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Tool use and social homophily among male bottlenose dolphins

SUPPLEMENTARY INFORMATION

1. Data Restrictions

We included only independent/weaned males and excluded dependent calves (1). Only males observed more than nine times and identified as spongers or non-spongers were included in our analyses. Sex was identified either genetically (see below) or behaviourally by several observations of alliance-typical behaviour (being observed regularly travelling side-by-side, engaging in synchronous surfacing, consorting of females, or inter-group aggression with other males; *cf.,* (2,3)). Furthermore, in order to assess males with similar association opportunities, we restricted our analyses to comparisons of male spongers with non-sponging males that also met habitat use criteria based on depth and home range overlap derived from data on sponging males.

Water depth All 129 surveys that included sponging events by known male spongers took place in water depths between 8 and 16 m (mean = 12 m, and 16 m representing the deepest parts of our study area). Thus, we only included males in our data set whose average survey depth was deeper than 8 m, with water depth as a proxy for sponging habitat (4).

Home range We also included only male non-spongers whose home ranges overlapped substantially with the pooled home range of all sponging males. To calculate home ranges, we considered only the last location per day for each individual to reduce dependencies between data points. For each male non-sponger within our study population, and for the pooled sightings of male spongers, we defined home ranges using 95% kernel density estimates (KDE; epanechnikov kernel; adehabitatHR (5)), subtracting inaccessible areas, *i.e.*, landmasses (see below for further details on the choice of smoothing factor and subtracting landmasses). We then estimated the overlap of individual male non-sponger home ranges with the pooled home range of all male spongers using the method of volume of intersection (VI; adehabitatHR (5)). The dyadic VI is represented by a single symmetric measure of the volume of overlap based on the compared KDEs. When considering individual home range overlaps of male non-spongers with the pooled home range of male spongers, we included only male non-spongers whose home ranges had above 45% overlap with the pooled home range of all male spongers. We used 45% as a threshold, because when considering the overlaps of non-spongers with the pooled spongers, 45% represented the average overlap. The threshold was further justified as it lay well within the mean and minimal home range overlap (95% KDEs) observed exclusively among male spongers (min = 23%, mean = 59%). Figure S1 shows the pooled home range of the male spongers (n = 13) and the pooled home range resulting from all the included male non-spongers (n = 24).

Smoothing factor We used kernel density estimates to define individual home ranges, as detailed in (6). Thereby, accuracy of the home ranges is greatly influenced by the choice of a suitable smoothing factor (7). The commonly used smoothing factor h_{ref} (reference bandwidth) often leads to over-smoothed, *i.e.* inflated, home ranges (8), while other approaches such as least-square cross validation (LSCV) tend to under-smooth, *i.e.* fragmentise, home ranges (9). Other approaches, such as least-square cross validation (LSCV), make no assumptions about the underlying distribution, but tend to under-smooth *i.e.*, fragmentise home ranges. We therefore selected a subjective smoothing parameter based on visual inspection (7) setting a lower and upper limit (1,000; 4,000) for the reference bandwidth h_{ref} and then calculating an adjusted smoothing factor for each individual as $h = 0.5(h_{ref}) + 1{,}500$, where $1{,}000 \leq h_{ref} \leq 4000$.

Removal of landmasses To restrict home ranges to water only, we multiplied the individual utilisation distributions with a grid (100 m resolution) coding land-cells as 0 and water-cells as 1. We subsequently re-weighted each grid cell within an individual's home range to ensure that the sum of the weights over the utilisation distribution of each individual added up to 1 again (*cf.* (6)).

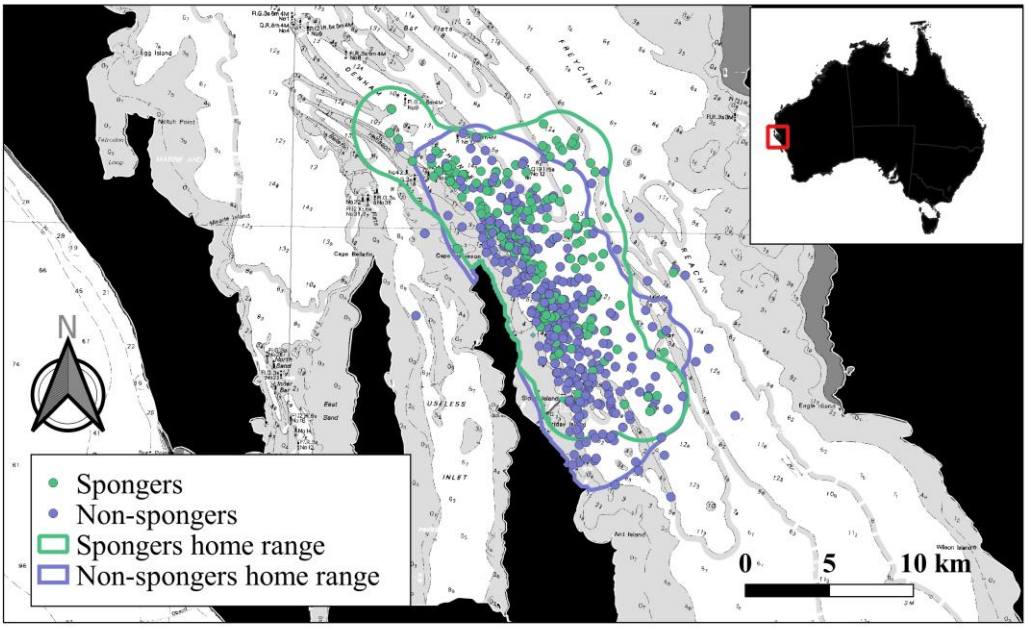

Figure S1: Western gulf of Shark Bay showing the distribution of sightings of male dolphins in the restricted data set (n = 37). Pooled home ranges (based on 95% KDEs) of spongers (n = 13) and non-spongers (n = 24) were visualized using QGIS (10).

2. Ethogramm

During boat-based surveys of dolphin groups, within the first five minutes, we recorded GPS position, environmental parameters (including sea state, water depth and temperature), group size and composition, as well as predominant group activity (rest, travel, forage, socialise, or unknown). The behavioural states were defined according to a long-established protocol used in the field since the founding of this study site in 2007, but was itself based on an ethogram developed much earlier still (11). The categories are as follows:

Rest – Dolphins in a tight group, moving very slowly with regular, peduncle or tail-out dives, or snagging – dives and surfacing bouts are often synchronous during resting behaviour. During rest, there is no evidence of foraging or socialising (although juveniles/calves may socialise while adults rest). Resting groups regularly snag en masse.

Travel - Individuals clustered or line abreast and moving in one general direction (i.e., no rapid changes in direction) for a period of several minutes or during consecutive surfacing bouts.

Social – Social behaviour is characterised by body contact, rubbing and petting, often accompanied by splashing, surface activity and acoustic behaviour. Bottlenose dolphins also perform a variety of synchronous behaviours.

Forage - Foraging is one of the most frequently observed behaviours, often involving lone dolphins or widely dispersed groups. An exception to this occurs when one or more dolphins remain in close proximity to a foraging dolphin for social reasons (i.e., when males are herding a female). Both inter-individual geometry (iig) and dive type are important in determining whether or not dolphins are foraging, independent of observations of feeding. Movement and iig are usually characterised by milling or meandering during foraging. In deeper water, foraging usually involves multiple breath surfacing bouts, culminating in a tail-out or peduncle dive. In shallower water, regular bottom grubbing, rapid surfaces and (fish) chases are often observed.

3. Activity Budget

To investigate differences in activity budgets (proportions of resting, travelling, foraging and socialising behaviour) between male spongers and non-spongers, we conducted a multivariate analysis of variance (MANOVA) with the sole predictor of whether an individual was classified as a sponger or a non-sponger (hereafter: foraging technique). While investigating the data structure of the multivariate activity budgets, we identified five outliers from the combined normal distribution. Table S1 shows the results of the analysis when outliers are retained.

Table S1: Supplementary analysis including the five multivariate outliers. The analysis demonstrates the same reported pattern as that excluding the outliers. Significant p-values are shaded in light grey. Sp = spongers, N.Sp = non-spongers

N	Manova	Proportion	Post hoc t test				
			Mean (SD) Sp	Mean (SD) N.Sp	t(df)	r	p
37	(V = 0.70, F(4,32) = 18.30, p < 0.001)	Forage	0.44 (0.02)	0.21 (0.02)	- 9.24 (30.51)	0.86	<0.001
		Rest	0.17 (0.01)	0.28 (0.02)	4.94 (33.44)	0.65	<0.001
		Travel	0.16 (0.02)	0.30 (0.02)	4.68. (29.25)	0.65	<0.001
		Socialise	0.15 (0.01)	0.12 (0.01)	-1.89 (33.14)	0.31	0.134

4. Social Network Structure

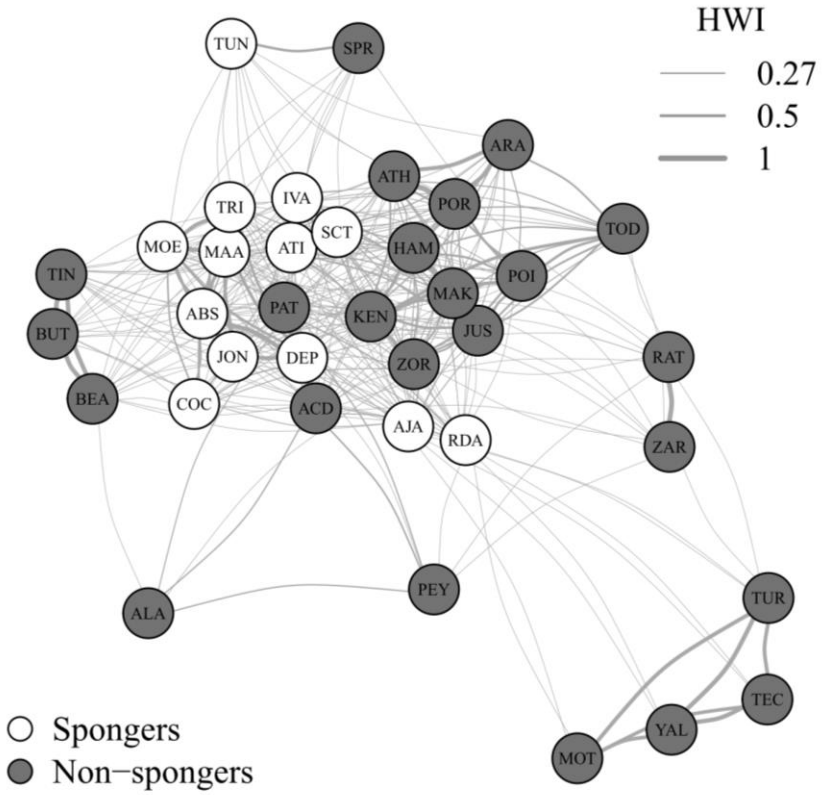

Figure S2: Social network of male dolphins in the restricted data set (n = 37). The nodes represent individuals and are coloured according to foraging technique. All edges (lines) are shown and edge thickness corresponding to dyadic association strengths (HWI). The figure was plotted with the force directed Fruchterman-Reingold algorithm implemented in the 'igraph' package (12).

5. MRQAP-DSP including all males

Between 2007 and 2015, we identified 118 male dolphins with at least 10 sightings and that were classified as spongers (n = 13) and non-spongers (n = 105). We computed 'half weight association indices' (HWIs) from 1,649 survey records over the nine-year study period.

Among the 118 males, the overall mean HWI was 0.03 (1,000 bootstraps: SE = 0.01), including the zeros of no associations. Considering only non-zero associations, the more conservative measure, the mean HWI was 0.12 (1,000 bootstraps: SE = 0.04). The generated network based on the dyadic association indices represented a non-random social structure (daily sampling period, 10,000 permutations, 1,000 switches; SD_{obs} = 0.10, SD_{random} = 0.07, $p < 0.01$). Thus, over all sampling periods, some males were observed more often in association than expected by chance alone.

In the MRQAP-DSP test, we only included males for which there were genetic data available (spongers: n = 9, non-spongers: n = 52). Pairwise relatedness (mean = 0.04, SE = 0.001) was estimated based on 27 microsatellite loci (see below), similarity matrices were coded as described in the methods section of the main paper and dyadic home range overlaps were based on 95% kernel density estimates (KDE; epanechnikov kernel; 'adehabitatHR' package (5)), subtracting landmasses.

Since the two predictor matrices relatedness and home range overlap were correlated (Mantel test, z = 26.13; $p < 0.01$), we decided to conduct the test three times: (I) retaining all predictors: home range overlap, pairwise relatedness, and similarity matrices; (II) retaining only pairwise relatedness and the similarity matrices, and (III) retaining only home range overlap and the similarity matrices. All MRQAP regression models showed that sponging was a significant predictor of male association patterns, even after correcting for pairwise relatedness and home range overlap (Table S3). Model (II), using only relatedness had the lowest fit, which is in line with our findings presented in the main paper.

Table S3: MRQAP-DSP model including only genotyped males (n = 61, 1,830 dyadic relationships). Significant p-values are indicated in bold print.

variable	coefficient	p
(I) Used predictors: dyadic home range overlap, pairwise relatedness, similarity matrices		
sponger similarity	0.10	<**0.001**
non-sponger similarity	-0.02	0.03
relatedness	-0.11	<**0.01**
Home range overlap	0.18	<**0.001**
F(4, 1826) = 204.2, adjusted R^2 = 0.31, p-value < **0.001,** AIC = -538.5275		
(II) Used predictors: pairwise relatedness, similarity matrices		
sponger similarity	0.20	<**0.001**
non-sponger similarity	0.03	<**0.01**
relatedness	0.07	0.08
F(3, 1827) = 91.36, adjusted R^2 = 0.13, p-value < **0.001,** AIC = -390.8307		
(III) Used predictors: dyadic home range overlap, similarity matrices		
sponger similarity	0.10	<**0.001**
non-sponger similarity	-0.02	**=0.01**
Home range overlap	0.17	<**0.001**
F(3, 1827) = 267.6, adjusted R^2 = 0.30, p-value < **0.001,** AIC = -477.1827		

6. Definition of alliances

In Shark Bay, multiple levels of nested male alliances have been documented (3). Two to three males cooperate in '1st-order' alliances to consort single oestrus females (13). Almost all 1st-order alliances are also part of '2nd-order' alliances which comprise 4-14 males that cooperate to take females from rival alliances and to defend against such attacks (13).

6.1 Defining a threshold to detect structure of 2nd order alliances

To identify an appropriate HWI threshold value for our study population, we used the dyadic associations of all males in western Shark Bay that were seen at least 10 times (n = 124) and subsequently used this threshold on our data set. We conducted a change point analysis employing the Pruned Exact Linear Time (PELT) method specified in the 'changepoint' package (14). Change points are identified by sorting the dyadic HWIs according to size and are located where the statistical properties of the HWI series to either side differ, as is expected for different levels of social structures in multi-level societies (*e.g.* (15),(16)). Based on an elbow plot where we plotted penalty values against the numbers of change points, we chose the segmentation of the social structure with only one change point defining the threshold for 2nd-order alliances.

6.2 Structure of 2nd-order alliances when all males in dataset were included

We also re-did the analysis defining 2nd-order alliances including all males within our study set with at least ten sightings and classified into spongers (n = 13) and non-spongers (n = 105). The average linkage agglomerative cluster analysis (17) resulted in a tree diagram which represented the underlying data well with a cophenetic correlation coefficient of 0.98 (18,19). The analysis showed that the homophily of male spongers is reflected in 2nd-order alliance compositions. It identified 27 potential 2nd-order alliances based on association patterns alone, only one alliance consisting of a mixed composition (sponger and non-sponger), two alliances consisting of exclusively spongers and 24 alliances consisted of exclusively non-spongers. Nineteen individuals (spongers: n = 3, non-spongers: n = 16) could not be assigned to a 2nd-order alliance (Figure 3S).

6.3 First-order alliance structure: Quantitative analysis of consortship compositions

Males in 1st-order alliances cooperate to consort single oestrus females. To investigate whether the composition of males consorting females was in line with the observed homophilous tendencies of male spongers, we quantitatively assessed recorded consortships of the males in our study set. We recorded a consortship if we observed a sub-group of one female and two to three males that were >10m apart from other sub-groups combined with one or more of the following criteria: (I) one hour of observation of the males with the consorted female, (II) the group was sighted again in the same composition more than one hour after the initial sighting, (III) one or several males elicited popping sounds (20), (IV) the female tried to escape, (V) display of aggressive behaviour of males towards the female (e.g. charge, hit, head jerk), or (VI) the capture or the theft of the female was witnessed.

During the period between 2012 and 2015, when consortships have been systematically noted at this study site, for the 37 males chosen for this study, 11 males (spongers: n = 6, non-spongers: n = 5) were involved in 11 consortships. In all these consortships, males consorting together were either all spongers or all non-spongers. Six of the observed consortships consisted exclusively of male spongers and the remaining five exclusively of male non-spongers. There was no obvious pattern regarding the choice of females consorted (spongers or non-spongers).

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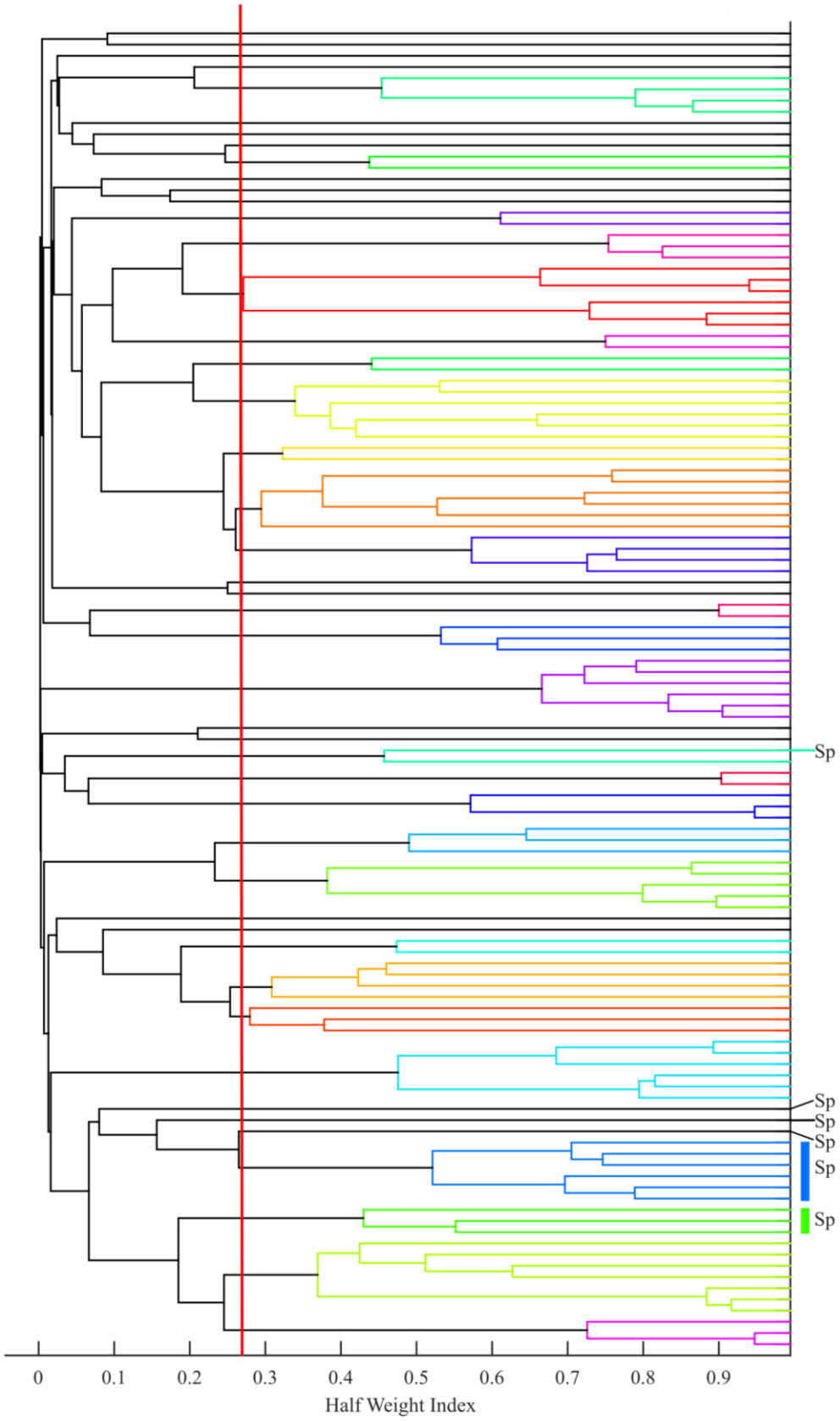

Figure S3: Hierarchical cluster diagram based on dyadic HWI measures. 0.27 was used as a cut-off value (red line) to define communities (2^{nd}-order alliances).Only branches of spongers are labelled (Sp), unlabelled branches represent non-spongers.

7. Laboratory procedures and genetic data analysis

After obtaining biopsy samples from dolphins, we stored them in saturated NaCl/20% dimethyl sulphoxide solution (DMSO) (21) in the field at 4 °C and at minus 80 °C in the laboratory for further use. We extracted the DNA in the laboratory using Qiagen's DNeasy kit according to the manufacturer's protocol for minute samples and Qiagen's Gentra Puregen Mouse Tail kit for larger samples. With the latter we used the following modifications: we added Proteinase K multiple times if the tissue was not dissolved after intervals of three hours; and we used 300µl 70% ethanol instead of 300µl isopropanol in the second purifying step to extract DNA. After the DNA clean-up, we measured DNA concentrations using NanoDrop 1,000 V3.3 (Technologies Inc. Wilmington, DE) and diluted the extracted DNA concentration to 20 ng/µl with DNA Hydration Solution (Qiagen).

Molecular sexing Sex was determined by PCR amplification of the two sex chromosome specific loci ZFX and SRY using the primers P1-5EZ, P2-3EZ (22) and Y53-3C, Y53-3D (23), respectively. We used gel electrophoresis in combination with stained DNA bands (GelRedTM), UV light and an AlphaImager CCD camera system (Alpha Innotech, San Leandro, CA) to visually determine sex.

Pairwise relatedness estimations We genotyped each individual for 27 hyper-variable microsatellite loci (Table S4) using three different multiplex PCR setups. We diluted PCR products twenty-fold with ddH2O and then 10µl HiDi formamide, and added 0.07µl size standard (GenScanTM500LizTM, Applied Biosystems). We analysed the PCR products via capillary electrophoresis using an ABI 3730 DNA Sequencer (Applied Biosystems) and scored it with the GeneMapper 4.0 software (Applied Biosystems). We chose a background population including only individuals sampled in the sponging area (95% KDE of all observed sponging events; *cf.* (24)), relative to which dyadic relatedness scores were calculated. The background population totalled 160 individuals by only including samples with less than 13 missing loci. Pairwise relatedness estimators assume Hardy-Weinberg equilibrium (HWE), no linkage disequilibrium and negligible chance of null alleles for all markers within the background population. We controlled for these assumptions using GENEPOP 4.5.1 (25) and by applying sequential Bonferroni corrections and excluding the offspring of known mother-offspring pairs (n_{dyads} = 9). All 27 markers were in HWE (Table S5), four locus pairs were significantly in linkage disequilibrium (Table S6) and three markers showed evidence for possible null alleles (Table S5). However, according to previous research (26), linkage disequilibrium and null alleles are not consistent across subareas of western Shark Bay and, therefore, all 27 markers were used to calculate pairwise relatedness. The discriminatory power of the genetic markers in the western Shark Bay study population was assessed in detail elsewhere (26). To calculate dyadic relatedness, we chose the triadic likelihood estimator proposed by Wang (27) as it correlated best with both simulated dyads produced through COANCESTRY 1.0.1.2 (28) and the known mother-offspring pairs to calculate dyadic relatedness.

Table S4: Summary of the 27 microsatellites used in this research.

Locus	Allele size range (bp)	No of Alleles	Repeat Motif	Multiple x	Reference
E12	260–280	6	tetra	1	(29)
MK6	145-189	13	di	1	(30)
Tur4_105	367–403	6	tetra	1	(29)
Tur4_108	258–270	2	tetra	1	(29)
Tur4_111	287–307	4	tetra	1	(29)
Tur4_117	175–187	4	tetra	1	(29)
Tur4_128	295–311	4	tetra	1	(29)
Tur4_66	185–201	5	tetra	1	(29)
Tur4_98	192–196	2	tetra	1	(29)
D22	NA	4	di	2	(31)
D8	322-342	3	tetra	2	(29)
Tur4_F10	374–390	4	tetra	2	(29)
Tur4_138	207-227	6	tetra	2	(29)
Tur4_141	218-286	10	tetra	2	(29)
Tur4_87	178–194	5	tetra	2	(29)
Tur4_91	207-235	8	tetra	2	(29)
Tur4_162	403-407	3	tetra	3	(29)
MK9	168-180	6	di	3	(30)
MK5	201-221	5	di	3	(30)
Tur4_132	326-334	3	tetra	3	(29)
KWM12	NA	14	di	3	(32)
EV37	NA	11	di	3	(33)
Tur4_80	287–335	7	tetra	3	(29)
MK3	139-171	6	di	3	(30)
Tur4_142	330-346	4	tetra	3	(29)
Tur4_153	330-346	2	tetra	3	(29)
MK8	87-119	8	di	3	(30)

Table S5: Summary of the 27 loci used for pairwise relatedness estimates and results of testing for deviation from HWE, estimated null allele frequency (34) within the reference population (n = 151), and empirical error rate. Empirical error rates were based on repeatedly genotyped loci (in total 4,290 calls). F_{IS} values were calculated according to Weir and Cockerham (35). Bonferroni corrected significant p-values ($p_{crit.}$ = 0.002) of the F_{IS} are indicated in bolt.

Locus	F_{IS}	*p*	Est. Null Allele Freq.	Error Rate
E12	0.0421	0.0994	0.0230	0
MK6	0.0039	0.4350	0.0000	0
Tur4_105	-0.0299	0.3522	0.0250	0
Tur4_108	0.0583	0.2986	0.5710	0
Tur4_111	-0.0041	0.3330	0.0388	0
Tur4_117	0.0285	0.3669	0.0000	0
Tur4_128	0.0576	0.1842	0.0991	0
Tur4_66	0.0269	0.5031	0.0000	0
Tur4_98	-0.0367	0.7410	0.3431	0
D22	0.0663	0.0393	0.0356	0
D8	0.1091	0.0431	0.1601	0
Tur4_F10	0.0867	0.0696	0.0426	0
Tur4_138	-0.0527	0.8406	0.0402	0
Tur4_141	-0.0295	0.9109	0.0144	0
Tur4_87	-0.0019	0.1258	0.0000	0
Tur4_91	0.0329	0.3176	0.0211	0
Tur4_162	-0.0944	0.8084	0.3133	0
MK9	0.0031	0.4487	0.0000	0
MK5	0.0358	0.1632	0.0088	0
Tur4_132	-0.0334	0.6616	0.0000	0
KWM12	-0.0678	0.7294	0.0000	0
EV37	0.0676	0.0327	0.0155	0.012
Tur4_80	0.0331	0.3097	0.0000	0.011
MK3	0.1487	0.0217	0.0453	0
Tur4_142	-0.0584	0.9083	0.0000	0
Tur4_153	-0.2023	0.9978	0.0636	0
MK8	0.0087	0.1285	0.0000	0

Table S6: Loci found to be in linkage disequilibrium within the chosen reference population (n = 151). Bonferroni corrected significant p-values ($p_{crit.}$ = 0.002) are indicated in bolt.

Locus 1	Locus 2	*p*
Tur4_108	Tur4_117	**<0.002**
Tur4_108	Tur4_128	**<0.002**
MK8	F10	**<0.002**
Tur4_142	MK8	**<0.002**

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References

1. Mann J, Connor RC, Barré LM, Heithaus MR. Female reproductive success in bottlenose dolphins (*Tursiops* sp.): life history, habitat, provisioning, and group-size effects. Behav Ecol. 2000;11(2):210–9. Available from: http://dx.doi.org/10.1093/beheco/11.2.210
2. Connor RC, Smolker R, Bejder L. Synchrony, social behaviour and alliance affiliation in Indian Ocean bottlenose dolphins, *Tursiops aduncus*. Anim Behav. 2006 Dec;72(6):1371–8. Available from: http://dx.doi.org/10.1016/j.anbehav.2006.03.014
3. Connor RC, Krützen M. Male dolphin alliances in Shark Bay: Changing perspectives in a 30-year study. Anim Behav. 2015;103:223–35. Available from: http://dx.doi.org/10.1016/j.anbehav.2015.02.019
4. Tyne JA, Loneragan NR, Kopps AM, Allen SJ, Krützen M, Bejder L. Ecological characteristics contribute to sponge distribution and tool use in bottlenose dolphins *Tursiops* sp. Mar Ecol Prog Ser. 2012 Jan;444:143–53. Available from: http://dx.doi.org/10.3354/meps09410
5. Calenge C. The package "adehabitat" for the R software: a tool for the analysis of space and habitat use by animals. Ecol Modell. 2006;197(3–4):516–9. Available from: https://doi.org/10.1016/j.ecolmodel.2006.03.017
6. Wild S, Allen S, Gerber L, Hoppitt W, King S, Krützen M. Multi-network Network-based Diffusion Analysis reveals vertical cultural transmission of sponge tool use within dolphin matrilines. Biol Lett Rev. 2019;
7. Jones MC, Marron JS, Sheather SJ. A brief survey of bandwidth selection for density estimation. J Am Stat Assoc. 1996;91(433):401–7.
8. Silverman BW. Density estimation: For statistics and data analysis. Routledge. 2018. 1-175 p. Available from: http://dx.doi.org/10.1201/9781315140919
9. Calenge C. The package adehabitat for the R software: a tool for the analysis of space and habitat use by animals. Ecol Modell. 2006;197(3–4):516–9. Available from: https://doi.org/10.1016/j.ecolmodel.2006.03.017
10. QGIS Development Team. QGIS 3.6.0-Noosa, Geographic Information System. Open Source Geospatial Foundation Project; 2019. Available from: http://qgis.osgeo.org
11. Smolker RA, Richards AF, Connor RC, Pepper JW. Sex Differences in Patterns of Association among Indian Ocean Bottlenose Dolphins. Behaviour. 1992;123(1–2):38–69. Available from: https://doi.org/10.1163/156853992X00101
12. Csárdi G, Nepusz T. The igraph software package for complex network research. 2014.
13. Connor RC, Smolker RA, Richards AF. Two levels of alliance formation among male bottlenose dolphins (*Tursiops* sp.). Proc Natl Acad Sci. 1992;89(3):987–90. Available from: http://dx.doi.org/10.1073/pnas.89.3.987
14. Killick R, Haynes K, Eckley I, Fearnhead P, Lee J. "changepoint": Methods for Changepoint Detection. 2016. Available from: https://cran.r-project.org/web/packages/changepoint/changepoint.pdf
15. Wittemyer G, Douglas-Hamilton I, Getz WM. The socioecology of elephants: analysis of the processes creating multitiered social structures G. Anim Behav. 2004;69(6):1357–71. Available from: http://dx.doi.org/10.1016/j.anbehav.2004.08.018
16. Patzelt A, Kopp GH, Ndao I, Kalbitzer U, Zinner D, Fischer J. Male tolerance and male–male bonds in a multilevel primate society. Proc Natl Acad Sci. National Acad Sciences; 2014;111(41):14740–5.
17. Whitehead H. Analyzing animal societies: quantitative methods for vertebrate social analysis. Chicago: University of Chicago Press; 2008. 161-168 p. Available from: http://public.eblib.com/choice/publicfullrecord.aspx?p=408184.
18. Bridge PD. Classification. In: Fry J, editor. Biological data analysis. Oxford, UK: Oxford University Press; 1993. p. 219–42.
19. Whitehead H. SOCPROG: program for analyzing social structure. Behav Ecol Sociobiol. 2009;63(5):765–78. Available from: http://dx.doi.org/10.1007/s00265-008-0697-y%0A
20. Smolker RA, Connor RC. "Pop" Goes the Dolphin: a Vocalization Male Bottlenose Dolphins Produce During Consortships. Behaviour. 1996;133(9–10):643–62. Available from: http://dx.doi.org/10.1163/156853996X00404
21. Amos B, Hoelzel AR. Long-term preservation of whale skin for DNA analysis. In: Hoelzel AR, editor. Genetic ecology of whales and dolphins Report of the International Whaling Commission. 1991. p. 99–103.
22. Aasen E, Medrano JF. Amplification of the Zfy and Zfx Genes for Sex Identification in Humans, Cattle, Sheep and Goats : Nature Biotechnology. Nature Publishing Group; 1990. p. 1279–81. Available from: http://www.nature.com/nbt/journal/v8/n12/abs/nbt1290-1279.html
23. Gilson A, Syvanen M, Levine K, Banks J. Deer gender determination by polymerase chain reaction: Validation study and application to tissues, bloodstains, and hair forensic samples from California. Calif Fish Game. 1998;84(4):159–69.
24. Krützen M, Mann J, Heithaus MR, Connor RC, Bejder L, Sherwin WB. Cultural transmission of tool use in bottlenose dolphins. Proc Natl Acad Sci. 2005;102(25):8939–43. Available from: https://doi.org/10.1073/pnas.0500232102
25. Raymond M, Rousset F. GENEPOP (Version 1.2): Population Genetics Software for Exact Tests and Ecumenicism. J Hered. 1995;86(3):248–9. Available from: http://dx.doi.org/10.1093/oxfordjournals.jhered.a111573
26. Kalberer S. Relatedness patterns in male bottlenose dolphin alliances in Shark Bay , Western Australia (Master thesis). University of Zurich; 2012.
27. Wang J. Triadic IBD coefficients and applications to estimating pairwise relatedness. Genet Res. 2007;89(3):135–53. Available from: https://doi.org/10.1017/S0016672307008798
28. Wang J. Coancestry: A program for simulating, estimating and analysing relatedness and inbreeding coefficients. Mol Ecol Resour. 2010;11(1):141–5. Available from: https://doi.org/10.1111/j.1755-0998.2010.02885.x
29. Nater A, Kopps AM, Krützen M. New polymorphic tetranucleotide microsatellites improve scoring accuracy in the bottlenose dolphin *Tursiops aduncus*. Mol Ecol Resour. 2009;9(2):531–4. Available from: http://dx.doi.org/10.1111/j.1755-0998.2008.02246.x
30. Krützen M, Valsecchi E, Connor RC, Sherwin WB. Characterization of microsatellite loci in *Tursiops aduncus*. Mol Ecol Notes. Wiley Online Library; 2002;1(3):170–2. Available from: https://doi.org/10.1046/j.1471-8278.2001.00065.x
31. Shinohara M, Domingo-Roura X, Takenaka O. Microsatellites in the bottlenose dolphin *Tursiops truncatus*. Mol Ecol. 1997;6(7):695–6. Available from: https://doi.org/10.1046/j.1365-294X.1997.00231.x
32. Hoelzel A, Dahlheim M, Stern S. Low genetic variation among killer whales (*Orcinus orca*) in the eastern North Pacific and genetic differentiation between foraging specialists. J Hered. Am Genetic Assoc; 1998;89(2):121–8.
33. Valsecchi E, Amos W. Microsatellite markers for the study of cetacean populations. Mol Ecol. Wiley Online Library; 1996;5(1):151–6. Available from: https://doi.org/10.1111/j.1365-294X.1996.tb00301.x
34. Brookfield J. A simple new method for estimating null allele frequency from heterozygote deficiency. Mol Ecol. Wiley Online Library; 1996;5(3):453–5. Available from: https://doi.org/10.1046/j.1365-294X.1996.00098.x

35. Weir BS, Cockerham CC. Estimating F-Statistics for the Analysis of Population Structure. Evolution (N Y). JSTOR; 1984;38(6):1358. Available from: https://doi.org/10.1111/j.1558-5646.1984.tb05657.x