

Diversity in the breadfruit complex (*Artocarpus*, Moraceae): genetic characterization of critical germplasm

Nyree Zerega · Tyr Wiesner-Hanks · Diane Ragone ·
Brian Irish · Brian Scheffler · Sheron Simpson · Francis Zee

Received: 29 August 2014 / Revised: 7 October 2014 / Accepted: 2 December 2014
© Springer-Verlag Berlin Heidelberg 2015

Abstract Breadfruit (*Artocarpus altilis*, Moraceae) is a traditional staple crop in Oceania and has been introduced throughout the tropics. This study examines important germplasm collections of breadfruit and its closest wild relatives and aims to (1) characterize genetic diversity, including identification of unknown and duplicate accessions, (2) evaluate genetic structure and hybridization within the breadfruit complex, and (3) compare utility of microsatellite markers to previously reported amplified fragment length polymorphism (AFLP) and isozyme markers in differentiating among cultivars. Data for 19 microsatellite loci were collected for 349 individuals

(representing 255 accessions) including breadfruit (*A. altilis*), two wild relatives (*Artocarpus camansi* and *Artocarpus mariannensis*), and putative hybrids (*A. altilis* × *A. mariannensis*). Accessions were of mixed ploidy and regional origin, but predominantly from Oceania. Microsatellite loci collectively had a polymorphic information content (PIC) of 0.627 and distinguished 197 unique genotypes sorted into 129 different lineages, but a single genotype accounts for 49 % of all triploid breadfruit examined. Triploid hybrids and diploid *A. altilis* exhibited the highest levels of diversity as measured by allele number and gene diversity. Most accessions (75 %) of unknown origin matched either a known genotype or lineage group in the collection. Putative hybrids all had genetic contributions from *A. mariannensis* but ranged in the level of genetic contribution from *A. altilis*. Microsatellite markers were found to be more informative than isozyme markers and slightly less informative, with regard to accession discrimination, than AFLP markers. This set of microsatellite markers and the dataset presented here will be valuable for breadfruit germplasm management and conservation.

Communicated by W. Ratnam

Electronic supplementary material The online version of this article (doi:10.1007/s11295-014-0824-z) contains supplementary material, which is available to authorized users.

N. Zerega (✉) · T. Wiesner-Hanks
Plant Biology and Conservation, Northwestern University, Evanston,
IL 60208, USA
e-mail: nzerega@chicagobotanic.org

N. Zerega
Department of Plant Science, Chicago Botanic Garden, Glencoe,
IL 60022, USA

D. Ragone
Breadfruit Institute, National Tropical Botanical Garden, Kalaheo,
HI 96741, USA

B. Irish
Tropical Agriculture Research Station, USDA-ARS,
Mayagüez 00680, Puerto Rico

B. Scheffler · S. Simpson
Genomics and Bioinformatics Research Unit, USDA-ARS,
Stoneville, MS 38776-0350, USA

F. Zee
Pacific Basin Agricultural Research Center (PBARC), USDA-ARS,
Hilo, HI 96720, USA

Keywords Breadfruit · *Artocarpus* · Germplasm conservation management · Microsatellites · Plant genetic resources · Underutilized crops

Introduction

Breadfruit (*Artocarpus altilis* (Parkinson) Fosberg, Moraceae) is a traditional staple in Oceania and has many uses ranging from construction, medicine, and animal feed to insect repellent (Ragone 1997; Jones et al. 2011). However, it is principally grown as a starch food crop and is an important component of agroforestry systems. Breadfruit has been recognized as a crop with great potential for increasing food security and

alleviating malnutrition in Oceania, the Caribbean, tropical Africa, and beyond (Wootton and Tumaalii 1984; Morton 1990; Adebowale et al. 2005; Omubuwojo 2007; Roberts-Nkrumah 2007; Taylor et al. 2009; Jones et al. 2011). It produces large starchy compound fruits with high yields and high levels of minerals and provitamin A carotenoids as compared to other important staples like wheat, maize, and rice (Jones et al. 2011, 2013). Although most cultivars produce fruit from August to January, growing a set of cultivars with diverse fruit-bearing seasons could allow for year-round harvest of nutrient-rich fruit (Fownes and Raynor 1991; Jones et al. 2010, 2013). Since Europeans first encountered breadfruit in the Pacific nearly 400 years ago (Markham 1904), a small number of cultivars have been introduced to tropical regions throughout the tropics, including the Caribbean (Powell 1977; Leakey 1977; Roberts-Nkrumah 2007), Africa (Omubuwojo 2007), and India (Ragone 1997). The genetic diversity and importance of breadfruit, however, remain greatest in Oceania, where breadfruit was domesticated (Zerega et al. 2004, 2005, 2006).

Breadfruit's wild relatives have been identified as *Artocarpus camansi* Blanco and *Artocarpus mariannensis* Trécul, and hybrids also exist (Fosberg 1960; Zerega et al. 2004, 2005). Over millennia, Pacific Islanders have selected and named hundreds of traditional cultivars based on fruiting season, fruit shape, color and texture of the flesh and skin, presence or absence of seeds, flavor, cooking and storage qualities, leaf shape, and horticultural needs (Wilder 1928; Ragone 1997). Cultivars include vegetatively propagated seedless triploids, vegetatively or seed-propagated fertile diploids, and diploid and triploid hybrids (Ragone 2001, 2007; Zerega et al. 2004). Over 2000 names have been collected for breadfruit cultivars in Oceania, where breadfruit was domesticated (Ragone 1991). Because names are typically based on morphological traits of the tree and fruit, which may be environmentally influenced across islands (Ragone and Wiseman 2007), and because the same name may be used for different types or different names may be used for the same type, genetic characterization of cultivars is important.

To help conserve, study, and improve breadfruit, many germplasm collections have been assembled over the last several decades throughout the tropics (Ragone 1997). Most cultivars are seedless, and even when seeds are present, they are recalcitrant and cannot be dried, frozen, or stored, so collections must be maintained as living trees in field genebanks. This is a time-consuming and expensive task, and for this reason, many collections are no longer being maintained (Ragone 1997). Understanding the genetic diversity of collections is critical for informed germplasm management and prioritizing conservation efforts and is an important step in the increased utilization of this crop.

The largest and most extensive collection of breadfruit is housed at the Breadfruit Institute at the National Tropical Botanical Garden (NTBG). It contains breadfruit, hybrids, and wild relatives from throughout Oceania and beyond (Table 1, see also Online Resource 1 and [www.http://ntbg.org/breadfruit](http://www.ntbg.org/breadfruit)). USDA-ARS National Plant Germplasm System (NPGS) repositories (Pacific Basin Agricultural Research Center (PBARC) in Hilo, HI, and Tropical Agriculture Research Station (TARS) in Mayagüez, Puerto Rico) include duplicates from NTBG as well as some additional unique accessions (http://www.ars-grin.gov/npgs/acc/acc_queries.html) (Table 1). This study analyzed the accessions from these important breadfruit collections. Previous genetic diversity studies conducted on a subset of these collections used isozymes (Ragone 1991) and amplified fragment length polymorphism (AFLPs) (Zerega et al. 2004, 2005, 2006). The present study characterized 349 breadfruit individuals and wild congeners from the NTBG and NPGS collections using 19 microsatellite loci (Witherup et al. 2013) in order to (1) characterize genetic diversity, including identification of unknown and replicate accessions, (2) evaluate genetic structure and hybridization within the breadfruit complex, and (3) compare microsatellite utility to AFLP and isozyme markers in assessing breadfruit diversity and differentiating among cultivars.

Methods and materials

Plant materials

Leaf tissue samples from 349 individuals (representing 255 accessions) were collected on silica for 229 *A. altilis*, 70 *A. altilis* × *A. mariannensis* hybrids, 36 *A. camansi*, and 14 *A. mariannensis* specimens. Samples came from the following sites: NTBG (Kahanu Garden, Maui, and the McBryde, Allerton, and Limahuli Gardens in Kauai, HI), USDA-ARS, TARS, and USDA-ARS, PBARC (Table 1). Provenance localities were predominantly in Oceania (Fig. 1, Table 1). Taxon name, cultivar name, and additional provenance information are also publicly available for most of the accessions through NTBG (<http://ntbg.org/breadfruit/database>) and the ARS's Germplasm Resources Information Network (GRIN) databases (http://www.ars-grin.gov/npgs/acc/acc_queries.html). An accession number was frequently represented by only one tree; however, in some cases, multiple individuals were represented from the same base accession number (the six-digit number preceding the decimal point). If individuals shared the same base accession number, it indicated that they were either vegetatively propagated from the same parent tree or that they were grown from seed from the same mother tree.

Table 1 Accession data for specimens used in study

Taxon	Accession number	Lin	Gn	Pl	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa	TARS 6732	1	1	3n	TARS	Mayaguez	Plot 22	Unk	Caribbean	Barbados		H	4
Aa	890472.001	1	1	3n	NTBG	Kahanu	W4	Enua	E Poly	Cook Islands		H	4
Aa	890472.002	1	1	3n	NTBG	Kahanu	U4	Enua	E Poly	Cook Islands		H	4
Aa	900256.001	1	1	3n	NTBG	Kahanu	T3	Enua	E Poly	Cook Islands		H	4
Aa	HART 56	1	1	3n	HART	Hilo		Kahaluu	E Poly	Hawaii		H	4
Aa	070101.001	1	1	3n	NTBG	Kahanu	O10	Ulu	E Poly	Hawaii		H	4
Aa	070882.002	1	1	3n	NTBG	McBryde	AU2	Ulu	E Poly	Hawaii		H	4
Aa	090739.001	1	1	3n	NTBG	Limahuli	LMI	Ulu	E Poly	Hawaii		H	4
Aa	100346.001	1	1	3n	NTBG	Allerton	AG4	Ulu	E Poly	Hawaii		H	4
Aa	100347.001	1	1	3n	NTBG	Allerton	AG1	Ulu	E Poly	Hawaii		H	4
Aa	100348.001	1	1	3n	NTBG	Allerton	AG2	Ulu	E Poly	Hawaii		H	4
Aa	100349.001	1	1	3n	NTBG	Allerton	AG3	Ulu	E Poly	Hawaii		H	4
Aa	970274.001	1	1	3n	NTBG	McBryde	CG1	Ulu	E Poly	Hawaii		H	4
Aa	900240.001	1	1	3n	NTBG	Kahanu	M8	Mei kauhiva	E Poly	Marquesas		H	4
Aa	900238.001	1	1	3n	NTBG	Kahanu	F5	Mei kiahahi	E Poly	Marquesas		H	4
Aa	890462.001	1	1	3n	NTBG	Kahanu	U2	Mei puau	E Poly	Marquesas		H	4
Aa	HART 23 (900237)	1	1	3n	HART	Hilo		Mei puou	E Poly	Marquesas		H	4
Aa	780332.001	1	1	3n	NTBG	Kahanu	33	Aarue	E Poly	Society Islands		H	4
Aa	040051.001	1	1	3n	NTBG	McBryde	McB8	Afara	E Poly	Society Islands		H	4
Aa	780325.001	1	1	3n	NTBG	Kahanu	32	Afara	E Poly	Society Islands		H	4
Aa	910267.001	1	1	3n	NTBG	Kahanu	V8	Afara	E Poly	Society Islands		H	4
Aa	780333.001	1	1	3n	NTBG	Kahanu	30	Ahani	E Poly	Society Islands		H	4
Aa	900249.002	1	1	3n	NTBG	Kahanu	I8	Anahonaho	E Poly	Society Islands		H	4
Aa	890157.001	1	1	3n	NTBG	Kahanu	42	Apu	E Poly	Society Islands		H	4
Aa	HART 29 (890157)	1	1	3n	HART	Hilo		Apu	E Poly	Society Islands		H	4
Aa	900232.001	1	1	3n	NTBG	Kahanu	A7	Atu	E Poly	Society Islands		H	4
Aa	890147.001	1	1	3n	NTBG	Kahanu	21	Aue	E Poly	Society Islands		H	4
Aa	890147.002	1	1	3n	NTBG	Kahanu	L7	Aue	E Poly	Society Islands		H	4
Aa	780335.001	1	1	3n	NTBG	Kahanu	23	Aumee	E Poly	Society Islands		H	4
Aa	780330.001	1	1	3n	NTBG	Kahanu	56	Fafai	E Poly	Society Islands		H	4
Aa	890154.001	1	1	3n	NTBG	Kahanu	Y1	Hamoia	E Poly	Society Islands		H	4
Aa	HART 32 (890154)	1	1	3n	HART	Hilo		Hamoia	E Poly	Society Islands		H	4
Aa	780291.001	1	1	3n	NTBG	Kahanu	47	Havana pataitai	E Poly	Society Islands		H	4
Aa	900245.001	1	1	3n	NTBG	Kahanu	G8	Huero	E Poly	Society Islands		H	4
Aa	890150.001	1	1	3n	NTBG	Kahanu	Y8	Ioio	E Poly	Society Islands		H	4
Aa	HART 42 (890150)	1	1	3n	HART	Hilo		Ioio	E Poly	Society Islands		H	4
Aa	890149.001	1	1	3n	NTBG	Kahanu	46	Mamaha	E Poly	Society Islands		H	4

Table 1 (continued)

Taxon	Accession number	Lin	Gn	Pl	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa	890464.001	1	1	3n	NTBG	Kahanu	P7	Ouo	E Poly	Society Islands		H	4
Aa	780328.001	1	1	3n	NTBG	Kahanu	14	Puaa	E Poly	Society Islands		H	4
Aa	890460.001	1	1	3n	NTBG	Kahanu	T6	Puaa	E Poly	Society Islands		H	4
Aa	900244.001	1	1	3n	NTBG	Kahanu	S8	Puaa	E Poly	Society Islands		H	4
Aa	HART 18 (900244)	1	1	3n	HART	Hilo		Puaa	E Poly	Society Islands		H	4
Aa	HART 16	1	1	3n	HART	Hilo		Puero	E Poly	Society Islands		H	4
Aa	890152.001	1	1	3n	NTBG	Kahanu	W1	Puurea	E Poly	Society Islands		H	4
Aa	890152.002	1	1	3n	NTBG	Kahanu	S7	Puurea	E Poly	Society Islands		H	4
Aa	780329.001	1	1	3n	NTBG	Kahanu	29	Rare	E Poly	Society Islands		H	4
Aa	040518.001	1	1	3n	NTBG	McBryde	McB14	Rare autia	E Poly	Society Islands		H	4
Aa	780345.001	1	1	3n	NTBG	McBryde	BB1	Raumae	E Poly	Society Islands		H	4
Aa	780338.001	1	1	3n	NTBG	Kahanu	51	Tapehaa	E Poly	Society Islands		H	4
Aa	100370.001	1	1	3n	NTBG	Limahuli	LM3	Tuutou	E Poly	Society Islands		H	4
Aa	900246.001	1	1	3n	NTBG	Kahanu	H7	Tuutou auena	E Poly	Society Islands		H	4
Aa	900247.001	1	1	3n	NTBG	Kahanu	I6	Tuutou ooa	E Poly	Society Islands		H	4
Aa	890186.001	1	1	3n	NTBG	Kahanu	Z2	Tuutou taatoc	E Poly	Society Islands		H	4
Aa	890186.002	1	1	3n	NTBG	Kahanu	B7	Tuutou taatoc	E Poly	Society Islands		H	4
Aa	890165.001	1	1	3n	NTBG	Kahanu	Z3	Mei chon	Micro	Chuuk, FSM		H	4
Aa	HART 51 (890165)	1	1	3n	HART	Hilo		Mei chon	Micro	Chuuk, FSM		H	4
Aa	890162.001	1	1	3n	NTBG	Kahanu	X4	Lemae	Micro	Mariana Islands		H	4
Aa	890159.001	1	1	3n	NTBG	Kahanu	X5	Meriaur	Micro	Palau		H	4
Aa	HART 35 (890159)	1	1	3n	HART	Hilo		Meriaur	Micro	Palau		H	4
Aa	890478.001	1	1	3n	NTBG	Kahanu	R5	Meikalak	Micro	Pohnpei, FSM		H	4
Aa	890478.002	1	1	3n	NTBG	Kahanu	O4	Meikalak	Micro	Pohnpei, FSM		H	4
Aa	790497.002	1	1	3n	NTBG	Kahanu	49	Meinuwe	Micro	Pohnpei, FSM		H	4
Aa	890167.001	1	1	3n	NTBG	Kahanu	41	Meisaip	Micro	Pohnpei, FSM		H	4
Aa	890167.002	1	1	3n	NTBG	Kahanu	R6	Meisaip	Micro	Pohnpei, FSM		H	4
Aa	890479.001	1	1	3n	NTBG	Kahanu	Q7	Meisei	Micro	Pohnpei, FSM		H	4
Aa	890479.002	1	1	3n	NTBG	Kahanu	O7	Meisei	Micro	Pohnpei, FSM		H	4
Aa	030033.001	1	1	3n	NTBG	McBryde	McB16	Meitehid	Micro	Pohnpei, FSM		H	4
Aa	910273.002	1	1	3n	NTBG	Kahanu	G9	Meitehid	Micro	Pohnpei, FSM		H	4
Aa	910271.001	1	1	3n	NTBG	Kahanu	D9	Meiuhpw	Micro	Pohnpei, FSM		H	4
Aa (Aa × Am)	890480.003	1	1	3n	NTBG	Kahanu	R8	Lipet 2	Micro	Pohnpei, FSM		H	4
Aa	810290.002	1	1	3n	NTBG	Kahanu	45	White	Micro	Pohnpei, FSM		H	4
Aa	Unknown	1	1	3n	TARS	Mayaguez	Plot 18	Unk	Seychelles	Seychelles		H	4
Aa	910290.001	1	1	3n	NTBG	Kahanu	L10	Unk 04	Unk (Epoly or Micro)	Unknown		H	4
Aa	910287.001	1	1	3n	NTBG	Kahanu	R9	Unk 09	Unk (Epoly or Micro)	Unknown		H	4

Table 1 (continued)

Taxon	Accession number	Lin	Gn	Pl	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa	910286.001	1	1	3n	NTBG	Kahanu	T8	Unk 11	Unk (Epoly or Micro)	Unknown		H	4
Aa	800269.001	1	3	3n	NTBG	Kahanu	36	Mahani	E Poly	Society Islands		H	4
Aa	TARS 17990	1	3	3n	TARS	Mayaguez	Plot 5	Samoa	W Poly	Samoa		H	4
Aa	900242.001	1	5	3n	NTBG	Kahanu	B8	Mei kopumoko	E Poly	Marquesas		H	4
Aa	HART 28 (900242)	1	5	3n	HART	Hilo		Mei kopumoko	E Poly	Marquesas		H	4
Aa	900249.001	1	5	3n	NTBG	Kahanu	I7	Anahonaho	E Poly	Society Islands		H	4
Aa	030028.001	1	5	3n	NTBG	McBryde	McB13	White	Seychelles	Seychelles		H	4
Aa	810290.001	1	5	3n	NTBG	Kahanu	43	White	Seychelles	Seychelles		H	4
Aa	810290.003	1	5	3n	NTBG	Kahanu	48	White	Seychelles	Seychelles		H	4
Aa	810290.004	1	5	3n	NTBG	McBryde	BB3	White	Seychelles	Seychelles		H	4
Aa	810289.001	1	5	3n	NTBG	McBryde	BB2	Yellow	Seychelles	Seychelles		H	4
Aa	810289.002	1	5	3n	NTBG	Kahanu	38	Yellow	Seychelles	Seychelles		H	4
Aa	890158.001	1	6	3n	NTBG	Kahanu	Z6	Apuapua	E Poly	Society Islands		H	4
Aa	890158.002	1	6	3n	NTBG	Kahanu	H8	Apuapua	E Poly	Society Islands		H	4
Aa	HART 34 (890158)	1	6	3n	HART	Hilo		Apuapua	E Poly	Society Islands		H	4
Aa	900243.001	1	7	3n	NTBG	Kahanu	D7	Araahaari	E Poly	Society Islands		H	4
Aa	780330.002	1	9	3n	NTBG	Kahanu	40	Fafai	E Poly	Society Islands		H	4
Aa	100369.001	1	9	3n	NTBG	Limahuli	LM2	Fafai (unk)	E Poly	Society Islands		H	4
Aa	890162.002	1	14	3n	NTBG	Kahanu	S4	Lemae	Micro	Mariana Islands		H	4
Aa	HART 38 (890162)	1	14	3n	HART	Hilo		Lemae	Micro	Mariana Islands		H	4
Aa	890459.001	1	15	3n	NTBG	Kahanu	R7	Maire	E Poly	Society Islands		H	4
Aa	HART 44 (890459)	1	15	3n	HART	Hilo		Maire	E Poly	Society Islands		H	4
Aa	HART 54 (890459)	1	15	3n	HART	Hilo		Maire	E Poly	Society Islands		H	4
Aa	900267.001	1	15	3n	NTBG	Kahanu	N8	Maire (unk 05)	E Poly (unk)	Society Islands (unk)		H	4
Aa	890459.002	1	16	3n	NTBG	Kahanu	J7	Maire	E Poly	Society Islands		H	4
Aa	900262.001	1	17	3n	NTBG	Kahanu	M6	Manua	W Poly	Samoa		H	4
Aa	900241.001	1	18	3n	NTBG	Kahanu	F6	Mei aueka	E Poly	Marquesas		H	4
Aa	900239.001	1	19	3n	NTBG	Kahanu	T5	Mei maoi	E Poly	Marquesas		H	4
Aa	HART 11 (900239)	1	19	3n	HART	Hilo		Mei maoi	E Poly	Marquesas		H	4
Aa	900237.001	1	20	3n	NTBG	Kahanu	B6	Mei puou	E Poly	Marquesas		H	4
Aa	080439.001	1	23	3n	NTBG	Kahanu	Heiau1	Ulu	E Poly	Hawaii		H	4
Aa	080858.001	1	23	3n	NTBG	Kahanu	CG3	Ulu	E Poly	Hawaii		H	4
Aa	080863.001	1	23	3n	NTBG	Kahanu	CG8	Ulu	E Poly	Hawaii		H	4
Aa	080881.001	1	23	3n	NTBG	Kahanu	CG11	Ulu	E Poly	Hawaii		H	4
Aa	070132.001	1	23	3n	NTBG	Kahanu	CG1	Ulu	E Poly	Hawaii		H	4
Aa	080440.001	1	23	3n	NTBG	Kahanu	Heiau2	Ulu	E Poly	Hawaii		H	4
Aa	890159.002	1	23	3n	NTBG	Kahanu	V4	Meriaur	Micro	Palau		H	4

Table 1 (continued)

Taxon	Accession number	Lin	Gn	PI	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa	000534.001	1	23	3n	NTBG	Kahanu	GG4	Meikalak en meikuet	Micro	Pohnpei, FSM		H	4
Aa	790493.001	1	25	3n	NTBG	Kahanu	4	Meitehid	Micro	Pohnpei, FSM		H	4
Aa	HART 57	1	27	3n	HART	Hilo		Poahomo Prickly	E Poly			H	4
Aa	030035.001	1	32	3n	NTBG	McBryde	McB7	Otea	E Poly	Society Islands		H	4
Aa	780327.001	1	32	3n	NTBG	Kahanu	39	Otea	E Poly	Society Islands		H	4
Aa	910266.001	1	36	3n	NTBG	Kahanu	H9	Piipia	E Poly	Society Islands		H	3
Aa	30037.001	1	37	3n	NTBG	McBryde	McB10	Porohiti	E Poly	Society Islands		H	4
Aa	790486.001	1	38	3n	NTBG	Kahanu	20	Roithaa	E Poly	Society Islands		H	4
Aa	890465.001	1	39	3n	NTBG	Kahanu	V7	Teahimatoa	E Poly	Society Islands		H	4
Aa	HART 40 (890465)	1	39	3n	HART	Hilo		Teahimatoa	E Poly	Society Islands		H	4
Aa	070882.001	1	42	3n	NTBG	McBryde	AU1	Ulu	E Poly	Hawaii		H	4
Aa	080859.001	1	43	3n	NTBG	Kahanu	CG4	Ulu	E Poly	Hawaii		H	4
Aa	080864.001	1	44	3n	NTBG	Kahanu	CG9	Ulu	E Poly	Hawaii		H	4
Aa	080864.002	1	44	3n	NTBG	Kahanu	CG10	Ulu	E Poly	Hawaii		H	4
Aa	900268.001	1	46	3n	NTBG	Kahanu	E4	Unk 01	Unk	Unknown		H	4
Aa	900225.001	1	48	3n	NTBG	Kahanu	S5	Unk 10	Unk	Unknown		H	4
Aa	890148.001	1	50	3n	NTBG	Kahanu	Y6	Mei puou (unk 14)	E Poly	Marquesas (Society)		H	4
Aa	070883.001	1	52	3n	NTBG	McBryde	Pump6	Unk	E Poly	Unknown		H	4
Aa	910265.001	1	69	3n	NTBG	Kahanu	V9	Rotuma	E Poly	Society Islands		H	4
Aa	Unknown	2	2	3n	TARS	Mayaguez	Plot 11	Unk	Epoly (unk)	Society Islands (unk)		G	3
Aa	890151.001	2	10	3n	NTBG	Kahanu	X7	Fafai	E Poly	Society Islands		G	3
Aa	880690.001	2	13	3n	NTBG	Kahanu	P8	Kea	W Poly	Tonga		G	3
Aa	890153.002	2	33	3n	NTBG	Kahanu	2	Paea	E Poly	Cook Islands		G	3
Aa	890463.001	2	34	3n	NTBG	Kahanu	V3	Patara	E Poly	Society Islands		G	3
Aa	890463.002	2	34	3n	NTBG	Kahanu	G6	Patara	E Poly	Society Islands		G	3
Aa	HART 33 (890463)	2	34	3n	HART	Hilo		Patara	E Poly	Society Islands		G	3
Aa	790485.001	2	34	3n	NTBG	Kahanu	16	Puupuu	E Poly	Society Islands		G	3
Aa	790491.001	2	34	3n	NTBG	Kahanu	13	Tuutou	E Poly	Society Islands		G	3
Aa	900236.001	3	4	3n	NTBG	Kahanu	D6	Abareba	Mela	Solomon Islands		C	3
Aa	HART 45 (890186)	4	8	3n	HART	Hilo		Tuutou	E Poly	Society Islands		F	4/1
Aa	HART 30 (900245)	6	12	2n	HART	Hilo		Huero	E Poly	Society Islands		E	3
Aa	900266.001	7	21	3n	NTBG	Kahanu	M5	Meiarephe	Micro	Pohnpei, FSM		D	4
Aa	900266.002	7	21	3n	NTBG	Kahanu	E5	Meiarephe	Micro	Pohnpei, FSM		D	4
Aa	000531.001	8	24	3n	NTBG	Kahanu	58	Meipwet	Micro	Pohnpei, FSM		D	4
Aa	030042.001	11	40	3n	NTBG	McBryde	McB11	Tonono	E Poly	Society Islands		F	4
Aa	790488.001	11	41	3n	NTBG	Kahanu	12	Tonono	E Poly	Society Islands		F	4
Aa	30044.001	12	45	3n	NTBG	Off site	Off site	Ulu tala	W Poly	Samoa		E	3

Table 1 (continued)

Taxon	Accession number	Lin	Gn	Pl	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa	770524.001	12	45	3n	NTBG	Kahanu	54	Ulu tala	W Poly	Samoa		E	3
Aa	910288.001	13	47	3n	NTBG	Kahanu	Q8	Uto vula (unk 08)	Mela (unk)	Fiji (unk)		F	2
Aa	890476.002	13	51	3n	NTBG	Kahanu	S6	Uto vula	Mela	Fiji		F	2
Aa	020354.001	25	76	2n	NTBG	Kahanu	KM1	Aveloloa	W Poly	Samoa		G	3
Aa	910278.001	26	77	2n	NTBG	Kahanu	M9	Forari	Mela	Vanuatu		E	3
Aa	910279.001	26	103	2n	NTBG	Kahanu	E9	Siviri 2	Mela	Vanuatu		E	3
Aa	910279.002	26	103	2n	NTBG	Kahanu	C8	Siviri 2	Mela	Vanuatu		E	3
Aa	890470.002	27	78	2n	NTBG	Kahanu	N5	Furau	Mela	Rotuma		G	2
Aa	040063.001	27	78	2n	NTBG	McBryde	McB15	Ulu fti	Mela	Rotuma		G	2
Aa	890258.001	27	78	2n	NTBG	Kahanu	35	Ulu fti	Mela	Rotuma		G	2
Aa	890458.001	27	78	2n	NTBG	Kahanu	T4	Ulu fti	Mela	Rotuma		G	2
Aa	890458.002	27	78	2n	NTBG	Kahanu	Q4	Ulu fti	Mela	Rotuma		G	2
Aa	900368.001	27	78	2n	NTBG	Kahanu	C4	Ulu fti	Mela	Rotuma		G	2
Aa	970236.001	27	78	2n	NTBG	Kahanu	AA4	Ulu fti	Mela	Rotuma		G	2
Aa	890469.002	27	78	2n	NTBG	Kahanu	S3	Kukumu tasi	Mela	Solomon Islands		G	2
Aa	020353.001	27	78	2n	NTBG	Kahanu	KM5	Unk 18	W Poly	Samoa		G	2
Aa	020353.002	27	78	2n	NTBG	Kahanu	KM4	Unk 18	W Poly	Samoa		G	2
Aa	900260.001	27	110	2n	NTBG	Kahanu	K7	Ulu fti, Samoan	Mela	Rotuma		G	2
Aa	900260.002	27	110	2n	NTBG	Kahanu	K5	Ulu fti, Samoan	Mela	Rotuma		G	2
Aa	890470.001	28	79	2n	NTBG	Kahanu	V6	Furau	Mela	Rotuma		G	2
Aa	890469.001	28	79	2n	NTBG	Kahanu	V1	Kukumu tasi	Mela	Solomon Islands		G	2
Aa	HART 26	29	80	2n	HART	Hilo		G. Wilder-Brash	E Poly			E	3
Aa	900248.001	30	81	2n	NTBG	Kahanu	G5	Huero ninamu	E Poly	Society Islands		E	3
Aa	890461.001	30	89	2n	NTBG	Kahanu	W2	Mei kakano	E Poly	Marquesas		E	3
Aa	890461.002	30	89	2n	NTBG	Kahanu	C7	Mei kakano	E Poly	Marquesas		E	3
Aa	HART 21 (890461)	30	89	2n	HART	Hilo		Mei kakano	E Poly	Marquesas		E	3
Aa	890457.001	31	82	2n	NTBG	Kahanu	W5	Karawa	Mela	Rotuma		G	2
Aa	020356.001	31	82	2n	NTBG	Kahanu	KM7	Ulu fti	Mela	Rotuma		G	2
Aa	900265.001	32	83	2n	NTBG	Kahanu	C5	Karawa	Mela	Fiji		F	2
Aa	900224.001	32	83	2n	NTBG	Kahanu	U7	Karawa (unk 12)	Mela (unk)	Fiji (unk)		F	2
Aa	900234.001	32	102	2n	NTBG	Kahanu	N6	Samoan 1	Mela	Fiji		F	2
Aa	070659.021	33	84	2n	NTBG	McBryde	CHC1	Ma'afala	W Poly	Samoa		E	3
Aa	070659.022	33	84	2n	NTBG	McBryde	HQ1	Ma'afala	W Poly	Samoa		E	3
Aa	770517.001	33	84	2n	NTBG	Kahanu	55	Ma'afala	W Poly	Samoa		E	3
Aa	HART 1	33	85	2n	HART	Hilo		Ma'afala				E	3
Aa	890454.001	33	93	2n	NTBG	Kahanu	W6	Niue	E Poly	Cook Islands		E	3
Aa	900231.001	33	93	2n	NTBG	Kahanu	L6	Niue	E Poly	Cook Islands		E	3

Table 1 (continued)

Taxon	Accession number	Lin	Gn	Pl	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa	890153.001	33	93	2n	NTBG	Kahanu	25	Niue (Paca)	E Poly	Cook Islands		E	3
Aa	900259.001	34	86	2n	NTBG	Kahanu	C6	Malphang	Mela	Vanuatu		G	3
Aa	890473.001	35	87	2n	NTBG	Kahanu	U5	Manang	Mela	Vanuatu		E	3
Aa	900263.001	36	88	2n	NTBG	Kahanu	J8	Masee	W Poly	Samoa		E	3
Aa	890156.001	37	90	2n	NTBG	Kahanu	Y4	Tahitian	E Poly	Cook Islands		E	3
Aa	890156.002	37	90	2n	NTBG	Kahanu	W7	Tahitian	E Poly	Cook Islands		E	3
Aa	890477.001	37	90	2n	NTBG	Kahanu	R4	Uto samoa	Mela	Fiji		E	3
Aa	910275.001	37	90	2n	NTBG	Kahanu	K8	Puou	Mela	Vanuatu		E	3
Aa	960575.001	37	90	2n	NTBG	McBryde	LF1	Mos en Samoa	Micro	Kosrae		E	3
Aa	070246.002	37	90	2n	NTBG	Kahanu	61	Meiuhpw en Samoa	Micro	Pohnpei, FSM		E	3
Aa	030039.001	37	90	2n	NTBG	McBryde	McB17	Puou	W Poly	Samoa		E	3
Aa	770520.001	37	90	2n	NTBG	Kahanu	53	Puou	W Poly	Samoa		E	3
Aa	890474.001	37	98	2n	NTBG	Kahanu	U6	Puou	W Poly	Samoa		E	3
Aa	910289.001	37	98	2n	NTBG	Kahanu	O9	Puou (unk 06)	Wpoly (unk)	Samoa (unk)		E	3
Aa	880691.001	37	99	2n	NTBG	Kahanu	O8	Puou	W Poly	Tonga		E	3
Aa	770519.001	38	91	2n	NTBG	Kahanu	57	Momolega	W Poly	Samoa		E	3
Aa	HART 50 (890469)	39	92	2n	HART	Hilo	F8	Kukumu Tasi	Mela	Solomon Islands		C	3
Aa	910266.002	40	94	2n	NTBG	Kahanu	9	Pipitia	E Poly	Society Islands		E	4
Aa	790492.001	41	95	2n	NTBG	Kahanu	F7	Porohiti	E Poly	Society Islands		C	1
Aa	900261.001	42	96	2n	NTBG	Kahanu	P9	Samoaan 2	Mela	Fiji		G	2
Aa	900261.002	42	96	2n	NTBG	Kahanu	W3	Samoaan 2	Mela	Fiji		G	2
Aa	890471.001	42	96	2n	NTBG	Kahanu	MV4	Uto dina	Mela	Fiji		G	2
Aa	890471.002	42	96	2n	NTBG	McBryde	8	Uto dina	Mela	Fiji		G	2
Aa	900233.002	42	96	2n	NTBG	Kahanu	J6	Pulupulu	Mela	Rotuma		G	2
Aa	900233.001	42	97	2n	NTBG	Kahanu	P5	Pulupulu	Mela	Rotuma		G	2
Aa	900226.001	42	114	2n	NTBG	Kahanu	KM3	Unk 07	Mela (unk)	Fiji (unk)		G	2
Aa	020352.001	42	117	2n	NTBG	Kahanu	A8	Unk 17	W Poly	Samoa		G	2
Aa	900264.001	42	121	2n	NTBG	Kahanu	A6	Uto ni viti	Mela	Fiji		G	2
Aa	900257.001	43	100	2n	NTBG	Kahanu	S9	Rauulu	Mela	Rotuma		F	2
Aa	890475.002	44	101	2n	NTBG	Kahanu	K9	Sagosago	W Poly	Samoa		E	3
Aa	910276.001	45	104	2n	NTBG	Kahanu	E8	Siviri 3	Mela	Vanuatu		E	3
Aa	910277.001	46	105	2n	NTBG	Kahanu	P4	Tedailir	Mela	Vanuatu		G	3
Aa	900281.001	47	106	2n	NTBG	Kahanu	D4	Tehelewa	Mela	Solomon Islands		C	3
Aa	900281.002	47	107	2n	NTBG	Kahanu	T7	Tehelewa	Mela	Solomon Islands		C	3
Aa	890456.001	48	108	2n	NTBG	Kahanu	52	Toro	Mela	Solomon Islands		C	3
Aa	770521.001	49	109	2n	NTBG	Kahanu	17	Ulu ea	W Poly	Tokelau		E	3
Aa	890155.001	50	111	2n	NTBG	Kahanu		Ulu sina	W Poly	Samoa		E	3

Table 1 (continued)

Taxon	Accession number	Lin	Gn	PI	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa	890155.002	50	111	2n	NTBG	Kahanu	K6	Ulu sina	W Poly	Samoa		E	3
Aa	900235.001	52	113	2n	NTBG	Kahanu	L5	Unk 03	Mela	Solomon Islands		E	3
Aa	020347.001	53	115	2n	NTBG	Kahanu	KM2	Unk 15	W Poly	Samoa		C	3
Aa	020348.001	54	116	2n	NTBG	Kahanu	KM8	Unk 16	W Poly	Samoa		C	3
Aa	020355.001	55	118	2n	NTBG	Kahanu	KM6	Unk 19	W Poly	Samoa		E	3
Aa	020498.001	56	119	2n	NTBG	Kahanu	59	Unk 21	Mela	Solomon Islands		F	3
Aa	020500.001	57	120	2n	NTBG	Kahanu	60	Unk 22	Mela	Solomon Islands		F	3
Aa×Am	HART 37 (890160)	4	8	3n	HART	Hilo		Ebechab	Micro	Palau		F	4/1
Aa×Am	890183.001	4	26	3n	NTBG	Kahanu	Y3	Midolab	Micro	Palau	0.735 (0.579–0.863)	F	4/1
Aa×Am	890183.003	4	26	3n	NTBG	McBryde	MV3	Midolab	Micro	Palau	0.752 (0.586–0.883)	F	4/1
Aa×Am (Aa)	HART 46 (890183)	4	26	3n	HART	Hilo		Midolab	Micro	Palau	0.845 (0.693–0.953)	F	4/1
Aa×Am	890183.002	4	67	3n	NTBG	Kahanu	L8	Midolab	Micro	Palau	0.775 (0.615–0.9)	F	4/1
Aa×Am (Aa)	790487.001	5	11	3n	NTBG	Kahanu	27	Unk (Huehue)	Micro (E Poly)	Pohnpei, FSM (Society)	0.722 (0.566–0.853)	D	5
Aa×Am	HART 52 (890480)	5	29	3n	HART	Hilo		Lipet	Micro	Pohnpei, FSM	0.76 (0.59–0.894)	D	4
Aa×Am (Aa)	790489.001	5	35	3n	NTBG	Kahanu	1	Unk (Pipitia)	Micro (E Poly)	Pohnpei, FSM (Society)	0.768 (0.617–0.887)	D	4
Aa×Am	030034.001	5	54	3n	NTBG	McBryde	McB9	Meinpadahk	Micro	Pohnpei, FSM	0.768 (0.617–0.887)	D	4
Aa×Am	030034.002	5	54	3n	NTBG	McBryde	McB18	Meinpadahk	Micro	Pohnpei, FSM	0.251 (0.115–0.433)	D	4
Aa×Am	790494.001	5	54	3n	NTBG	Kahanu	Z9	Meinpadahk	Micro	Pohnpei, FSM	0.752 (0.586–0.883)	D	4
Aa×Am	030041.001	5	55	3n	NTBG	McBryde	McB20	Rotuma	E Poly	Society Islands	0.773 (0.625–0.89)	D	5/4
Aa×Am	890163.002	5	61	3n	NTBG	Kahanu	U9	Lemae	Micro	Mariana Islands	0.802 (0.652–0.915)	D	5/4
Aa×Am	890163.001	5	62	3n	NTBG	Kahanu	X2	Lemae	Micro	Mariana Islands	0.802 (0.652–0.915)	D	4
Aa×Am	890480.001	5	63	3n	NTBG	Kahanu	O6	Lipet	Micro	Pohnpei, FSM	0.694 (0.5–0.856)	D	4
Aa×Am	890480.002	5	63	3n	NTBG	Kahanu	N7	Lipet	Micro	Pohnpei, FSM	0.812 (0.668–0.92)	D	4
Aa×Am	910270.001	5	64	3n	NTBG	Kahanu	D8	Lipet	Micro	Pohnpei, FSM	0.27 (0.131–0.451)	D	4
Aa×Am	890467.001	5	66	3n	NTBG	Kahanu	W9	Meinpuuht	Micro	Pohnpei	0.845 (0.693–0.953)	D	4
Aa×Am	790490.001	5	68	3n	NTBG	Kahanu	15	Rotuma	E Poly	Society Islands	0.775 (0.615–0.9)	D	5/4
Aa×Am	910272.001	9	28	3n	NTBG	Kahanu	F9	Meinpohtsakar	Micro	Pohnpei, FSM	0.734 (0.573–0.867)	D	5
Aa×Am	910272.002	9	28	3n	NTBG	Kahanu	E6	Meinpohtsakar	Micro	Pohnpei, FSM	0.719 (0.562–0.85)	D	5
Aa×Am	HART 49 (910272)	9	28	3n	HART	Hilo		Meinpohtsakar	Micro	Pohnpei, FSM	0.815 (0.667–0.924)	D	5
Aa×Am	HART 53 (910274)	10	30	3n	HART	Hilo		Nahnmwal	Micro	Pohnpei, FSM	0.768 (0.601–0.898)	D	4
Aa×Am	910274.001	10	31	3n	NTBG	Kahanu	Q10	Nahnmwal	Micro	Pohnpei, FSM	0.804 (0.647–0.922)	D	4
Aa×Am	980210.001	15	53	3n	NTBG	Kahanu	EE3	Ebechab	Micro	Palau	0.768 (0.617–0.887)	F	4
Aa×Am	890160.001	15	58	3n	NTBG	Kahanu	X8	Ebechab	Micro	Palau	0.736 (0.575–0.866)	F	4
Aa×Am	910652.001	15	59	3n	NTBG	Kahanu	U8	Errud	Micro	Palau	0.792 (0.644–0.905)	F	4
Aa×Am	910268.001	16	56	3n	NTBG	Kahanu	J9	Meion	Micro	Chuuk, FSM	0.85 (0.701–0.955)	D	4
Aa×Am	990781.001	16	56	3n	NTBG	McBryde	LF2	Meion (unk)	Micro	Pohnpei, FSM	0.798 (0.637–0.919)	D	4
Aa×Am	030045.001	17	57	3n	NTBG	McBryde	McB19	Yap	Micro	Palau	0.804 (0.647–0.922)	F	4

Table 1 (continued)

Taxon	Accession number	Lin	Gn	Pl	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa×Am	900250.001	17	74	3n	NTBG	Kahanu	A5	Yap	Micro	palau	0.845 (0.693–0.953)	F	4
Aa×Am	910269.001	18	60	3n	NTBG	Kahanu	A9	Faine	Micro	Chuuk, FSM	0.768 (0.617–0.887)	D	4
Aa×Am	900255.001	19	65	3n	NTBG	Kahanu	B5	Meinpwah	Micro	Pohnpei, FSM	0.773 (0.625–0.89)	D	5
Aa×Am	890164.001	20	70	3n	NTBG	Kahanu	22	Sewan	Micro	Chuuk, FSM	0.717 (0.553–0.853)	D	4
Aa×Am	890164.002	20	70	3n	NTBG	Kahanu	T9	Sewan	Micro	Chuuk, FSM	0.491 (0.319–0.667)	D	4
Aa×Am	890468.002	21	71	3n	NTBG	Kahanu	L9	Te bukiraro	Micro	Kiribati	0.555 (0.378–0.726)	D	4
Aa×Am	890453.001	22	72	3n	NTBG	Kahanu	Q5	Ulu afa	W Poly	Tokelau	0.356 (0.2–0.539)	D	5
Aa×Am	890185.001	23	73	3n	NTBG	Kahanu	28	Ulu elise	W Poly	Tokelau	0.428 (0.264–0.607)	D	5
Aa×Am	890161.001	24	75	3n	NTBG	Kahanu	19	Yuley	Micro	Yap	0.783 (0.629–0.902)	F	4/5
Aa×Am	890452.001	58	122	2n	NTBG	Kahanu	U3	Te mai	Micro	Kiribati	0.926 (0.791–0.997)	D	5
Aa×Am	890184.001	59	123	2n	NTBG	Kahanu	10	Luthar	Micro	Yap	0.263 (0.127–0.44)	C	5
Aa×Am	900253.002	60	124	2n	NTBG	Kahanu	J5	Mei chocho	Micro	Chuuk, FSM	0.254 (0.122–0.427)	D	5
Aa×Am	900253.001	61	125	2n	NTBG	Kahanu	Q6	Mei chocho	Micro	Chuuk, FSM	0.204 (0.085–0.378)	D	5
Aa×Am	890166.001	62	126	2n	NTBG	Kahanu	Y7	Mei koeng	Micro	Chuuk, FSM	0.768 (0.617–0.887)	D	5
Aa×Am	890466.002	63	127	2n	NTBG	Kahanu	Q9	Mei koeng	Micro	Chuuk, FSM	0.752 (0.586–0.883)	D	5
Aa×Am	890177.003	64	128	2n	NTBG	Kahanu	3	Ulu afa	W Poly	Tokelau	0.491 (0.319–0.667)	D	5
Aa×Am	890178.001	65	129	2n	NTBG	Kahanu	26	Ulu afa	W Poly	Tokelau	0.366 (0.212–0.543)	D	5
Aa×Am	890177.001	66	130	2n	NTBG	Kahanu	31	Ulu afa	W Poly	Tokelau	0.649 (0.469–0.807)	D	5
Aa×Am	890172.001	67	131	2n	NTBG	Kahanu	37	Ulu afa	W Poly	Tokelau	0.432 (0.266–0.611)	D	5
Aa×Am	890179.001	68	132	2n	NTBG	Kahanu	44	Ulu afa	W Poly	Tokelau	0.462 (0.293–0.64)	D	5
Aa×Am	890257.001	69	133	2n	NTBG	Kahanu	X3	Ulu afa	W Poly	Tokelau	0.213 (0.095–0.376)	D	5
Aa×Am	890171.001	70	134	2n	NTBG	Kahanu	X6	Ulu afa	W Poly	Tokelau	0.628 (0.465–0.776)	D	5
Aa×Am	890176.001	71	135	2n	NTBG	Kahanu	Z8	Ulu afa	W Poly	Tokelau	0.527 (0.351–0.702)	D	5
Aa×Am	890174.001	72	136	2n	NTBG	Kahanu	18	Ulu afa	W Poly	Tokelau	0.274 (0.14–0.446)	D	5
Aa×Am	890172.002	73	137	2n	NTBG	Kahanu	ZZ5	Ulu afa	W Poly	Tokelau	0.429 (0.265–0.608)	D	5
Aa×Am	890176.002	74	138	2n	NTBG	Kahanu	ZZ8	Ulu afa	W Poly	Tokelau	0.588 (0.409–0.757)	D	5
Aa×Am	890168.002	75	139	2n	NTBG	Kahanu	ZZ9	Ulu afa	W Poly	Tokelau	0.814 (0.644–0.934)	D	5
Aa×Am	890177.002	76	140	2n	NTBG	Kahanu	5	Ulu afa 1	W Poly	Tokelau	0.622 (0.442–0.787)	D	5
Aa×Am	890171.002	77	141	2n	NTBG	Kahanu	7	Ulu afa 2	W Poly	Tokelau	0.556 (0.379–0.728)	C	5/3
Aa×Am	890181.002	78	142	2n	NTBG	Kahanu	B4	Ulu afa 3	W Poly	Tokelau	0.554 (0.377–0.726)	D	5
Aa×Am	890181.001	79	143	2n	NTBG	Kahanu	Z4	Ulu afa 4	W Poly	Tokelau	0.491 (0.32–0.668)	D	5
Aa×Am	890173.001	80	144	2n	NTBG	Kahanu	11	Ulu afa elise	W Poly	Tokelau	0.685 (0.505–0.839)	D	5
Aa×Am	890173.002	81	145	2n	NTBG	Kahanu	ZZ7	Ulu afa elise	W Poly	Tokelau	0.428 (0.264–0.607)	D	5
Aa×Am	890175.001	81	147	2n	NTBG	Kahanu	34	Ulu afa haomoa	W Poly	Tokelau	0.429 (0.264–0.607)	D	5
Aa×Am	890182.002	81	149	2n	NTBG	Kahanu	ZZ6	Ulu elise 2	W Poly	Tokelau	0.554 (0.377–0.725)	D	5
Aa×Am	900230.001	82	146	2n	NTBG	Kahanu	6	Ulu afa haomoa	W Poly	Tokelau	0.511 (0.359–0.664)	C	5
Aa×Am	890182.001	83	148	2n	NTBG	Kahanu	X9	Ulu elise 1	W Poly	Tokelau	0.651 (0.471–0.811)	D	5

Table 1 (continued)

Taxon	Accession number	Lin	Gn	PI	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Aa×Am	890180.001	84	150	2n	NTBG	Kahanu	24	Ulu hamoa	W Poly	Tokelau	0.926 (0.791–0.997)	D	5
Aa×Am	890170.002	85	151	2n	NTBG	Kahanu	ZZ3	Ulu hamoa	W Poly	Tokelau	0.872 (0.728–0.966)	D	5
Aa×Am (Am)	000528.001	129	197	2n	NTBG	Kahanu	AA5	Meikole	Micro	Pohnpei, FSM	0.064 (0.01–0.188)	D	5
Ac	TARS 18009	86	152	2n	TARS	Mayaguez	Plot 14	Unk	Unk			A	1
Ac	960576.001	86	153	2n	NTBG	McBryde	MV2	Breadnut	C. Am.	Honduras		A	1
Ac	770444.001	86	153	2n	NTBG	Kahanu	50	Camansi	Epoly	Society Islands		A	1
Ac	980212.001	87	154	2n	NTBG	Kahanu	EE5	Camansi	Micro	Palau		A	1
Ac	910281.001	88	155	2n	NTBG	Kahanu	M10	Kamansi	Phil.			A	1
Ac	000390.002	89	156	2n	NTBG	McBryde	McB4	Kapiak	Mela	Papua New Guinea		A	1
Ac	000394.002	90	157	2n	NTBG	McBryde	McB2	Kapiak	Mela	Papua New Guinea		A	1
Ac	000395.002	91	158	2n	NTBG	McBryde	McB21	Kapiak	Mela	Papua New Guinea		A	1
Ac	000398.005	92	159	2n	NTBG	McBryde	McB25	Kapiak	Mela	Papua New Guinea		A	1
Ac	000499.003	93	160	2n	NTBG	McBryde	McB3	Kapiak	Mela	Papua New Guinea		A	1
Ac	000500.002	94	161	2n	NTBG	McBryde	McB23	Kapiak	Mela	Papua New Guinea		A	1
Ac	000501.004	95	162	2n	NTBG	McBryde	McB5	Kapiak	Mela	Papua New Guinea		A	1
Ac	000501.005	96	163	2n	NTBG	McBryde	McB1	Kapiak	Mela	Papua New Guinea		A	1
Ac	000502.002	97	164	2n	NTBG	McBryde	McB22	Kapiak	Mela	Papua New Guinea		A	1
Ac	000503.004	98	165	2n	NTBG	McBryde	McB24	Kapiak	Mela	Papua New Guinea		A	1
Ac	000398.001	99	166	2n	NTBG	Kahanu	F10	Kapiak	Mela	Papua New Guinea		A	1
Ac	000501.001	100	167	2n	NTBG	Kahanu	F11	Kapiak	Mela	Papua New Guinea		A	1
Ac	000398.003	101	168	2n	NTBG	Kahanu	H10	Kapiak	Mela	Papua New Guinea		A	1
Ac	000399.001	102	169	2n	NTBG	Kahanu	I10	Kapiak	Mela	Papua New Guinea		A	1
Ac	000398.002	103	170	2n	NTBG	Kahanu	K10	Kapiak	Mela	Papua New Guinea		A	1
Ac	000502.001	104	171	2n	NTBG	Kahanu	D10	Kapiak	Mela	Papua New Guinea		A	1
Ac	000501.001	105	172	2n	NTBG	Kahanu	EE4	Kapiak	Mela	Papua New Guinea		A	1
Ac	000499.001	106	173	2n	NTBG	Kahanu	EE7	Kapiak	Mela	Papua New Guinea		A	1
Ac	000503.001	107	174	2n	NTBG	Kahanu	E10	Kapiak	Mela	Papua New Guinea		A	1
Ac	000499.002	108	175	2n	NTBG	Kahanu	E11	Kapiak	Mela	Papua New Guinea		A	1
Ac	000395.001	109	176	2n	NTBG	Kahanu	FF6	Kapiak	Mela	Papua New Guinea		A	1
Ac	000390.001	110	177	2n	NTBG	Kahanu	FF7	Kapiak	Mela	Papua New Guinea		A	1
Ac	000503.002	111	178	2n	NTBG	Kahanu	G10	Kapiak	Mela	Papua New Guinea		A	1
Ac	000389.001	112	179	2n	NTBG	Kahanu	GG3	Kapiak	Mela	Papua New Guinea		A	1
Ac	000398.004	113	180	2n	NTBG	Kahanu	HH3	Kapiak	Mela	Papua New Guinea		A	1
Ac	000500.001	114	181	2n	NTBG	Kahanu	HH4	Kapiak	Mela	Papua New Guinea		A	1
Ac	910280.001	115	182	2n	NTBG	Kahanu	B9	Meikole	Micro	Pohnpei, FSM		A	1
Ac	HART 63	116	183	2n	HART	Hilo		N90-148	Malaysia			A	1
Ac	910283.001	117	184	2n	NTBG	Kahanu	R10	Timbul	Indonesia			A	1

Table 1 (continued)

Taxon	Accession number	Lin	Gn	Pl	GC	GS	Grid	Cultivar	Region	Island	Hybrid Index	UP	ST
Ac (Aa)	890455.001	14	49	2n	NTBG	Kahanu	V5	Ulu fatu	W Poly	Samoa		A	1
Ac (Aa)	900228.001	51	112	2n	NTBG	Kahanu	E7	Ulu fatu (unk 02)	W Poly	Samoa		A	1
Am	000523.002	118	185	2n	NTBG	McBryde	McB6	Dugdug	Micro	Mariana Islands		B	5
Am	900252.001	119	186	2n	NTBG	Kahanu	H5	Dugdug	Micro	Mariana Islands		B	5
Am	900252.002	120	187	2n	NTBG	Kahanu	A4	Dugdug	Micro	Mariana Islands		B	5
Am	900252.003	120	187	2n	NTBG	Kahanu	N9	Dugdug	Micro	Mariana Islands		B	5
Am	000522.002	121	188	2n	NTBG	Kahanu	BB6	Dugdug	Micro	Mariana Islands		B	5
Am	000523.001	122	189	2n	NTBG	Kahanu	CC4	Dugdug	Micro	Mariana Islands		B	5
Am	HART 67	122	198	2n	HART	Hilo		N05-8	Micro			B	5
Am	000521.002	123	190	2n	NTBG	Kahanu	CC5	Dugdug	Micro	Mariana Islands		B	5
Am	000521.001	124	191	2n	NTBG	Kahanu	CC7	Dugdug	Micro	Mariana Islands		B	5
Am	000522.001	125	192	2n	NTBG	Kahanu	DD5	Dugdug	Micro	Mariana Islands		B	5
Am	000521.003	126	193	2n	NTBG	Kahanu	DD6	Dugdug	Micro	Mariana Islands		B	5
Am	940010.002	126	194	2n	NTBG	McBryde	MV1	Dugdug	Micro	Mariana Islands		B	5
Am	970306.001	127	195	2n	NTBG	Kahanu	CC3	Kuru kimiti	Micro	Pohnpei, FSM		B	5
Am	970306.002	128	196	2n	NTBG	Kahanu	DD4	Kuru kimiti	Micro	Pohnpei, FSM		B	5

Taxon (Aa=*A. altitis*, Ac=*A. camansi*, Am=*A. mariannensis*, and Aa×Am=*A. altitis*×*A. mariannensis*), accession number (NTBG no. is given in parentheses if the collection is housed somewhere else but is a duplicate from NTBG), lineage (lin), genotype (gn), ploidy (pl), germplasm site (GS), grid location within germplasm (Grid), germplasm code (GC), cultivar name (Unk=unknown), region of provenance (*Mela* Melanesia, *Micro* Micronesia, *E Poly* Eastern Polynesia, *W Poly* Western Polynesia, *Phil.* Philippines, *C. Am.* Central America, *Unk* unknown), island group of provenance, hybrid index for putative hybrids (numbers closer to 1 indicate greater genetic contribution from *A. altitis*, and numbers closer to 0 indicate greater genetic contribution from *A. mariannensis*), UPGMA (UP), and STRU CTURE (ST) clusters are provided. For an individual to be assigned to a STRU CTURE cluster, it must have greater than 60% likelihood of being a member of the cluster. If an individual is assigned to two clusters, that individual had less than 60% likelihood of being a member of a single cluster. If a species or cultivar identification was changed as a result of the study, the previous misidentification is indicated in parentheses. For accessions that had unknown provenance or name, the likely identification is indicated based on lineage and/or genotype match, and “unk” is in parentheses

DNA extraction and SSR marker analysis

DNA extraction, PCR amplification of 19 microsatellite loci, electrophoresis, and scoring of the samples were performed as described in Witherup et al. (2013). Ploidy levels of some individuals were known from chromosome squashes (Ragone 2001). To assess ploidy in the other individuals, any individual with three alleles at multiple microsatellite regions was designated as triploid. Genotype-based ploidy assignment was considered to be accurate, as the vast majority of known triploid individuals displayed three alleles for at least three loci (deviations are discussed in “Results”). In addition, 174 of the individuals were known to be either seeded (indicating diploidy) or seedless (frequently indicating triploidy), and these data matched ploidy assignments in virtually all instances (deviations discussed in “Results”).

Data analyses

Resolving allele dosage in partial heterozygotes is challenging in triploid samples. The presence of two alleles at a given locus could signify either a genotype with allele *x* represented twice or allele *y* represented twice (i.e., *xx**y* vs. *xy**y*). Various methods exist for estimating allele copy number, but they are meant for samples with uniform, even-numbered ploidy and a known selfing rate (Clark and Jasieniuk 2011; De Silva et al. 2005), assume that either allele has an equal chance of being present in two copies (Hardy and Vekemans 2002; Tomiuk et al. 2009), or require data that can be normalized to complete heterozygotes (Esselink et al. 2004). Therefore, we did not impute a third allele in the case of partial heterozygotes (McGregor et al. 2000; Mengoni et al. 2000; Creste et al. 2004).

GenoDive (Meirmans and Van Tienderen 2004) was used to calculate genetic diversity measures and genetic distance. Genetic distances were calculated using the Bruvo method, a distance measure suited to codominant marker data in populations of mixed ploidy (Bruvo et al. 2004). UPGMA and neighbor-joining trees were then constructed using the neighbor program from PHYLIP package 3.69 (Felsenstein 2005) and visualized using FigTree 1.4.0. Analyses were run with all 349 samples and also excluding hybrids. Principal component analysis was performed in GenoDive (Meirmans and Van Tienderen 2004) and visualized using the ggplot2 visualization package for R (Wickham 2009).

The Bayesian clustering analysis software STRUCTURE v2.3.4 (Pritchard et al. 2000; Falush et al. 2007) was used to visualize genetic structure and subdivisions (number of genetic clusters, *K*) among samples. In order to analyze the allele data from both diploid and triploid samples simultaneously, the recessive alleles option in STRUCTURE was set to one to allow for ambiguity in genotypes, and diploid individuals were scored as missing data for the third allele. We carried

out 20 independent runs per *K* using a burn-in period of 100,000 and collected data for 100,000 iterations for *K*=1–15. The minimum value of *K* that can explain the data was assessed using the rate of change in the log likelihood probability of data between corresponding *K* values (DK) as detailed in Evanno et al. (2005).

The hybrid index, a quantitative estimate of the genetic contribution of two parental species, was calculated for hybrid collections using GenoDive (Meirmans and Van Tienderen 2004), extending the maximum likelihood approach of Buerkle (2005) to include polyploid individuals.

GenoDive (Meirmans and Van Tienderen 2004) was used to assign lineage groups and genotypes. Samples were assigned the same genotype only if they were identical at all loci. Since allele dosage in triploids with two alleles could not be reliably distinguished, any instances of such partial heterozygotes were considered identical, leading to a possible underestimation of unique genotypes. Samples were assigned the same lineage group if the genotype of one could be transformed to the genotype of the other within a threshold number of mutation steps. The threshold distance was set independently for each ploidy level, since the triploids were much less diverse. Pairwise genetic distances clustered as expected for both ploidy levels, with a distribution bounded at zero from those relationships that are putatively clonal and a normal distribution consisting of the distances between siblings. Thresholds were set to include all contiguous distances greater than 0 with frequency greater than 1. The lineage threshold for triploids was set at 16, while the diploid threshold was set at 8.

The polymorphic information content (PIC) for each locus was calculated as follows:

$$1 - \sum_{i=1}^n p_i^2 - \sum_{i=1}^{n-1} \sum_{j=i+1}^n 2p_i^2 p_j^2$$

where p_i and p_j are the frequency of two given alleles *i* and *j* (Botstein et al. 1980). In order to evaluate data from both ploidy levels concurrently, the allele frequency for a given allele *i* was calculated as $p_i = \frac{c_{i2} + c_{i3}}{N}$, where c_{in} is the number of occurrences of allele *i* among individuals with ploidy *n* and *N* is the number of all non-null alleles observed.

Results

Diversity characterization

When considering each taxon separately, the average number of alleles across all 19 loci ranged from 1.74 in

A. mariannensis to 7.37 in diploid breadfruit (Table 2). Alleles per locus ranged from 1 (in MAA3 for all taxa except *A. camansi*) to 16 (in MAA201 for *A. camansi*), and across all taxa, the average number of alleles was 11.63, with a range of 2 (in MAA3) to 24 (in MAA201) (Table 2). Gene diversity was greatest in triploid hybrids ($H_e=0.62$), followed by diploid *A. altilis* ($H_e=0.61$), triploid *A. altilis* ($H_e=0.52$), *A. camansi* ($H_e=0.41$), diploid hybrids ($H_e=0.38$), and *A. mariannensis* ($H_e=0.18$). Averaged across all taxa, gene diversity was 0.44 (Table 2).

Lineage and genotype groups

The 349 individuals analyzed represented 197 distinct genotypes sorted into 129 lineages (Table 1). All lineage groups and genotypes were comprised of individuals of the same species, with the exception of one lineage group (lineage 4) that included *A. altilis* as well as hybrids. Among the 189 triploid individuals (representing 140 accessions of *A. altilis* and hybrids) there were 74 genotypes, 50 of which were singletons present in only a single individual (Table 1). Among the 150 *A. altilis* triploid individuals (representing 112 accessions), there were 42 genotypes sorted into 8 lineage groups. However, there was a single dominant genotype

(genotype 1) that accounted for 75 individuals (representing 62 accessions) and was part of lineage group 1, which accounted for 130 individuals (representing 111 accessions). The members of this lineage group have provenance from Eastern Polynesia and Micronesia, with a few from Barbados, the Seychelles, or of unknown provenance. Among the 39 *A. altilis* × *A. mariannensis* triploid individuals (representing 27 accessions), there were 30 genotypes sorted into 15 lineage groups.

Compared to triploids, the diploid accessions were more readily distinguishable from one another based on microsatellite profiles (Table 1). Among the 160 diploid individuals (representing 113 accessions across all four taxa), there were 115 genotypes, 111 of which were represented by a single individual in the collection. The genotypes were sorted into 106 lineages, 89 of which contained only one accession. There were no genotypes or lineages shared across different taxa among the diploids. Among the 36 *A. camansi* individuals (representing 21 accessions), there were 35 genotypes sorted into 34 lineage groups. Among the 14 *A. mariannensis* individuals (representing 7 accessions), there were 13 genotypes sorted into 11 lineage groups. Among the 79 *A. altilis* diploid individuals (representing 63 accessions), there were 47 genotypes sorted into 33 lineage groups. Among the 31 diploid

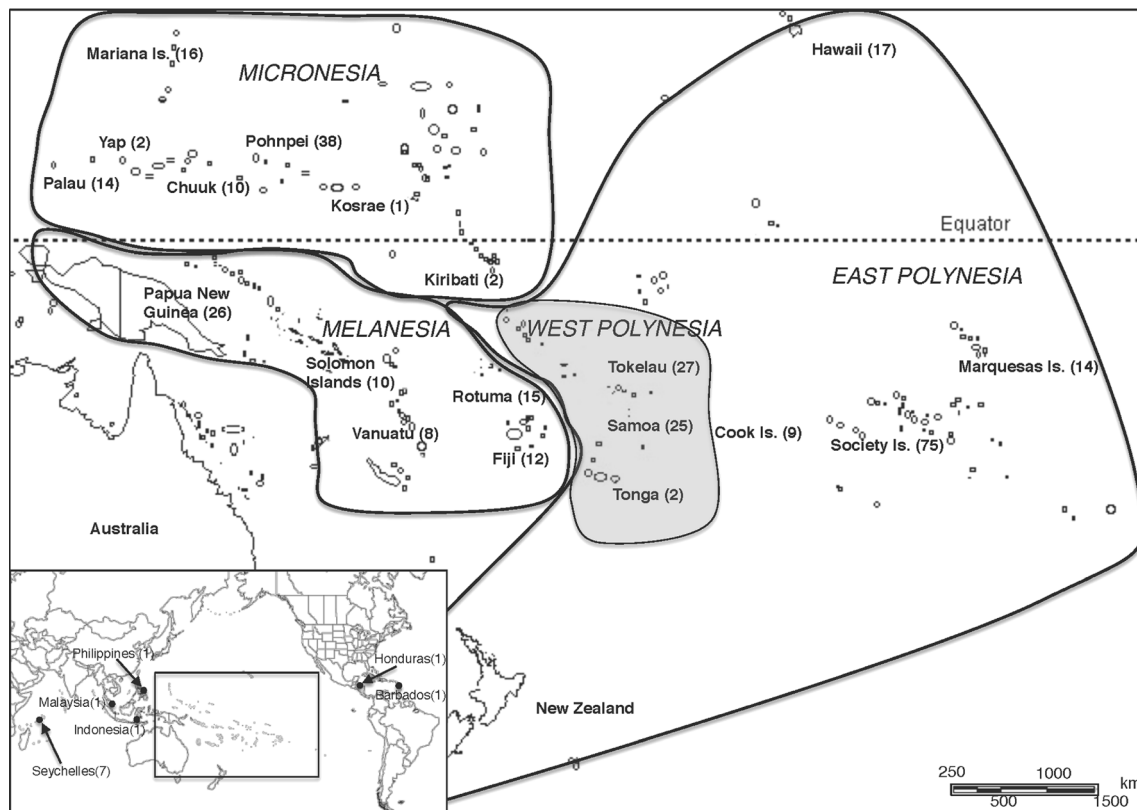


Fig. 1 Map of collections used in the study. *Inset in lower left* shows collections outside of Oceania with sample size in parentheses. The area highlighted in gray on the inset map is the focus of the larger Oceania

panel, which has island groups indicated and the following regions outlined: East Polynesia, West Polynesia, Micronesia, and Melanesia

Table 2 (continued)

MAA178a	5	211–245	0.16	0.104 (0)	2	223–245	0.304	0.207 (7.1)	14	0.741 (8.1)	0.477
MAA178b	6	241–257	0.621	0.519 (3.0)	3	251–255	0.588	0.367 (7.1)	10	0.648 (1.9)	0.661
MAA182	6	182–210	0.785	0.712 (0)	2	202–204	0.304	0.201 (0)	10	0.662 (1.6)	0.604
MAA201	16	268–312	0.925	0.808 (0)	3	266–276	0.405	0.274 (0)	24	0.838 (0.9)	0.695
MAA219	4	256–277	0.607	0.461 (0)	1	260	0	0 (0)	9	0.654 (0.4)	0.405
MAA287	2	179	0.028	0 (0)	1	183	0	0 (0)	11	0.632 (0.8)	0.319
MAA293	3	162–166	0.319	0.214 (0)	1	160	0	0 (0)	6	0.492 (1.1)	0.363
Average	4.895		0.407	0.318 (0.2)	1.737		0.176	0.109 (1.4)	11.632	0.627 (1.4)	0.441

N_a, Number of alleles; *ASR*, allele size range (bp); *H_e*=expected heterozygosity; *PIC* (%MD)=polymorphic information content (% missing data.)

hybrid individuals (representing 22 accessions), there were 31 genotypes sorted into 29 lineage groups.

Only a few lineages or genotypes were shared across regions. When considering only the breadfruit (*A. altilis*) and hybrids (*A. altilis* × *A. mariannensis*), one lineage was shared across all regions in Oceania (lineage 37); another lineage was shared across Western and Eastern Polynesia, Micronesia, and areas outside of Oceania (lineage 1); two lineages were shared between Western and Eastern Polynesia (lineages 2 and 33), two lineages were shared across Eastern Polynesia and Micronesia (lineages 4 and 5), and two lineages were shared across Western Polynesia and Melanesia (Lineages 27 and 42) (Fig. 2a). When considering genotypes, one genotype (lineage 37/genotype 90) was shared across all regions in Oceania; one genotype was shared across Micronesia and Eastern Polynesia (lineage 1/genotype 23); one genotype was shared across Eastern Polynesia, Micronesia, and areas outside of Oceania (lineage 1/genotype 1); one genotype was shared across Melanesia and Western Polynesia (lineage 27/genotype 78); one genotype was shared across Western and Eastern Polynesia (lineage 1/genotype 3); and one genotype was shared across Eastern Polynesia and areas outside of Oceania (lineage 1/genotype 5) (Fig. 2b).

Identification of unknowns

Twenty-eight accessions had unknown cultivar names, and 13 of them were also of unknown provenance. Among the 28 unknowns, 12 (43 %) shared identical genotypes to an accession(s) with known provenance and cultivar name(s) (Table 1). Nine (32 %) of the unknown accessions shared a lineage group but not a genotype with other known accessions, and seven (25 %) unknowns shared neither lineage nor genotypes with other accessions, suggesting they are genetically unique within the collection.

Cultivar names

There were 136 different cultivar names recorded for the cultivated breadfruit and hybrids analyzed in this study. Half of these names (68) were singletons, only represented by a single individual. Among the other 68 names with multiple individuals, 40 names were represented each by 2 individuals, 16 names were represented each by 3 individuals, 5 names were represented each by 4 individuals, and 3 names were represented each by 5 individuals. There were four different names that were represented by 6, 7, 13, and 18 individuals. Only one name occurred in more than one island group within a region: Puou (Tonga and Samoa in Western Polynesia). Puou also occurred in Vanuatu (Melanesia), but this is known to be an introduction from a Samoan variety. Of the 68 names represented by more than 1 individual, only 33 names consistently shared the same genotypes with other individuals

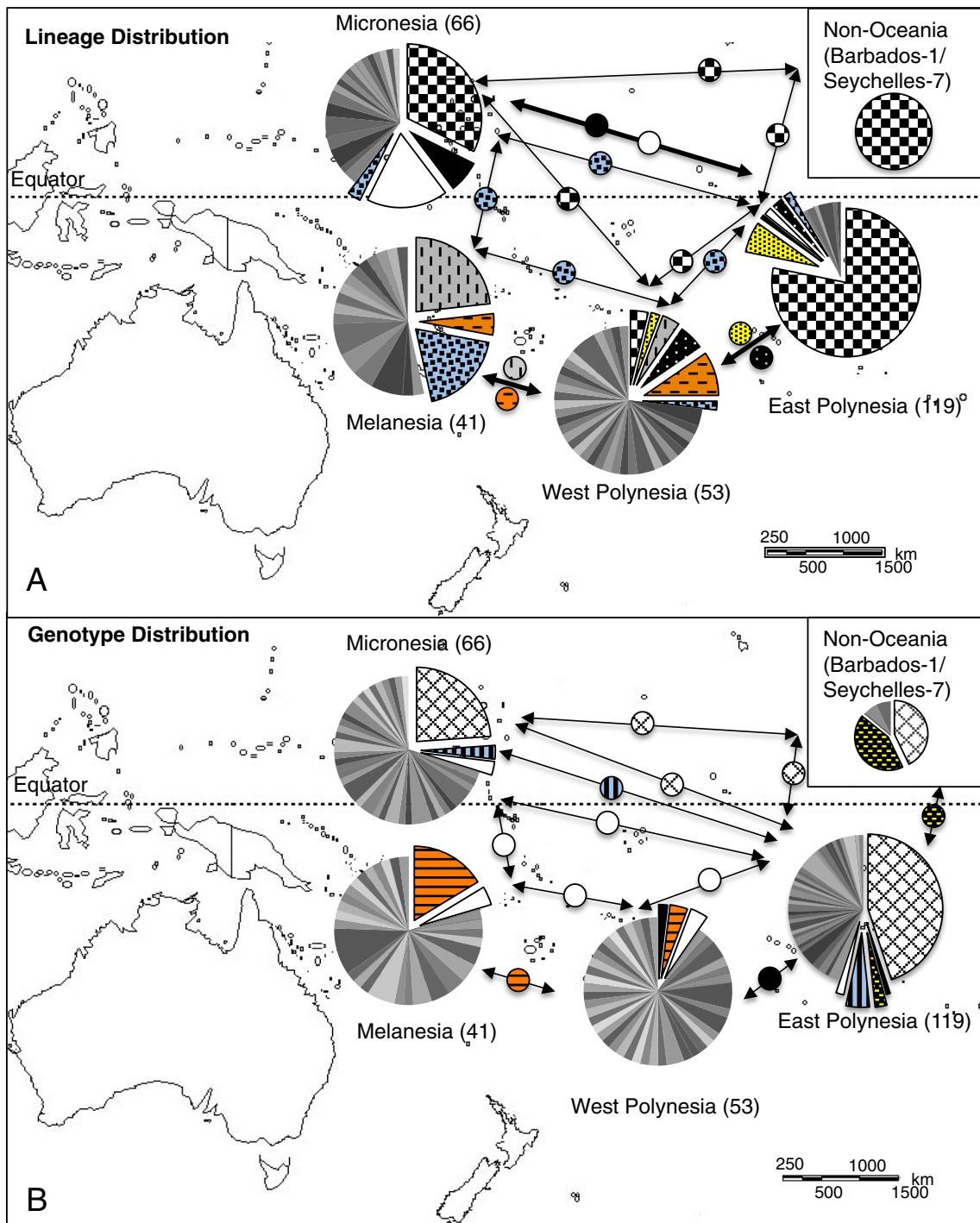


Fig. 2 Distribution across Oceania of lineages and genotypes in *A. altilis* and *A. altilis* × *A. mariannensis* hybrids based on microsatellite data from 19 loci. Pie charts show regional distributions of lineages (a) and genotypes (b). Within a pie chart, solid gray shades indicate unique genotypes that are not shared across regions. Pattered wedges that are

pulled out indicate lineages or genotypes that are shared across regions. Arrows indicate shared lineages or genotypes across regions (thin arrows=one shared group and thick arrows indicate two shared groups). Pattered circles on the arrows indicate the wedge(s) shared across regions. Sample sizes for each region are indicated in parentheses

bearing the same name, and 24 of those 33 were members of the same base accession number. Eight of those 33 names all shared the most ubiquitous lineage group and genotype (1/1). The remaining nine names, represented by more than one individual and consistently sharing the same genotype, share

their respective genotypes across different base accession numbers: Afara, Enuu, Mei chon, Meion, Meinpadahk, Niue, Otea, Puaa, and Ulu tala. Among the 75 individuals with the ubiquitous genotype (lineage 1/genotype 1), 44 different names were represented.

Genetic distance

The results of the UPGMA and neighbor-joining analyses were nearly identical and only the UPGMA tree is discussed here. Analyses without hybrids resulted in each taxon making up its own cluster with the *A. camansi* and *A. altilis* clusters together forming a larger cluster with the *A. mariannensis* cluster next to it (not shown). This is consistent with previous studies based on AFLP (Zerega et al. 2005). In the UPGMA tree with all samples included, each taxon largely clustered with other members of the same taxon, with hybrids largely clustered between *A. altilis* and *A. mariannensis* (Fig. 3). All samples identified as *A. camansi* clustered together with two specimens (890455.001 of unknown provenance and 900228.001 from Samoa) that had previously been classified as *A. altilis*. Upon closer examination of the two specimens, we determined that they share many characteristics with *A. camansi* and their identifications have been changed to *A. camansi*. The *A. camansi* cluster was split into two smaller clusters, one cluster made up of samples from Papua New Guinea and the other cluster made up of

samples from elsewhere. With the exception of one sample (000528.001 from Pohnpei), all specimens previously identified as *A. mariannensis* clustered together. The exception was nested in a cluster containing mostly hybrids as well as triploid and diploid *A. altilis* from islands throughout Oceania. Upon close examination of the specimen, we determined that his accession shares hybrid characteristics with greater contribution from *A. mariannensis*.

Principal component analysis

Plotting of the first two principal components of the microsatellite profiles shows segregation by species, with hybrids clustering to some degree between parental species (Fig. 4). When regional samples are analyzed separately, most Micronesian and Western Polynesian hybrids cluster between the parental species or cluster more closely to *A. mariannensis* (Fig. 5), and in Eastern Polynesia, hybrids cluster much more closely to *A. altilis* (Fig. 5).

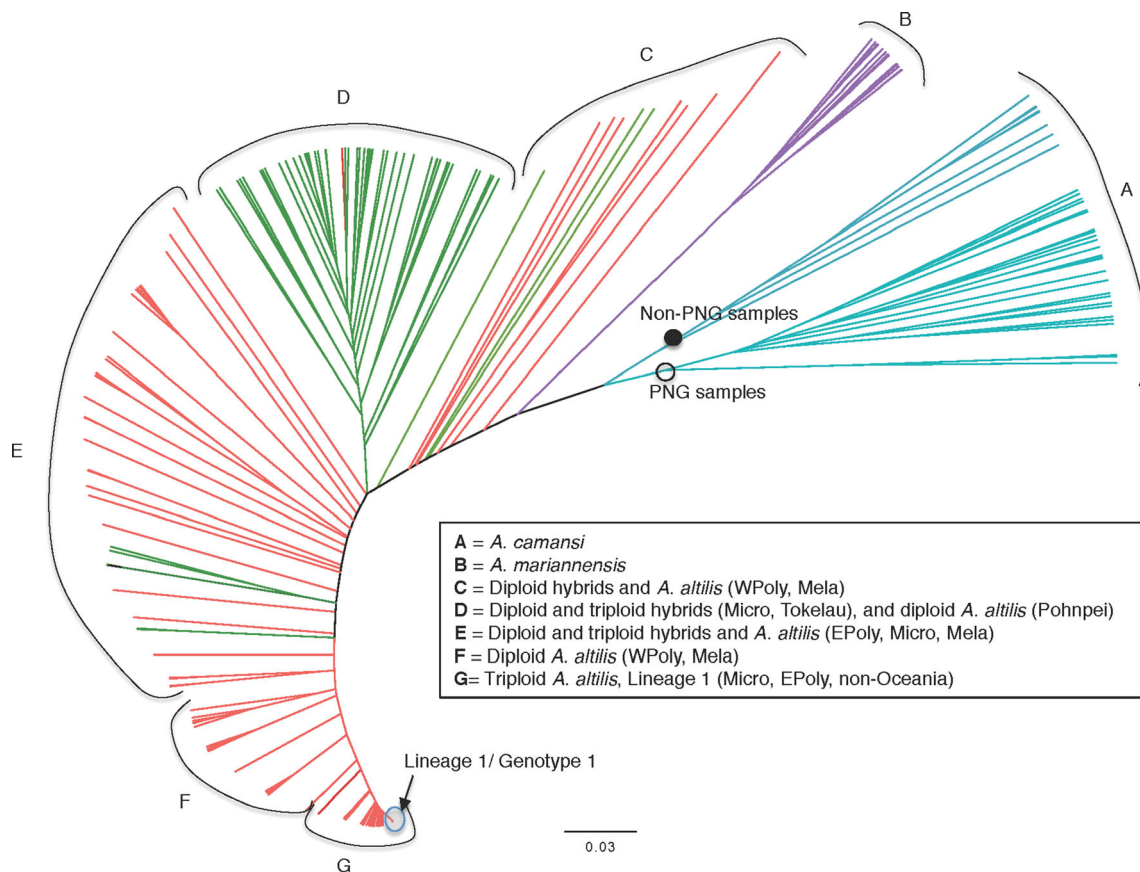
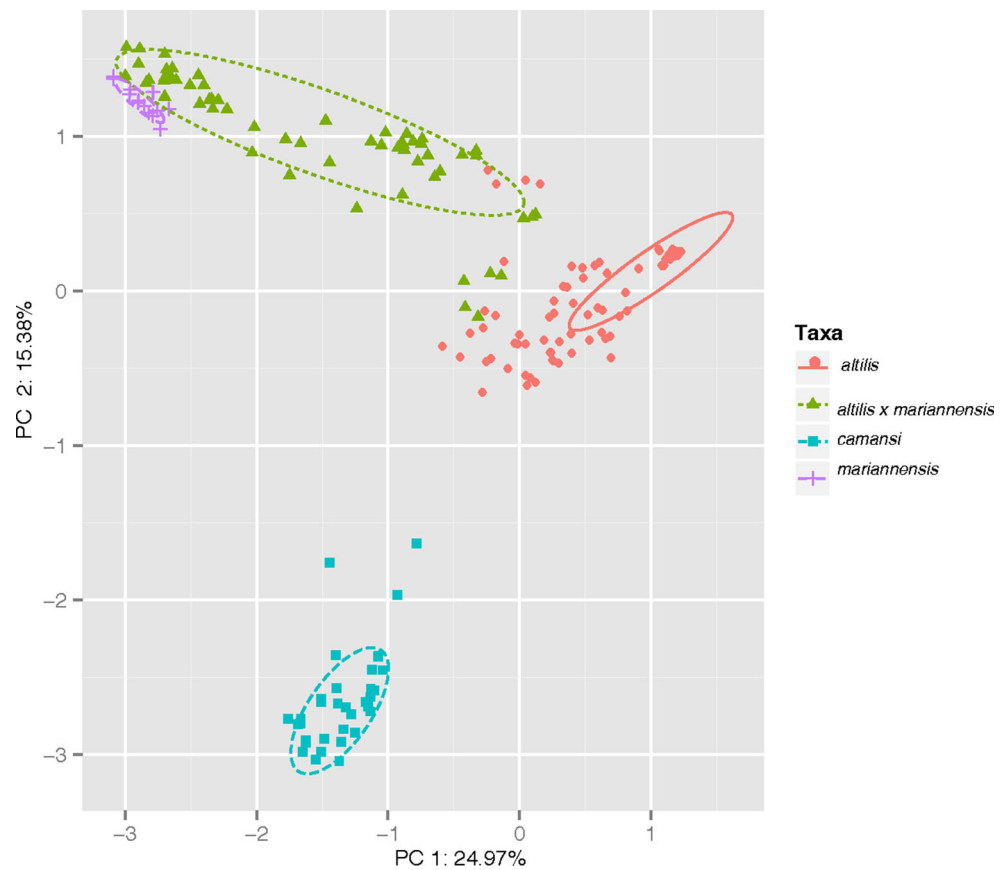


Fig. 3 UPGMA tree of *A. altilis*, *A. altilis* × *A. mariannensis*, *A. camansi*, and *A. mariannensis* based on data from 19 microsatellite loci. Cluster members are indicated in the legend. The area within cluster G that is in the shaded gray circle is the 75 individuals belonging to the ubiquitous

lineage 1/genotype 1. The filled circle on the node in cluster A denotes samples from outside of Papua New Guinea and the open circle denotes samples from Papua New Guinea. Cluster designations are also included in Table 1. Bar of Bruvo distance 0.03 shown for scale

Fig. 4 PCA results based on data from 19 microsatellite loci of breadfruit, hybrids, and wild relatives from throughout Oceania. Taxon coding is indicated in the legend, and 95 % bivariate confidence interval ellipses are shown for taxon clustering



Hybrid index

Examining the hybrid index (h) across the 69 individuals identified as hybrids based on morphological characters (Table 1) revealed a similar pattern evident from principal component analysis (PCA). Individuals showing greater contributions from *A. mariannensis* ($h < 0.4$) were from Micronesia and Tokelau in Western Polynesia. Individuals showing more equal contributions ($h = 0.4–0.7$) from both parental species were almost exclusively from Tokelau. Individuals showing greater contribution from *A. altilis* ($h > 0.7$) were predominantly from Micronesia and Eastern Polynesia.

Structure

In the STRUCTURE analysis, the modal value of the distribution of the true K identified a peak at $K=5$, which was supported by large shifts in $L(K)$ and $\text{Ln}'(K)$ from $K=5$ to $K=6$ associated with true value of K , as described in Evanno et al. (2005). All *A. mariannensis* individuals group together as do all the *A. camansi* individuals (Fig. 6). *A. altilis* specimens were subdivided into three groups that can generally be characterized as Melanesian breadfruit (predominantly diploid), breadfruit from throughout Oceania (diploid and

triploid), and triploid breadfruit from Eastern Polynesia and Micronesia. Diploid Micronesian hybrids almost exclusively shared the same group as *A. mariannensis*, while triploid Micronesian hybrids were admixtures of *A. mariannensis* and the triploid Micronesian/Eastern Polynesian breadfruit groups. Western Polynesian hybrids either had dominant contributions from *A. mariannensis* or were admixtures between *A. mariannensis* and the diploid/triploid trans-Oceania breadfruit group. Some of the diploid hybrids from Western Polynesia and Micronesia only shared the *A. mariannensis* group with no apparent admixture with *A. altilis*.

Utility of microsatellite markers

The PIC of the microsatellite loci across all taxa ranged from 0.007 to 0.838, with an average value of 0.627 (Table 2). The extent to which each locus was informative varied by taxon, with loci generally being the most informative for *A. altilis* and hybrids. A codominant marker is generally classified as highly informative if its $\text{PIC} > 0.5$ (Botstein et al. 1980; Ghislain et al. 2004). Using this criterion, 15 markers were highly informative within *A. altilis*, 13 within hybrids, 7 within *A. camansi*, and none within *A. mariannensis*. If only the genotypes from the 10 most informative loci were analyzed, 111 out of 123 unique diploid genotypes (90 %) and

100 % of 75 triploid genotypes could still be distinguished, and all taxa could be fully distinguished (i.e., no two accessions of different species shared the same microsatellite profile based on these 10 markers). Only primer MAA3 had zero distinguishing utility.

A previous study examined AFLP profiles of breadfruit and its relatives using three primer pairs across 313 individuals (Zerega et al. 2004), 181 of which were included in the present study. Among this 181 subset of individuals, 156 distinct AFLP and 110 distinct microsatellite genotypes were distinguished. Given the nature of the PIC equation,

comparing PICs between microsatellites and AFLPs is not informative. Because AFLP fragments are not mutually exclusive, the information of an AFLP primer pair can change drastically while the relative frequencies of fragments remain the same. On the basis of number of markers, the AFLP assays were generally more informative (Table 3).

Zymotypes established from the isozyme profiles of six enzymes were available for 140 of the individuals characterized here (Ragone 1991; Zerega et al. 2005). Among these 140 individuals, there were 54 distinct zymotypes and 88 distinct microsatellite genotypes. While the average enzyme

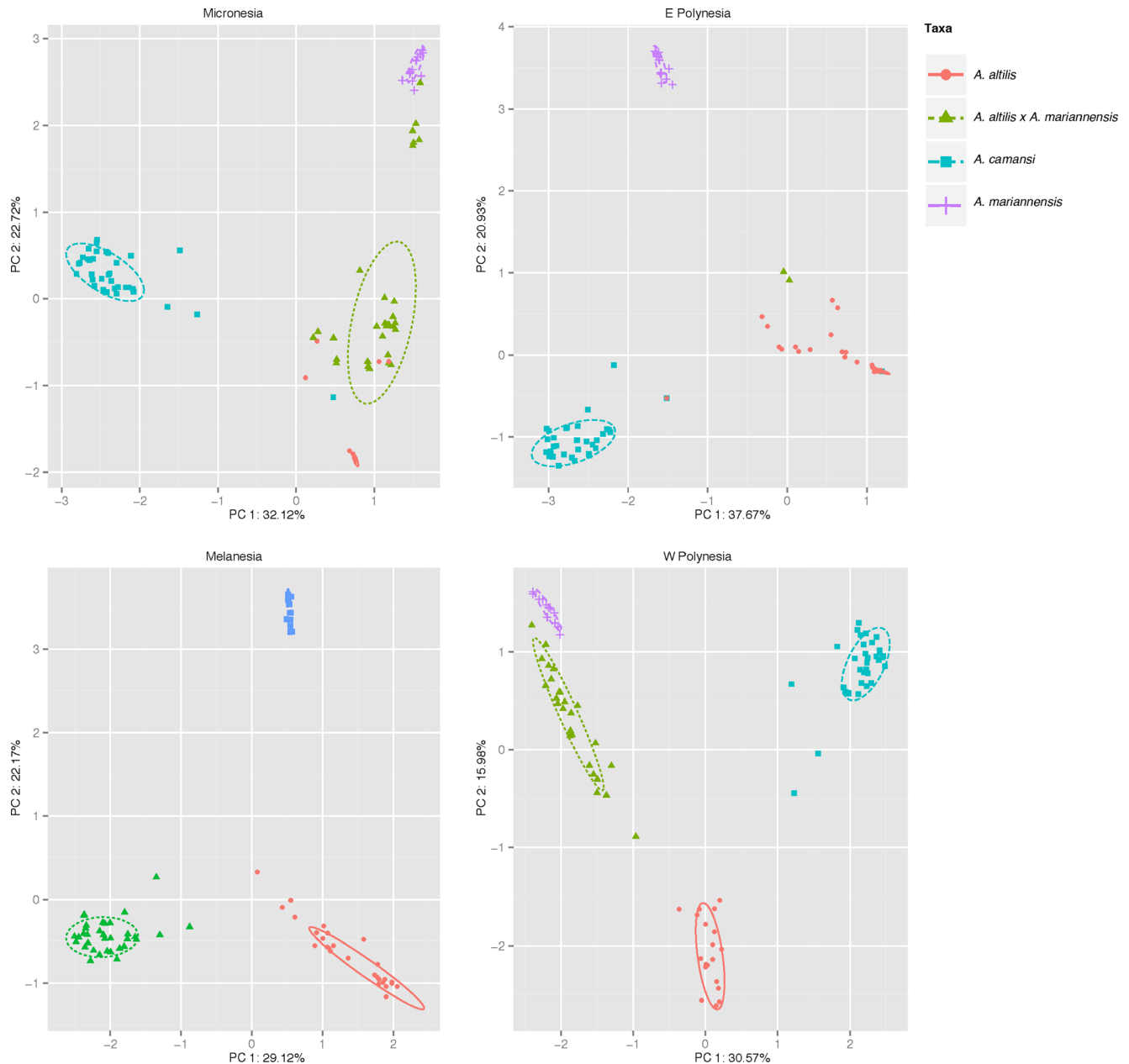


Fig. 5 PCA results based on data from 19 microsatellite loci of breadfruit, hybrids, and wild relatives from throughout Oceania. Samples were analyzed by region with *A. mariannensis* and *A. camansi*

included in each regional analysis. Taxon coding is indicated in the legend, and 95 % bivariate confidence interval ellipses are shown for taxon clustering

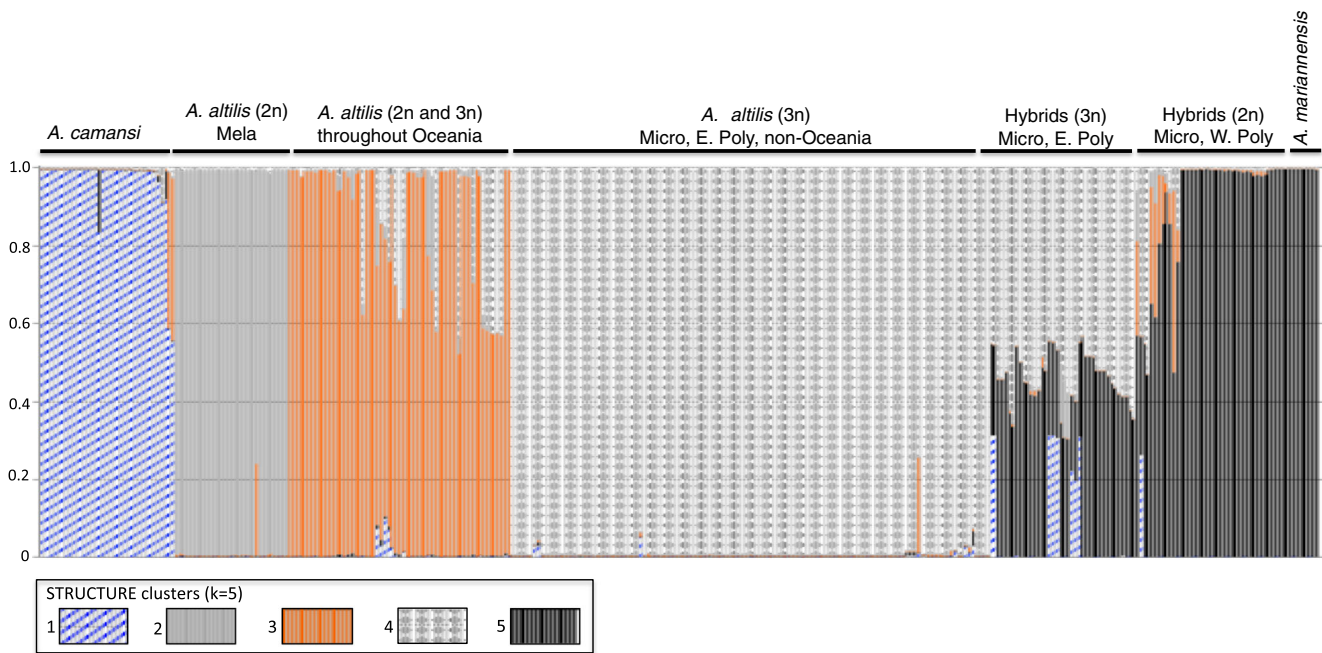


Fig. 6 Structure analysis of 349 samples of breadfruit, hybrids, and wild relatives based on data from 19 microsatellite loci. A total of five clusters ($K=5$) were identified as being the most likely to describe the data. Taxon names present in each cluster are indicated. In the case of breadfruit and hybrids, ploidy level and region(s) where the breadfruit came from are

also indicated. *Mela* Melanesia, *E. Poly* Eastern Polynesia, *W. Poly* Western Polynesia, *Micro* Micronesia. The five clusters are indicated below by color/pattern and assignment of individuals to a cluster is included in Table 1

was somewhat informative (Table 3), 51 of the 140 individuals shared the same zymotype profile. Zymotype profiles for overlapping individuals were less informative than microsatellites, as measured by both PIC and number of polymorphisms (Table 3).

Comparison of lineage and genotype groups across the same accession

Not all individuals of the same base accession number shared the same lineage or genotype group. This was especially true among the seed-propagated diploids, which never shared the same genotype, and frequently represented different lineage groups. This was expected given that the diploid accessions were largely seed-propagated and the product of sexual recombination. Triploids of the same accession nearly always shared the same lineage with other individuals of the same accession and predominantly shared the same genotype; however, there were a few exceptions. Accession NTBG 890160

was represented by two triploid individuals in this study, and they shared neither the same lineage nor genotype group. Two individuals of accession NTBG 890153 had different lineage groups, genotype groups, and ploidy levels. The diploid individual (NTBG 890153.001) shared the same lineage group and genotype with the cultivar Niue from the Cook Islands, and the triploid individual (890153.002) shared a lineage group with other Eastern Polynesian accessions, but had a unique genotype. This discrepancy was determined to be due to mislabeling and corrections have been made in the germplasm. There were four triploid individuals of accession NTBG 890480 (one of which is replicated as HART 52). Three of the 890480 individuals were triploid hybrids and shared identical lineage groups, and two of those also shared identical genotypes. The fourth individual (NTBG 890480.003) was also labeled as a hybrid, but had a different lineage group than the other three hybrids, and upon examination of the fruit, we determined that it was misidentified and the identification has been changed to *A. altilis*. Two

Table 3 Comparative utility of microsatellite, AFLP, and isozyme markers as measured by PIC and number of markers

Method	Number of primer pairs/enzyme systems	Average PIC	PIC range	Average number of markers	Range of number of markers
Microsatellites	19	0.627	0.007–0.820	11.68	2–24
AFLP	3	–	–	58.3	51–68
Isozymes	6	0.476	0.229–0.803	7.00	2–17

Microsatellite markers are alleles, AFLP markers are different fragment sizes, and isozyme markers are unique zymotypes

individuals of accession NTBG 910266 had different ploidy levels and this matched with data on seediness of their respective fruits (e.g., the diploid was seeded and the triploid was seedless), suggesting mislabeling. Two accessions of NTBG 900237 (one of which is replicated as HART 23) were both triploids and shared the same lineage group but have different genotypes.

There were some anomalies associated with microsatellite ploidy predictions and observed seediness. NTBG 900245.001 had three triploid loci and in the UPGMA tree it grouped with other triploids sharing its lineage and genotype group; however, it is recorded to occasionally produce seeds. Additionally, the same accession at HART had only diploid loci and produces seeds. NTBG 900236.001 had one triploid locus but produces many seeds and it clustered with other diploids in the UPGMA tree. The following accessions all had a single triploid locus, but they clustered with diploids in the UPGMA tree and are recorded as often seedless but they occasionally produce seeds: NTBG 030044.001, 770524.001, and 890476.002.

Discussion

Diversity characterization

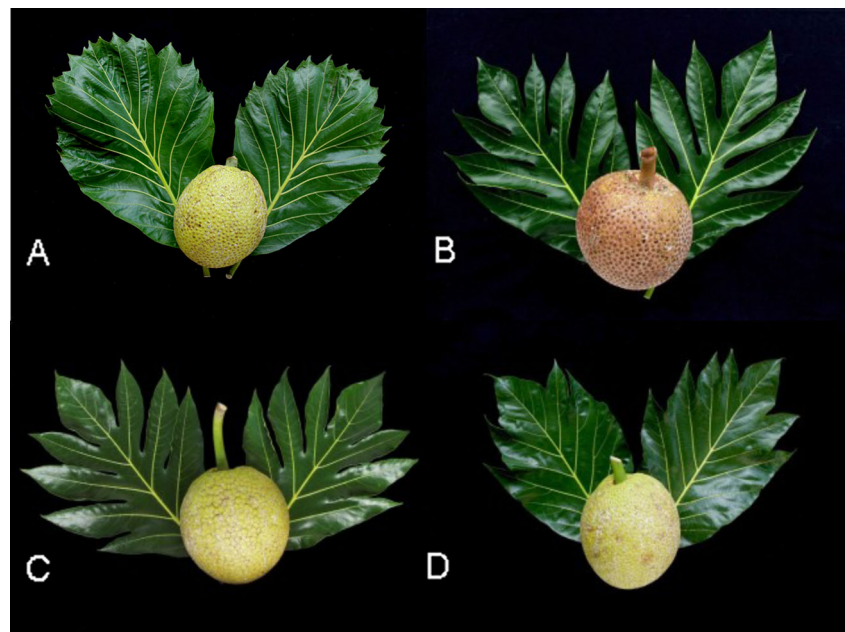
The wild relatives of breadfruit (*A. camansi* and *A. mariannensis*) harbor unique and valuable genetic diversity in breadfruit germplasm collections; among 49 wild individuals, 48 unique genotypes are represented. Nonetheless, the wild relatives display lower levels of genetic diversity than cultivated breadfruit (Table 2). This is likely due to variation in sample size and geographic distribution of the wild relatives housed in collections. There are only 14 *A. mariannensis* individuals, virtually all collected from the Northern Mariana Islands, and 36 *A. camansi* individuals, mostly collected from Papua New Guinea (PNG). *A. camansi* is believed to be native to the island of New Guinea and the Moluccas and has been introduced to other regions for its edible seeds. It is interesting to note that within the *A. camansi* cluster, all the PNG collections clustered together while the non-PNG collections clustered separately (Fig. 3). This suggests that there is variation among different regions and it will be important to expand the collections of these wild relatives. Consistent with previous research (Zerega et al. 2006), breadfruit samples from outside of Oceania displayed very little genetic diversity. Non-Oceanic samples included in this study came from Barbados and the Seychelles. The sample from Barbados belonged to the ubiquitous lineage 1/genotype 1 group that is prevalent throughout out Eastern Polynesia and Micronesia, so it is difficult to speculate specifically where it originally came from. However, most of the samples from the Seychelles belonged to the less common lineage 1/genotype 5, which

was also found in a few collections from the Society Islands and the Marquesas Islands, suggesting that they were originally introduced from one of those regions. This makes sense, given the historical role of the French in all of these regions.

The levels of genetic diversity in both diploid and triploid breadfruit and hybrids are higher than those reported for many annual crops and are comparable to levels found in several other perennial fruit tree crops like sweet cherry (*Prunus avium*, Mariette et al. 2010), cocoa tree (*Theobroma cacao*, Motilal et al. 2013), jackfruit (*Artocarpus heterophyllus*, Schnell et al. 2006; Witherup et al. 2013), mango (*Mangifera indica*, Hirano et al. 2010), walnuts (*Juglans*, Gunn et al. 2010), olives (*Olea europaea*, Trujillo et al. 2013), and others (Miller and Gross 2011). Given that long-lived perennials typically have lengthy juvenile phases, extensive outcrossing, widespread hybridization, and limited population structure compared to annuals, higher levels of genetic diversity is to be expected in perennial crops. Under domestication, these features of perennial fruit tree crops (combined with frequent clonal propagation, multiple origins, and ongoing gene flow between the crop and its wild relatives) typically contribute to more mild domestication bottlenecks in perennial fruit crops compared to annuals (Miller and Gross 2011). The diploid breadfruit cultivars and hybrids, which continue to undergo sexual reproduction, are more diverse and are readily distinguishable from one another, whereas some triploids are indistinguishable based on genetic characters. Even when the most stringent of criteria are used (i.e., classifying genotypes that differ by one mutation step as non-duplicates), many of the triploid accessions analyzed were found to be replicates (Table 1, Figs. 2 and 3). While identical microsatellite profiles are not proof of a clonal origin, they are at least indicative of a very close genetic relationship, given the high mutation rate of microsatellites (Lai and Sun 2003).

Some level of replication in germplasm collections provides a necessary safeguard, but it is also important to consider the most effective way to maximize diversity while minimizing redundancy and resources (i.e., personnel, land, funding). Genetic data can be used to help prioritize collections (Motilal et al. 2013); however, the results of this study need to be considered with caution. The vast majority of the replication is accounted for by a single genotype that is represented by 75 triploid individuals. Many individuals with this genotype display different fruit and leaf morphological characteristics that are apparent even when grown under the same conditions in a single field genebank (Fig. 7). This result is also common among vegetatively propagated seedless *Musa* cultivars (de Jesus et al. 2013; Irish et al. 2014). The markers in this study were unable to detect these differences at the genetic level, some of which may be due to somatic mutations that have occurred and were selected for over millennia of vegetative propagation. The inability to discriminate partial heterozygotes among the triploids may also lead to the apparent lack of genetic diversity across triploid accessions

Fig. 7 Morphological variation among triploid breadfruit with the ubiquitous lineage 1, genotype 1. **a** Meitehid, NTBG 030033.001, **b** Afara, NTBG 040051.001, **c** Meinuwe, NTBG 790497.002, and **d** Hamoa, NTBG 890154.001. Photos © Jim Wiseman



from throughout the Pacific Islands. Interestingly, Jones et al. (2011) were also unable to reliably distinguish many of these same accessions using morphological descriptors. Exploring the variance in nutritional content or seasonality within these genotype groups, for which some data already exist (Ragone and Cavaletto 2006; Jones et al. 2010, 2011, 2013), may provide insight into the range of phenotypic diversity and plasticity in breadfruit. Ultimately, phenotypic data should be integrated with the genetic data to evaluate the best way to capture both genetic and phenotypic diversity in breadfruit collections.

Genetic structure

Despite the inability to distinguish among many of the triploid accessions, the triploids display relatively high levels of heterozygosity (Table 2). Triploids have been vegetatively propagated for millennia and are essentially snapshots from the past. They capture the diversity of preferentially selected phenotypes, which were perpetuated over generations without genetic recombination. This is also apparent in the genetic structure that exists among breadfruit and its wild relatives. In UPGMA, PCA, and STRUCTURE analyses, *A. camansi* and *A. mariannensis* clearly represent distinct lineages and *A. altilis* and hybrids exhibit a degree of genetic structure (Figs. 3–6). Many accessions recognized as hybrids cluster between *A. altilis* and *A. mariannensis* in the UPGMA, PCA, and STRUCTURE analyses and have hybrid indices that indicate varying contributions from both parental species. Triploid Micronesian and Eastern Polynesian hybrids are clear admixtures of *A. mariannensis* and *A. altilis* (Fig. 6),

while diploid Western Polynesian and Micronesian hybrids have higher levels of contribution from *A. mariannensis*. In fact, some individuals appear to have nearly 100 % grouping with *A. mariannensis* (Fig. 6), although morphologically they share characters of both *A. mariannensis* and *A. altilis*. These diploid hybrids are likely the result of greater introgression with *A. mariannensis*. The Tokelau hybrids are known to be the result of fairly recent introductions dating back to the mid-nineteenth to early twentieth century when seeded *A. altilis* from Samoa and seeded *A. mariannensis* (and possibly hybrids) from Tuvalu (Ellice Islands, no samples included in study) were likely introduced. Thus, hybrids in Tokelau may have greater opportunity to cross back to *A. mariannensis*.

Among *A. altilis*, three very distinct clusters are associated with varying geographic regions and ploidy levels: (1) Melanesian diploids; (2) diploids and triploids from throughout Oceania; and (3) Micronesian, Eastern Polynesian, and non-Oceania triploids (Fig. 6). The latter group appears to be the most prevalent component of admixture in other clusters, suggesting that members of this group have been moved around by humans to a greater extent and have contributed the most to breadfruit diversity in other regions, especially the Micronesian hybrids. Interestingly, there is little overlap of the *A. camansi* group in any of the breadfruit or hybrids (Fig. 6), although it is thought to be the wild progenitor of non-hybrid breadfruit (Zerega et al. 2004, 2005). When hybrids were removed from the UPGMA analysis, however, there was a clear sister relationship between *A. altilis* and *A. camansi* (not shown).

Identification of unknowns and detection of errors

Of the 28 accessions lacking either a cultivar name or provenance information, likely cultivar names and/or areas of origin could be assigned to 21 of them as they shared genotypes or lineage groups with known accessions (Table 1). The remaining seven unknown accessions represent unique diversity within the collection and their provenance remains uncertain, emphasizing the value of germplasm even without provenance information.

Characterizing the genetic profiles of germplasm collections also allows for the identification of errors in labeling. As would be expected among vegetatively propagated triploids, most members of the same accession shared the same lineage group and typically the same genotype. However, some accessions did not meet these criteria as described in the results. Upon close examination of these trees, several were discovered to be mislabeled, an error that most likely occurred in the propagation facility. In addition, some individuals grouped with lineage groups or genotypes that were inconsistent with their provenance information. For example, NTBG 790487 and NTBG 790489 were labeled as *A. altilis* from the Society Islands; however, they grouped with lineage 5, which otherwise contained only hybrids from Pohnpei. Upon examination of these two accessions, it was determined that they are actually hybrids. Also, replication of accessions between collections may not always be properly recorded. For example, the triploid NTBG 890160 is housed at NTBG (Kahanu Garden, grid X8) and recorded as a hybrid, but it is different from another individual housed at HART (HART 37) where it is listed as *A. altilis* and recorded to have come from NTBG 890160. Appropriate corrections have been made (Table 1), and this points to the importance of genetic data in identifying such errors. It is worth noting that labeling and misidentification errors were minimal in the breadfruit collections.

Cultivar names

Shared names were not found to be reliable indicators of shared genotypes (Table 1). Many cultivar names were found to encompass only one genotype, while some genotypes encompassed multiple names. Even among sterile vegetatively propagated triploids, only eight names consistently shared the same genotype and the single most ubiquitous genotype shared by 75 triploid individuals encompassed 44 different names. This suggests that names are not systematically passed on and applied from island to island. These findings may be due to a variety of causes. Cultivar names for breadfruit in the Pacific Islands can vary widely across villages and islands and typically reflect morphology and preferred usage of the fruit and tree, rather than any strict lineage (Ragone 1991). The

disconnect between vernacular names and microsatellite profiles corroborates studies of high name variability (Ragone 1995, 1997; Ragone et al. 2004). In some areas, accessions are simply referred to by a generic name. For example, all *A. altilis* accessions referred to as “Ulu” were collected in Hawaii, while all *A. altilis* × *A. mariannensis* accessions referred to as “Ulu afa” (half-caste ulu) were collected in Tokelau (Table 1). In Samoa, diverse names are often indicative of the cultural and dietary importance of breadfruit, as well as how much traditional knowledge of breadfruit has been retained there, though knowledge of cultivar names varies widely among individuals (Ragone et al. 2004). The use of more generic names in some regions may be indicative of a loss of local knowledge in the uses or advantages of different cultivars or reflect the small base of cultivars for a particular island or island group. The existence of numerous synonyms for a given cultivar can cause problems when one attempts to compare results in different breadfruit-growing regions. This makes the collection and identification of breadfruit cultivars difficult and not only presents an obstacle in the communication and exchange of material but also makes the maintenance of collections very costly in terms of space, time, and money. The microsatellite markers could be used to address some of these problems.

Utility of microsatellites markers

In terms of discriminating among individuals, the microsatellite loci were much more informative than isozyme data (Ragone 1991) and slightly less informative than AFLP data (Zerega et al. 2005) (Table 3). Microsatellites have several key pragmatic advantages over AFLP and isozymes as molecular markers. Isozymes are highly tissue- and phenophase-specific and are generally not a practical high-information molecular marker (Moe et al. 2012). The codominant nature of microsatellites allows one to infer ploidy level with fairly high confidence (Hoshino et al. 2012). Microsatellite data are highly replicable and simple to use, making them desirable for analysis of large amounts of germplasm (Korir et al. 2013). For these reasons, microsatellites have been a standard platform for genetic fingerprinting and cultivar characterization in many other tree crops, including apple (Baric et al. 2010), pear (Wünsch and Hormaza 2007), poplar (Rajora and Rahman 2003), almond (Szikriszt et al. 2011), European plum (Xuan et al. 2011), peach (Wünsch et al. 2006), banana (de Jesus et al. 2013), and common fig (Giraldo et al. 2008). As next generation sequencing platforms and techniques are further developed, other genotypic markers (e.g., SNPs) will also play an important role in more detailed characterization of *Artocarpus* germplasm.

Conclusion

Ex situ germplasm collections serve a critical role in safeguarding and studying plant genetic resources. As a crop that must be preserved in a living field genebank, maintaining a breadfruit collection involves a significant commitment. Given limited space and other resources, making informed decisions about breadfruit germplasm management is essential. Understanding genetic diversity along with morphological and agronomic characters of a collection will aid in decisions regarding the selection of how to add novel diversity to the collection, breeding, or the reduction in duplicate accessions. Breadfruit's phenotypic plasticity and the variability of its uses and names (Ragone 1995; Ragone et al. 2004) mean that morphology, common names, or common usages are not always effective proxies for genetic diversity. There are several small collections (<10 accessions) scattered throughout the Pacific and Caribbean where provenance data are often not available, and many collections have been critically neglected (Ragone 2007). Particularly where accession data are missing or unreliable, the microsatellite markers and data presented here will be valuable in identifying unique accessions from these smaller collections that can be incorporated into larger, well-maintained, and documented collections such as at the NTBG and those of the NPGS and thereby fill gaps in these important collections. These tools will also be important for linking genetic, morphological, and agronomic data and understanding patterns of historical breadfruit distribution.

Acknowledgments The authors thank the Breadfruit Institute and the USDA/ARS National Plant Germplasm System for use of plant material, Ian Cole for collecting the samples at Kahanu Garden at NTBG, the Trustees and Fellows of NTBG for their support of the Breadfruit Institute, and two reviewers who provided valuable feedback that improved the manuscript. The research was made possible in part by National Science Foundation Grant DEB-0919119 and support from USDA/ARS and the Chicago Botanic Garden.

Conflict of interest The authors declare that they have no conflict of interest.

Data archiving statement Information on germplasm materials used in this study are publicly available online through NTBG (<http://ntbg.org/breadfruit/database>) and the ARS's GRIN databases (http://www.ars-grin.gov/npgs/acc/acc_queries.html).

References

- Adebowale KO, Olu-Oqolabi BI, Olawumi EK, Lawal OS (2005) Functional properties of native, physically and chemically modified breadfruit (*Artocarpus altilis*) starch. *Ind Crops Prod* 21:343–351
- Baric S, Wagner J, Storti A, Dalla Via J (2010) Application of an extended set of microsatellite DNA markers for the analysis of presumed synonym cultivars of apple. *Acta Hort* 918:303–308
- Botstein D, White RL, Skolnick M, Davis RW (1980) Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *Am J Hum Genet* 32:314–331
- Bruvo R, Michiels NK, D'Souza TG, Schulenburg H (2004) A simple method for the calculation of microsatellite genotype distances irrespective of ploidy level. *Mol Ecol* 13:2101–2106. doi:10.1111/j.1365-294X.2004.02209.x
- Buerkle CA (2005) Maximum-likelihood estimation of a hybrid index based on molecular markers. *Mol Ecol Notes* 5:684–687
- Clark LV, Jasieniuk M (2011) POLYSAT: an R package for polyploid microsatellite analysis. *Mol Ecol Resour* 11:562–566. doi:10.1111/j.1755-0998.2011.02985.x
- Creste S, Neto AT, Vencovsky R, De OSilva S, Figueira A (2004) Genetic diversity of *Musa* diploid and triploid accessions from the Brazilian banana breeding program estimated by microsatellite markers. *Genet Resour Crop Evol* 51:723–733
- de Jesus ON, de Oliveira S, Amorim EP, Ferreira CF, Salabert JM, de Campos, de Gaspari-Silva G, Figueira A (2013) Genetic diversity and population structure of *Musa* accessions in ex situ conservation. *BMC Plant Biol* 13:41. doi:10.1186/1471-2229-13-41
- De Silva HN, Hall AJ, Rikkerink E, McNeilage MA, Fraser LG (2005) Estimation of allele frequencies in polyploids under certain patterns of inheritance. *Heredity* 95:327–334. doi:10.1038/sj.hdy.6800728
- Esselink GD, Nybom H, Vosman B (2004) Assignment of allelic configuration in polyploids using the MAC-PR (microsatellite DNA allele counting-peak ratios) method. *Theor Appl Genet* 109:402–408. doi:10.1007/s00122-004-1645-5
- Evanno G, Regnaut S, Goudet J (2005) Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. *Mol Ecol* 14(8):2611–20
- Falush D, Stephens M, Pritchard JK (2007) Inference of population structure using multilocus genotype data: dominant markers and null alleles. *Mol Ecol Notes* 7:574–578
- Felsenstein J (2005) PHYLIP (Phylogeny Inference Package) version 3.6. Distributed by the author. Department of Genome Sciences, University of Washington, Seattle.
- Fosberg FR (1960) Introgression in *Artocarpus* in Micronesia. *Brittonia* 12:101–113
- Fownes JH, Raynor WC (1991) Seasonality and yield of breadfruit cultivars in the indigenous agroforestry system of Pohnpei, Federated States of Micronesia. *Trop Ag (Trinidad)* 70(2):103–9
- Ghislain M, Spooner DM, Rodriguez F, Villamon F, Nuñez J, Vásquez C, Waugh R, Bonierbale M (2004) Selection of highly informative and user-friendly microsatellites (SSRs) for genotyping of cultivated potato. *Theor Appl Genet* 108:881–890. doi:10.1007/s00122-003-1494-1497
- Giraldo E, Lopez-Corales M, Hornaza JI (2008) Optimization of the management of an ex-situ germplasm bank in common fig with SSRs. *J Am Soc Hort Sci* 133(1):69–77
- Gunn BF, Aradhya M, Salick JM, Miller AJ, Yang YP, Liu L, Hai X (2010) Genetic variation in walnuts (*Juglans regia* L. and *J. sigillata* Dode, Juglandaceae): species distinctions, human impacts, and the conservation of agrobiodiversity in Yunnan, China. *Am J Bot* 97:60–671
- Hardy OJ, Vekemans X (2002) SPAGeDi: a versatile computer program to analyse spatial genetic structure at the individual or population levels. *Mol Ecol Notes* 2:618–620. doi:10.1046/j.1471-8278.2002.00305.x
- Hirano R, Oo TH, Watanabe KN (2010) Myanmar mango landraces reveal genetic uniqueness over common cultivars from Florida, India, and Southeast Asia. *Genome* 53:321–330
- Hoshino AA, Bravo JP, Nobile PM, Morelli KA (2012) Microsatellites as tools for genetic diversity analysis. In: Caliskan M (ed) Genetic diversity in microorganisms. In Tech, Croatia, pp 149–170
- Irish BM, Cuevas HE, Simpson SA, Scheffler BE, Sardos J, Ploetz R, Goenaga R (2014) *Musa* spp. germplasm management:

- microsatellite fingerprinting of USDA–ARS national plant germplasm system collection. *Crop Sci* 54:2140–2151
- Jones AMP, Murch SJ, Ragone D (2010) Diversity of breadfruit (*Artocarpus altilis*, Moraceae) seasonality: a resource for year-round nutrition. *Econ Bot* 64(4):340–351
- Jones AMP, Ragone D, Aiona K, Lane WA, Murch SJ (2011) Nutritional and morphological diversity of breadfruit (*Artocarpus*, Moraceae): identification of elite cultivars for food security. *J Food Compos Anal* 24:1091–1102. doi:10.1016/j.jfca.2011.04.002
- Jones AMP, Baker R, Ragone D, Murch SJ (2013) Identification of provitamin A carotenoid-rich cultivars of breadfruit (*Artocarpus*, Moraceae). *J Food Compos Anal* 31(1):51–61. doi:10.1016/j.jfca.2013.03.003
- Korir NK, Li Y, Leng XP, Wu Z, Wang C, Fan JG (2013) A novel and efficient strategy for practical identification of tomato (*Solanum lycopersicon*) varieties using modified RAPD fingerprints. *Genet Mol Res* 12(2):1816–1828
- Lai Y, Sun F (2003) The relationship between microsatellite slippage mutation rate and the number of repeat units. *Mol Biol Evol* 20(12):2123–2131
- Leakey CLA, (1977) Breadfruit reconnaissance study in the Caribbean region. CIAT/InterAmerican Development Bank
- Mariette S, Tavaud M, Arunyawat U, Capdeville G, Millan M, Salin F (2010) Population structure and genetic bottleneck in sweet cherry estimated with SSRs and the gametophytic self-incompatibility locus. *BMC Genet* 11:77. doi:10.1186/1471-2156-11-77
- Markham C (1904) The voyages of Pedro Fernandez de Quiros. Hakluyt Society, London, pp 1595–1606, Vol. I
- McGregor CE, Lambert CA, Greyling MM, Louw JH, Warnich L (2000) A comparative assessment of DNA fingerprinting techniques (RAPD, ISSR, AFLP and SSR) in tetraploid potato (*Solanum tuberosum* L.) germplasm. *Euphytica* 113:135–144
- Meimans PG, Van Tienderen PH (2004) Genotype and genotype: two programs for the analysis of genetic diversity of asexual organisms. *Mol Ecol Notes* 4:792–794. doi:10.1111/j.1471-8286.2004.00770.x
- Mengoni A, Gori A, Bazzicalupo M (2000) Use of RAPD and microsatellite (SSR) variation to assess genetic relationships among populations of tetraploid alfalfa, *Medicago sativa*. *Plant Breed* 119:311–317
- Miller A, Gross BL (2011) From forest to field: perennial fruit crop domestication. *Am J Bot* 98(9):1389–414
- Moe KT, Kwon SW, Park YJ (2012) Trends in genomic and molecular marker systems for the development of some underutilized crops. *Genes Genom* 34:451–466
- Morton JF (1990) Under-exploited fruit-vegetables can enhance the world food supply. *Acta Hort* 275:401–408
- Motilal LA, Zhang D, Mischke S, Meinhardt LW, Umaharan P (2013) Microsatellite-aided detection of genetic redundancy improves management of the international Cocoa Genebank, Trinidad. *Tree Genet Genomes* 9:1395–1411
- Omubuwajo TO (2007) Breadfruit as a key component of sustainable livelihoods in Nigeria: prospects, opportunities and challenges. *Acta Hort* 757:121–124
- Powell D (1977) Voyage of the plant nursery, HMS providence. *Econ Bot* 31:387–431, 1791–1793
- Pritchard JK, Stephens M, Donnelly P (2000) Inference of population structure using multilocus genotype data. *Genetics* 155(2):945–59
- Ragone D (1991) Collection, establishment and evaluation of a germplasm collection of Pacific Island breadfruit. Dissertation, University of Hawaii
- Ragone D (1995) Description of Pacific Island breadfruit cultivars. *Acta Hort* 413:93–98
- Ragone D (1997) Breadfruit: *Artocarpus altilis* (Parkinson) Fosberg. Promoting the conservation and use of underutilized and neglected crops 10. IPGRI, Inter Plant Genetic Resources Inst, Rome
- Ragone D (2001) Chromosome numbers and pollen stainability of three species of Pacific Island breadfruit (*Artocarpus*, Moraceae). *Am J Bot* 88(4):693–696
- Ragone D (2007) Breadfruit: diversity, conservation, and potential. *Acta Hort* 757:19–30
- Ragone D, Cavaletto CG (2006) Sensory evaluation of fruit quality and nutritional composition of 20 breadfruit (*Artocarpus*, Moraceae) cultivars. *Econ Bot* 60(5):335–346
- Ragone D, Wiseman J (2007) Developing and applying descriptors for breadfruit germplasm. *Acta Hort* 757:71–80
- Ragone D, Tavana G, Stevens JM, Stewart PA, Stone R, Cox PM, Cox PA (2004) Nomenclature of breadfruit cultivars in Samoa: saliency, ambiguity, and monomiality. *J Ethnobiol* 24(1):33–49
- Rajora OP, Rahman MH (2003) Microsatellite DNA and RAPD fingerprinting, identification and genetic relationships of hybrid poplar (*Populus* × *canadensis*) cultivars. *Theor Appl Genet* 106:470–477
- Roberts-Nkrumah LB (2007) An overview of breadfruit (*Artocarpus altilis*) in the Caribbean. *Acta Hort* 757:51–60
- Schnell RJ, Brown JS, Olano CT, Meerow AW, Campbell RJ, Khun DN (2006) Mango genetic diversity analysis and pedigree inferences for Florida cultivars using microsatellite markers. *J Am Soc Hort Sci* 131(2):214–224
- Szikriszt B, Hegedüs A, Halász J (2011) Review of genetic diversity studies in almond (*Prunus dulcis*). *Acta Agronom* 59(4):379–395
- Taylor M, Kete T, Tuia V (2009) Underutilized species in the Pacific: an untapped source of nutritional and economic wealth. *Acta Hort* 806:235–240
- Tomiuk J, Guldbrandtsen B, Loeschke V (2009) Genetic similarity of polyploids: a new version of the computer program POPDIST (version 1.2.0) considers intraspecific genetic differentiation. *Mol Ecol Resour* 9:1364–1368. doi:10.1111/j.1755-0998.2009.02623.x
- Trujillo I, Ojeda MA, Urdiroz NM, Potter D, Barranco D, Rallo L, Diez CM (2013) Identification of the Worldwide Olive Germplasm Bank of Córdoba (Spain) using SSR and morphological markers. *Tree Genet Genomes*. doi:10.1007/s11295-013-0671-3
- Wickham H (2009) ggplot2: elegant graphics for data analysis. Springer, New York
- Wilder GP (1928) Breadfruit of Tahiti. B.P. Bishop museum. Bulletin 50, Honolulu
- Witherup C, Ragone D, Wiesner-Hanks T, Irish B, Scheffler B, Simpson, Zee F, Zuberi MI, Zerega NJC (2013) Development of microsatellite loci in *Artocarpus altilis* (Moraceae) and cross-amplification in congeneric species. *Appl Plant Sci* 1(7):1200423. doi:10.3732/apps.1200423
- Wootton M, Tumaalii F (1984) Breadfruit production, utilisation and composition—a review. *Food Tech* 37(10):464–465
- Wünsch A, Hormaza JI (2007) Characterization of variability and genetic similarity of European pear using microsatellite loci developed in apple. *Scientia Hort* 113(1):37–43
- Wünsch A, Carrera M, Hormaza JI (2006) Molecular characterization of local Spanish peach [*Prunus persica* (L.) Batsch] germplasm. *Genet Resour Crop Ev* 53:925–932
- Xuan H, Ding Y, Spann D, Möller O, Büchele M, Neumüller M (2011) Microsatellite markers (SSR) as a tool to assist in identification of European plum (*Prunus domestica*). *Acta Hort* 918:689–692
- Zerega NJC, Ragone D, Motley TJ (2004) Complex origins of breadfruit (*Artocarpus altilis*, Moraceae): implications for human migrations in Oceania. *Am J Bot* 91(5):760–6
- Zerega NJC, Ragone D, Motley TJ (2005) Systematics and species limits of breadfruit (*Artocarpus*, Moraceae). *Sys Bot* 30:603–15
- Zerega NJC, Ragone D, Motley TJ (2006) Breadfruit origins, diversity, and human-facilitated distribution. In: Motley TJ, Zerega NJC, Cross H (eds) Darwin's harvest: new approaches to the origins, evolution, and conservation of crops. Columbia University Press, New York, pp 213–238