Ecography

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Appendix 4:

Figure A2: Population structure based on ISSR loci, inferred from Bayesian assignment tests, showing that *Lantana camara* populations in KNP's Sabie-Sand catchment consists of two genetic clusters and that the 'outgroup' collected at Kloof Station in KwaZulu Natal (South Africa) shares no genetic information with these populations. Vertical bars show each individual divided into coloured sections that represent the proportional membership of its genome to a particular K cluster.

Figure A3: PCA of populations collected along the Sabie-Sand catchment in KNP and one population collected at Kloof Station (KwaZulu Natal, South Africa), based on ISSR loci. Axis 1 and 2 explains 58 % and 19 % of the variation respectively.

References

Appendix 1: Methods for determining ploidy levels

Before genetic analyses, we needed to determine whether there are differences in ploidy levels associated with *Lantana camara* in KNP. Polyploidisation is common in *L. camara* (Spies 1984, Sanders 1987, Brandao et al. 2007), with chromosome numbers of 2n = 22, 33, 44, 55, and 66 reported from South Africa (Spies 1984). Furthermore, frequent hybridization between different ploidy levels in *L. camara* is known to occur, indicating that reproductive isolation between these levels is not complete (Spies 1984, Vardien et al. 2012). Such variable ploidy levels would affect amplification, and vastly influence population genetic analyses and interpretation of results.

Using the two-step flow cytometry procedure described by Suda and Trávniček (2006), relative fluorescent intensities of a subset of 95 *L. camara* individuals from across the study area were determined. *Bellis perennis* L. was selected as an internal reference standard and flow histograms were evaluated using Partec Flomax v2.4 (Partec GmbH, Műnster, Germany). Fluorescent intensity was then used to determine the amount of nuclear DNA in all samples analysed. These estimates were compared to Ohri et al.'s (2004) observation that haploid *L. camara* individuals contain ~ 2.75 pg of nuclear DNA. Inferred ploidy levels were confirmed by chromosome counts using a standard root-tip squash technique (Hanson et al. 2005).

Appendix 2: Results of ploidy levels

All *L. camara* individuals analysed for ploidy levels contained the same amount of nuclear DNA (5.54 pg, see Table A1 for raw fluorescent intensity ratios and Fig. S1 for chromosome counts), corresponding to diploidy. Consequently, the influence of varying ploidy levels was not considered in further analyses.

Appendix 3: Additional table and figure from ploidy level determination

Table A1: Fluorescent intensities and DNA amounts for *Lantana camara* samples derived from flow cytometry.

Sample ID	Latitude	Longitude	Route	Flower colour	DAPI ratio	DNA amount (pg)
M1	-24.965	31.692	Skukuza to Lower Sabie	pink	1.66	5.61
M2	-24.963	31.708	Skukuza to Lower Sabie	no flowers	1.64	5.56
M3	-24.995	31.769	Skukuza to Lower Sabie	yellow/pink	1.65	5.59
M4	-24.996	31.769	Skukuza to Lower Sabie	no flowers	1.65	5.57
M5	-24.997	31.769	Skukuza to Lower Sabie	yellow/pink, very light	1.64	5.55
M6	-25.000	31.773	Skukuza to Lower Sabie	orange	1.64	5.55
M7	-25.000	31.773	Skukuza to Lower Sabie	pink/yellow	1.67	5.65
M8	-25.000	31.773	Skukuza to Lower Sabie	pink	1.66	5.61
M9	-25.061	31.819	Skukuza to Lower Sabie	yellow (little pink)	1.67	5.65
M10	-25.066	31.826	Skukuza to Lower Sabie	yellow/pink, very light	1.68	5.66
M11	-25.067	31.831	Beyond Lower Sabie camp	pink/yellow	1.68	5.68
M12	-25.137	31.944	Beyond Lower Sabie camp	pink/yellow	1.70	5.73
M13	-25.137	31.944	Beyond Lower Sabie camp	pink/yellow	1.67	5.65
M14	-25.137	31.944	Beyond Lower Sabie camp	no flowers	1.66	5.61
M15	-25.141	31.942	Beyond Lower Sabie camp	pink/yellow	1.68	5.68
M16	-25.146	31.940	Beyond Lower Sabie camp	pink/yellow	1.70	5.74
M17	-25.149	31.940	Beyond Lower Sabie camp	no flowers	1.70	5.73
M18	-25.151	31.941	Around Lower Sabie camp	no flowers	1.69	5.71
M19	-25.117	31.914	Around Lower Sabie camp	pink/yellow	1.68	5.67
M20	-25.118	31.916	Around Lower Sabie camp	pink/yellow	1.72	5.80
M21	-25.119	31.916	Around Lower Sabie camp	pink/yellow	1.68	5.68
M22	-25.119	31.916	Around Lower Sabie camp	pink/yellow	1.66	5.62
M23	-25.119	31.917	Around Lower Sabie camp	pink/yellow	1.69	5.70
M24	-25.120	31.919	Around Lower Sabie camp	pink/yellow	1.67	5.63
M25	-25.118	31.916	Around Lower Sabie camp	pink/yellow	1.69	5.70
M26	-25.121	31.919	Around Lower Sabie camp	pink/yellow	1.70	5.74
M27	-24.990	31.612	Close to Skukuza	pink/yellow	1.67	5.63
M28	-24.991	31.610	Close to Skukuza	no flowers	1.67	5.65
M34	-24.969	31.348	Kruger gate to Sabie town	pink-white	1.65	5.57
M35	-24.969	31.348	Kruger gate to Sabie town	yellow/pink	1.71	5.78
M36	-24.963	31.313	Kruger gate to Sabie town	pink-white	1.70	5.74
M37	-24.964	31.293	Kruger gate to Sabie town	yellow/pink	1.64	5.56
M38	-24.964	31.292	Kruger gate to Sabie town	pink-white	1.70	5.76
M39	-25.028	31.216	Kruger gate to Sabie town	pink-white	1.65	5.59
M40	-25.034	31.200	Kruger gate to Sabie town	pink-white	1.64	5.54
M41	-25.034	31.200	Kruger gate to Sabie town	pink-white	1.65	5.59
M42	-25.034	31.200	Kruger gate to Sabie town	yellow/pink	1.67	5.65
M43	-25.028	31.172	Kruger gate to Sabie town	pink-white	1.66	5.61
M44	-25.028	31.172	Kruger gate to Sabie town	yellow/pink	1.66	5.62
M45	-25.041	31.139	Kruger gate to Sabie town	yellow/pink	1.65	5.56
M46	-25.064	31.128	Kruger gate to Sabie town	yellow/pink	1.66	5.61
M47	-25.052	31.134	Kruger gate to Sabie town	yellow/pink	1.65	5.57
M48	-25.052	31.134	Kruger gate to Sabie town	yellow/pink	1.64	5.56
M49	-25.049	31.128	Kruger gate to Sabie town	pink-white	1.68	5.66
M50	-25.044	31.110	Kruger gate to Sabie town	pink-white	1.67	5.66
M51	-25.041	31.089	Kruger gate to Sabie town	pink-white	1.69	5.71

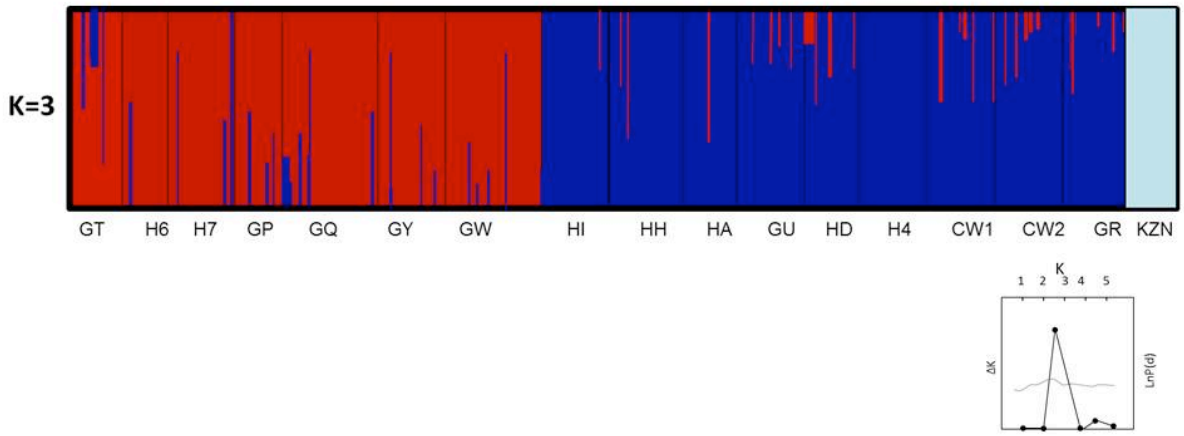
M52	-25.041	31.089	Kruger gate to Sabie town	yellow/pink	1.66	5.62
M53	-25.032	31.039	Kruger gate to Sabie town	white	1.70	5.73
M54	-25.032	31.039	Kruger gate to Sabie town	yellow/pink	1.66	5.60
M55	-25.038	31.008	Kruger gate to Sabie town	white	1.67	5.63
M56	-25.038	31.008	Kruger gate to Sabie town	yellow-pink	1.67	5.65
M57	-25.047	30.961	Kruger gate to Sabie town	pink-white	1.71	5.76
M58	-25.051	30.956	Kruger gate to Sabie town	white	1.66	5.62
M59	-25.051	30.956	Kruger gate to Sabie town	yellow-pink	1.69	5.70
M60	-25.051	30.957	Kruger gate to Sabie town	red/yellow	1.69	5.72
M61	-25.051	30.955	Kruger gate to Sabie town	red/yellow	1.67	5.64
M62	-25.067	30.928	Kruger gate to Sabie town	white	1.68	5.68
M64	-25.067	30.894	Kruger gate to Sabie town	red/yellow	1.70	5.73
M65	-25.067	30.894	Kruger gate to Sabie town	yellow/pink	1.68	5.67
M66	-25.076	30.845	Kruger gate to Sabie town	yellow/pink	1.67	5.63
M67	-25.072	30.855	Kruger gate to Sabie town	dark pink/light yellow	1.65	5.57
M68	-25.072	30.854	Kruger gate to Sabie town	red/orange-yellow	1.66	5.60
M69	-25.098	30.785	Kruger gate to Sabie town	red/yellow	1.65	5.57
M70	-25.098	30.785	Kruger gate to Sabie town	dark pink/light yellow	1.71	5.77
M71	-25.067	30.930	Kruger gate to Sabie town	white	1.65	5.56
M72	-25.056	30.950	Kruger gate to Sabie town	white	1.66	5.61
M73	-25.056	30.950	Kruger gate to Sabie town	red/yellow	1.68	5.67
M74	-25.043	30.988	Kruger gate to Sabie town	white	1.69	5.71
M75	-25.039	31.075	Kruger gate to Sabie town	white	1.66	5.60
M76	-25.035	31.125	Kruger gate to Sabie town	pink-white	1.65	5.56
M77	-25.025	31.230	Kruger gate to Sabie town	pink-white	1.70	5.74
M79	-24.991	31.590	Kruger gate to Phabeni gate	pink	1.69	5.70
M80	-24.990	31.457	Kruger gate to Phabeni gate	pink	1.66	5.61
M81	-24.990	31.457	Kruger gate to Phabeni gate	pink	1.69	5.70
M82	-24.990	31.457	Kruger gate to Phabeni gate	pink	1.65	5.58
M83	-24.989	31.443	Kruger gate to Phabeni gate	pink - pink/yellow	1.64	5.54
M84	-24.989	31.443	Kruger gate to Phabeni gate	pink	1.68	5.69
M85	-24.988	31.436	Kruger gate to Phabeni gate	pink	1.65	5.58
M86	-24.988	31.436	Kruger gate to Phabeni gate	yellow/pink	1.68	5.69
M87	-24.988	31.436	Kruger gate to Phabeni gate	yellow/pink	1.69	5.71
M88	-24.988	31.437	Kruger gate to Phabeni gate	pink	1.67	5.63
M89	-24.988	31.437	Kruger gate to Phabeni gate	yellow/pink	1.70	5.73
M90	-24.988	31.437	Kruger gate to Phabeni gate	white	1.65	5.57
M91	-24.984	31.301	Kruger gate to Phabeni gate	pink-white	1.65	5.57
M92	-24.986	31.296	Kruger gate to Phabeni gate	pink-white	1.70	5.73
M93	-24.986	31.296	Kruger gate to Phabeni gate	pink-white	1.69	5.72
M94	-24.986	31.296	Kruger gate to Phabeni gate	pink-white	1.67	5.63
M95	-24.986	31.296	Kruger gate to Phabeni gate	yellow/pink	1.70	5.75
M96	-24.989	31.294	Kruger gate to Phabeni gate	pink	1.65	5.57
M97	-24.988	31.294	Kruger gate to Phabeni gate	yellow/pink	1.66	5.59
M98	-24.988	31.293	Kruger gate to Phabeni gate	pink	1.66	5.61
M99	-25.028	31.265	Kruger gate to Phabeni gate	pink	1.65	5.58
M100	-25.028	31.265	Kruger gate to Phabeni gate	yellow/pink	1.65	5.59
M101	-25.027	31.251	Kruger gate to Phabeni gate	pink	1.69	5.72

Figure A1: Microphotograph of a mitotic metaphase plate for *Lantana camara* (2n=22).

Appendix 4: Population genetic analysis including an ‘outgroup’ population.

We genotyped individuals for all ISSR markers from a lantana population collected from Kloof Station (GPS: -29.7913 ; 30.8332) in the Kwa-Zulu Natal Province. Bayesian assignment tests and principle components analysis were done as described in the full main manuscript.

Figure A2: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GW
HI
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CW1
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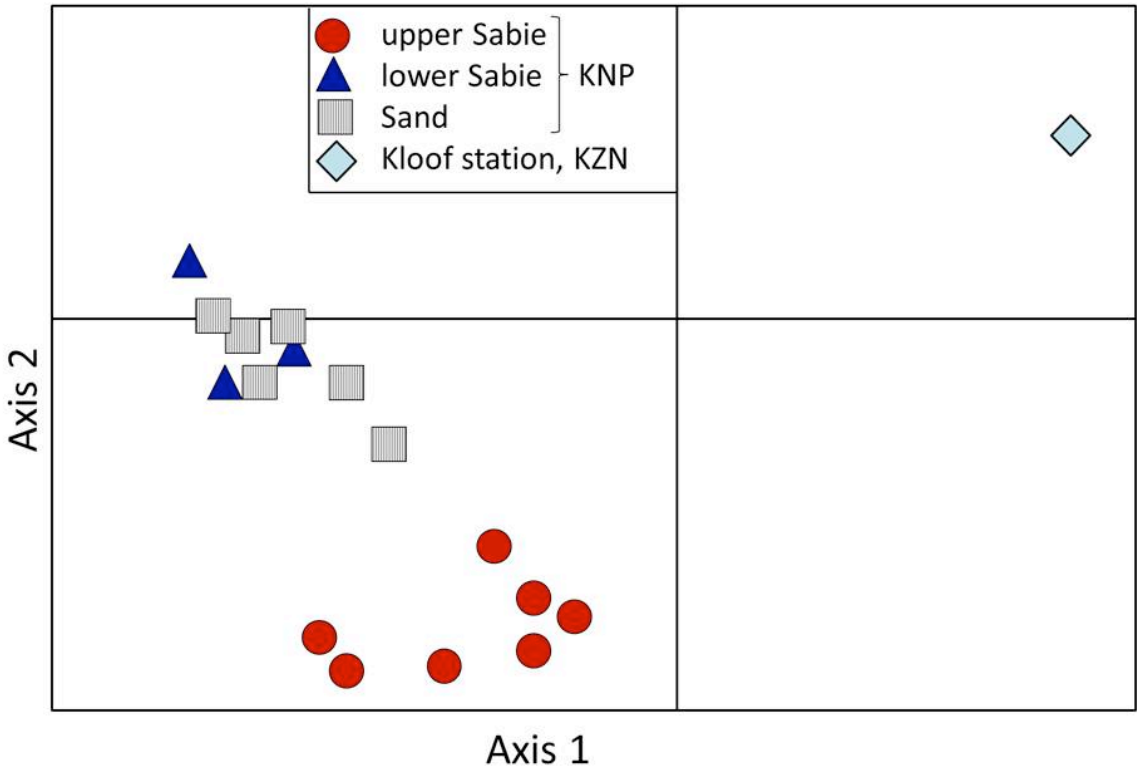
ΔK
LnP(d)

Figure A3:


upper Sabie
lower Sabie
KNP
Sand
Kloof station, KZN
Axis 2
Axis 1

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