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Supplementary Material

Factors influencing the germination, establishment and distribution of *Micromyrtus minutiflora* (Myrtaceae), in western Sydney, New South Wales

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Description	Date of BioNet record(s)	Survey method	Survey effort	Outcome
Australian Defence Industries (ADI) site (now Wianamatta Regional Park)	2002-2004	Random meanders in proximity to BioNet records	Three people over one day in 2011	 <i>M. minutiflora</i> not recorded. Records towards the centre of the site have likely been removed as part of the development. Habitat along Ropes Creek degraded and was not consistent with microhabitats observed at other locations where the species has been recorded.
Two BioNet records in Mulgoa	2014 (accuracy of 10–20 m)	Parallel transects and random meanders in proximity to BioNet record(s) (licensed user search) and neighbouring mapped Castlereagh Scribbly Gum Woodland using hand- held GPS unit with coordinates downloaded	Three people over one day in April 2022	Two disjunct records of the species in the Mulgoa locality were identified approximately 16 km south-west of the species known distribution range. Reviewing the BioNet database information and subsequent discussions with DPE staff revealed that the records were never verified and no voucher specimens were provided. As these records represented a substantial extension of the species otherwise narrow distribution, they were visited to confirm the species presence. Prior to inspecting the record locations, plants were identified as flowering strongly at both the CNR and WSU reference populations in March 2022. <i>M. minutiflora</i> was not recorded. Two sympatric species (being <i>Baeckea brevifolia</i> and <i>Leptospermum parvifolium</i>) recorded at BioNet record locations. Microhabitats on site were not consistent with that observed at other locations where the species has been recorded. Suspected that records may be misidentifications and thus excluded from extent of occurrence.
One BioNet record in St Marys	2004		Two people over half a day in 2011	<i>M. minutiflora</i> not recorded. Habitat along South Creek degraded and was not consistent with microhabitats observed at other locations where the species has been recorded. Due to the above and seeing as the record has since been removed from the BioNet database this record has been excluded from extent of occurrence.

Table S1. Locations surveyed in addition to the six main study areas where *M. minutiflora* presence was not confirmed.

Description	Date of BioNet record(s)	Survey method	Survey effort	Outcome
One BioNet record in Vineyard	1996		Two people over half a day in 2011	<i>M. minutiflora</i> not recorded. Habitat was not consistent with microhabitats observed at other locations where the species has been recorded. Due to the above and seeing as the record has since been removed from the BioNet database this record has been excluded from extent of occurrence.
Windsor Downs Nature Reserve	No BioNet records, potential habitat suspected	Random meanders through reserve	Two people over two days in 2011	<i>M. minutiflora</i> not recorded. Habitat was largely not consistent with microhabitats observed at other locations where the species has been recorded.

	Agnes Banks Nature Reserve										
Patch	Area (ha)	Number of quadrats per patch	Mean density (plants m ⁻²)	Total plants in population	Mean stem length (cm)						
А	0.02	5	1.19	285	105						
В	0.05	4	0.84	390	101						
С	0.10	5	0.39	0.39 384							
Total / Mean	0.17	14	0.81 ± 0.23	1,364 ± 33.95	93.3 ± 9.6						

 Table S2. Estimation of abundance of Micromyrtus minutiflora for Agnes Banks Nature Reserve

in 2010–11 with ± s.e.

Patch, discrete location within which *M. minutiflora* was found; Area, area of occupation of *M*.

minutiflora.

Castlereagh Nature Reserve										
Patch	Area (ha)	Number of quadrats per	Mean density (plants m ⁻²)	Total plants in population	Mean stem length (cm)					
A	5.15	patch 10	0.6	30,900	120					
B	9.20	10	0.63	57,500	120					
C	10.63	10	1.56	166,016	111					
D	5.60	10	0.89	49,700	68					
E	4.40	10	1.12	49,225	118					
F	3.20	10	0.31	9,800	131					
G	2.70	5	0.59	15,863	96					
H	2.88	5	0.69	19,766	123					
I	37.40	30	1.32	493,992	113					
J	2.55	5	0.31	7,969	102					
K	5.60	5	2.80	156,800	120					
L	3.75	5	1.73	64,688	131					
М	2.40	5	0.23	5,400	127					
Ν	0.90	5	2.33	20,925	120					
0	0.23	5	0.96	2,166	124					
Р	0.01	С	0.17	10	125					
Q	2.08	5	0.30	6,240	174					
R	0.01	5	4.00	216	113					
S	0.23	5	0.90	2,052	146					
Т	0.28	5	1.53	4,194	89					
U	0.66	5	0.31	2,063	-					
Total / Mean	99.83	156	1.11 ± 0.21	1,105,254 ±	118.3 ± 4.7					

Table S3. Estimation of abundance of Micromyrtus minutiflora for Castlereagh Nature Reserve

in 2010–11 with ± s.e.

Patch, discrete location within which *M. minutiflora* was found; Area, area of occupation of *M*.

24,210

minutiflora; C, complete census.

2010–11 with ± s.e.

Londonderry Drop Zone									
Patch	ch Area (ha) Number quadrats		Mean density (plants m ⁻²)	Total plants in population	Mean stem length (cm)				
		patch		I I I I I I I					
А	0.27	5	0.80	2,160	89				
В	0.05	1	1.56	781	24				
С	0.23	3	0.27	609	28				
D	0.03	1	0.13	38	21				
Е	0.01	1	0.81	81	48				
F	2.60	5	1.68	43,550	54				
G	4.40	5	1.08	47,300	61				
Н	0.60	5	1.79	10,725	73				
Ι	< 0.01	С	0.47	14	102				
J	0.32	4	0.86	2,707	39				
Κ	1.95	5	0.68	13,163	56				
L	0.20	3	0.58	1,138	38				
М	3.14	5	0.86	27,083	90				
Ν	5.00	10	0.59	29,375	89				
0	0.08	С	0.02	18	67				
Р	0.41	3	0.44	1,805	123				
Q	0.05	1	0.56	281	95				
R	0.11	3	0.25	275	89				
S	0.06	1	0.94	563	75				
Т	1.40	5	0.49	6,825	76				
U	2.50	10	0.40	10,000	70				
V	0.40	5	3.26	13,050	83				
W	1.82	3	1.33	24,267	47				
Total / Mean	25.62	86	0.86 ± 0.15	220,934 ±	66.9 ± 5.6				
				2,990					

Patch, discrete location within which *M. minutiflora* was found; Area, area of occupation of *M*.

minutiflora; C, complete census.

with ± s.e.

		Marso	den Park		
Patch	Area (ha)	Number of quadrats per patch	Mean density (plants m ⁻²)	Total plants in population	Mean stem length (cm)
А	0.01	1	0.31	31	77
В	0.27	5	0.77	2081	85
С	0.06	1	1 1.13 675		89
D	0.14	5	0.38	525	83
Е	1.60	5	0.13	2000	89
F	0.06	1	0.38	225	64
G	0.60	5	0.02	125	86
Н	0.01	1	0.69	34	65
Ι	< 0.01	С	1.69 27		60
Total / Mean	2.75	25	$\textbf{0.61} \pm \textbf{0.18}$	$16{,}721\pm276$	77.6 ± 3.9

Patch, discrete location within which *M. minutiflora* was found; Area, area of occupation of *M*.

minutiflora; C, complete census.

Wianamatta Nature Reserve									
Patch	Area	Number of	Mean density	Total plants	Mean stem				
	(ha)	quadrats per	(plants m ⁻²)	in population	length (cm)				
		patch							
А	5.76	10	4.22	243,000	85				
В	3.41	10	3.27	111,464	80				
С	5.46	10	0.43	23,546	114				
D	3.24	10	2.16	70,065	50				
E	3.08	10	0.57	17,518	51				
F	2.47	10	2.79	69,006	67				
G	1.81	10	9.61	173,506	54				
Н	4.00	10	8.64	345,500	52				
Ι	4.49	10	6.96	311,988	44				
J	2.28	10	4.30	98,040	47				
K	4.18	10	9.31	389,001	38				
L	3.75	10	9.81	367,969	45				
М	5.20	10	6.53	339,625	38				
Ν	< 0.01	1	5.44	214	75				
0	0.58	5	0.17	967	68				
Р	0.08	1	0.19	150	70				
Q	1.68	10	3.41	57,225	64				
R	5.51	10	8.03	442,178	65				
S	7.25	10	7.55	547,375	53				
Т	0.96	5	0.67	6,400	73				
U	0.09	1	1.00	900	64				
V	2.45	5	2.29	56,146	77				
W	1.44	5	0.34	4,843	56				
Total / Mean	69.16	183	4.25 ± 0.71	2,936,812 ± 35,962	62.2 ± 3.6				

Table S6. Estimation of abundance of *Micromyrtus minutiflora* for Wianamatta Nature Reserve

in 2010–11 with ± s.e.

 35,962

 Patch, discrete location within which *M. minutiflora* was found; Area, area of occupation of *M*.

minutiflora.

Table S7. Estimation of abundance of Micromyrtus minutiflora for Western Sydney University

(Hawkesbury campus) in 2010–11 with ± s.e.

Western Sydney University (Hawkesbury campus)									
Patch	Patch Area (ha) Numbo quadrat patc		Mean density (plants m ⁻²)	Total plants in population	Mean stem length (cm)				
Total / Mean 1.25		50	0.54 ± 0.12 6,781		70.3 ± 1.5				
Area area of occ	unation of M	minutiflong		,					

Area, area of occupation of *M. minutiflora*.

 Table S8. Total location area, area of occupancy, estimated total number of *Micromyrtus minutiflora* plants, 95% confidence interval and percentage relative precision for total number

 of plants found at each of the six study areas (locations) in 2010–11.

Location	LocationArea of occupancyPercentage of total numberTotal number(ha)(ha)area (%)of plants		area occupancy of total number inte			
Agnes Banks Nature	114	0.17	0.15	1,364	± 922	61
Reserve Castlereagh Nature Reserve	495	99.83	20.17	1,105,254	± 386,060	37
Londonderry Drop Zone	212	25.62	12.08	220,934	$\pm 64,811$	35
Marsden Park	20	2.75	13.75	16,721	$\pm 9,009$	61
Wianamatta Nature Reserve	181	69.16	38.21	2,936,812	± 843,974	33
Western Sydney University	330	1.25	0.38	6,781	± 2,987	44
Total / Mean	1,352	198.78	14.70	4,287,758	± 1,308,188	30

Extent of occurrence (EOO) estimate	EOO area (ha)	Proportion of Bangel <i>et al.</i> 2022 area of occupancy (AOO) within each EOO	Comments
BioNet 2011	17,319	1.1%	EOO based on 2010 BioNet record prior to field validation surveys completed in 2010–11 and 2022.
BioNet 2022	26,452	0.8%	EOO based on 2022 BioNet record prior to field validation surveys completed in 2010–11 and 2022.
Bangel <i>et al</i> . 2022	13,136	1.5%	Based on a combination of field validated BioNet records and the removal of BioNet records considered to be misidentifications, to have been cleared and records where the species was not confirmed present during 2010– 2011 and 2022 surveys.

Table S9. Micromyrtus minutiflora extent of occurrence and area of occupancy estimates

Site	Latitude	Longitude	Time since fire (years)	Number of fires in past 20 years	Cover score (%)	S	R	J	Μ	Fl	Fr	Vegetation community
FireTrail Road, Castlereagh	-33.66331	150.7002	1.5	3	0.4				у	у		CSGW
Govt Road Berkshire Park	-33.68652	150.7795	4	3	0.1				у			CSGW
Deerubbin land opposite 173 Devlin Road, Castlereagh	-33.65993	150.6815	11	2	2	У			У			CSGW
Torkington Road, Londonderry	-33.64393	150.7225	19	2	0.1				у	у		CSGW
Deerubbin land, west of Nutt Road, Londonderry	-33.645	150.723	7	2	5				У	У		CSGW
WSU campus, The Driftway, Richmond	-33.62674	150.7515	2	1	0.1			у				CSGW
Torkington Road, Londonderry	-33.64324	150.7219	19	2	0.1				у			CSGW
Deerubbin land, Nutt Road, Londonderry	-33.64476	150.7217	7	2	0.1				У			CSGW
Torkington Road, Londonderry	-33.64284	150.7224	19	1	2				у	У		CSGW
Wianamatta Nature Reserve	-33.69562	150.7251	4	2	0.2				у	У		CSGW
Wianamatta Nature Reserve	-33.69639	150.7159	14	1	0.1				у	У		CSGW
outside Castlereagh Nature Reserve boundary west of Northern Road	-33.68158	150.7421	4	2	0.5				у	у		CSGW
Castlereagh Nature Reserve - perimeter trail	-33.68757	150.7469	4	2	0.2				У	у		CSGW
Castlereagh Nature Reserve - perimeter trail	-33.68759	150.7479	4	2	1				У	У		CSGW
Top of Nutt Road, Londonderry	-33.63924	150.725	10	1	0.2				У	У		CSGW
Top of Nutt Road, Londonderry	-33.63884	150.7246	10	1	0.5				У		У	CSGW

Table S10. *Micromyrtus minutiflora* life history and fire response observations in 2020–21 flora surveys .

Site	Latitude	Longitude	Time since fire (years)	Number of fires in past 20 years	Cover score (%)	S	R	J	М	Fl	Fr	Vegetation community
outside Castlereagh Nature Reserve boundary west of Northern Road	-33.6823	150.7413	4	2	0.1				У	у		CSGW
Smeeton Road (East), Cranebrook	-33.68334	150.7184	11	3	0.1				У			CSGW
Smeeton Road (West), Londonderry	-33.67844	150.6985	1.5	3	0.4	У			У	У		CSGW
Smeeton Road (East), Cranebrook	-33.68456	150.72	14	2	0.1				У	У		CSGW
Smeeton Road (East), Cranebrook	-33.68406	150.7275	14	1	0.2				у		У	CSGW
Rickards Road, Agnes Banks	-33.65045	150.6798	7	2	0.1				у	у		CIF
Castlereagh Nature Reserve	-33.68225	150.7497	2	3	0.1				у	у		CSGW
Castlereagh Nature Reserve	-33.68494	150.7452	4	2	0.1				у	у		CSGW
Tadmore Road, Cranebrook	-33.67926	150.7115	11	2	0.1				у			CSGW

S, seedling, R, resprouting, J, juvenile, M, mature, Fl, flowering, Fr, fruiting. CSGW, Castlereagh Scribbly Gum Woodland, CIF, Cooks River–Castlereagh Ironbark Forest. GPS datum GDA94.

 Table S11. Soil plant available nutrients in locations where *Micromytrus minutiflora* is present

 and absent in 2010–11.

Plant available nutrient (mg kg ⁻¹)	Mean across all areas (mg kg ⁻¹ ± s.e)	Mean – M. minutiflora present (mg kg ⁻¹)	Mean – M. minutiflora absent (mg kg ⁻ ¹)	Significant difference
Nitrate (NO_3)	0.17 ± 0.05	0.1	0.0	NS
Potassium (K)	5.91 ± 1.02	1.0	0.5	NS
Calcium (Ca)	12.60 ± 2.86	5.9	5.9	NS
Magnesium (Mg)	23.18 ± 5.76	11.7	5.8	NS
Phosphate (PO_4^{3-})	0.78 ± 0.27	11.7	13.5	NS
Sulphate (SO_4^{2-})	8.70 ± 2.40	32.2	14.2	NS
Iron (Fe)	73.75 ± 11.82	89.8	57.7	NS
Boron (B)	0.06 ± 0.02	1.1	0.5	NS
Manganese (Mn)	0.76 ± 0.35	0.4	0.3	NS
Zinc (Zn)	0.33 ± 0.08	0.1	0.1	NS
Copper (Cu)	0.15 ± 0.06	0.1	0.1	NS

NS, not significant.

Table 12. Soil physical properties in locations where Micromytrus minutiflora is present or
absent in 2010–11.

Physical soil property	ABNR (present)	ABNR (absent)	CNR (present)	CNR (absent)	WNR (present)	WNR (absent)	WSU (present)	WSU (absent)
Texture	sandy loam	loamy sand	fine sandy loam	fine sandy loam	fine sandy clay	fine sandy clay	sandy loam	fine sandy loam
Typical clay content (%)	10-20	5-10	10-20	10-20	20-30	10-20	10-20	10-20
Size Gravel content	fine not gravelly	fine not gravelly	fine gravelly	fine gravelly	medium gravelly	- gravelly	fine not gravelly	fine not gravelly
Aggregate strength	weak	weak	weak	weak	moderate	massive	weak	weak
Structural unit	crumb	crumb	crumb	crumb	crumb	-	crumb	crumb
Potential infiltration rate	rapid	very rapid	rapid	rapid	moderate	rapid	rapid	rapid
Permeability (mm h^{-1})	60-120	>120	60-120	60-120	5-20	60-120	60-120	60-120

Dashes represent no value reported by Sydney Environmental and Soil Laboratory. Agnes Banks

Nature Reserve (ABNR), Castlereagh Nature Reserve (CNR) and Western Sydney University

(WSU).

pH, EC & Exchangeable Acidity	Mean across all areas (g m ⁻²)	Mean <i>M</i> . <i>minutiflora</i> present soils	Mean <i>M</i> . <i>minutiflora</i> absent soils	Significant difference
pH (H ₂ O)	6.5	6.5	6.5	NS
pH (CaCl ₂)	6	6	6	NS
Salinity (dS m ⁻¹)	0.001	0.001	0.001	NS
Sodium (mg kg^{-1})	25.19	27.60	22.70	NS
Sum of base cations $(meq/100 g^{-1})$	1.16	1.20	1.13	NS
Effective cation exchange capacity (eCEC)	1.16	1.20	1.13	NS
Base Saturation (%)	100	100	100	NS

Table S13. Soil pH, electrical conductivity (EC) and exchangeable acidity in locations where *Micromytrus minutiflora* is present or absent in 2010–11.

NS, not significant.

Table S14. Soil cation balance in locations where *Micromytrus minutiflora* is present and absent in 2010–11

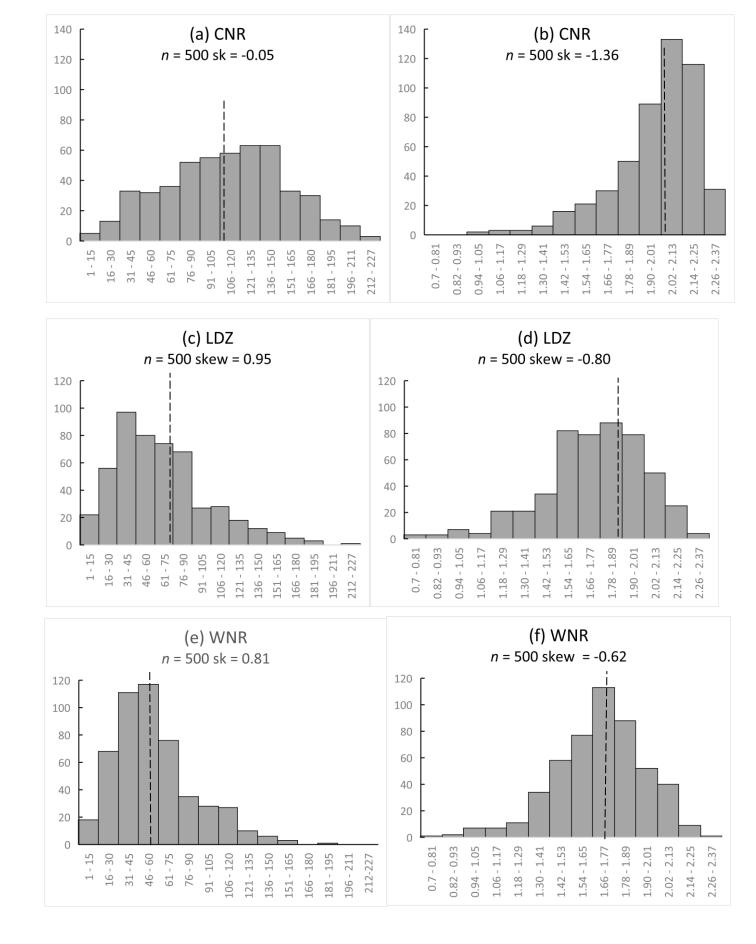
Cation Balance	Mean across all areas (mg/km)	Mean <i>M</i> . <i>minutiflora</i> present soils	Mean <i>M</i> . <i>minutiflora</i> absent soils	Significant difference	
Ca:Mg	0.5	0.3	0.7	*	
Mg:K	19.9	32.7	7.1	NS	
K/(Ca + Mg)	0.1	0.1	0.1	NS	
K:Na	1.6	1.5	1.8	NS	

Probabilities between locations are significant at: *, $\alpha = 0.05$. NS, not significant

Size class distribution in *M. minutiflora* populations

Methods

The field sampling methodology varied, depending on the numbers of plants and their extent at each study area, and varied from a census for small populations, to simple random sampling of patches for intermediate populations, to stratified random sampling for large populations (strata; patches within strata; quadrats within patches). At three study areas with very large populations, a random sample of patches was selected from the total population to give n = 500 plants. At the three remaining study areas, data from all plants were used (range n = 89-236 plants). Data for stem length were used to construct size class distributions for the raw data and log transformed data. Descriptive statistics (median, skewness, kurtosis) were calculated for raw and transformed data. The cumulative percentage of numbers with increasing log size class was calculated and used to identify the size at which the percentage of the population started to increase rapidly, compared to smaller size classes.

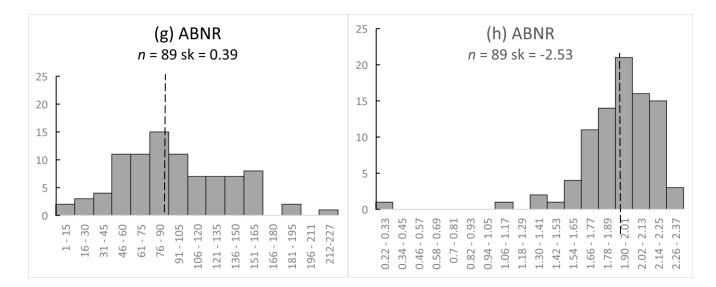


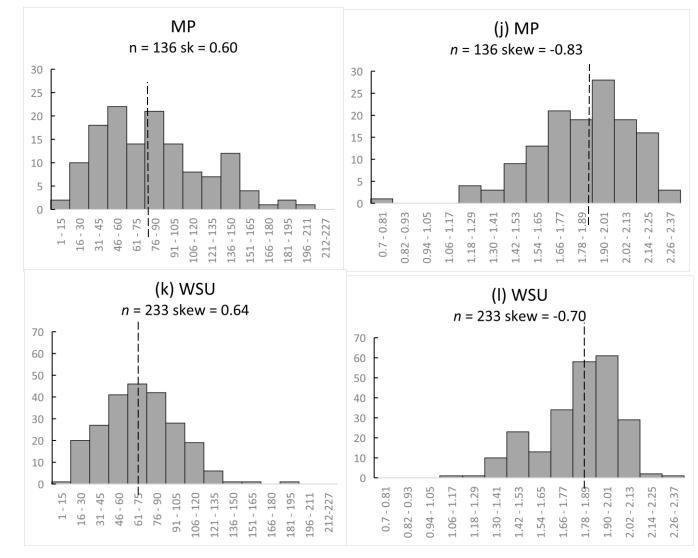
Stem length (cm)

Log stem length (cm)

Fig. S1. Distribution of stem length size classes of the longest stem of *Micromyrtus minutiflora* in 2010–11 raw data (lefthand column) and log data (right hand column) for (a, b) CNR (Castlereagh Nature Reserve); (c, d) LDZ (Londonderry Drop Zone); (e,f) WNR (Wianamatta Nature Reserve); (g, h) ABNR (Agnes Banks Nature Reserve); (i, j) MP (Marsden Park) and (k, l) WSU (Western Sydney University). Sample size (*n*) and skewness value (sk) are shown. Median shown as dashed vertical line on each plot.

Number





Stem length (cm)

Log stem length (cm)

Fig. S1. (Cont.)

Number

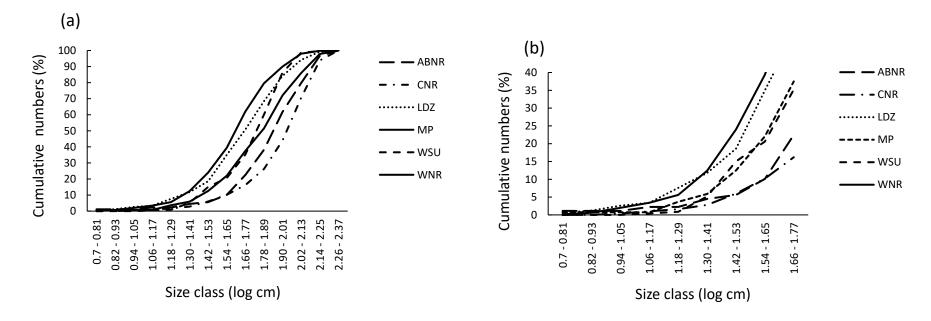


Fig. S2. Cumulative percentage of *Micromyrtus minutiflora* populations in 2010–11 in increasing log stem size classes of the longest stem for (a) total populations and (b) for first nine increasing size classes. Legend: ABNR (Agnes Banks Nature Reserve), CNR (Castlereagh Nature Reserve), LDZ (Londonderry Drop Zone), MP (Marsden Park), WNR (Wianamatta Nature Reserve) and WSU (Western Sydney University).