

*The following supplement accompanies the article*

# **Speciation of two stingrays with antitropical distributions: low levels of divergence in mitochondrial DNA and morphological characters suggest recent evolution**

**A. Le Port<sup>1,2,\*</sup>, M. D. M. Pawley<sup>3</sup>, S. D. Lavery<sup>1,2</sup>**

<sup>1</sup>Leigh Marine Laboratory, University of Auckland, PO Box 349, Warkworth 0941, New Zealand

<sup>2</sup>School of Biological Sciences, University of Auckland, Private Bag 92019, Auckland, New Zealand

<sup>3</sup>Institute of Information and Mathematical Sciences, Massey University, Auckland 0745, New Zealand

\*Email: agnes.leport@gmail.com

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**Supplement.** Details of specimens and characters used in analyses, and raw morphological data.

Table S1. List of specimens used in molecular and morphometric and meristic analyses. \*x-ray data available

Species	Country	Sampling location	Sample code/voucher number	Sex	mtDNA markers			
					COI	cytb	dloop	
<b>Ingroups</b>								
<i>Dasyatis breviceaudata</i> (Short-tailed stingray)	Australia	Melbourne, SW Pacific, VIC	Dbr05ME01					
			Dbr05ME02		X	X	X	
			Dbr05ME03					
			Dbr05ME04					
			Dbr07ME04					
			Storm Bay, Tasmania Gascoyne Seamount, Tasman Sea Rottneest Island, Eastern Indian Ocean, WA	Dbr07Tas01/H6257-01*	F		X	
				Dbr07Tas02/H6312-19*	M			
				Dbr07WA01/H6340-17*	F			
				Dbr07WA02/H6346-25*	F		X	X
				Dbr07WA06				
	New Zealand	Manukau Harbour, Tasman Sea	Dbr04Ma02					
			Dbr07Ma02					
			Dbr04Le01					
			Dbr04MOO2					
			Dbr04MO05					
			Bay of Islands, SW Pacific Marlborough Sounds, SW Pacific Poor Knights Islands, SW Pacific	Dbr05BI01		X	X	X
				Dbr05MS02				
				Dbr05PK22				
				Dbr05PK29				
				Dbr05Wa01				
South Africa	Waitemata Harbour, SW Pacific Coromandel Peninsula, SW Pacific Parengarenga Harbour, SW Pacific False Bay, Western Indian Ocean	Dbr06CO02						
		Dbr06Pa03			X	X		
		Dbr06SA03		X	X	X		
		Dbr06SA04						
		Dbr06SA05						
	St Francis Bay, Western Indian Ocean Struis Bay, Western Indian Ocean	Dbr06SA06			X	X		
Dbr08SA01			X	X	X			
		Dbr08SA03						
<i>Dasyatis matsubarai</i> (Pitted stingray)	Japan	Miura Peninsula, Kanagawa Prefecture Nomozaki, Nagasaki	Dmat06MP01		X	X	X	
			Dmat06NA01		X	X	X	
<b>Outgroups</b>								
<i>Dasyatis thetidis</i> (Longtail stingray)	New Zealand	SW Pacific	Dth04Pa01			X	X	
			Dth05PK03					
	South Africa	Western Indian Ocean Unknown locality	Dth06SA01		X	X	X	
			Dth07SA02		X	X	X	
<i>Dasyatis lata</i> (Brown stingray)	USA	Central Pacific	Dlat06US01		X	X	X	
<i>Dasyatis sabina</i> (Atlantic stingray)	USA	Atlantic Ocean	Dsab06US01		X	X	X	

Table S2. *Dasyatis brevicaudata* and *D. matsubarai*. Material examined for morphometric and meristic analyses. \*x-ray data available; <sup>n</sup>tissue samples also available and used in molecular analyses (<sup>1</sup>Dbr07Tas02, <sup>2</sup>Dbr07Tas01, <sup>3</sup>Dbr07WA01, <sup>4</sup>Dbr07WA02, <sup>5</sup>Dth06SA01; see Table S1), <sup>-</sup>samples not included in morphometric analyses

Species	Collecting location or coordinates	Country	Voucher no.	Sex	Disc width (mm)
<i>D. brevicaudata</i>	Manukau Harbour, Tasman Sea	New Zealand	P34697*	F	540.0
	Ninety Mile Beach, Tasman Sea		P34853	F	290.0
	Hauraki Gulf, SW Pacific		DMN2 4681*	F	327.0
	Hauraki Gulf, SW Pacific		DM 4681*	F	430.5
	Whangaparoa Peninsula, SW Pacific		DM 5544*	F	530.0
	Gascoyne Seamount, Tasman Sea	Australia	H6312-191*	M	673.0
	Storm Bay, Tasmania		H6257-012*	F	492.0
	Eden, Tasman Sea, NSW		H1003-1*	F	365.0
	Point Dover/Low Point, Eastern Indian Ocean, WA		CA3520	M	345.0
	Rottneest Island, Eastern Indian Ocean, WA		H6340-173*	F	325.0
	Rottneest Island, Eastern Indian Ocean, WA		H6346-254	F	506.0
	Western Indian Ocean (-35°10.3'S; 21°09.9'E)	South Africa	SAM 36411*	M	560.0
	Western Indian Ocean (-35°4.0'S; 20°11.0'E)		SAM 32486*	M	430.0
	Unknown locality		SAM 35599*	F	461.0
	Unknown locality		SAM 35915*--	M	642.0
	Unknown locality		SAM 34865*--	F	620.0
	Unknown locality		SAM 36891*--	F	390.0
	Western Indian Ocean (-34°41'06''S; 21°14'54''E)	SAIAB 26470*	M	582.0	
	Western Indian Ocean (-35°13'024''S; 20°39'048''E)		SAIAB 27444*	F	750.0
	Unknown locality		SAIAB 26469*	M	425.5
<i>D. matsubarai</i>	Minamikayabe, Hokkaido	Japan	HUMZ 70808*	F	470.0
	SE coast Osima Peninsula, Hokkaido		HUMZ 18144*	M	422.0
	Numazu City, Shizuoka Prefecture		HUMZ 106269*	F	395.5
	Numazu City, Shizuoka Prefecture		HUMZ 105481*	F	383.5
	Numazu City, Shizuoka Prefecture		HUMZ 105477*	M	387.5
	Numazu City, Shizuoka Prefecture		HUMZ 105478*	M	400.0
	Numazu City, Shizuoka Prefecture		HUMZ 105482*	F	367.0
	Unknown locality		HUMZ 65256*	F	416.0
	Unknown locality		HUMZ 35022*	F	395.0
	Unknown locality		HUMZ _85.4.23D*	M	390.0

Table S3. Definitions of 33 morphological characters measured and presented in Table S4

<b>Abbreviation</b>	<b>Morphometric character</b>	<b>Methodology</b>
TL	Total length	Greatest straight line distance between snout tip and apex of tail
DW	DW - Disc width	Maximum straight line distance between wing tips
DL1	Disc length excluding pelvics	Distance from mid point of anterior margin to free rear tip of pectoral fin
DL2	Disc length including pelvics	Distance from mid point of anterior margin to free rear tip of pelvic fin
POB	Preorbital snout length	Distance from snout tip to fleshy anterior margin of orbit
SWP	Snout to max width of pectorals	Distance from snout tip to maximum width of pectoral fins
PSP	Prespine length	Distance from snout to base of 1st sting
STF	Snout to ventral tailfold	Distance from snout tip to base of ventral tailfold
VTH	Ventral tailfold height	Greatest vertical height from ventral tailfold base to edge
VTL	Ventral tailfold length	Distance from origin to end of ventral tailfold
IPD	Interpectoral distance	Shortest distance between pectoral fin insertions
HED	Horizontal eye diameter	Length of eye including eye socket
IOW	Interorbital width	Distance between soft interorbit taken at mid-length of eye
SPL	Spiracle length	Maximum width of opening
POR	Preoral length	Distance from snout tip to upper jaw
PNL	Prenasal length	Distance from snout tip to anterior edge of nostril
PGL	Prebranchial length	Distance from snout tip to upper edge of the first gill slit
HDL	Head length	Distance from snout tip to upper edge of the fifth gill slit
GS1/5	Distance between 1st and 5th gill slit	Shortest distance between outer edge of first and fifth gill slits
IGS1	Distance between 1st gill slits	Shortest distance between first gill slits
GS1	First gill slit height	Vertical height of first gill slit
GS5	Fifth gill slit height	Vertical height of fifth gill slit
SVL	Pre-vent length	Distance from snout tip to anterior end of cloaca
SPV	Prepelvic length	Distance from snout tip to pelvic fin origin (use finger to find origin)
MOW	Mouth width	Maximum width of mouth measured internally
NSL	Nostril length	Maximum length of opening
INW1	Internasal width (anterior)	Shortest distance between the two nostrils (taken at anterior edge of nostrils)
INW2	Internasal width (posterior)	Shortest distance between the two nostrils (taken at posterior edge of nostrils)
NCL	Nasal curtain length	Maximum distance from base to outer tip of nasal curtain
NCW	Nasal curtain width	Maximum width of internasal flap
PVL	Pelvic fin length	Distance from anterior margin of vent to free rear tip of pelvic fin
TAL1	Tail length 1	Distance from anterior margin of cloaca to apex of tail
TAL2	Tail length 2	Distance from posterior margin of cloaca to apex of tail

Table S4. Morphological dataset used in CAP analysis (33 characters). Measurements of *Dasyatis brevicaudata* and *D. matsubarai* presented as a percentage of disc width

Species	Country	Sample code	Sex	DW	%DW																															
					TL	DL1	DL2	POB	SWP	PSP	STF	VTH	VTL	IPD	HED	IOW	SPL	POR	PNL	PGL	HDL	GS1/5	IGS1	GS1	GS5	SVL	SPV	MOW	NSL	INW1	INW2	NCL	NCW	PVL	TAL1	TAL2
<i>Dasyatis brevicaudata</i>	Australia	H6312-19	M	673	166.9	82.8	90.5	19.0	38.6	112.2	114.4	0.8	29.5	15.1	3.3	13.2	6.0	18.7	14.1	29.3	37.7	12.0	21.2	3.6	2.5	71.6	70.7	10.0	3.1	10.9	13.5	6.1	12.3	6.7	95.1	89.2
<i>Dasyatis brevicaudata</i>	Australia	CA3520	M	345	149.3	81.2	88.4	16.2	35.7	113.0	115.9	0.8	30.4	15.3	5.7	11.0	6.2	20.3	14.5	29.0	41.2	12.1	22.0	2.2	1.8	71.0	71.6	10.4	3.5	11.9	14.1	6.0	11.6	11.4	76.8	71.9
<i>Dasyatis brevicaudata</i>	Australia	H6257-01	F	492	168.7	78.5	87.0	15.2	31.9	122.6	90.9	1.0	29.6	18.8	4.1	13.4	6.4	16.7	10.8	25.8	37.2	13.3	23.9	2.8	2.4	67.5	65.0	9.8	3.2	11.2	14.6	4.8	11.0	7.0	101.6	97.6
<i>Dasyatis brevicaudata</i>	Australia	H1003-1	F	365	169.9	84.9	90.4	16.4	30.1	104.7	115.1	0.8	26.6	18.0	6.1	12.1	6.7	17.8	14.5	29.0	41.1	13.2	23.6	3.6	2.7	76.7	71.2	10.7	4.0	11.2	14.7	5.3	12.6	10.0	95.9	91.2
<i>Dasyatis brevicaudata</i>	Australia	H6340-17	F	325	179.7	86.2	88.9	17.2	41.2	114.5	117.5	1.2	30.6	16.6	5.0	12.1	7.1	19.1	14.2	31.4	40.3	14.0	22.3	2.2	1.9	72.0	69.5	11.3	3.9	12.1	14.7	4.9	12.6	8.8	103.1	98.5
<i>Dasyatis brevicaudata</i>	Australia	H6346-25	F	506	161.1	80.4	88.3	17.2	32.0	118.6	118.6	0.7	29.2	15.8	4.0	13.3	6.1	17.8	13.4	28.1	39.9	11.9	22.2	3.0	2.3	70.6	69.8	9.6	3.8	11.6	14.8	5.4	12.7	10.1	89.9	84.4
<i>Dasyatis brevicaudata</i>	New Zealand	P34797	F	540	166.7	85.2	94.1	19.4	34.3	104.4	118.9	1.1	36.9	18.0	3.1	16.8	6.4	20.0	15.2	30.2	42.4	12.2	22.3	4.7	2.6	74.1	74.8	9.8	7.2	12.4	14.4	5.4	12.5	5.7	93.5	88.3
<i>Dasyatis brevicaudata</i>	New Zealand	DMN2 4681	F	327	191.1	86.5	95.1	19.5	37.9	120.2	121.7	1.2	30.7	19.5	5.9	12.8	7.3	19.1	15.0	28.5	43.3	13.3	23.1	2.3	2.2	77.4	75.2	10.2	3.3	12.6	14.8	5.0	12.6	10.3	108.0	102.1
<i>Dasyatis brevicaudata</i>	New Zealand	DM 5544	F	530	156.0	82.1	89.4	16.3	34.9	96.8	110.2	1.1	30.0	18.1	3.1	14.2	6.1	17.0	13.2	27.7	37.1	12.0	20.3	3.2	2.4	70.6	70.8	9.5	3.5	11.3	13.6	4.5	12.2	11.0	84.2	80.4
<i>Dasyatis brevicaudata</i>	New Zealand	DM 4681	F	430.5	157.7	72.7	90.6	17.7	61.1	105.9	116.8	0.9	29.0	19.3	4.2	12.5	6.3	17.7	14.2	28.7	41.6	12.1	21.0	2.7	1.6	72.5	73.4	8.7	3.2	11.6	12.9	4.7	11.5	9.4	84.1	79.3
<i>Dasyatis brevicaudata</i>	New Zealand	P34853	F	290	193.1	86.6	93.1	16.2	42.4	121.6	112.8	1.3	35.9	20.0	6.0	14.8	7.6	16.6	10.1	26.5	39.8	14.1	23.4	3.0	2.4	75.3	73.8	14.5	4.5	13.4	15.5	6.4	13.4	10.9	118.1	111.5
<i>Dasyatis brevicaudata</i>	South Africa	SAM 36411	M	560	159.8	80.5	86.4	18.0	36.6	101.3	112.0	1.2	30.4	16.6	4.4	12.3	6.3	17.5	12.9	27.3	38.8	13.1	35.4	3.3	2.4	68.2	67.9	9.3	3.4	10.8	13.8	5.2	11.2	7.1	89.5	84.3
<i>Dasyatis brevicaudata</i>	South Africa	SAM 35915	M	642	170.6	83.3	86.8	17.1	35.8	103.9	108.3	0.7	31.3	15.4	5.5	12.1	5.2	14.6	10.4	25.9	38.3	13.5	21.5	3.6	2.6	70.1	67.2	9.3	3.5	11.2	14.4	6.3	12.2	5.8	98.6	93.5
<i>Dasyatis brevicaudata</i>	South Africa	SAM 34865	F	620	151.0	85.2	91.9	18.9	39.8	111.0	113.9	1.1	33.1	17.6	5.1	13.2	6.0	30.6	14.4	29.7	40.7	13.4	21.8	3.5	2.4	72.3	72.6	9.8	3.7	12.4	14.4	5.9	12.5	8.1	77.4	71.8
<i>Dasyatis brevicaudata</i>	South Africa	SAM 36891	F	390.5	175.9	85.5	89.8	17.9	33.5	111.7	112.4	1.0	27.9	15.0	5.4	11.4	6.6	17.3	13.1	27.8	39.7	13.2	22.4	3.4	2.6	71.7	68.6	10.1	3.3	11.9	15.3	5.0	11.9	7.6	100.6	96.8
<i>Dasyatis brevicaudata</i>	South Africa	SAM 32486	M	430	180.2	81.4	87.4	17.7	34.4	111.6	115.9	0.9	28.2	18.0	4.7	11.4	6.6	15.8	12.4	27.0	39.0	12.6	21.5	3.1	2.7	68.1	61.6	9.4	3.1	11.3	13.6	5.6	11.5	6.4	108.1	102.1
<i>Dasyatis brevicaudata</i>	South Africa	SAM 35599	F	461	169.1	83.1	88.9	15.4	37.4	111.9	114.5	1.3	32.2	17.7	5.0	11.0	6.5	16.8	12.1	27.7	39.5	13.1	20.8	3.0	2.5	71.6	71.1	9.2	3.3	11.4	14.3	5.3	11.8	6.5	97.0	91.3
<i>Dasyatis brevicaudata</i>	South Africa	SAIAB 26470	M	582	173.5	82.1	86.9	16.8	45.0	110.0	114.1	1.5	35.3	17.0	4.7	11.9	6.5	17.7	13.1	28.2	40.2	12.9	20.3	3.8	2.8	69.8	68.4	9.1	3.1	11.2	14.3	5.3	11.8	8.2	101.5	96.6
<i>Dasyatis brevicaudata</i>	South Africa	SAIAB 27444	F	750	139.6	78.8	82.7	16.7	33.7	103.3	103.5	1.2	25.1	18.1	3.8	11.8	6.2	16.3	11.9	25.6	36.7	12.6	19.8	3.1	1.8	65.6	65.3	8.5	3.1	10.8	13.2	4.6	10.9	8.5	72.5	66.7
<i>Dasyatis brevicaudata</i>	South Africa	SAIAB 26469	M	425.5	169.2	81.8	89.5	16.8	34.3	112.1	113.7	1.6	30.6	18.8	4.6	11.6	6.6	19.0	14.1	29.1	41.1	13.6	22.1	3.4	2.0	72.4	72.2	10.2	2.9	11.9	14.1	5.6	11.2	6.1	94.0	89.3
<i>Dasyatis matsubarai</i>	Japan	HUMZ 35022	F	395	196.7	86.3	92.4	17.7	36.2	115.4	115.2	1.3	37.6	18.4	5.8	12.4	6.8	18.5	14.0	28.6	40.5	13.4	22.0	3.4	2.2	74.4	72.3	9.5	3.8	11.8	12.1	6.2	11.6	10.7	122.5	118.5
<i>Dasyatis matsubarai</i>	Japan	HUMZ 70808	F	470	175.1	81.9	89.0	17.2	39.9	110.2	109.1	1.1	30.7	15.6	5.0	13.1	6.4	14.7	10.9	24.3	37.2	13.3	22.3	3.0	2.3	71.3	71.6	10.0	3.9	10.9	12.7	5.2	11.5	10.5	103.2	96.5
<i>Dasyatis matsubarai</i>	Japan	HUMZ 106269	F	395.5	163.6	85.5	90.6	15.8	42.2	115.8	116.3	0.9	32.3	16.4	5.8	15.1	6.2	15.7	12.1	30.0	41.3	12.9	22.4	2.6	2.4	73.3	71.3	9.7	3.6	11.6	14.2	4.0	11.6	10.6	89.8	83.7
<i>Dasyatis matsubarai</i>	Japan	HUMZ 105481	F	383.5	177.1	86.4	92.0	17.6	43.9	88.3	114.7	1.1	40.3	13.9	5.9	12.5	6.5	19.2	13.0	28.7	41.5	13.8	22.8	3.4	2.5	73.7	70.7	9.8	4.3	11.8	14.8	6.0	12.5	8.0	102.7	97.0
<i>Dasyatis matsubarai</i>	Japan	HUMZ 18144	M	422	192.1	84.1	92.9	17.8	38.3	91.0	114.1	1.4	37.4	17.4	5.4	13.6	6.8	19.5	14.8	29.9	41.9	13.9	22.3	3.1	2.0	75.6	71.6	9.0	5.1	11.2	13.3	5.5	12.3	8.3	116.4	112.1
<i>Dasyatis matsubarai</i>	Japan	HUMZ 105477	M	387.5	151.2	81.0	85.5	16.8	39.7	104.5	107.9	1.1	35.1	12.8	5.1	11.4	6.9	17.4	12.6	26.5	39.0	10.8	21.1	3.1	2.4	67.5	67.0	9.4	3.8	11.5	13.7	5.8	12.0	11.0	82.3	76.9
<i>Dasyatis matsubarai</i>	Japan	HUMZ 105478	M	400	179.6	84.8	88.0	15.0	42.3	110.5	108.3	0.8	32.1	13.2	5.5	11.2	6.5	17.0	11.2	26.3	38.9	13.3	20.9	3.4	2.4	70.0	70.0	9.6	3.8	11.6	14.5	5.3	12.0	11.1	108.5	104.1
<i>Dasyatis matsubarai</i>	Japan	HUMZ 65256	F	416	187.5	85.8	91.7	16.0	37.0	114.2	112.7	1.1	32.6	16.7	5.4	13.2	6.3	16.8	13.9	25.8	39.7	13.5	21.9	3.4	2.4	72.8	71.6	10.4	3.7	11.4	13.2	5.6	11.3	10.1	113.0	108.2
<i>Dasyatis matsubarai</i>	Japan	HUMZ 105482	F	367	165.4	82.2	86.4	17.2	39.6	104.1	106.5	1.3	32.5	12.5	5.7	12.8	6.4	16.3	12.0	26.0	37.6	12.7	21.2	3.2	1.9	67.0	65.7	9.0	5.1	11.2	13.5	5.7	11.9	8.6	97.0	92.6
<i>Dasyatis matsubarai</i>	Japan	HUMZ_85.4.23.D	M	390	141.0	82.9	87.2	16.7	39.5	105.1	109.2	1.3	31.3	13.3	6.8	11.5	5.7	19.5	14.4	26.0	40.3	12.2	20.6	3.0	2.5	69.5	67.4	9.4	3.4	11.2	13.6	6.0	11.9	10.4	70.0	66.9

Fig. S1. A constrained ordination comparing *Dasyatis brevicaudata* ( $n = 20$ ) and *D. matsubarae* ( $n = 10$ ) ('Species' grouping variable) using canonical analysis of principal coordinates (CAP) on 33 morphological characters

