	9	11	12
Pichia cactophila	.2	.018	0	.167	.438	.045	0	0
P. norvegensis	0	.200	0	0	0	0	0	0
P. sp. "B"	.2	.109	0	.167	0	0	0	0
P. kluyveri	0	0	0	0	0	.182	0	0
P. nakasei	0	0	0	0	0	.045	0	.333
P. membranaefaciens	0	.055	0	.167	0	.182	0	0
P. amethionina var. *ameth.*	0	0	0	0	0	.045	0	0
P. amethionina var. "f"	.2	.036	0	.167	.063	0	0	0
Issatchenkia terricola	0	.036	0	0	0	0	0	0
Candida sonorensis	0	.036	0	0	.063	.045	0	.200
C. guilliermondii	0	.073	0	0	.188	0	0	0
C. parapsilosis	0	.018	0	0	0	0	0	0
C. stellata	0	.018	0	0	0	0	0	0
C. krusei	0	.127	0	0	0	0	0	.067
Clavispora sp.	.2	.055	1	0	.25	.091	0	0
Torulaspora delbrueckii	0	.018	0	0	0	0	0	0
Saccharomyces cerevisiae	0	.018	0	0	0	0	0	0
Kloeckera apiculata	0	0	0	0	0	.273	.688	.400
K. apis	0	.145	0	.167	0	0	0	0
Hanseniaspora sp.	0	0	0	.167	0	0	0	0
Candida curvata	.2	0	0	0	0	0	0	0
Cr. macerans	0	0	0	0	0	.045	0	0
Cr. hungaricus	0	0	0	0	0	.045	0	0
Cr. infirmo-miniatus	0	0	0	0	0	0	.313	0
Rhodotorula graminis	0	.018	0	0	0	0	0	0
Prototheca zopfii	0	.018	0	0	0	0	0	0
Total no. of isolates	5	55	2	6	16	22	16	15

less frequent species were found only or more often in fruits (*P. membranaefaciens* and *C. guilliermondii*), while *Clavispora* sp. was infrequent in both cladodes and fruits in the Caribbean. In Australia, *Clavispora* sp. was common in cladode rots, sometimes representing 50% or more of the total yeast present in an individual rot. In summary, Table 4 lists a qualitative comparison of the overlap between the common yeasts (>5%) in the two communities and two regions. This table was constructed by summing all isolates from each locality within a region by substrate, then normalizing by the total number of isolates. The data of Barker et al. [3, 4] were included. It should be noted that both *P.* sp. "B" and *Cr. cereanus* complex have been isolated from *O. stricta* cladodes by Barker et al. [3, 4] in Australia. Each of these species accounted for 4% of the isolates from *O. stricta* cladodes in their large survey.

Statistical Comparisons

Table 5 contains several levels of comparative information: (1) within and between substrates (cladodes and fruits), (2) within and between regions (Ca-

Table 4. Qualitative comparison of the overlap between the common yeasts (75% of the community) of the cladode and fruit communities

	Only in cladodes	Mainly in cladodes	Both	Mainly in fruit	Only in fruit
Caribbean	*Cr. cereanus* complex	*P. cactophila*	*P. amethionina* (var. "f")	*P. norvegensis*	
		C. sonorensis	*Clavispora*	*P.* sp. "B"	
		Prototheca		*P. membranae-faciens*	
				C. guillier-mondii	
				C. krusei	
				K. apis	
Australia	*P. opuntiae*	*P. cactophila*	*P. kluyveri*[a]	*K. apiculata*[a]	*P. nakasei*
		C. sonorensis	*P. amethio-nina* var. a.	*Cr. infirmo-miniatus*	
					P. membranae-faciens
		Clavispora sp.			

[a] Killer yeasts

ribbean and Australia), and (3) seasonal and between years. Our purpose here was to establish the extent of similarity between regions and ascertain what factors may be responsible for similarities and differences in the structure of the yeast communities.

Yeast Communities of Cactus Cladodes within Regions. Previous work [15] on *Opuntia phaeacantha, O. ficus-indica,* and *O. lindheimeri* showed that yeast communities collected from the same cactus species at the same time of the year but from separate localities have similarity coefficients ranging from about $r = 0.65$–0.95. Similarities between yeast communities from different *Opuntia* species were somewhat lower (approximately 0.60). The data of Barker et al. [3] were analyzed in a similar manner and the average similarity across seasons within one locality (0.78 ± 0.04; from all six season by season similarities in Table 5) was essentially the same as similarities measured across localities in one season (Australia, 1984; $r = 0.74 \pm 0.06$; Table 5). This indicates that spatial and temporal differences in community composition are of similar magnitudes for the introduced yeast communities found in Australia. Year-to-year similarity for the Caribbean communities was estimated at 0.71 ± 0.05 (Table 5) which is similar to within-year similarity of 0.79 and 0.73 ± 0.07 (Table 5) for the same region. The Australian data were also compared across years. This comparison showed an average similarity of 0.65 ± 0.06 (Table 5) for the years 1977 and 1984. It is worth noting that the 1984 data were collected in the winter and spring and showed an average similarity of 0.83 ± 0.06 (Table 5) with the 1977 spring data. In both regions we therefore see about the same amount of yeast community similarity from locality to locality, season to season, and year to year.

Table 5. Average similarity ($r \pm$ SE) among yeast communities of *O. stricta* cladodes and fruits collected from two regions (Caribbean and Australia)

	Caribbean cladodes		Australian cladodes							
			1977 (Seasons)				1977	1984	Caribbean	Australia
	May 1982	Nov 1983	Summer	Autumn	Winter	Spring	Year	Winter–spring	Fruit	Fruit
No. of localities	2	5	1	1	1	1	1	5	2	3
Caribbean cladodes										
May 1982	0.79	0.71	0.72	0.43	0.58	0.64	0.62	0.50	0.37	0.00
		±0.05	±0.16	±0.17	±0.16	±0.14	±0.18	±0.06	±0.20	±0.04
Nov 1983		0.73	0.81	0.65	0.79	0.65	0.77	0.49	0.37	0.03
		±0.07	±0.04	±0.09	±0.10	±0.03	±0.06	±0.03	±0.08	±0.02
Australian cladodes										
1977 Summer			1	0.78	0.86	0.78	0.92	0.57	0.41	0.05
								±0.07	±0.40	±0.06
Autumn				1	0.88	0.64	0.93	0.46	0.19	0.06
								±0.05	±0.25	±0.07
Winter					1	0.72	0.94	0.53	0.25	0.08
								±0.04	±0.29	±0.07
Spring						1	0.86	0.83	0.33	0.06
								±0.06	±0.31	±0.07
1977 Total							1	0.65	0.31	0.07
								±0.06	±0.33	±0.07
1984 Winter–spring								0.74	0.28	0.12
								±0.06	±0.08	±0.03
Caribbean fruit									0.12	0.004
										±0.04
Australia fruit										0.59
										±0.02

Yeast Communities of Cactus Cladodes between Regions. The similarity between yeast communities from the Caribbean region and Australia is significantly lower for each Caribbean collection of 1982 and 1983 compared to the 1984 Australian collection (0.50 ± 0.06 and 0.49 ± 0.03, respectively; Table 5) but relatively higher compared to the 1977 Australian collection (0.62 ± 0.18 and 0.77 ± 0.06, respectively; Table 5). The decline for the 1984 Australian collection mentioned above is primarily due to the low frequency of *P. cactophila* and *Cr. cereanus* complex coupled with an increase in the frequency of *Clavispora* sp. in that collection.

Yeast Communities of Cactus Fruit within and between Regions. The data for fruit were not obtained over seasons or years. To a large extent fruit are seasonal and this precluded collection during certain times of the year (i.e., May 1982; Caribbean region). The Caribbean data are limited to two localities with adequate sample size, one of which (Cayman Brac) was characterized by rotting young green fruit still on the plant. These rots were dark yellow to brown, most of which had not been opened by birds as is typical of ripe rotting fruit. Both Caribbean yeast communities from fruit showed little similarity with those observed in Australia ($r = 0.004 \pm 0.04$; Table 5). Within Australia the similarity among communities in fruit (0.59 ± 0.02; Table 5) was comparable, although lower, to within-region (1984) similarities among yeast communities in cladodes (0.74 ± 0.06; Table 5). It therefore appears that the yeast communities of fruits in the two regions are distinct and share little similarity.

Yeast Communities of Cactus Cladodes and Cactus Fruits. In both regions two localities yielded data for which yeast communities of fruits and cladodes could be compared within the same locality. This within locality across substrate comparison showed similarities (not shown in Table 5) of 0.33 (Great Inagua), 0.57 (Cayman Brac), 0.33 (O'Hara), and 0.24 (Trinkey). This illustrates the overlap between the substrates within a locality and in the case of the Cayman Brac collection, shows the effect of green fruit on the overlap.

Comparisons among localities within regions show that the Caribbean yeast communities of fruits and cladodes do have significant similarities (0.37 ± 0.20 and 0.37 ± 0.08; Table 5). By contrast, the Australian yeast communities show little across substrate similarities for any level of comparison (seasonal data, $r = 0.05$–0.08; two years, $r = 0.07$–0.12; Table 5). The Australian yeast communities of cactus fruit thus appear to be less linked to the yeast communities of cactus cladodes, as compared to those in the Caribbean.

Yeast Community Physiology. On the basis of physiology (Fig. 1), the yeast communities associated with cladodes in the Caribbean appeared very homogeneous, and showed much resemblance with the Australian community sampled at Breeza. The position of cladodes samples in Haiti in the dendrogram is attributed to their lower proportion of *Candida sonorensis,* their higher proportion of *P. mexicana,* and to the isolation from these samples of three representatives of *Cryptococcus.* The yeasts isolated from cladodes at the other five Australian sites (other than Breeza) formed a separate cluster, characterized by a higher-than-expected utilization of xylose, xylitol, and ribitol, and to some

degree, salicin. In most of the remaining communities studied, these compounds were poorly utilized, or their utilization was not unusual. This is obviously linked with the frequent isolation, in Australian cladodes, of *P. opuntiae* (only salicin), *C. boidinii,* or *C. mucilagina.* It is doubtful that this trend could represent a difference in cactus chemistry. In may, however, be linked to differences in neighboring plants which could serve as inoculation sources.

The fruit yeast communities appeared less structured physiologically, as indicated by their sporadic appearance in several clusters (Fig. 1). In general, they differed from typical cladode yeasts in their decreased use of lactic acid, ethanol, and methanol, and to a certain extent, in the maximum growth temperatures. The yeasts found in cladodes tended to exhibit more positive responses than expected for these five characteristics. Such patterns were much less pronounced among fruit yeasts. The outlying position of the fruit community from Metz Gorge is simply a reflection of its very unusual yeast composition (Table 3). The high frequency of *K. apiculata* maintained the generally low resource utilization characteristic typical of cactus yeast florae [10], but a large proportion of *Cr. infirmo-miniatus* in this community gave it a unique profile with respect to traits that are specific to that yeast and to basidiomycetous yeasts in general.

Discussion

It is clear from the analysis of the data (Table 5) that yeast communities of cladodes and fruits are distinct, and yet some overlap does occur. Earlier analyses of *Drosophila* habitats had indicated that *Opuntia* cladode yeast communities were more similar to yeast communities of columnar cacti [14] than to those from fruits. However, the fruits studied were not cactus fruits and could have been distinct for other reasons. It is now apparent that the cactus fruit yeast community, even when the fruits are physically next to the cladode habitat, is quantitatively and qualitatively different from the cladode yeast community. The reasons for the difference are not entirely clear, although it is presumably based on the chemical nature of the two substrates. All of the yeasts recovered from the two substrates can grow on either fruit or cladode tissue when inoculated on them. The community difference is thus not a chemical inhibitor present in the different tissues. Species-species interactions may be important in that fruit yeasts tend to carry "killer factors" more often than cladode yeasts. All of the isolates (Tables 2–3) were tested for killer activity against a standard sensitive strain (*C. glabrata* Y55), and the following species tested positive for killer activity: (1) *P. kluyveri,* all strains; (2) *K. apiculata,* most strains; (3) *P. opuntiae,* all strains from locality #13 (Hemmant) but not elsewhere; (4) *H. californica,* from locality #11.

It is therefore possible that community structure of the fruit habitat is partially determined by killer activity of the resident yeasts. This is feasible because the low pH of fruit (pH = 3–4) coincides with the optimum pH for killer toxin [7]. However, we have not systematically tested resistance/sensitivity patterns of yeasts restricted to the cladode habitat (i.e., *Cr. cereanus* complex and *P. opuntiae*) to the common killer fruit yeast *K. apiculata* and to *P. kluyveri* which

lives in both habitats. Furthermore, the pH of cladode rots rapidly rises from about 4 to 8–9 where the killer toxins are ineffective.

It is not surprising that the cladode communities of the two regions are similar, especially because of the introduction into Australia of cladode microbiota in such large amounts during the biological control program. The differences between the regions for yeast communities of fruits, however, could be based on the extrinsic microbiota, i.e., yeast community of other fruits and their yeast communities resident in Australia. Presumably most, if not all, of the rotting materials and "cut portions of prickly pear" [8] shipped to Australia for the biological control program were cladodes and not fruit.

In addition, other noncactus breeding drosophilids as well as bees, wasps, and other insects are feeding and/or breeding in the *O. stricta* fruit, thus providing a vectoring passage for extrinsic microorganisms. Even birds feeding on cactus fruit could introduce yeasts from other fruit where present. This is not the case for the cladodes because *Drosophila buzzatii* and *D. aldrichi*, introduced during biological control efforts, are both restricted to breeding in cladodes, and apparently have exclusive use among the drosophilids of this habitat [1]. In this sense the cactus-microorganism-*Drosophila* system is closed with respect to the cladodes but uncoupled and open with respect to the fruits. This supposition is reinforced by the seasonal nature of fruit production, which causes a hiatus every year in the continuation of rot-to-rot community dynamics.

In terms of overall physiological specificity, the cladode yeast communities described in this study were rather typical of other cactophilic yeast communities. The analysis of 679 cactophilic yeast strains (or isolates) by the same method [10] had shown clearly that they differ from random yeast assemblages by their very high degree of nutritional specialization, giving higher than expected responses for growth on primary alcohols and organic acids, and of course, for growth at 37°C. At variance with the results found previously [10], significantly high fermentative abilities prevailed in almost half the collections analyzed here. Interestingly, most collections exhibiting this special feature were from Australian sites.

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