## Supplementary material

- **Table S1.** Details for the populations of *Corymbia hamersleyana* (*n*=20) sampled across the Pilbara region of north-western Australia.
- 3 Haplotype ID, Haplotype diversity and Genbank accession numbers shown. Sequence alignment accession: 10.6084/m9.figshare.11846940

Location	Pop.	Latitude	Longitude	Hap #	Hap. diversity	Genbank accession rpl16	ndhC-trnV	trnG
Bennett	BEN	22° 57' 33.7" S	117° 51' 52.2" E	1,2	0.571	MN539542	MN539548	MN539557;MN539558
Coppin	COP	20° 55' 17.5" S	119° 58' 49.8" E	3,4	0.25	MN539543	MN539549	MN539559;MN539560
Dawson	DAW	21° 34' 34.3" S	117° 12' 47.0" E	3,5	0.536	MN539542:MN539543	MN539549;MN539550	MN539557;MN539559
Deepdale	DEE	21° 38' 33.0" S	116° 05' 18.2" E	6,7	0.429	MN539542;MN539543;MN539544	MN539549;MN539551;MN539552	MN539557
Dinner	DIN	22° 34' 32.3" S	118° 18' 03.3" E	1,3	0.25	MN539542	MN539548	MN539557;MN539559
East Munjina	EMU	22° 29' 22.5" S	118° 44' 10.3" E	1,3	0.25	MN539542;MN539543	MN539548;MN539549	MN539557;MN539559
Gallop	GAL	21° 26' 53.4" S	120° 04' 24.8" E	1,3	0.25	MN539542;MN539543	MN539548;MN539549	MN539557;MN539559
Gidge	GID	22° 29' 19.8" S	119° 01' 09.5" E	1,3,5	0.679	MN539542;MN539543	MN539548;MN539549;MN539550	MN539557;MN539559
Hamersley	HAM	22° 15' 10.5" S	117° 40' 24.7" E	1,3,8	0.679	MN539542;MN539543	MN539548;MN539549;MN539553	MN539557;MN539559
Hooley	HOO	21° 49' 58.3" S	117° 55' 39.6" E	1,3	0.429	MN539542;MN539543;MN539545	MN539548;MN539549	MN539557;MN539559
Kangan	KAN	21° 02' 06.2" S	118° 39' 45.3" E	1,7,9	0.607	MN539542;MN539546	MN539548;MN539552;MN539554	MN539557;MN539561
Karratha	KAR	20° 44' 34.6" S	116° 52' 19.8" E	10	0.25	MN539542	MN539548	MN539557;MN539562
McKay	MCK	22° 25' 16.9" S	119° 59' 29.4" E	1,3,11	0.607	MN539542;MN539543;MN539545	MN539548;MN539549;MN539555	MN539557;MN539559
Minnie	MIN	21° 55' 48.9" S	115° 16' 23.1" E	1	0	MN539542	MN539548;MN539551	MN539557
Nullagine	NUL	21° 53' 19.6" S	120° 05' 55.6" E	1,3,11	0.607	MN539542;MN539543	MN539548;MN539549;MN539555	MN539557;MN539559
Ord	ORD	20° 19' 36.2" S	119° 10' 36.2" E	1	0	MN539542	MN539548	MN539557
Pamelia	PAM	23° 09' 24.7" S	119° 20' 55.5" E	1,3	0.429	MN539542;MN539543	MN539548;MN539549	MN539557;MN539559
Rubin	RUB	20° 23' 58.1" S	119° 58' 45.5" E	3,12	0.536	MN539543;MN539546	MN539549	MN539557
Shaw	SHA	20° 42' 35.1" S	119° 19' 06.2" E	5,9,13, 14	0.75	MN539542;MN539546;MN539547	MN539550;MN539554;MN539555;MN539556	MN539557;MN539561
Weeli Wolli	WEE	22° 55' 32.6" S	119° 11' 49.7" E	1,2,3	0.464	MN539542;MN539543	MN539548;MN539549	MN539557;MN539558

Location	Pop.	Latitude	Longitude	Hap #	Hap. diversity	Genbank accessionndhF-rpl32	rpl32-trnL	trnS-trnG	psbD-trnT
Angelas	ANG	23° 05' 05.9" S	118° 41' 31.1" E	1,2,3,4	0.821	MN539563	MN539575;MN539576; MN539577	MN539585;MN539586; MN539587;MN539588	MN539603
Bungaroo	BUN	21° 51' 52.9" S	116° 26' 18.7" E	4,5,6,7	0.786	MN539563;MN539564	MN539576;MN539577	MN539588;MN539589	MN539603
Coondiner	COO	22° 42' 46.6" S	119° 41' 03.9" E	8,9,10	0.714	MN539563;MN539565	MN539577;MN539578	MN539588;MN539590	MN539603;MN539604
Corbay	COR	21° 57' 56.1" S	118° 03' 05.9" E	8,11,12	0.607	MN539563;MN539566; MN539567	MN539577;MN539578	MN539590;MN539591	MN539603;MN539604
Hamersley	HAM	22° 15' 10.5" S	117° 40' 24.7" E	4,8,13	0.571	MN539563	MN539576;MN539578; MN539579	MN539588;MN539590; MN539592	MN539603
Hardy	HAR	22° 57' 14.2" S	117° 18' 39.6" E	4	0	MN539563	MN539576	MN539588	MN539603
Harper	HRP	21° 52' 16.3" S	117° 37' 15.5" E	8	0	MN539563	MN539578	MN539590	MN539603
Hesta	HES	22° 13' 31.7" S	119° 00' 53.6" E	8,14,15,16	0.714	MN539563;MN539568; MN539569	MN539578;MN539580; MN539581	MN539590;MN539593	MN539603
Hillditch	HIL	23° 13' 41.3" S	118° 45' 32.6" E	7,9,17,18	0.821	MN539563;MN539570	MN539577	MN539588;MN539590; MN539594	MN539603;MN539604
Mesa G	MSJ	21° 39' 10.2" S	116° 08' 07.5" E	22,23	0.429	MN539563;MN539571	MN539576;MN539584	MN539588;MN539595	MN539605
Metawandy	MET	22° 40' 10.3" S	116° 36' 07.9" E	7,18,19,20,21	0.857	MN539563	MN539577;MN539581; MN539582;MN539583	MN539588;MN539590; MN539594	MN539603
Nameless	NAM	22° 43' 38.7" S	117° 44' 57.5" E	3,24,25	0.679	MN539563	MN539576;MN539577	MN539587;MN539588; MN539596	MN539606;MN539607
Ophthalmia	OPH	23° 20' 33.2" S	119° 49' 51.4" E	8,10,26,27	0.643	MN539563;MN539565	MN539577;MN539578; MN539581;MN539582	MN539588;MN539590; MN539592;MN539594	MN539604
Oxer	OXE	22° 34' 33.3" S	118° 13' 43.1" E	3,4,28,8	0.607	MN539563	MN539576;MN539577; MN539578	MN539587;MN539588; MN539590;MN539597	MN539603
Rhodes	RHO	23° 03' 18.4" S	119° 15' 30.2" E	7,31	0.571	MN539563;MN539572	MN539577;MN539578	MN539588;MN539590	MN539603
Rio	RIO	22° 11' 38.6" S	117° 57' 19.9" E	4,18	0.536	MN539563;MN539573	MN539576;MN539577	MN539588;MN539590	MN539603
Roy Hill	RHL	22° 23' 52.1" S	119° 59' 27.9" E	8,9,29,30	0.786	MN539563	MN539577;MN539578	MN539588;MN539590; MN539598;MN539599	MN539603;MN539604
Spearhole	SPE	23° 21' 30.9" S	119° 06' 41.2" E	3,18	0.571	MN539563	MN539577	MN539587;MN539600	MN539603
Stewart	STE	21° 57' 16.1" S	119° 33' 36.5" E	8	0	MN539563	MN539578	MN539590	MN539603
Warrawanda	WAR	23° 43' 56.9" S	119° 43' 35.7" E	10,18,32,33	0.714	MN539563;MN539565; MN539569	MN539577	MN539587;MN539588; MN539590;MN539600	MN539603
Western Range	WRA	23° 10' 19.1" S	117° 26' 06.7" E	4,34	0.714	MN539563;MN539574	MN539576	MN539588	MN539603

6 **Table S2.** Details for the populations of *Acacia pruinocarpa* (*n*= 23) sampled across the Pilbara region of north-western Australia. Haplotype

number, Haplotype diversity and Genbank accession numbers shown. Sequence alignment accession: 10.6084/m9.figshare.11846937

	Yampire	YAM	22° 27' 18.6" S	118° 27' 22.1" E	13,35,36	0.429	MN539563;MN539566	MN539578;MN539579	MN539590;MN539601	MN539603	
	Yarrintree	YAR	22° 36' 49.8" S	118° 42' 18.7" E	8,9,18,37	0.75	MN539563	MN539577;MN539578	MN539590;MN539595; MN539602	MN539603	-
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22 Table S3. Primer sequences and characteristics of microsatellite loci developed for Corymbia hamersleyana (CH) and Acacia pruinocarpa (AP).

23 Loci were used to genotype 24 individuals from 20 populations. Values indicate population mean. N, number of individuals analysed, NA, allelic

24 richness, Ho, observed heterozygosity, HE, expected heterozygosity, FIS, inbreeding coefficient. Annealing temperature for all loci was 60°C.

25 Genotype table accession C.hamersleyana: 10.6084/m9.figshare.11846904; A. pruinocarpa: 10.6084/m9.figshare.11846952

Locus	Primer Sequence (5'-3')	Repeat motif	Size (bp)	Ν	NA	Но	HE	Fis
CH1	F: TGTTCGCAAGAGCTTCATGT	$(\mathbf{A}_{\mathbf{C}})$	02.00	22.45	2.2	0.00	0.2(0	0.022
CHI	R: TTGCTAATGTCCCAAGGAGAA	(AG)5	82-98	23.45	3.3	0.26	0.209	0.055
CH6	F: CTGGTCCGAGACTGAAGCAT	$(\mathbf{A}\mathbf{C})$	106 110	24	2 45	0.006	0.110	0 196
	R: ATTTCTCCACGAACCCGAC	(AU)7	100-118	24	2.43	0.090	0.118	0.180
CUO	F: AAGACCCATCTCCACAGTCG	$(\mathbf{A}\mathbf{C})$	120 100	22.05	12 15	0.626	0.956	0.257
СПУ	R: GGAGCACACCCAACATCTCT	(AU)7	126-198	23.03	15.15	0.030	0.830	0.237
CUIA	F: GCCATGCTTATTCGGCTTT	$(\mathbf{A}_{\mathbf{C}})$	120 126	23.8	2.95	0.372	0.395	0.057
CHIU	R: CCGACGTCTTCGTCTTCACT	(AU) <sub>6</sub>	130-130					0.057
CH11	F: TGCTCGATCATCCGACTATG	$(\mathbf{A}_{\mathbf{C}})$	120 177	22	12.05	0.685	0.966	0.200
	R: TTAGGGCTTTCCAGAGCAAA	(AU)9	130-177	23			0.800	0.209
01114	F: AAGGTCAAGCATGGACTTTCA	$(\Lambda C)$	147 152	24	2	0.121	0.129	0.059
CH14	R: ATTCACGAGTTCCCATCTGC	(AC)5	14/-135				0.128	
CU17	F: TCGTTGCCGACATAAGTCCT	$(\mathbf{A},\mathbf{C})$	149-167	23.95	3.55	0.474	0.487	0.027
CHI/	R: AAACGCTCCGAGCTCTTTCT	(AG) <sub>7</sub>						0.027
CU10	F: AATCTTGAGCATCGCCAATC	$(\Lambda CC)$	1(1 107	24	5.65	0.750	0.00	0.102
СПІб	R: CAGGAATCTCGCCATCAACT	(AGG)7	101-185			0.732	0.082	-0.105
CUM	F: GTTATACCGTTCACGCCACC	$(\Lambda C)$	101 227	22 75	12.7	0.010	0.854	0.041
СП22	R: CAGCCAGGTCTCGAAGTAGG	$(AO)_{10}$	181-227	25.15	12.7	0.818	0.834	0.041
C1125	F: GCGTCGAAGAAGATGACGAT		250 200	23.75	4.65	0.467	0.521	0 122
СНЭЭ	R: CGGTAGCACTGCTTTCCAA	(ACG)8	258-288				0.551	0.122
C1120	F: ATCGTGGTCGCTCTTCATCT	$(\Lambda C)$	100 120	22.4	0.45	0.704	0.007	0.140
CH39	R: TTGTCTCCGCCCATGATAA	(AG) <sub>12</sub>	100-130	23.4	9.45	0.704	0.827	0.148

CH42	F: ATGTCATCAACGCGTCTTGT	$(\mathbf{A} \mathbf{C})$	125 157	10.45	5 5	0.100	0 6 4 4	0.601
	R: TCCAGTATCCAGATTATGCGG	(AC) <sub>8</sub>	155-15/	19.43	5.5	0.199	0.044	0.091
CII47	F: TTGTGCCTCTGTTTGCTGTT		101 220	22.25	0.2	0.256	0.705	0 546
СП4/	R: CCTTGAATTCAGAGCTTGGG	(AU) <sub>11</sub>	191-229	22.23	8.5	0.550	0.785	0.340
CH51	F: GCCGAATCCTAGGACAGAGA	$(\mathbf{A}\mathbf{C})$	200 242	22.6	10.45	0.900	0.022	0.016
	R: TGTATCCAAACATCGAAAGGC	(AG)9	299-343	23.0	10.45	0.809	0.825	0.016
A D05	F: GCTTCTCCTGTTGCTTGTCC	$(\Lambda \Lambda C)$	106 114	<u></u>	17	0.060	0.069	0.016
AP05	R: TTGTGGACCTCTTGGTCTGG	$(AAC)_6$	100-114	23.22	1./	0.069	0.068	-0.016
A D 1 1	F: TGTTGTCCATACCTGAGGGC	$(\mathbf{AC})$	115 114	<u></u>	1 97	0 429	0.650	0.251
AFII	R: GGTCCGCTTTCCCATTCTAT	(AG)9	115-144	23.22	4.8/	0.428	0.039	0.331
A D1 4	F: TGGATCAATCCTGTTACCCA	$(\Lambda \Lambda C)$	124 140	<u></u>	2.06	0.518	0.408	0.04
AI 14	R: GCATCCACTTGGACAGATCC	(AAO) <sub>6</sub>	134-149	23.22	2.90	0.318	0.498	-0.04
AP15	F: GGCAAGCAACTCTTACCGAA	$(\Lambda CT)$	144 150	<u>, , , , , , , , , , , , , , , , , , , </u>	2.04	0.068	0.452	0.207
	R: ACATTCCGTGAATCGAAAGG	$(ACI)_6$	144-150	23.22	2.04	0.008	0.432	0.207
AP21	F: GCGATGAATATCGTCGAACA	$(\Lambda G)_{in}$	171-201	23 13	017	0.844	0.823	-0.025
	R: CCCTTCCTTTGCCCAACTAT	$(AU)_{10}$	1/1-201	23.13	9.17	0.844	0.823	-0.023
A <b>D</b> 37	F: CTTCTCCCATTCTTTGCTCG	$(\Lambda \Lambda G)_{c}$	86-107	22.04	3 13	0.417	0.416	-0.003
AI 57	R: TTACTATGCCCTCCTGCCTG	(AAO)6	80-107	22.04	5.45	0.417	0.410	-0.003
A <b>D</b> 38	F: GCTTATCTTTGATGCAGGGC	$(\Lambda G)_{ii}$	03-117	23 13	6 78	0.608	0.741	0.18
AI 30	R: TTGGTGGACCCACTGATGT	(AU)II	<i>yy</i> <b>-</b> 11 <i>1</i>	23.13	0.78	0.008	0.741	0.10
A P 3 9	F: GATTCACTGTCCGGCCTCT	(AG) <sub>e</sub>	104-110	23.22	27	0.252	0 249	-0.012
111 37	R: AGTATAACGCCGTTGGCAAT	(110)8	107-110	<i>23.22</i>	2.1	0.232	0.277	0.012
A P43	F: AAATGCGATGTTGGATGAGG	(AAG)	122-140	22.96	3 57	0 314	0 554	0.433
1 11 TJ	R: TTGACAAGTCCAAGGAACCC	(1110)6	122 170	22.70	5.51	0.217	0.557	0.TJJ
AP44	F: GAACCCACAAGGTTCTCCAA	(AGG)₄	131-134	23.22	17	0 089	0.093	0 049
111 77	R: CCAACACGTAGGGATGGAGT	(2100)6	151-154	<i>23.22</i>	1./	0.007	0.075	0.047
A P48	F: GCTTGGTATCAACGCAGATG	$(AAT)_{12}$	138-190	21 43	8 78	0 461	0 798	0 422
111 TU	R: CTCCCAACAAGTGCTCTTCC	(1111)13	150-170	21.TJ	0.70	0.701	0.790	0.722
AP52	F: GGTCTGTCGTTGTTGCTGAG	$(AC)_7$	190-200	23.22	3 87	0.538	0 548	0.018
AF32	R: ACCCAATATCGGAAACACCA		170-200		5.07	0.550	0.570	0.010

A D56	F: CCGACCGAAACTTGAGCTAC	$(\mathbf{A}\mathbf{C})$	106 222	22.22	0.06	0 776	0.010	0.051
AP56	R: CGGTGGCCACTATCACTGTA	$(AC)_{16}$ 1	190-232	23.22	9.90	9.90 0.770 0.818	0.051	
	F: GCTTTCGACAGTTCTCAGGA		269.279	22.22	2 20	0.275	0 272	0.000
AP60	R: CTCCCAAGGAAGAACAACCA	(AG)7	268-278	23.22	5.39	0.375	0.373	-0.006

## **Table S4.** Estimates of global Fst of Weir (1996) both using and without using the ENA correction described in Chapuis and Estoup (2007).

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	<i>Corymbia</i>	hamersleyana	Acacia	pruinocarpa
locus	ENA	FST with ENA	ENA	FST with ENA
1	0.024953	0.033828	0.068962	0.080862
2	0.037304	0.034806	0.051368	0.052736
3	0.063402	0.063841	0.053933	0.056890
4	0.039019	0.035099	0.029996	0.042152
5	0.039403	0.038888	0.076366	0.074141
6	0.038115	0.035858	0.064094	0.063249
7	0.043063	0.058053	0.086067	0.074754
8	0.005898	0.035318	0.050109	0.049813
9	0.053859	0.052749	0.076608	0.077406
10	0.113549	0.108202	0.053923	0.049598
11	0.032972	0.033751	0.062431	0.062763
12	0.061974	0.049563	0.070483	0.069669
13	0.047541	0.037192	0.039838	0.039787
14	0.05375	0.051641	0.041530	0.042595
all loci	0.049884	0.047828	0.060318	0.058601
95% lower Cl	0.040322	0.039145	0.051635	0.051644
95% upper Cl	0.062223	0.059460	0.068624	0.065321



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**Fig. S1**. Results of STRUCTURE analyses of *C. hamersleyana* showing Delta K vs K and bar plots of individual assignment to clusters K=2, 6,

36 8, 13 and 15.



37

**Fig. S2**. Results of STRUCTURE analysis of *A. pruinocarpa* showing Delta K vs K and bar plots of individual assignment to clusters K=2, 6, 8,

39 13 and 15.

- 40 Fig. S3. Principal Co-ordinates Analysis based on microsatellite data for individuals of A)
- 41 *Corymbia hamersleyana* and B) *Acacia pruinocarpa* sampled across the Pilbara region of
  - ♦ BEN
    ♦ COP a) DAW DEE DIN ♦ EMU PCoA2 (4.8%) ♦ GAL ♦ GID ♦ HAM HOO KAN KAR MCK MIN NUL ORD PAM ♦ RUB ♦ SHA PCoA1 (5.4%) **♦WEE** ANG b) BUN COO COR HAM ♦ HAR PCoA2 (6.6%) HES ♦ HIL HRP MES MET NAM ♦ OPH OXE • RHL RHO RIO SPE STE + WAR + WRA ♦ YAM ♦ YAR PCoA1 (7.1%)
- 42 Western Australia.



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