

Supplementary material

Figure S1. Correlation of forearm sizes and latitude of *P. fulvus* and *P. gymnonotus*.

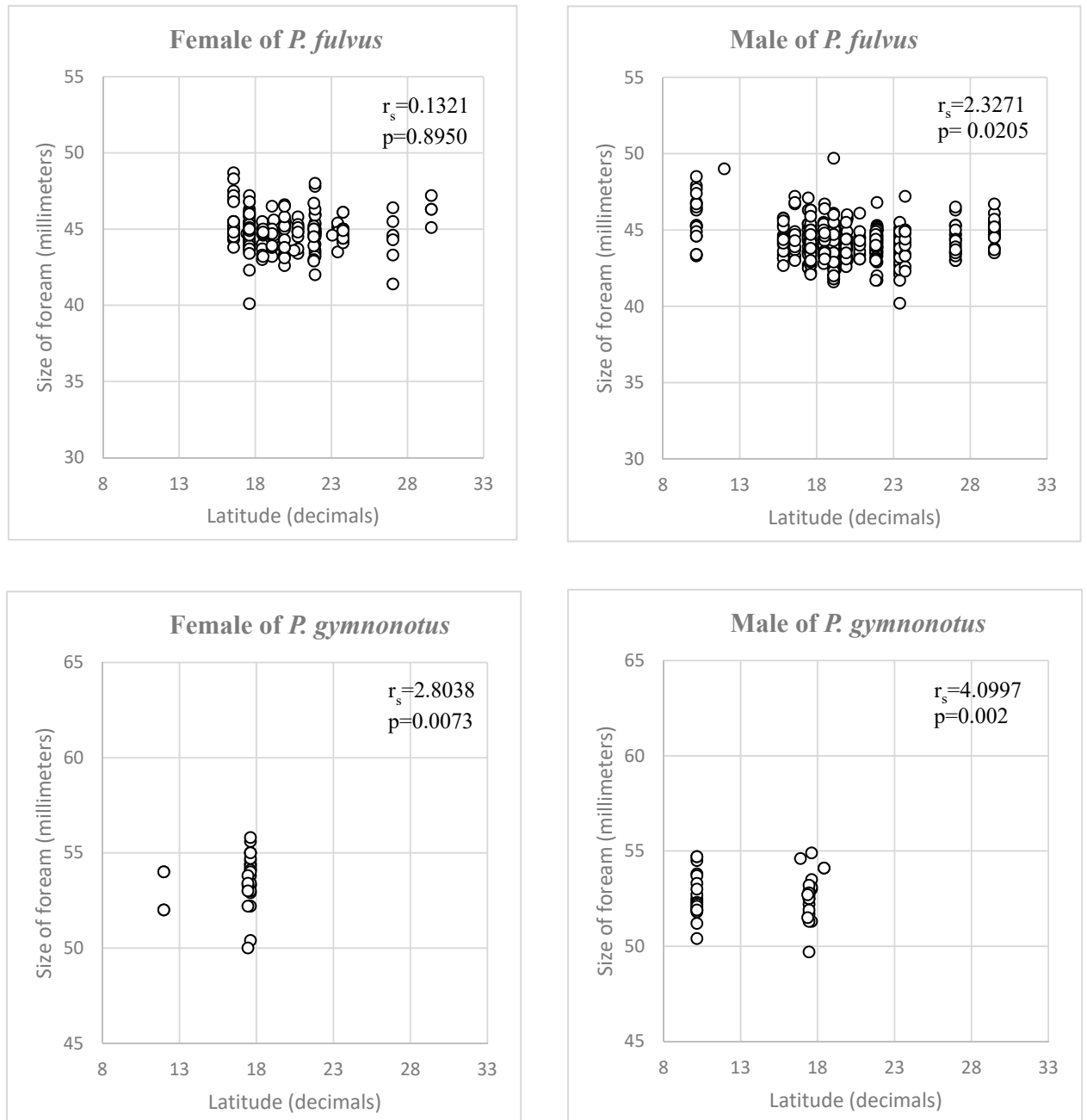


Figure S2. Bayesian reconstruction with divergence time for DNanc+DNamt in BEAST. Bars show the 95% interval for the high posterior density (HPD). Divergence time value above each branch, time in millions of years ago. Down with Posterior probability.

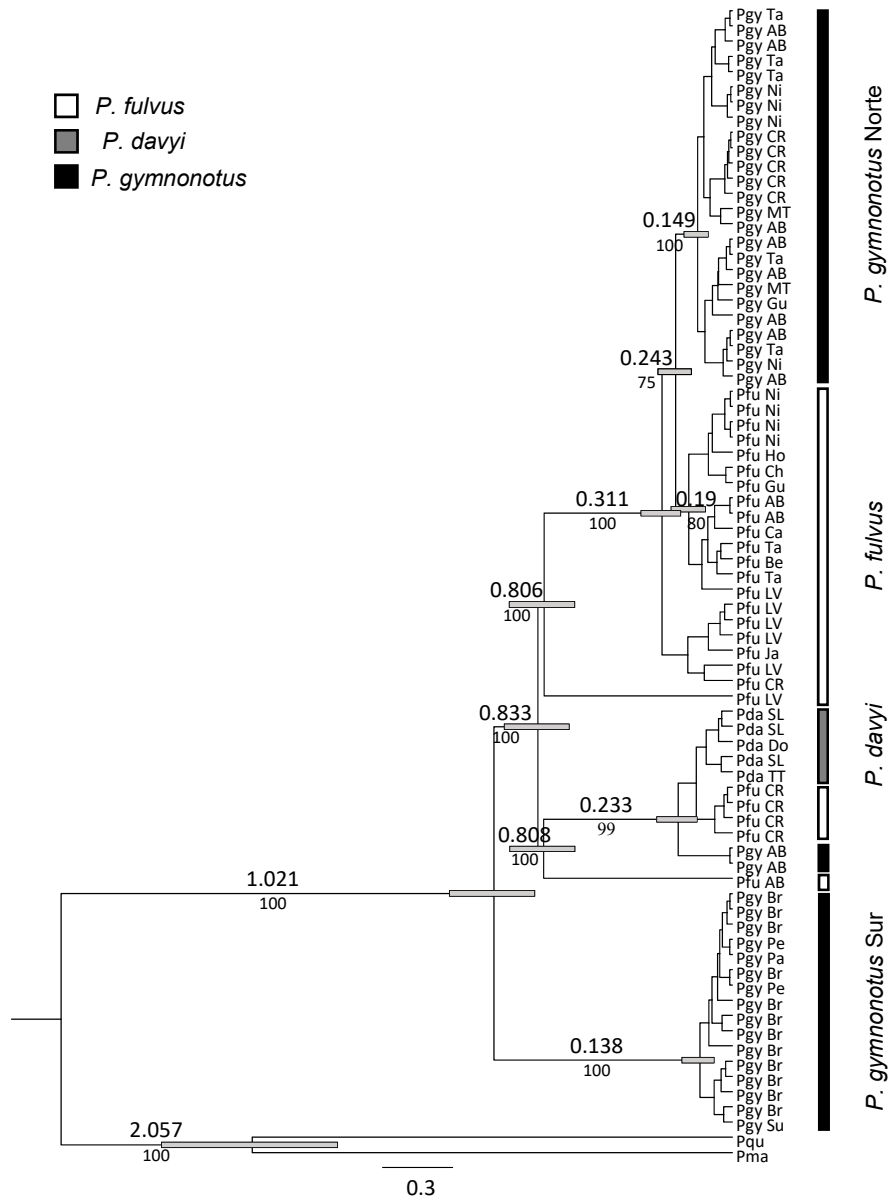


Figure S3. Distribution haplotype for **a.** *P. fulvus* and **b.** *P. gymnonotus*.

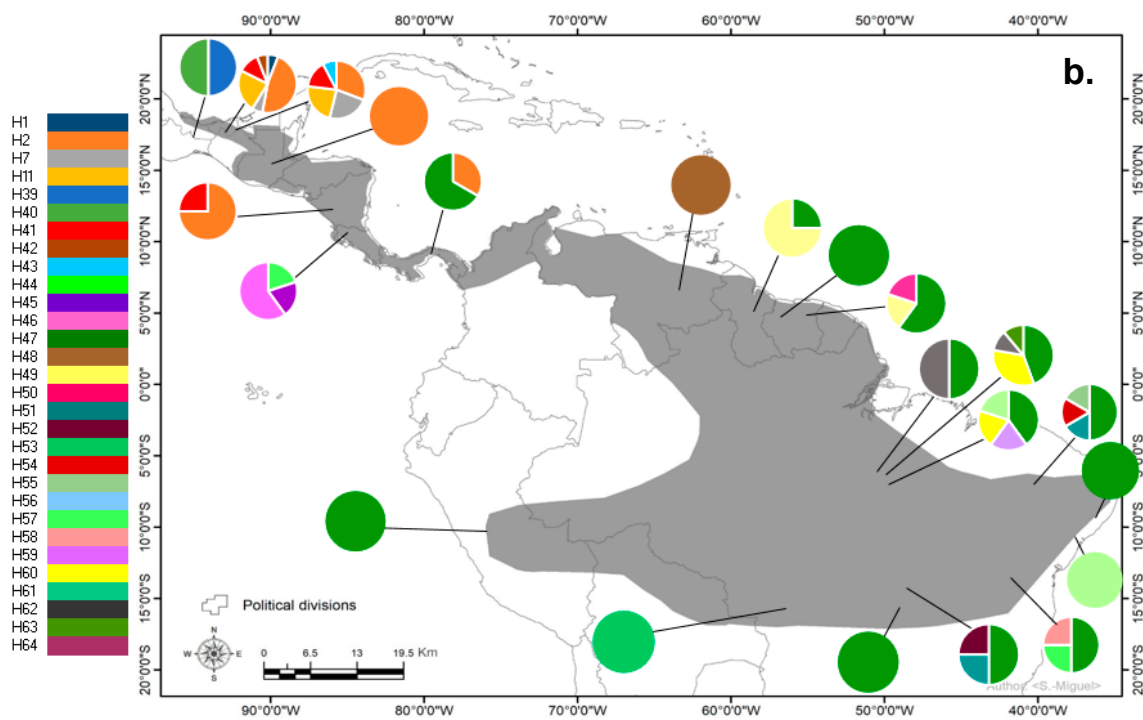
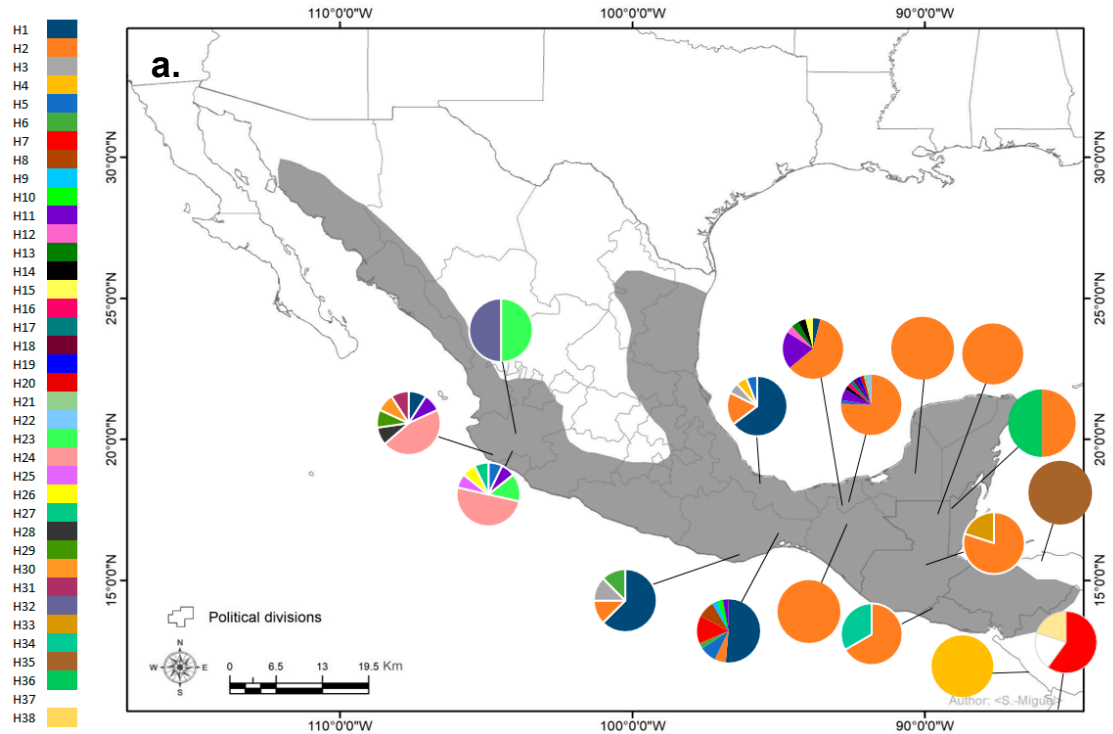


Table S1. Study localities type of amplified marker (B=COI; C=PRKC1; D= STAT5A; F=RAG2; M=Microsatellites) and GenBank access number.

Study site	Species	Marker	ID	GenBank	Citation
Laguna Encantada, Veracruz, Mexico LE	<i>P. fulvus</i>	B	PfLE01	MT860955	This studio
		B	PfLE02	MT860956	
		B	PfLE03	MT860957	
		B	PfLE04	MT860958	
		B	PfLE05	MT860959	
		B	PfLE06	MT860960	
		B	PfLE07	MT860961	
		B	PfLE08	MT860962	
		B	PfLE09	MT860963	
		B	PfLE10	MT860964	
		B	PfLE11	MT860965	
		B	PfLE12	MT860966	
		B	PfLE13	MT860967	
		B	PfLE14	MT860968	
		B	PfLE15	MT860969	
		B	PfLE16	MT860970	
		B	PfLE17	MT860971	
Cerro Huatulco, Oaxaca, Mexico CH	<i>P. fulvus</i>	BM	PfCH01	MT860972	This studio
		BM	PfCH02	MT860973	
		BM	PfCH03	MT860974	
		BM	PfCH04	MT860975	
		BM	PfCH05	MT860976	
		BM	PfCH06	MT860977	
		BM	PfCH07	MT860978	
		BM	PfCH08	MT860979	
		BM	PfCH09p	MT860980	
		BM	PfCH10	MT860981	
		BM	PfCH11	MT860982	

		BM	PfCH12	MT860983	
		BM	PfCH13	MT860984	
		BM	PfCH14	MT860985	
		BM	PfCH15	MT860986	
		BM	PfCH16	MT860987	
La Venta, Oaxaca, Mexico LV	<i>P. fulvus</i>	BM	PfLV01	MT860920	This studio
		BM	PfLV02	MT860921	
		BM	PfLV03	MT860922	
		BM	PfLV04	MT860923	
		BM	PfLV05	MT860924	
		BM	PfLV06	MT860925	
		BM	PfLV07	MT860926	
		BM	PfLV08	MT860927	
		BM	PfLV09	MT860928	
		BM	PfLV10	MT860929	
		BM	PfLV11	MT860930	
		BM	PfLV12	MT860931	
		BM	PfLV13	MT860932	
		BM	PfLV14	MT860933	
		BCDFM	PfLV15	MT860934, MT555040, MT549059, MT536984	
		BM	PfLV16	MT860935	
		BM	PfLV17	MT860936	
		BM	PfLV18	MT860937	
		BM	PfLV19	MT860938	
		BM	PfLV20	MT860939	
		BM	PfLV21	MT860940	
		BM	PfLV22	MT860941	
		BM	PfLV23	MT860942	
		BM	PfLV24	MT860943	
		BCDFM	PfLV25	MT860944, MT555041, MT549060, MT536985	
		B	PfLV26	MT860945	
		BM	PfLV27	MT860946	

		BM BCDFM BCDFM BCDFM BM BM BM BCDFM	PfLV28 PfLV29 PfLV30 PfLV31 PfLV32 PfLV33 PfLV34 PfLV35	MT860947 MT860948, MT555042, MT549061, MT536986 MT860949, MT555043, MT549062, MT536987 MT860950, MT555044, MT549063, MT536988 MT860951 MT860952 MT860953 MT860954, MT555045, MT549064, MT536989	
Martínez de la Torre, Oaxaca, Mexico MT	<i>P. gymnonotus</i>	BCDF BCDF	PgMT01 PgMT02	MT863662, MT559922, MT555024, MT547969 MT863663, MT559923, MT555025, MT547970	[55]
Tapijulapa, Tabasco, Mexico Ta	<i>P. fulvus</i>	M M BM BM BM BM M BM BM BM BM B BCDFM M BM BM BM BM M BM	PfTa01 PfTa02 PfTa03 PfTa04 PfTa05 PfTa06 PfTa07 PfTa08 PfTa09 PfTa10 PfTa11 PfTa12 PfTa13 PfTa14 PfTa15 PfTa16 PfTa17 PfTa18 PfTa19 PfTa20	- - MT860895 MT860896 MT860897 MT860898 - MT860899 MT860900 MT860901 MT860902 MT860903 MT860904, MT555038, MT549057, MT536982 - MT860905 MT860906 MT860907 MT860908 - MT860909	This studio

		BM BM M BM BM M BM BM BM BM BM BCDFM	PfTa21 PfTa22 PfTa23 PfTa24 PfTa25 PfTa26 PfTa27 PfTa28 PfTa29 PfTa30 PfTa31 PfTa32	MT860910 MT860911 - MT860912 MT860913 - MT860914 MT860915 MT860916 MT860917 MT860918 MT860919, MT555039, MT549058, MT536983	
Tapijulapa, Tabasco, Mexico Ta	<i>P. gymnonotus</i>	BM BCDFM BM BM BM BM BCDFM BCDFM BCDFM BCDFM BM BM BM BM BM BM BM	PgTa01 PgTa02 PgTa03 PgTa04 PgTa05 PgTa06 PgTa07 PgTa08 PgTa09 PgTa10 PgTa11 PgTa12 PgTa13 PgTa14 PgTa15 PgTa16 PgTa17	MT863645 MT863646, MT559917, MT555019, MT547964 MT863647 MT863648 MT863649 MT863650 MT863651, MT559918, MT555020, MT547965 MT863652, MT559919, MT555021, MT547966 MT863653, MT559920, MT555022, MT547967 MT863654, MT559921, MT555023, MT547968 MT863655 MT863656 MT863657 MT863658 MT863659 MT863660 MT863661	This studio
Agua Blanca, Tabasco, Mexico	<i>P. fulvus</i>	BM BM BM	PfAB01 PfAB02 PfAB03	MT559958 MT559959 MT559960	This studio

AB		B	PfAB04	MT559961	
		B	PfAB05	MT559962	
		BM	PfAB06	MT559963	
		BM	PfAB07	MT559964	
		BM	PfAB08	MT559965	
		BM	PfAB09	MT559966	
		BM	PfAB10	MT559967	
		BM	PfAB11	MT559968	
		BM	PfAB12	MT559969	
		BM	PfAB13	MT559970	
		BM	PfAB14	MT559971	
		BM	PfAB15	MT559972	
		BM	PfAB16	MT559973	
		BM	PfAB17	MT559974	
		BM	PfAB18	MT860863	
		BM	PfAB19	MT860864	
		BM	PfAB20	MT860865	
		BM	PfAB21	MT860866	
		BM	PfAB22	MT860867	
		BM	PfAB23	MT860868	
		BM	PfAB24	MT860869	
		BM	PfAB25	MT860870	
		BM	PfAB26	MT860871	
		BM	PfAB27	MT860872	
		BM	PfAB28	MT860873	
		BM	PfAB29	MT860874	
		BM	PfAB30	MT860875	
		BM	PfAB31	MT860876	
		BM	PfAB32	MT860877	
		BCDFM	PfAB33	MT860878, MT555035, MT549054, MT536979	
		M	PfAB34	-	
		BCDFM	PfAB35	MT860879, MT555036, MT549055, MT536980	

		BCDFM BM BM BM BM BM BM BM BM BM BM M BM BM BM M M	PfAB36 PfAB37 PfAB38 PfAB39 PfAB40 PfAB41 PfAB42 PfAB43 PfAB44 PfAB45 PfAB46 PfAB47 PfAB48 PfAB49 PfAB50 PfAB51 PfAB52 PfAB53	MT860880, MT555037, MT549056, MT536981 MT860881 MT860882 MT860883 MT860884 MT860885 MT860886 MT860887 MT860888 MT860889 MT860890 MT860891 - MT860892 MT860893 MT860894 - -	
Agua Blanca, Tabasco, Mexico AB	<i>P. gymnonotus</i>	M BM BM B BM BM BM BCDFM BCDFM BM BCDF BCDFM M BCDFM	PgAB02 PgAB03 PgAB04 PgAB05 PgAB06 PgAB07 PgAB08 PgAB09 PgAB10 PgAB11 PgAB12 PgAB13 PgAB14 PgAB15	- MT863619 MT863620 MT863621 MT863622 MT863623 MT863624 MT863625, MT559907, MT555009, MT547954 MT863626, MT559908, MT555010, MT547955 MT863627 MT863628, MT559909, MT555011, MT547956 MT863629, MT559910, MT555012, MT547957 - MT863630, MT559911, MT555013, MT547958	This studio

		BCDFM BM BM BCDFM BM BM BM BM BCDFM BM BM BCDFM BCDFM BM	PgAB16 PgAB17 PgAB18 PgAB19 PgAB20 PgAB21 PgAB22 PgAB23 PgAB24 PgAB25 PgAB26 PgAB27 PgAB28 PgAB29	MT863631, MT559912, MT555014, MT547959 MT863632 MT863633 MT863634, MT559913, MT555015, MT547960 MT863635 MT863636 MT863637 MT863638 MT863639, MT559914, MT555016, MT54761 MT863640 MT863641 MT863642, MT559915, MT555017, MT547962 MT863643, MT559916, MT555018, MT547963 MT863644	
Los Ortices, Colima, Mexico LO	<i>P. fulvus</i>	B BM BM BM BM BM BM BM BM BM BM BM BM BM	PfLO01 PfLO02 PfLO03 PfLO04 PfLO05 PfLO06 PfLO07 PfLO08 PfLO09 PfLO10 PfLO11 PfLO12 PfLO13 PfLO14	MT559944 MT559945 MT559946 MT559947 MT559948 MT559949 MT559950 MT559951 MT559952 MT559953 MT559954 MT559955 MT559956 MT559957	This studio
Playa de Oro, Colima, Mexico	<i>P. fulvus</i>	BM BM BM BM	PfPO01 PfPO02 PfPO03 PfPO04	MT559933 MT559934 MT559935 MT559936	This studio

PO		BM BM BM BM BM BM B	PfPO05 PfPO06 PfPO07 PfPO08 PfPO09 PfPO10 PfPO11	MT559937 MT559938 MT559939 MT559940 MT559941 MT559942 MT559943	
Jalisco, Mexico Ja	<i>P. fulvus</i>	BCD B	PJa01 PJa02	KX590178; KX590469; KX589689 KX590180	[31,34]
Campeche, Mexico Ca	<i>P. fulvus</i>	BCD	PCa01	JF447313; KX590455; KX589675	[31]
Chiapas, Mexico Ch	<i>P. fulvus</i>	BCDF	PCh01	KX590181; KX590471; KX589691; KX590385	[31]
Tikal, El Petén, Guatemala Gu1	<i>P. fulvus</i>	BCD	PGu06	JF446816; KX590459; KX589679	[31]
Grutas de Lanquín, Alta Verapaz, Guatemala Gu2	<i>P. fulvus</i>	B B B B B	PfGu01 PfGu02 PfGu03 PfGu04 PfGu05	JF446821 JF446820 JF446819 JF446818 JF446817	[29]
Grutas de Lanquín, Alta Verapaz, Guatemala Gu2	<i>P. gymnonotus</i>	BCDF	PgGu01	KX590174; KX590457; KX589677; KX590384	[31]

El Refugio, Ahuachapan, El Salvador ES	<i>P. fulvus</i>	B B B	PfES05 PfES06 PfES07	JF446542 JF446541 JF446540	[30]
Colón, Honduras Ho	<i>P. fulvus</i>	BCD	PfHo01	KX590179; KX590470; KX589690	[31]
Gallon Jug, Distrito de Orange Walk, Belize Be	<i>P. fulvus</i>	B BCD	PfBe01 PfBe02	KX590084 KX590085; KX590412; KX589631	[31]
Nidiri, Masaya Volcano Park, Nicaragua Ni	<i>P. fulvus</i>	BCDF BCDF BCDF BCDF	PfNi01 PfNi02 PfNi03 PfNi04	MT860854, MT559898, MT555000, MT547945 MT860855, MT559899, MT555001, MT547946 MT860856, MT559900, MT555002, MT547947 MT860857, MT559901, MT555003, MT547948	This studio
Nidiri, Masaya Volcano Park, Nicaragua Ni	<i>P. gymnonotus</i>	BCDF BCDF BCDF BCDF	PgNi01 PgNi02 PgNi03 PgNi04	MT871939, MT559924, MT555026, MT547971 MT871940, MT559925, MT555027, MT547972 MT871941, MT559926, MT555028, MT547973 MT871942, MT559927, MT555029, MT547974	This studio
Barra Honda National Park, Costa Rica CR	<i>P. fulvus</i>	BCDF BCDF BCDF BCDF BCDF	PfCR01 PfCR02 PfCR03 PfCR04 PfCR05	MT860858, MT559902, MT555004, MT547949 MT860859, MT559903, MT555005, MT547950 MT860860, MT559904, MT555006, MT547951 MT860861, MT559905, MT555007, MT547952 MT860862, MT559906, MT555008, MT547953	This studio
Barra Honda National Park, Costa Rica CR	<i>P. gymnonotus</i>	BCDF BCDF BCDF BCDF	PfCR01 PfCR02 PfCR03 PfCR04	MT862768, MT559928, MT555030, MT547975 MT862769, MT559929, MT555031, MT547976 MT862770, MT559930, MT555032, MT547977 MT862771, MT559931, MT555033, MT547978	This studio

		BCDF	PfCR05	MT862772, MT559932, MT555034, MT547979	
Altos de Campana National Park, Panama Pa	<i>P. gymnonotus</i>	BCD B B	PgPa01 PgPa02 PgPa03	JF447432; KX590462; KX589682 JF447431 JF459487	[30,31,54]
Hato la Florida, Bolivar, Venezuela Ve	<i>P. gymnonotus</i>	B	PgVe01	JF447854	[31]
Saint Joseph, Dominica Do	<i>P. davyi</i>	CD	PdDo03	KX590467; KX589687	[34]
Arena Reserve, Nariva, Trinidad and Tobago TT	<i>P. davyi</i>	BCD	PdTT01	KX590177; KX590468; KX589688	[34]
Castries, Santa Lucia SL	<i>P. davyi</i>	BCD BCDF BCD	PdSL02 PdSL06 PdSL08	KX590183; KX590472; KX589692 KX590193; KX590473; KX589693; KX590386 KX590195; KX590474; KX589694	[31]
Iwokrama Reserve, Potaro-Siparuni, Guyana Gy	<i>P. gymnonotus</i>	B B B B	PgGy01 PgGy02 PgGy03 PgGy04	EF080591 EF080589 EF080590 KC011684	[30,31,53]

Brokopondo, Surinam Su1	<i>P. gymnonotus</i>	BCD B B B B	PgSu02 PgSu03 PgSu04 PgSu05 PgSu06	KX590198; KX590477; KX589697 EU096898 EU096899 EU096901 JF447711	[30,31,52]
Bakhluis, Sipaliwini, Surinam Su2	<i>P. gymnonotus</i>	B	PgSu01	EU096900	[31]
Huanuco, Peru Pe	<i>P. gymnonotus</i>	BCD BCD	PgPe01 PgPe02	KX590196; KX590475; KX589695 KX590197; KX590476; KX589696	[31,34]
Caverna do Bigode, Goiás, Brazil Go	<i>P. gymnonotus</i>	B B B B	PgGo01 PgGo02 PgGo03 PgGo04	MT862764 MT862765 MT862766 MT862767	This studio
Barro Alto, Goiás, Brazil Br1	<i>P. gymnonotus</i>	BCD	PgBr02	KX590169; KX590453; KX589673	[31]
Tapirapé- Aquiri, Marabá, Pará, Brazil Br2	<i>P. gymnonotus</i>	BCDF B	PgBr32 PgBr33	KX590138; KX590441; KX589658; KX590381 KX590140	[31]
Carajás, Parauapebas, Pará, Brazil Br3	<i>P. gymnonotus</i>	B B B B BCD BCD B	PgBr23 PgBr24 PgBr25 PgBr26 PgBr27 PgBr28 PgBr29	KX590354 KX590357 KX590358 KX590359 KX590360; KX590524; KX589745 KX590364; KX590525; KX589746 KX590368	[31]

		B B	PgBr30 PgBr31	KX590370 KX590372	
Canaã dos Carajás, Pará, Brazil Br4	<i>P. gymnonotus</i>	BCD B B B B	PgBr18 PgBr19 PgBr20 PgBr21 PgBr22	KX590312; KX590519; KX589740 KX590313 KX590329 KX590339 KX590341	[31]
Chapada Diamantina, Bahía, Brazil Br5	<i>P. gymnonotus</i>	B BCDF B BCD	PgBr14 PgBr15 PgBr16 PgBr17	KX590307 KX590308; KX590517; KX589738; KX590398 KX590309 KX590310; KX590518; KX589739	[31]
Usina Serra Grande, Alagoas, Brazil Br6	<i>P. gymnonotus</i>	B	PgBr13	KX590306	[31]
Itabaiana, Sergipe, Brazil Br7	<i>P. gymnonotus</i>	B B BCD BCD	PgBr09 PgBr10 PgBr11 PgBr12	KX590294 KX590296 KX590304; KX590515; KX589736 KX590305; KX590516; KX589737	[31]
Aiuba Ecological Station, Aiuba, Ceará, Brazil Br8	<i>P. gymnonotus</i>	B BCDF B B BCD B	PgBr03 PgBr04 PgBr05 PgBr06 PgBr07 PgBr08	KX590284 KX590285; KX590510; X589731; KX590396 KX590286 KX590287 KX590288; KX590511; KX589732 KX590293	[31]

Jangada, Mato Grosso, Brazil Br9	<i>P. gymnonotus</i>	BCD	PgBr01	KX590156; KX590450; KX589669	[31]
Westmoreland; Revival; Monarva Cave, Jamaica	<i>P. macleayi</i>	BCDF	Pma	KX590077, KX590407, KX589626, KX590375	[31]
St. Anns Parish, Jamaica	<i>P. quadridens</i>	BCDF	Pqu	KX590268, KX590502, KX589722, KX590394	[31]

Table S2. Primers used for the amplification of fragments of COI, RAG2, and intrones (PRKC1 and STAT5A)

Locus	Primer 5'-3'	pb	Cita
COI VF1d VR1d	F: TTCTCAACCAACCACAARGAYATYGG R: TAGACTTCTGGGTGGCCRAARAAYCA	607	[58]
RAG2 RAG2-F1B RAG2-R1	F: ATCCTGCCCCACTGGAGTTTTTC R: AACYTGYTTATTGTCTCCTGGTATGC	717	[57]
Intron PRKC1	F: CTTGTCAATGATGATGAGG R: CCTATTTTAAAATATGAAAGAAATC	422	[59]
Intron STAT5A	F: CTGCTCATCAACAAGCCCGA R: GGCTTCAGGTTCCACAGGTTGC	475	[59]

Table S3. New microsatellite loci

For library construction, genomic DNA was extracted from the muscular tissue of one *P. psilotis* individual (Santiago Ixcuintla, Nayarit, Mexico; voucher UAM-I RLW140304Ppe74) using a salt-extraction protocol [114]. The sample was sent to the Georgia Genomics Facility (University of Georgia, Athens, USA). DNA was sheared using Covaris S2. Fragmented DNA was ligated to Illumina universal TruSeq adapters containing 10 custom nucleotide indexes [115, 116] and were enriched through polymerase chain reaction (PCR), purified and normalized, and sequenced using an Illumina HiSeq 2000 to produce paired-end 100-bp nucleotide reads. The resulting FASTQ files were demultiplexed and the readings were filtered and executed through the PALfinder pipeline to identify adequate microsatellites and for the design of primers [117]. More than 11 million 100-bp readings were obtained, and we registered 8321 loci with primer of which 8 were selected.

PCR amplification was performed using a T100 thermal cycler (Bio-Rad Laboratories, Hercules, USA). The amplification was carried out for each locus separately. The 20- μ L reaction volume contained 20 ng template DNA, 2 μ L PCR buffer (20 mM Tris-HCl, pH 8.4, 50 mM KCl), 0.28 mM dNTPs, 0.2 mM of each primer, 3.0 mM magnesium chloride (MgCl₂) and 0.12 U of *Taq* DNA polymerase (Invitrogen, Carlsbad, USA). Cycling conditions were: 4 min denaturation at 94°C; 26 cycles of denaturation at 94°C for 30 s, annealing for 30 s with temperatures varying by locus and an extension at 72°C for 45 s; others 12 cycles at 94°C for 45 s, 53°C for 45 s, 72°C for 45 s and final extension of 4 min at 72°C, using a MultiGene thermocycler (Labnet International, USA).

Locus	Primer sequence (5' to 3')	Ta (°C)	SSR motif	Fluorescent	GenBank accession number
Pps10	F: CCCACAGTCCAAGTTGAATGG R: CATCCAGGCGTTCTACAGGG	52	(AC) ₂₄	6FAM	MT505408
Pps11	F: GCTCCTCACAATTTGGCAGG R: ACACTTCCAAGTTTGCCCA	59.5	(AG) ₆	6FAM	MT754936
Pps12	F: TGTTCTCAGTTTCGTCAGGG R: TCCTGGTTCCTGTCCAGTA	59	(AT) ₆	PET	MT754937
Pps13	F: CACTCATAACGCCCTCCACA R: TGCAGCTAAACCACAGAGCA	59	(AG) ₇	VIC	MT754938
Pps14	F: CCCTTAGGCTTGAGCTGCTT R: TGAAGAGGCCTGAATGTGGG	60	(AG) ₈	6FAM	MT754939
Pps15	F: AGCCGTGAAATCTCCTTCCC R: GGGAAATGAGGAGGCTGACA	59	(AC) ₈	VIC	MT757398

Pps16	F: GCATGGAGTAGCCTGTTGGA R: TTGTGGCTCTGGGAAATCTGT	59	(AC) ₆	NED	MT754940
Pps17	F: TCTGGATGCTTCGTGCCAAA R: GATGCCGCTGGAAGGGATT	60	(AG) ₇	NED	MT754941

Table S4. Haplotypes list from *P. fulvus*, *P. davyi* and *P. gymnonotus* for PRKC1 + STAT5A, RAG2 and COI. The number of haplotypes, the studied site and the number of individuals corresponding to each locality are shown in parenthesis

	Intrones (PRKC1 + STAT5A)
Haplotypes	Species and locality
H1	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR01).
H2	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCRO02). <i>P. davyi</i> . Castries, Santa Lucía (PdSL08).
H3	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR03 and PfCR04).
H4	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR05). <i>P. fulvus</i> . Jalisco, Mexico (PfJa01). <i>P. fulvus</i> . Masaya Volcano Park, Nicaragua (PfNi01 and PfNi03).
H5	<i>P. fulvus</i> . Gallon Jug, Distrito de Orange Walk, Belize (PfBe02); Grutas de Lanquin, Alta Verapaz, Guatemala (PfGu06); Agua Blanca, Tabasco, Mexico (PfAB33, PfAB35 and PfAB36); Tapijulapa, Tabasco, Mexico (PfTa13, PfTa32); La Venta, Oaxaca, Mexico (PfLV15, PfLV25, PfLV29, PfLV30, PfLV31); Masaya Volcano Park, Nicaragua (PfNi02 and PfNi04).
H6	<i>P. fulvus</i> . Colón, Honduras (PfHo01).
H7	<i>P. fulvus</i> . Chiapas, Mexico (PfCh01).
H8	<i>P. fulvus</i> . Campeche, Mexico (PfCa01).
H9	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PfLV35).
H10	<i>P. davyi</i> . St. Joseph, Dominica (PdDo03); Castries, Santa Lucia (PdSL06).
H11	<i>P. davyi</i> . Arena Reserve, Trinidad and Tobago (PdTT01); Castries, Santa Lucía (PdSL02).

H12	<i>P. gymnonotus</i> . Jangada, Mato Grosso, Brazil (PgBr01); Barro Alto, Goiás, Brazil (PgBr02); Itabaiana, Sergipe, Brazil (PgBr12); Chapada Diamantina, Bahia, Brazil (PgBr15); Carajás, Parauapebas, Pará, Brazil (PgBr27); Tapirapé-Aquiri, Marabá, Pará, Brazil (PgBr32); Huanuco, Perú (PgPe01 and PgPe02); Altos de Campana National Park, Panamá (PgPa01); Barra Honda National Park, Costa Rica (PgCr01, PgCr02, PgCr03 and PgCr05); Grutas de Lanquin, Alta Verapaz, Guatemala (PgGu01); Agua Blanca, Tabasco, Mexico (PgAB10, PgAB13, PgAB15, PgAB16, PgAB24, PgAB27, PgAB28); Tapijulapa, Tabasco, Mexico (PgTa02, PgTa07 and PgTa09).
H13	<i>P. gymnonotus</i> . Aiuba, Ceará, Brazil (PgBr04 and PgBr07); Carajás, Parauapebas, Pará, Brazil (PgBr28); Brokopondo, Surinam (PgSu02).
H14	<i>P. gymnonotus</i> . Itabaiana, Sergipe, Brazil (PgBr11); Chapada Diamantina, Bahia, Brazil (PgBr17); Barra Honda National Park, Costa Rica (PgCr04); Agua Blanca, Tabasco, Mexico (PgAB12); Tapijulapa, Tabasco, Mexico (PgTa08); Martínez de la Torre, Oaxaca, Mexico (PgMT01).
H15	<i>P. gymnonotus</i> . Canaã dos Carajás, Pará, Brazil (PgBr18).
H16	<i>P. gymnonotus</i> . Masaya Volcano Park, Nicaragua (PgNi01, PgNi02 and PgNi03).
H17	<i>P. gymnonotus</i> . Masaya Volcano Park, Nicaragua (PgNi04).
H18	<i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB09 and PgAB19); Tapijulapa, Tabasco, Mexico (PgTa10); Martínez de la Torre, Oaxaca, Mexico (PgMT02).

	RAG2
Haplotypes	Species and locality
H1	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCr01, PfCr02 and PfCr04); La Venta, Oaxaca, Mexico (PflV35). <i>P. davyi</i> . Castries, Santa Lucia (PdSL06).
H2	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCr03 and PfCr05); Agua Blanca, Tabasco, Mexico (PfAB35 and PfAB36); Tapijulapa, Tabasco, Mexico (PfTa32). La Venta, Oaxaca, Mexico (PflV15, PflV29 and PflV30). <i>P. gymnonotus</i> . Aiuba, Ceará, Brazil (PgBr04); Chapada Diamantina, Bahía, Brazil (PgBr15); Tapirapé-Aquiri, Marabá, Pará, Brazil (PgBr32); Barra Honda National Park, Costa Rica (PgCR01, PgCR02, PgCR03, PgCR04 and PgCR05); Masaya Volcano Park, Nicaragua (PgNi01, PgNi02, PgNi03 and PgNi04); Grutas de Lanquin, Alta Verapaz, Guatemala (PgGu01); Agua Blanca, Tabasco, Mexico (PgAB10, PgAB12, PgAB13, PgAB15, PgAB16 and PgAB28); Tapijulapa, Tabasco, Mexico (PgTa02, PgTa07, PgTa08, PgTa09 and PgTa10); Martínez de la Torre, Oaxaca, Mexico (PgMT01).
H3	<i>P. fulvus</i> . Chiapas, Mexico (PfCh01).
H4	<i>P. fulvus</i> . Agua Blanca, Tabasco, Mexico (PfAB33).
H5	<i>P. fulvus</i> . Tapijulapa, Tabasco, Mexico (PfTa13).
H6	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV25).
H7	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV31).

H8	<i>P. fulvus</i> . Masaya Volcano Park, Nicaragua (PfNi01, PfNi02, PfNi03 and PfNi04)
H9	<i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB09, PgAB19, PgAB24 and PgAB27); Martínez de la Torre Oaxaca, Mexico (PgMT02).

	ADNnc (PRKC1 + STAT5A+ RAG2)
Haplotypes	Species and locality
H1	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR01).
H2	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR02). <i>P. davyi</i> . St. Joseph, Dominica (PdDo03); Castries, Santa Lucia (PdSL06 and PdSL08).
H3	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR03).
H4	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR04).
H5	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR05); Jalisco, Mexico (PfJa01).
H6	<i>P. fulvus</i> . Gallon Jug, Distrito de Orange Walk, Belize (PfBe02); Grutas de Lanquin, Alta Verapaz, Guatemala (PfGu06); Agua Blanca, Tabasco, Mexico (PfAB35); Tapijulapa, Tabasco, Mexico (PfTa32); La Venta, Oaxaca, Mexico (PfLV15, PfLV29 and PfLV30).
H7	<i>P. fulvus</i> . Colón, Honduras (PfHo01).
H8	<i>P. fulvus</i> . Chiapas, Mexico (PfCh01).
H9	<i>P. fulvus</i> . Campeche, Mexico (PfCa01).
H10	<i>P. fulvus</i> . Agua Blanca, Tabasco, Mexico (PfAB33 and PfAB36).

H11	<i>P. fulvus</i> . Tapijulapa, Tabasco, Mexico (PfTa13).
H12	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV25).
H13	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV31).
H14	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV35).
H15	<i>P. fulvus</i> . Masaya Volcano Park, Nicaragua (PfNi01, PfNi02, PfNi03 and PfNi04).
H16	<i>P. davyi</i> . Arena Reserve, Trinidad and Tobago (PdTT01).
H17	<i>P. davyi</i> . Castries, Santa Lucía (PdSL02).
H18	<i>P. gymnonotus</i> . Jangada, Mato Grosso, Brazil (PgBr01); Barro Alto, Goiás, Brazil (PgBr02); Itabaiana, Sergipe, Brazil (PgBr12); Chapada Diamantina, Bahía, Brazil (PgBr15); Carajás, Parauapebas, Pará, Brazil (PgBr27); Tapirapé-Aquiri, Marabá, Pará, Brazil (PgBr32); Huanuco, Perú (PgPe01 and PgPe02); Altos de Campana National Park, Panamá (PgPa01); Barra Honda National Park, Costa Rica (PgCR01, PgCR02, PgCR03, PgCR05); Grutas de Lanquin, Alta Verapaz, Guatemala (PgGu01); Agua Blanca, Tabasco, Mexico (PgAB10, PgAB13, PgAB15, PgAB16 and PgAB28); Tapijulapa, Tabasco, Mexico (PgTa02, PgTa07 and PgTa09).
H19	<i>P. gymnonotus</i> . Aiuba, Ceará, Brazil (PgBr04 and PgBr07); Canaã dos Carajás, Pará, Brazil (PgBr28); Brokopondo, Surinam (PgSu02).
H20	<i>P. gymnonotus</i> . Itabaiana, Sergipe, Brazil (PgBr11); Chapada Diamantina, Bahía, Brazil (PgBr17); Barra Honda National Park, Costa Rica (PgCR04); Agua Blanca, Tabasco, Mexico (PgAB12); Tapijulapa, Tabasco, Mexico (PgTa08); Martínez de la Torre Oaxaca, Mexico (PgMT01).
H21	<i>P. gymnonotus</i> . Canaã dos Carajás, Pará, Brazil (PgBr18).
H22	<i>P. gymnonotus</i> . Masaya Volcano Park, Nicaragua (PgNi01, PgNi02 and PgNi03)

H23	<i>P. gymnonotus</i> . Masaya Volcano Park, Nicaragua (PgNi04)
H24	<i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB09, PgAB19 and PgAB24); Tapijulapa, Tabasco, Mexico (PgTa10); Martínez de la Torre Oaxaca, Mexico (PgMT02).
H25	<i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB27).

	COI
Haplotypes	Species and locality
H1	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR01).
H2	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR02, PfCR03 and PfCR04); La Venta, Oaxaca, Mexico (PfLV35). <i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB15 and PgAB24).
H3	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR05).
H4	<i>P. fulvus</i> . Gallon Jug, Distrito de Orange Walk, Belize (PfBe02).
H5	<i>P. fulvus</i> . Colón, Honduras (PfHo01).
H6	<i>P. fulvus</i> . Tikal, El Peten, Guatemala (PfGu06); Chiapas, Mexico (PfCh01); Campeche, Mexico (PfCa01); Tapijulapa, Tabasco, Mexico (PfTa13); Masaya Volcano Park, Nicaragua (PfNi01, PfNi02, PfNi03 and PfNi04). <i>P. gymnonotus</i> . Masaya Volcano Park, Nicaragua (PgNi01, PgNi02 and PgNi03); Alta Verapaz, Guatemala (PgGu01); Agua Blanca, Tabasco, Mexico (PgAB19 and PgAB28); Tapijulapa, Tabasco, Mexico (PgTa07, PgTa08 and PgTa10).
H7	<i>P. fulvus</i> . Jalisco, Mexico (PfJa01).
H8	<i>P. fulvus</i> . Agua Blanca, Tabasco, Mexico (PfAB33, PfAB35 and PfAB36).

	<i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB12 and PgAB16); Tapijulapa, Tabasco, Mexico (PgTa02).
H9	<i>P. fulvus</i> . Tapijulapa, Tabasco, Mexico (PfTa32).
H10	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV15).
H11	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV25, PflV30 and PflV31).
H12	<i>P. fulvus</i> . La Venta, Oaxaca, Mexico (PflV29).
H13	<i>P. davyi</i> . Arena Reserve, Nariva, Trinidad and Tobago (PdTT01).
H14	<i>P. davyi</i> . Castries, Santa Lucia (PdSL02).
H15	<i>P. davyi</i> . Castries, Santa Lucia (PdSL06).
H16	<i>P. davyi</i> . Castries, Santa Lucia (PdSL08).
H17	<i>P. gymnonotus</i> . Jangada, Mato Grosso, Brazil (PgBr01).
H18	<i>P. gymnonotus</i> . Barro Alto, Goiás, Brazil (PgBr02); Itabaiana, Sergipe, Brazil (PgBr11 and PgBr12); Chapada Diamantina, Bahía, Brazil (PgBr17); Huanuco, Perú (PgPe01 and PgPe02); Brokopondo, Surinam (PgSu02); Altos de Campana National Park, Panamá (PgPa01).
H19	<i>P. gymnonotus</i> . Aiuba, Ceará, Brazil (PgBr04).
H20	<i>P. gymnonotus</i> . Aiuba Ecological Station, Aiuba, Ceará, Brazil (PgBr07).
H21	<i>P. gymnonotus</i> . Chapada Diamantina National Park, Bahía, Brazil (PgBr15).
H22	<i>P. gymnonotus</i> . Canaã dos Carajás, Pará, Brazil (PgBr18).

H23	<i>P. gymnonotus</i> . Carajás, Parauapebas; Pará, Brazil (PgBr27).
H24	<i>P. gymnonotus</i> . Carajás, Parauapebas, Pará, Brazil (PgBr28).
H25	<i>P. gymnonotus</i> . Tapirapé-Aquiri, Marabá, Pará, Brazil (PgBr32).
H26	<i>P. gymnonotus</i> . Barra Honda National Park, Costa Rica (PgCR01).
H27	<i>P. gymnonotus</i> . Barra Honda National Park, Costa Rica (PgCR02).
H28	<i>P. gymnonotus</i> . Barra Honda National Park, Costa Rica (PgCR03).
H29	<i>P. gymnonotus</i> . Barra Honda National Park, Costa Rica (PgCR04 and PgCR05).
H30	<i>P. gymnonotus</i> . Masaya Volcano Park, Nicaragua (PgNi04); Agua Blanca, Tabasco, Mexico (PgAB09 and PgAB13); Tapijulapa, Tabasco, Mexico (PgTa09).
H31	<i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB10 and PgAB27).
H32	<i>P. gymnonotus</i> . Martínez de la Torre Oaxaca, Mexico (PgMT01 and PgMT02).

Table S5. Haplotype list for COI marker used for *Pteronotus fulvus* and *P. gymnonotus*. The number of haplotypes, the studied site and the number of individuals corresponding to each locality are

shown in parenthesis.

Haplotypes	Species and locality
H1	<p><i>P. fulvus</i>. Laguna Encantada, Veracruz, Mexico (PfLE01, PfLE02, PfLE06, PfLE07, PfLE08, PfLE10, PfLE11, PfLE12, PfLE13, PfLE16, PfLE17); Cerro Huatulco, Oaxaca (PfCH01, PfCH03, PfCH04, PfCH05, PfCH07, PfCH09, PfCH10, PfCH11, PfCH14 and PfCH16); La Venta, Oaxaca, Mexico (PfLV01, PfLV09, PfLV11, PfLV12, PfLV13, PfLV14, PfLV18, PfLV19, PfLV21, PfLV22, PfLV24, PfLV25, PfLV26, PfLV27, PfLV30, PfLV31, PfLV33, PfLV34); Tapijulapa, Tabasco, Mexico (PfTa06); Playa de Oro, Colima, Mexico (PfPO08);</p> <p><i>P. gymnonotus</i>. Tapijulapa, Tabasco, Mexico (PgTa15).</p>
H2	<p><i>P. fulvus</i>. Laguna Encantada, Veracruz, Mexico (PfLE03, PfLE04 and PfLE05); Cerro Huatulco, Oaxaca (PfCH06 and PfCH08); La Venta, Oaxaca, Mexico (PfLV10 and PfLV16); Tapijulapa, Tabasco, Mexico (PfTa09, PfTa10, PfTa11, PfTa12, PfTa13, PfTa15, PfTa16, PfTa17, PfTa18, PfTa20, PfTa22, PfTa25, PfTa29, PfTa30 and PfTa31); Agua Blanca, Tabasco, Mexico (PfAB01, PfAB02, PfAB03, PfAB04, PfAB05, PfAB08, PfAB09, PfAB10, PfAB11, PfAB12, PfAB14, PfAB15, PfAB16, PfAB17, PfAB19, PfAB21, PfAB22, PfAB24, PfAB25, PfAB26, PfAB27, PfAB28, PfAB29, PfAB32, PfAB37, PfAB39, PfAB40, PfAB41, PfAB42, PfAB43, PfAB44, PfAB45, PfAB46, PfAB47, PfAB49, PfAB50, PfAB51); Campeche, Mexico (PfCa01); Chiapas, Mexico (PfCh01); Grutas de Lanquin, Alta Verapaz, Guatemala (PfGu01, PfGu02, PfGu03 and PfGu05); Tikal, El Peten, Guatemala (PfGu06); El Refugio; Ahuachapan, El Salvador (PfES06 and PfES07); Gallon Jug, Distrito de Orange Walk, Belize (PfBe01); Masaya Volcano Park, Nicaragua (PfNi01, PfNi02, PfNi03 and PfNi04),</p> <p><i>P. gymnonotus</i>. Tapijulapa, Tabasco, Mexico (PgTa04, PgTa05, PgTa07, PgTa08, PgTa10, PgTa11, PgTa12 and PgTa13); Agua Blanca, Tabasco, Mexico (PgAB04, PgAB08, PgAB11, PgAB19, PgAB21, PgAB22, PgAB23 and PgAB28); Grutas de Lanquin, Alta Verapaz, Guatemala (PgGu01); Masaya Volcano Park, Nicaragua (PgNi01, PgNi02 and PgNi03); Altos de Campana National Park, Panamá (PgPa03).</p>
H3	<i>P. fulvus</i> . Laguna Encantada, Veracruz, Mexico (PfLE09); Cerro Huatulco, Oaxaca (PfCH13 and PfCH15).
H4	<i>P. fulvus</i> . Laguna Encantada, Veracruz, Mexico (PfLE14).
H5	<i>P. fulvus</i> . Laguna Encantada, Veracruz, Mexico (PfLE15); La Venta, Oaxaca, Mexico (PfLV17, PfLV28 and PfLV32); Agua Blanca, Tabasco, Mexico (PfAB07); Los Ortices, Colima, Mexico (PfLO07).

H6	<i>P. fulvus.</i>); Cerro Huatulco, Oaxaca (PfCH02 and PfCH12); La Venta, Oaxaca, Mexico (PflV23).
H7	<i>P. fulvus.</i> La Venta, Oaxaca, Mexico (PflV02, PflV03, PflV04, PflV06, PflV35); Barra Honda National Park, Costa Rica (PfCR02, PfCR03 and PfCR04). <i>P. gymnonotus.</i> Tapijulapa, Tabasco, Mexico (PgTa03); Agua Blanca, Tabasco, Mexico (PgAB07, PgAB15, PgAB17, PgAB18, PgAB20 and PgAB24).
H8	<i>P. fulvus.</i> La Venta, Oaxaca, Mexico (PflV05 and PflV29); Los Ortices, Colima, Mexico (PflO01).
H9	<i>P. fulvus.</i> La Venta, Oaxaca, Mexico (PflV08).
H10	<i>P. fulvus.</i> La Venta, Oaxaca, Mexico (PflV15).
H11	<i>P. fulvus.</i> La Venta, Oaxaca, Mexico (PflV20); Tapijulapa, Tabasco, Mexico (PfTa04, PfTa05, PfTa08, PfTa21 and PfTa24); Agua Blanca, Tabasco, Mexico (PfAB33, PfAB35 and PfAB36); Los Ortices, Colima, Mexico (PflO06); Playa de Oro, Colima, Mexico (PfPO11). <i>P. gymnonotus.</i> Tapijulapa, Tabasco, Mexico (PgTa01, PgTa02, PgTa06 and PgTa17); Agua Blanca, Tabasco, Mexico (PgAB03, PgAB05, PgAB06, PgAB12, PgAB16 and PgAB26).
H12	<i>P. fulvus.</i> Tapijulapa, Tabasco, Mexico (PfTa03).
H13	<i>P. fulvus.</i> Tapijulapa, Tabasco, Mexico (PfTa27).
H14	<i>P. fulvus.</i> Tapijulapa, Tabasco, Mexico (PfTa28); Agua Blanca, Tabasco, Mexico (PfAB31).
H15	<i>P. fulvus.</i> Tapijulapa, Tabasco, Mexico (PfTa32).
H16	<i>P. fulvus.</i> Agua Blanca, Tabasco, Mexico (PfAB06).
H17	<i>P. fulvus.</i> Agua Blanca, Tabasco, Mexico (PfAB13).
H18	<i>P. fulvus.</i> Agua Blanca, Tabasco, Mexico (PfAB18).
H19	<i>P. fulvus.</i> Agua Blanca, Tabasco, Mexico (PfAB20).
H20	<i>P. fulvus.</i> Agua Blanca, Tabasco, Mexico (PfAB23).
H21	<i>P. fulvus.</i> Agua Blanca, Tabasco, Mexico (PfAB30).
H22	<i>P. fulvus.</i> Agua Blanca, Tabasco, Mexico (PfAB38).
H23	<i>P. fulvus.</i> Los Ortices, Colima, Mexico (PflO02, PflO12); <i>P. fulvus.</i> Jalisco, Mexico (PfJa01).

H24	<i>P. fulvus</i> . Los Ortices, Colima, Mexico (PfLO03, PfLO04, PfLO05, PfLO08, PfLO09, PfLO10 and PfLO11); Playa de Oro, Colima, Mexico (PfPO01, PfPO02, PfPO06, PfPO07 and PfPO09).
H25	<i>P. fulvus</i> . Los Ortices, Colima, Mexico (PfLO07).
H26	<i>P. fulvus</i> . Los Ortices, Colima, Mexico (PfLO13).
H27	<i>P. fulvus</i> . Los Ortices, Colima, Mexico (PfLO14).
H28	<i>P. fulvus</i> . Playa de Oro, Colima, Mexico (PfPO03).
H29	<i>P. fulvus</i> . Playa de Oro, Colima, Mexico (PfPO04).
H30	<i>P. fulvus</i> . Playa de Oro, Colima, Mexico (PfPO05).
H31	<i>P. fulvus</i> . Playa de Oro, Colima, Mexico (PfPO10).
H32	<i>P. fulvus</i> . Jalisco, Mexico (PfJa02).
H33	<i>P. fulvus</i> . Alta Verapaz, Grutas De Lanquin, Guatemala (PfGu04).
H34	<i>P. fulvus</i> . El Refugio; Ahuachapan, El Salvador (PfES05).
H35	<i>P. fulvus</i> . Colón, Honduras (PfHo01).
H36	<i>P. fulvus</i> . Gallon Jug, Distrito de Orange Walk, Belize (PfBe02).
H37	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR01).
H38	<i>P. fulvus</i> . Barra Honda National Park, Costa Rica (PfCR05).
H39	<i>P. gymnonotus</i> . Martínez de la Torre, Oaxaca (PfMT01).
H40	<i>P. gymnonotus</i> . Martínez de la Torre, Oaxaca (PfMT02).
H41	<i>P. gymnonotus</i> . Tapijulapa, Tabasco, Mexico (PgTa09 and PgTa16); Agua Blanca, Tabasco, Mexico (PgAB09, PgAB13, PgAB25 and PgAB29); Masaya Volcano Park, Nicaragua (PgNi04).
H42	<i>P. gymnonotus</i> . Tapijulapa, Tabasco, Mexico (PgTa14).
H43	<i>P. gymnonotus</i> . Agua Blanca, Tabasco, Mexico (PgAB10 and PgAB27).
H44	<i>P. gymnonotus</i> . Barra Honda National Park, Costa Rica (PgCR01).
H45	<i>P. gymnonotus</i> . Barra Honda National Park, Costa Rica (PgCR02).
H46	<i>P. gymnonotus</i> . Barra Honda National Park, Costa Rica (PgCR03, PgCR04, and PgCR05).

H47	<i>P. gymnonotus</i> . Altos de Campana National Park, Panamá (PgPa01 and PgPa02); Iwokrama Reserve, Potaro-Siparuni, Guyana (PgGy03); Bakhuis, Sipaliwini, Surinam (PgSu01); Brokopondo, Surinam (PgSu02, PgSu03 and PgSu04); Huánuco, Perú (PgPe01 and PgPe02); Caverna do Bigode, Goiás, Brazil (PgGo03 and PgGo04); Barro Alto, Goiás, Brazil (PgBr02); Aiuba, Ceará, Brazil (PgBr03, PgBr05, PgBr06); Itabaiana, Sergipe, Brazil (PgBr09, PgBr10, PgBr11 and PgBr12); Chapada Diamantina, Bahía, Brazil (PgBr14 and PgBr17); Canaã dos Carajás, Pará, Brazil (PgBr19 and PgBr22); Carajás, Parauapebas, Pará, Brazil (PgBr24, PgBr26, PgBr29 and PgBr31); Tapirapé-Aquiri, Marabá, Pará, Brazil (PgBr33).
H48	<i>P. gymnonotus</i> . Hato la Florida, Bolívar, Venezuela (PgVe01).
H49	<i>P. gymnonotus</i> . Iwokrama Reserve, Potaro-Siparuni, Guyana (PgGy01, PgGy02 and PgGy04); Brokopondo, Surinam (PgSu06).
H50	<i>P. gymnonotus</i> . Brokopondo, Surinam (PgSu05).
H51	<i>P. gymnonotus</i> . Caverna do Bigode, Goiás, Brazil (PgGo01); Aiuba, Ceará, Brazil (PgBr04).
H52	<i>P. gymnonotus</i> . Caverna do Bigode, Goiás, Brazil (PgGo02).
H53	<i>P. gymnonotus</i> . Jangada, Mato Grosso, Brazil (PgBr01).
H54	<i>P. gymnonotus</i> . Aiuba, Ceará, Brazil (PgBr07).
H55	<i>P. gymnonotus</i> . Aiuba, Ceará, Brazil (PgBr08);
H56	<i>P. gymnonotus</i> . Usina Serra Grande, Alagoas, Brazil (PgBr13).
H57	<i>P. gymnonotus</i> . Chapada Diamantina, Bahía, Brazil (PgBr15).
H58	<i>P. gymnonotus</i> . Chapada Diamantina, Bahía, Brazil (PgBr16).
H59	<i>P. gymnonotus</i> . Canaã dos Carajás, Pará, Brazil (PgBr18).
H60	<i>P. gymnonotus</i> . Canaã dos Carajás, Pará, Brazil (PgBr20); Carajás, Parauapebas, Pará, Brazil (PgBr23, PgBr27 and PgBr30).
H61	<i>P. gymnonotus</i> . Canaã dos Carajás, Pará, Brazil (PgBr21).
H62	<i>P. gymnonotus</i> . Carajás, Parauapebas, Pará, Brazil (PgBr25); Tapirapé-Aquiri, Marabá, Pará, Brazil (PgBr32).
H63	<i>P. gymnonotus</i> . Carajás, Parauapebas, Pará, Brazil (PgBr28)

Table S6. Genetic diversity statistics of microsatellites for each study site of *P. fulvus* and *P. gymnonotus*. Samples number (*N*); summary and mean of the alleles per locus (*Na*); exclusive alleles (*Np*); observed heterozygosity (*HO*), expected heterozygosity (*HE*); summary and mean of allelic

richness (AR). Study localities abbreviations correspond to the locations presented in Table 1.

Species	Study site	<i>N</i>	<i>Na</i>	<i>Np</i>	<i>Ho</i>	<i>HE</i>	<i>AR</i>
<i>P. fulvus</i>	PO	10	$\Sigma = 73$ $\bar{X} = 5.615$	3	0.612	0.646	$\Sigma = 54.699$ $\bar{X} = 4.208$
	LO	13	$\Sigma = 94$ $\bar{X} = 7.231$	4	0.598	0.644	$\Sigma = 55.756$ $\bar{X} = 4.289$
	AB	51	$\Sigma = 131$ $\bar{X} = 10.077$	4	0.523	0.660	$\Sigma = 55.016$ $\bar{X} = 4.232$
	Ta	31	$\Sigma = 124$ $\bar{X} = 9.538$	3	0.538	0.656	$\Sigma = 55.977$ $\bar{X} = 4.306$
	LV	34	$\Sigma = 118$ $\bar{X} = 9.077$	5	0.556	0.668	$\Sigma = 55.27$ $\bar{X} = 4.251$
	CH	16	$\Sigma = 102$ $\bar{X} = 7.846$	4	0.598	0.662	$\Sigma = 54.334$ $\bar{X} = 4.333$
<i>P. gymnonotus</i>	AB	27	$\Sigma = 82$ $\bar{X} = 6.308$	5	0.549	0.568	$\Sigma = 43.303$ $\bar{X} = 3.331$
	Ta	17	$\Sigma = 86$ $\bar{X} = 6.615$	8	0.574	0.622	$\Sigma = 49.448$ $\bar{X} = 3.804$

Table S7. Calculation of pairwise F_{ST} above by with COI and R_{ST} down by microsatellites. * $p < 0.05$

FST RST	<i>P. fulvus</i> CH	<i>P. fulvus</i> LV	<i>P. fulvus</i> Ta	<i>P. fulvus</i> AB	<i>P. fulvus</i> LO	<i>P. fulvus</i> PO	<i>P. gymnonotus</i> Ta	<i>P. gymnonotus</i> AB
<i>P. fulvus</i> CH	-	0.05518	0.47379*	0.63961*	0.26679*	0.26252*	0.14164*	0.16849*
<i>P. fulvus</i> LV	0.13077*	-	0.23620*	0.34647*	0.09121*	0.08271	0.05061	0.05011
<i>P. fulvus</i> Ta	0.06633*	0.01236	-	0.06784*	0.58659*	0.55582*	0.07302*	0.16718*
<i>P. fulvus</i> AB	0.02808	0.02752*	-0.00508	-	0.72548*	0.70976*	0.17350*	0.25954*
<i>P. fulvus</i> LO	0.02488	0.08888*	0.07581*	0.04164*	-	-0.06616	0.20040*	0.18635*
<i>P. fulvus</i> PO	0.15251*	0.21900*	0.21090*	0.15131*	-0.00698	-	0.16031*	0.15681*
<i>P. gymnonotus</i> Ta	0.07433*	0.12288*	0.11402*	0.07148*	-0.01649	0.03458	-	-0.01675
<i>P. gymnonotus</i> AB	0.19964*	0.22451*	0.22689*	0.16416*	0.02641*	0.05038*	0.02114	-

Table S8. Number of individuals identified as pure *P. fulvus* puro (Pfu); *P. gymnonotus* (Pgy); unclassified hybrids (SC); and hybrids according to STRUCTURE and NEWHYBRIDS with to criterion 2 of Burgarella et al. [87]. The percentage of the total number of individuals sampled by locality is included in parenthesis.

	NEWHYBRIDS	STRUCTURE
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	Pfu	Pgy	SC	Hybrid	Pfu	Pgy	SC	Hybrid
PO	10 (100)	0 (0)	0 (0)	0 (0)	9 (90)	0 (0)	0 (0)	1 (10)
LO	13 (100)	0 (0)	0 (0)	0 (0)	13 (100)	0 (0)	0 (0)	0 (0)
AB	49 (62.82)	23 (29.48)	6 (7.69)	0 (0)	49 (62.82)	27 (34.61)	0 (0)	2 (2.56)
Ta	31 (64.58)	10 (20.83)	4 (8.33)	3 (6.25)	31 (64.58)	16 (33.33)	0 (0)	1 (2.08)
LV	34 (100)	0 (0)	0 (0)	0 (0)	34 (100)	0 (0)	0 (0)	0 (0)
CH	15 (93.75)	0 (0)	1 (6.25)	0 (0)	14 (87.5)	0 (0)	0 (0)	2 (12.5)
Total	152	33	11	3	150	43	0	6