Supporting Information

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SI Materials and Methods

Field Sampling. Bats were captured in a broad range of habitats, including ravine forest, submontane grassland, submontane forest, evergreen lowland forest, and savannas during the dry season between November and December 2006 in Côte d'Ivoire, in December 2008 in Guinea (Convention on International Trade in Endangered Species Exportation permits 449 and 450), and between November and December 2010 in Liberia (Permit no. AMD/030/053–11) (Fig. 1, Fig. S1, and Table S2). Different types of mist nets were used at ground and canopy level. We took forearm length (in millimeter) and body mass (in grams) of all individuals, as well as standard measurements of voucher specimens (head and body, tail, ear, hind foot, tibia). Reproductive status, age class, and sex (1) were additionally recorded. All techniques used for capture and preservation complied with standard methods for measuring and monitoring mammal diversity (2). For field identification of bat species, a number of identification keys were used, mainly Rosevear (3) and Hayman and Hill (4). Taxonomy follows Simmons (5) and Nesi (6) except for *Miniopterus villiersi* and *Pipistrellus* aff. *grandidieri*. Koopman (7) lists *M. villiersi* as a subspecies of *Miniopterus schreibersii*, but genetic results indicate cryptic diversity (8) and West African populations probably represent a distinct species. The West African *Pipistrellus* aff. *grandidieri* might represent a yet undescribed species (9).

Blood samples were obtained from a puncture of either the antebrachial or interfemoral vein. Blood sampling followed approved animal care and protocols (1). Our study adhered to the guidelines of the American Society of Mammalogists for the use of wild mammals in research (1, 2). For morphological identification of the parasites, thin blood smears were prepared. Wing punches were collected from each captured individual using the nonlethal method after Worthington Wilmer and Barratt (10) and stored in 90% (vol/vol) ethanol. Wing punches, blood dots on filter paper, or an aliquot of the blood smear sample were used for subsequent molecular analysis. The majority of captured bats were released at the field sites, and only few voucher specimens were collected to document species occurrences and verify identifications. These specimens are deposited in the Zoological Museum A. Koenig (specimens from Guinea) and the research collection of J. Fahr (Liberia and Côte d'Ivoire).

Microscopy. Thin blood smears were fixed in 100% (vol/vol) methanol and stained with the Romanowsky–Giemsa staining and examined for blood parasites via light microscopy at a magnification of 1,000× for a minimum of 15 min. Examinations were repeated at least once to minimize false-negative records. The entire slide was scanned for asexual and sexual blood stages. Pictures of infected erythrocytes were processed using the SPOT Imaging Solutions software or Imaging for Windows. Parasitemias were recorded for each infected bat (Table S1) as follows. For each slide, an average number of erythrocytes per field was calculated by counting total erythrocytes observed in two fields and dividing by 2. Then, the number of parasites was counted across 40 fields of the slide or until one parasite cell was observed. Parasitemia is given as number of parasites per total estimated erythrocyte number. All positive samples were used for further genetic work.

DNA Extraction, Amplification, and Sequencing. All positive samples were subjected to parasite genotyping. A representative portion of the blood smear-negative samples was screened via PCR for confirmation. DNA was extracted from dried blood dots, wing punches, or blood that was scraped off the blood smear slides using the DNeasy extraction kit (Qiagen). The protocol for animal tissues was performed following the manufacturer's protocol with minor modifications. Samples were eluted in 50-µL buffer AE. PCR was performed using illustra* PuRe Taq Ready-To-Go PCR beads (GE Health Sciences), 2 µL of genomic DNA as template, and 1 µL of 10 mM of each primer. Because of the diversity of the taxa being studied, many novel primers were designed for this project and multiple combinations were tried to obtain optimal sequence reads. Occasionally, nested PCRs were performed, following previously published protocols (11, 12). All primers are listed in Table S3. PCR fragments were sequenced in both directions using BigDye v3.0 (Applied Biosystems) using the manufacturer's protocol and the original sequencing primers, and run on an ABI3730xl sequencer. Sequences included 1,823 nt of mitochondrial DNA [986 nt of the cytochrome *b* (*cytb*) and 837 nt of the cytochrome oxidase I (*cox1*) genes], 576 nt of the apicoplast caseinolytic protease (*clpc*) gene, and 523 nt of the nuclear elongation factor 2 (*ef2a*) gene. For chiropteran *Plasmodium* samples, additional sequences included 900 nt of *Actin1* (*act1*), 619 nt of *Actin2* (*act2*), 211 nt of adenylosuccinate lyase (*asl*), 667 nt of cysteine proteinase (*cyspro*), 596 nt of dihydrofolate reductase/thymidylate synthase (*dhfr*/*ts*), 356 nt of histone H2A, 460 nt of inosine monophosphate-dehydrogenase (*InMPDH*), 464 nt of ookinete surface protein (*P25*), and 454 nt of poly-ubiquitin (*UBI*).

SI Results

Parasite Classification. ***Plasmodium voltaicum.*** The growing gametocyte features a nucleus that is surrounded by a clear more transparent zone and pigment granules are visible in the cytoplasm (Fig. 3*A* and Fig. S3*A*). Other characteristics include: (*i*) the typical location of the nucleus at the margin of the parasite, (*ii*) a weaker staining of the microgametocyte cytoplasm compared with the macrogametocyte, (*iii*) an ambiguous boundary of the microgametocyte nucleus that occupies a large area of the parasite, and (*iv*) enlargement of erythrocytes that are occupied by mature gametocytes.

Plasmodium cyclopsi. Thin and fragile young ring forms were found in infected blood, with a vacuole limited by a narrow border of cytoplasm and a relatively large nucleus, resembling the description by Landau and Chabaud (13) (Fig. 3*A*). Schizonts are very compact, with small dense nuclei and a mass of granules that occasionally contain a large grain of black pigment. There were usually eight nuclei, although occasionally only six nuclei (Fig. S3*B*).

Polychromophilus. The parasite blood stages of *Polychromophilus* sp. are limited to gametocytes. When mature, these gametocytes fill the erythrocyte completely and cause a slight enlargement and a round or often oval-shaped form of the host cell with fine yellow-brown pigment grains scattered in the cytoplasm. The macrogametocyte features purple-blue–stained cytoplasm and a small distinct nucleus that is placed eccentrically and stains pink (Fig. 3*B* and Fig. S3*C*).

Nycteria. Only sexual stages were found in the blood, almost exclusively representing growing gametocytes. When immature, the gametocytes possess an oval shape (Fig. 3*C* and Fig. S3 *D* and *E*). Later, gametocytes adopt a spherical shape, and the brown pigment granules become scattered in the cytoplasm or are assembled in small groups. The growing forms feature a diffuse nucleus area, which only becomes homogeneous after maturity. Garnham and Heisch (14) described a well-condensed nucleus in fully mature gametocytes. However, we did not observe fully mature stages in our samples. Thus, it was difficult to distinguish the sexes of the gametocytes without the characteristic staining of these stages (14). The gametocytes found in *Rhinolophus alcyone* and *Rhinolophus landeri* resemble each other to a high degree. Both are oval-shaped parasites and contain several dark-stained granules, but no definite nucleus area. The cytoplasm does not appear amoeboid, and the parasite does not fill the entire erythrocyte. Hence, the host-cell margin remains detectable, even at the mature gametocyte stage, and the outline of the parasite membrane is clearly visible. The main morphological diagnostic features for species of *Nycteria* are the size and shape of the schizonts (15). Because in the present study only blood stages were available for morphological comparisons, parasites could not be classified further to the species level. We assigned the parasites found in *R. alcyone* and *R. landeri* to the genus *Nycteria* on the basis of the morphology of the gametocytes and due to their grouping as a distinct clade outside the *Hepatocystis*, *Plasmodium*, and *Polychromophilus* clades.

Hepatocystis. Blood stages of *Hepatocystis* spp. are limited to gametocyte stages and hemozoin was present in all infected erythrocytes (Fig. 3*D* and Fig. S3 *F–O*). In the majority of cases, the infectious stages found in the blood smears were fully mature gametocytes. The youngest forms were very small red-staining chromatin dots, as described by Desowitz (16) from *Hepatocystis* in monkeys. The more developed stages showed a solid nucleus and cytoplasm and appeared as rings resembling those of *Plasmodium* (Fig. 3*D*). The sexually dimorphic micro- and macrogametocytes were distinguished after Giemsa staining. The microgametocytes featured pale brown cytoplasm with evenly distributed hemozoin granulae. The nucleus consisted of a large portion that stained light red, and a smaller part within staining dense dark red. No granules were visible in the nuclear zone. The macrogametocytes, in contrast, stained blue, with a red staining nucleus of much smaller size in comparison with the nucleus of the microgametocyte. Pigment granules were dispersed in the whole cytoplasm. The appearance and size of the digestive vacuoles differed from total absence (Fig. S3*G*) to small size (Fig. S3 *F*, *I*, and *J*) and to vacuoles, which almost filled two quarter of the parasite (Fig. S3 *M* and *N*). Deformations of the host cells were common, resulting in amoeboid cell forms (Fig. S3 *K* and *L*). The morphology of the parasites isolated in the present study from *Epomops buettikoferi*, *Nanonycteris veldkampii*, *Micropteropus pusillus*, and *Epomophorus gambianus* is consistent with *Hepatocystis epomophori* (15). We note that the sequences of *Hepatocystis* species isolated from *Myonycteris leptodon* in Liberia display minor phylogenetic distances to the "*H. epomophori*" group and might therefore represent a distinct species.

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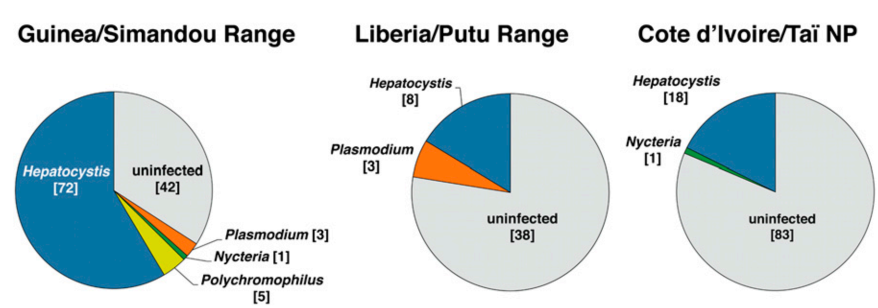

Fig. S1. Prevalences of hemosporidian genera across collecting sites. The pie charts for each country show the numbers of bat individuals that are infected with the respective hemosporidian parasite genus.

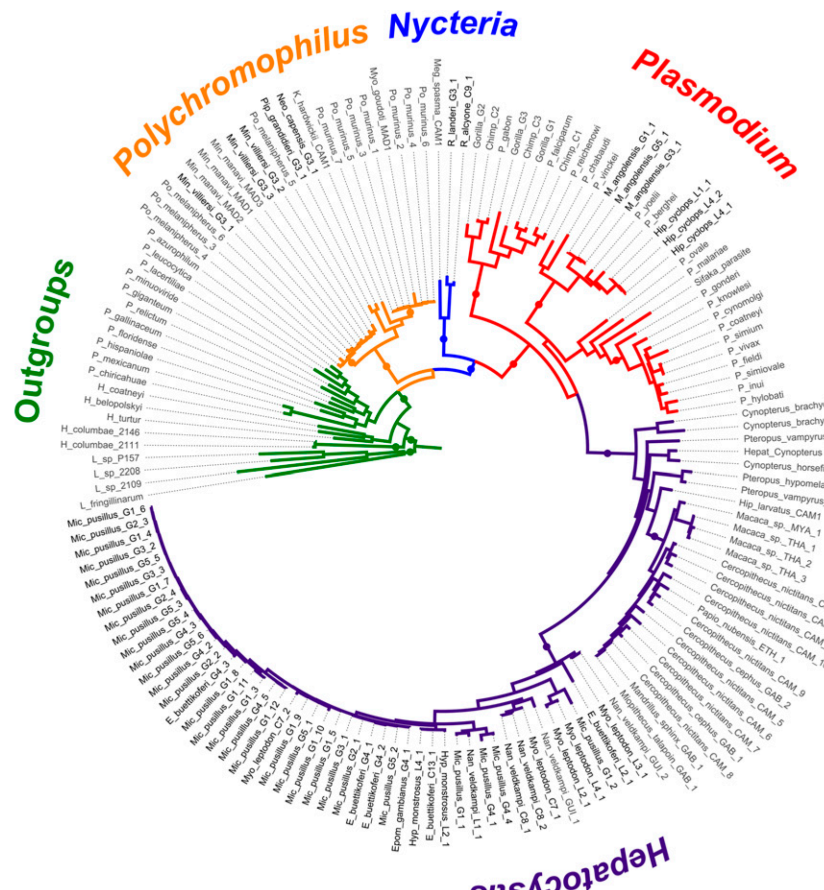

Fig. S2. Maximum-likelihood phylogeny of bat hemosporidian parasites in broad context of previously published sequences. Maximum-likelihood phylogeny of novel isolates of West African bat malaria parasites (black type) within context of large set of previously published taxa (gray type). Phylogeny was constructed with partitioned analysis of mitochondrial (*cytb*, *cox1*), apicoplast (*clpc*), and nuclear (*ef2a*) genes, rooted with *Leucocytozoon* taxa. Bootstrap support values ≥90% on key clades indicated with a dot.

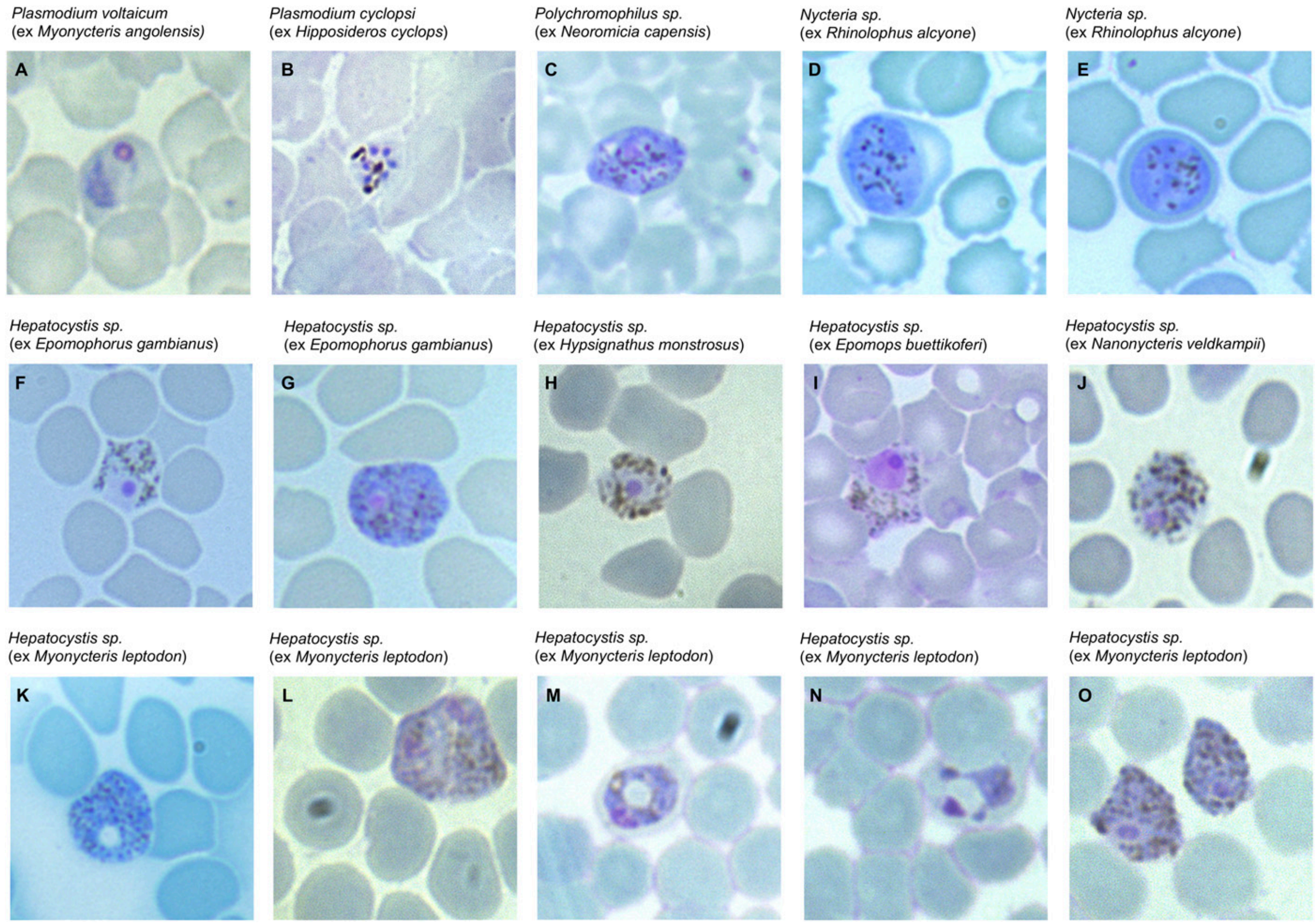

Fig. S3. Representative micrographs from Giemsa-stained blood films of infected Chiroptera. The parasite genus/species and the associated hosts are listed above the respective pictures. Sizes of erythrocytes of bats vary between 5 and 6.4 μm in diameter. Gametocytes slightly enlarge the host cells up to about 7 μm. (*A*) Growing gametocyte of *Plasmodium voltaicum*. (*B*) *Plasmodium cyclopsi* schizont including six nuclei. (*C*) Mature gametocyte stage of *Polychromophilus*, causing a round or oval-shaped form of the erythrocyte with yellow-brown pigment grains scattered in the cytoplasm. (*D* and *E*) Growing gametocytes of *Nycteria*. Note the rounded gametocytes that are not fully grown yet and the coarse grained hemozoin. (*F*, *H–J*) Male microgametocytes of *Hepatocystis* sp. (*G*, *K*, *L*, and *O*) female macrogametocytes of *Hepatocystis* sp. Note the double-infected erythrocyte in picture *L*. (*M* and *N*) Ring stages of *Hepatocystis* sp. Ring stages resemble *Plasmodium* ring stages, but show a larger nucleus. Micrographs were taken at 1,000× magnification.

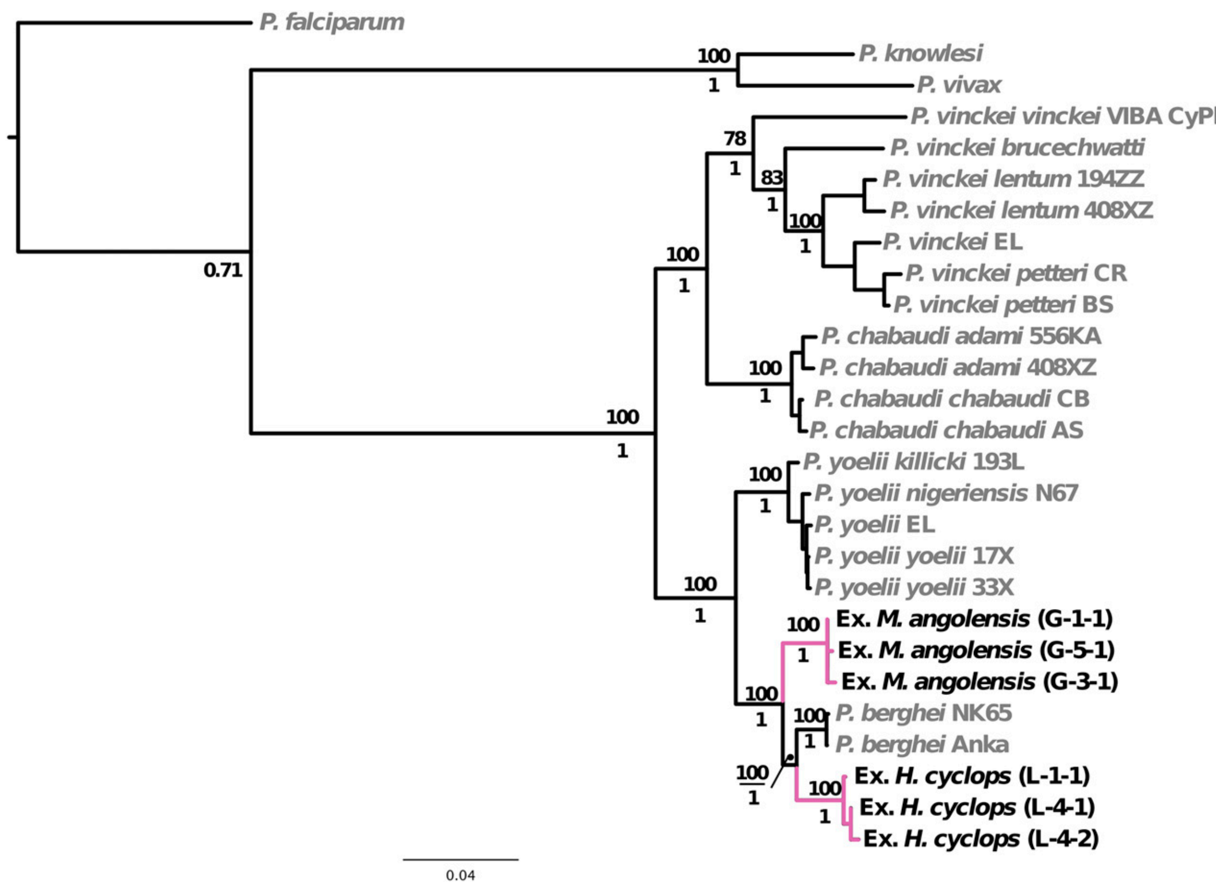

Fig. S4. Bat *Plasmodium* in context of 18 rodent *Plasmodium* lineages. Thirteen-gene maximum-likelihood phylogeny for the bat *Plasmodium* taxa from this study and rodent *Plasmodium* species, subspecies, and strains, including two *Plasmodium berghei*, five *Plasmodium yoelii,* four *Plasmodium chabaudi,* and seven *Plasmodium vinckei* lineages. Bootstrap support values above nodes and Bayesian posterior probability values are below nodes.

Table S1. Parasitemia, sex, and age of parasite-positive individuals

Parasite genus	Host species	Date collected*	Code†	Sex	Age‡	Parasitemia (%)§
Polychromophilus	*Miniopterus villiersi*	12 Dec 2008	G-3-2	*F*	Adult	0.03
		12 Dec 2008	G-3-3	*M*	Adult	< 0.001
		13 Dec 2008	G-3-1	*F*	Adult	< 0.001
	Neoromicia capensis	12 Dec 2008	G-3-1	*M*	Adult	< 0.001
	Pipistrellus aff. *grandidieri*	12 Dec 2008	G-3-1	*F*	Adult	0.02
Plasmodium	*Hipposideros cyclops*	21 Nov 2010	L-4-2	*F*	Adult	0.007
		21 Nov 2010	L-4-1	*M*	Adult	0.008
		01 Dec 2010	L-1-1	*M*	Adult	+
	Myonycteris angolensis	04 Dec 2008	G-1-1	*M*	Adult	< 0.001
		10 Dec 2008	G-5-1	*F*	Adult	< 0.001
		14 Dec 2008	G-3-1	*F*	Adult	< 0.001
Nycteria	*Rhinolophus alcyone*	19 Nov 2006	C-9-1	*M*	Adult	0.03
	Rhinolophus landeri	14 Dec 2008	G-3-1	*M*	Adult	< 0.001
Hepatocystis	*Epomophorus gambianus*	19 Dec 2008	G-4-1	*F*	Subadult/juvenile	< 0.001
	Epomops buettikoferi	19 Nov 2006	C-13-1	*F*	Adult	1.0
		19 Nov 2006	C-12-1	*F*	Adult	+
		27 Nov 2008	L-3-1	*M*	Subadult/juvenile	0.02
		18 Dec 2008	G-4-2	*F*	Adult	0.09
		19 Dec 2008	G-4-3	*F*	Adult (lactating)	< 0.001
		19 Dec 2008	G-4-1	*F*	Adult (lactating)	0.4
		12 Nov 2010	L-2-1	*F*	Adult	< 0.001
	Hypsignathus monstrosus	14 Nov 2006	C-5-1	*F*	Subadult/juvenile	< 0.001
		19 Nov 2006	C-12-1	*F*	Adult (lactating)	+
		23 Nov 2006	C-8-1	*F*	Subadult/juvenile	< 0.001
		19 Nov 2010	L-2-1	*M*	Adult	0.04
		23 Nov 2010	L-4-1	*F*	Adult	< 0.001
	Micropteropus pusillus	03 Dec 2008	G-1-8	*F*	Adult (lactating)	< 0.001
		03 Dec 2008	G-1-4	*F*	Subadult/juvenile	0.3
		03 Dec 2008	G-1-9	*M*	Adult	0.06
		03 Dec 2008	G-1-7	*F*	Adult	0.6
		03 Dec 2008	G-1-11	*F*	Adult	< 0.001
		03 Dec 2008	G-1-12	*F*	Adult	0.03
		03 Dec 2008	G-1-1	*M*	Adult	0.01
		03 Dec 2008	G-1-13	*F*	Adult (lactating)	< 0.001
		03 Dec 2008	G-1-3	*F*	Adult	0.01
		03 Dec 2008	G-1-6	*M*	Adult	0.1
		03 Dec 2008	G-1-10	*F*	Adult	< 0.001
		03 Dec 2008	G-1-14	*F*	Adult	+
		03 Dec 2008	G-1-2	*F*	Adult (lactating)	< 0.001
		04 Dec 2008	G-1-15	*F*	Adult	0.08
		04 Dec 2008	G-1-16	*F*	Adult (lactating)	0.02
		04 Dec 2008	G-1-17	*F*	Adult	< 0.001
		04 Dec 2008	G-1-5	*M*	Adult	0.07
Hepatocystis	*Micropteropus pusillus*	06 Dec 2008	G-2-4	*F*	Adult	+
		06 Dec 2008	G-2-1	*F*	Adult	0.04
		06 Dec 2008	G-2-2	*F*	Adult	0.06
		06 Dec 2008	G-2-3	*M*	Adult	0.06
		06 Dec 2008	G-2-5	*F*	Adult	0.02
		06 Dec 2008	G-2-6	*F*	Adult	0.05
		08 Dec 2008	G-5-7	*M*	Adult	0.01
		09 Dec 2008	G-5-5	*F*	Adult	< 0.001
		09 Dec 2008	G-5-8	*F*	Adult (lactating)	< 0.001
		09 Dec 2008	G-5-9	*F*	Adult (lactating)	0.02
		09 Dec 2008	G-5-2	*F*	Adult (lactating)	0.07
		09 Dec 2008	G-5-3	*F*	Adult (lactating)	< 0.001
		09 Dec 2008	G-5-1	*M*	Adult	0.03
		09 Dec 2008	G-5-6	*F*	Adult (lactating)	< 0.001
		10 Dec 2008	G-5-10	*M*	Adult	0.2
		10 Dec 2008	G-5-11	*F*	Adult	0.05
		10 Dec 2008	G-5-12	*F*	Adult	< 0.001
		10 Dec 2008	G-5-4	*F*	Adult	0.05
		13 Dec 2008	G-3-4	*F*	Adult (lactating)	< 0.001
		13 Dec 2008	G-3-1	*F*	Adult (lactating)	< 0.001

Table S1. Cont.

Parasite genus	Host species	Date collected*	Code[†]	Sex	Age[‡]	Parasitemia (%)[§]
		13 Dec 2008	G-3-3	*M*	Adult	0.02
		14 Dec 2008	G-3-2	*F*	Adult	0.01
		18 Dec 2008	G-4-6	*F*	Subadult/juvenile	< 0.001
		18 Dec 2008	G-4-7	*F*	Adult	< 0.001
		18 Dec 2008	G-4-8	*M*	Adult	+
		18 Dec 2008	G-4-9	*M*	Adult	< 0.001
		18 Dec 2008	G-4-10	*F*	Adult (lactating)	< 0.001
		18 Dec 2008	G-4-2	*F*	Subadult/juvenile	0.02
		18 Dec 2008	G-4-11	*F*	Adult	0.05
		18 Dec 2008	G-4-12	*F*	Subadult/juvenile	0.04
		18 Dec 2008	G-4-13	*F*	Subadult/juvenile	0.02
		18 Dec 2008	G-4-14	*F*	Adult	< 0.001
		18 Dec 2008	G-4-15	*M*	Adult	< 0.001
		18 Dec 2008	G-4-16	*F*	Adult (lactating)	0.02
		18 Dec 2008	G-4-17	*F*	Adult	0.06
		18 Dec 2008	G-4-4	*F*	Adult	0.05
		18 Dec 2008	G-4-18	*M*	Adult	0.05
		18 Dec 2008	G-4-1	*F*	Adult (lactating)	< 0.001
		18 Dec 2008	G-4-19	*F*	Adult	< 0.001
		18 Dec 2008	G-4-5	*F*	Adult (lactating)	0.05
		19 Dec 2008	G-4-20	*M*	Adult	0.07
		19 Dec 2008	G-4-21	*F*	Subadult/juvenile	< 0.001
		19 Dec 2008	G-4-22	*F*	Adult (lactating)	0.01
		19 Dec 2008	G-4-23	*F*	Adult (lactating)	< 0.001
	Micropteropus pusillus	19 Dec 2008	G-4-24	*F*	Adult (lactating)	0.01
		19 Dec 2008	G-4-3	*F*	Adult (lactating)	< 0.001
		19 Dec 2008	G-4-25	*F*	Subadult/juvenile	0.01
		19 Dec 2008	G-4-26	*F*	Subadult/juvenile	< 0.001
		19 Dec 2008	G-4-27	*F*	Adult (lactating)	< 0.001
		19 Dec 2008	G-4-1	*M*	Adult	0.06
		19 Dec 2008	G-1-18	*F*	Subadult/juvenile	< 0.001
	Myonycteris leptodon	08 Nov 2006	C-4-1	*F*	Adult	0.04
		08 Nov 2006	C-4-2	*M*	Adult	+
		08 Nov 2006	C-4-3	*M*	Adult	0.002
		18 Nov 2006	C-13-1	*F*	Adult	< 0.001
		18 Nov 2006	C- 13-2	*F*	Adult	+
		22 Nov 2006	C-8-1	*F*	Subadult/juvenile	+
		22 Nov 2006	C-7-1	*M*	Adult	< 0.001
		23 Nov 2006	C-7-2	*M*	Adult	+
		18 Nov 2010	L-2-1	*F*	Adult (pregnant)	< 0.001
		22 Nov 2010	L-4-1	*M*	Adult	0.03
		26 Nov 2010	L-3-1	*M*	Subadult/juvenile	0.8
	Nanonycteris veldkampii	10 Nov 2006	C-2-1	*F*	Adult	+
		22 Nov 2006	C-8-2	*M*	Subadult/juvenile	< 0.001
		23 Nov 2006	C-8-1	*M*	Adult	< 0.001
		24 Nov 2006	C-7-1	*F*	Adult	+
		01 Dec 2006	C-11-1	*F*	Adult	< 0.001
		01 Dec 2010	L-1-1	*M*	Adult	< 0.001

*All bats were captured between dusk (5:30 PM) and late night (3:00 AM).

[†]Coding of captured individuals is according to country (C, Cote d'Ivoire; G, Guinea; L, Liberia), the capture site (see Table S2) and the order of the same species at any given night.

[‡]Age was categorized into two groups, "adult" and "subadult/juvenile." Lactating/pregnant females are labeled in parenthesis.

[§]Parasitemia was determined by microscopic enumeration of infected/ total erythrocytes in Giemsa-stained blood films. +, parasite-positive, but parasite-density uncertain because of suboptimal preservation of blood-film. All microscopically identified infected individuals were confirmed by PCR.

Table S2. GPS coordinates of the sampling locations

Country_site	Latitude	Longitude	Sampling date
C_1 – C_13	05°50′00.0′′N	07°21′00.0′′W	Nov–Dec 2006
G_1	08°33′07.0′′N	08°53′57.1′′W	Dec 2008
G_2	08°37′08.2′′N	08°37′08.2′′W	Dec 2008
G_3	08°33′26.5′′N	08°55′16.5′′W	Dec 2008
G_4	08°35′35.7′′N	08°51′25.8′′W	Dec 2008
G_5	08°29′35.1′′N	08°53′48.2′′W	Dec 2008
L_1	05°39′08.3′′N	08°09′45.6′′W	Nov–Dec 2010
L_2	05°39′23.6′′N	08°11′10.8′′W	Nov–Dec 2010
L_3	05°41′53.7′′N	08°07′32.6′′W	Nov–Dec 2010
L_4	05°39′24.2′′N	08°11′26.0′′W	Nov–Dec 2010

C, Côte d'Ivoire; G, Guinea; L, Liberia. The country and site "code" corresponds with the codes in Fig. 2, and Figs. S2 and S4. The GPS coordinates (geographic reference) for each site are given in decimal degrees.

Table S3. Primer sequences

Primer name	Sequence (5′ – 3′) forward primer	Sequence (5′ – 3′) reverse primer
Cytb		
DW2/DW4	TAATGCCTAGACGTATTCCTGATTATCCAG	TGTTTGCTTGGGAGCTGTAATCATAATGTG
3932F/3932R	GGGTTATGTATTACCTTGGGGTC	GACCCCAAGGTAATACATAACCC
BatMalF3/BatHepR2	GGATTTAATGTAATGCCTAGACGT	AATGCTGTATCATACCCTAAAGGATT
BatMalF4/BatMalR5	AATCCTTTAGGGTATGATACAGC	ATGTTTGCTTGGGAGCTGTAATCAT
HepF3/HepR3	CTTACCTTGGGGACAAATGAGTTATT	CTCTAGCACCAAATGTCATTTTAAATTG
*Cox1**		
1989F/BMcox1R2	CCTGACATGGATGGATAATACTCG	GCTCCAATTGATAATACAAAATG
BMcox1F1/R1	TGGACATTATATCCTCCATTAAGTACATC	GCACCAATTGATAATACAAAATG
BMcox1F3/R3	TGGACATTATATCCACCTTTAAGTACATC	GCACCAATTGATAAAACAAAATG
BMcox1F4/R4	TGGACATTATATCCACCATTAAGTACATC	GCTCCAATAGATAAAACAAAATG
cox1F/R	CTATTTATGGTTTTCATTTTTATTTGGTA	AGGAATACGTCTAGGCATTACATTAAATCC
cox1inF/inR	ATGATATTTACARTTCAYGGWATTATTATG	GTATTTTCTCGTAATGTTTTACCAAAGAA
cox1midF/midR	TTATTCTGGTTTTTTGGTCATCCAG	CTGGATGACCAAAAAACCAGAATAA
Clpc		
clpcoutF/outR	GGTAAAACTGAATTAGCAAAAATATTA	GGACGAGCTCCATATAAAGGAT
clpcinF/ inR	TTAGCTAAACAATATTTGGTTCTG	GAGCTCCATATAAAGGATTATAAG
EF2		
EF2F1/R1	GTTCGTGAGATCATGAACAAAAC	CCTTGTAAACCAGAACCAAA
ACT1		
ACT1outF/outR	ATGGGTGACGAAGAAGTTCAAGC	TTAGAAGCATTTTCTGTGGACAATACTTGG
ACT1inF/inR	CAAGCTTTAGTTATTGATAATGG	CCCTTCCAGCCAAATCTAATCTCA
ACT2		
ACT2outF/outR	ATGCCAGAAGAATCAATAGC	TTAGAAGCACTTTCTGTGAACGATACTTGGG
ACT2inF/inR	CCAGCTATGTATGTTAGTATACAGG	CTTTAATTTTCATAGATGGCGG
ASL		
ASLoutF/outR	GSKAARTTTAATGGKGCTGTWGG	GGATTAAYTTTATGAGGCATTG
ASLinF/inR	GCTGATMAAAATRTTGATTGG	GAGGCATTGTACTACTWCC
CysPro		
CysPro_outF/outR	CAATGAAATGGTAGGTAAAAATGGT	CAATGAAATGGTAGGTAAAAATGGT
CysPro_inF/inR	GATTTTTCTAAGGAAGAATTTAATAG	TATTTTTTTATCAAACTGCTATCTAC
dhfr/ts		
dhfrF/R	CAAGATGATAGAACAGGTGTTGGTG	CCCAATACATGTATAAATTCAGCTG
H2A		
H2AoutF/outR	ATGTCAGCAAAAGGAAAAACCGG	TTAATAATCTTGGTTAGCTGTGGC
H2AinF/inR	GTCGCAAAAAAGCTGTTAAGGG	GTGGCACCTGATTTTAATTGGG
InMPDH		
InMPDH_F/R	GCKTCTAAAAGHVAAAAYAAACAATTAATTGTWGG	RAATYBTTTATTATACATRGCYTCCATACTSCCCAT
P25		
P25outF/outR	CTTTTCATATACGCTTTCCTTGG	TTAAATGATATTTGAAAATATTAG
P25inF/inR	GGCTCAAATGAGTAACCATTTAG	GAACATGCTATATATTGAATGTG
UBI		
UBIoutF/outR	ATGCAAATTTTTTGTGAAAACATTAAC	TTAGCAACCTCCTCTTAATCTTAAAAC
UBIinF/inR	GGAAAAACTATAACTCTTG	ATGTAAGGTTGATTCTTTTTG

ACT1, Actin1; *ACT2*, Actin2; *asl*, adenylosuccinate lyase; *clpc*, caseinolytic protease; *cox1*, cytochrome oxidase I; *cyspro*, cysteine proteinase; *Cytb*, cytochrome b; *dhfr/ts*, dihydrofolate reductase/thymidylate synthase; *ef2a*, elongation factor 2; F, forward primer; H2A, histone H2A; in/mid, inner primer for nested PCR; *InMPDH*, Inosine monophosphate-dehydrogenase; out, outer primer for nested PCR; *P25*, ookinete surface protein; R, reverse primer; *UBI*, poly-ubiquitin.
*Primers were used in different combinations of the available forward and reverse primers.

Table S4. GenBank accession numbers for *cytb*, *cox1*, *clpc*, and *ef2a*

Parasite	Host species	Abbreviation in Fig. 2*A* (and Fig. S2)	GenBank accession nos.			
			cytb	*cox1*	*clpc*	*ef2a*
Leucocytozoon						
L. fringillinarum	*Piplio chlorurus*	***Leucocytozoon* spp.**	**NC_012451**	**NC_012451**	—	—
L. sp.	*Buteo jamaicensis*	*Leucocytozoon* sp. (2109)	EU254518	EU254563	EU254609	—
L. sp.	*Accipiter brevipes*	***Leucocytozoon* spp. (157)**	**EU254519**	**EU254564**	**EU254610**	—
L. sp.	*Buteo lineatus*	*Leucocytozoon* sp. (2208)	EU254520	EU254565	EU254611	—
Haemoproteus						
H. columbae	*Columba livia*	***Haemoproteus columbae* (2111)**	**EU254548**	—	**EU254642**	—
H. columbae	*Columba livia*	*Haemoproteus columbae* (2146)	EU254553	—	EU254652	—
Haemoproteus (Parahaemoproteus)						
H. belopolskyi	*Sylvia curruca*	***Haemoproteus belopolskyi***	**DQ451408**	**EU254603**	**EU254657**	—
H. coatneyi	*Dendroica coronata*	***Haemoproteus coatneyi***	**EU254550**	**EU254595**	**EU254648**	—
H. turtur	*Streptopelia senegalensis*	***Haemoproteus turtur***	**DQ451425**	**EU254592**	**EU254644**	—
Plasmodium						
Avian *Plasmodium*						
P. gallinaceum	*Gallus gallus*	**Avian *Plasmodium* ssp. *P. gallinaceum***	**AY099029**	**AB564275**	**AB649424**	—
P. relictum	*Hemignathus virens*	**Avian *Plasmodium* ssp. *P. relictum***	**AY733090**	**AY733090**	**EU254633**	—
Saurian *Plasmodium*						
P. azurophilum	*Anolis oculatus*	*Plasmodium azurophilum*	EU254532	EU254575	EU254622	JN187876
P. chiricahuae	*Sceloporus jarrovi*	*Plasmodium chiricahuae*	AY099061	KF049536	KF049558	—
P. floridense	*Anolis oculatus*	**Saurian *Plasmodium* ssp. *P. floridense***	**EF079654**	**EF079654**	**EU254620**	**JN187874**
P. giganteum	*Agama agama*	*Plasmodium giganteum*	AY099053	EU254577	EU254624	—
P. hispaniolae	*Anolis distichus*	*Plasmodium hispaniolae*	JN187902	JN187863	—	JN187881
P. lacertiliae	*Emoia longicauda*	*Plasmodium lacertiliae*	EU834709	EU834714	—	—
P. leucocytica	*Anolis oculatus*	*Plasmodium leucocytica*	EU254533	EU254576	EU254623	JN187877
P. mexicanum	*Sceloporus occidentalis*	**Saurian *Plasmodium* ssp./ *P. mexicanum***	**EF079653**	**EF079653**	**EU254619**	—
P. minuoviride	*Prasinohema prehensicauda*	*Plasmodium minuoviride*	EU834703	EU834711	—	—
Mammalian *Plasmodium*						
Primate *Plasmodium*						
P. coatneyi	Thailand macaques	*Plasmodium coatneyi*	EU400407	AB354575	AB471872	—
P. cynomolgi	Old World Monkeys	***Plasmodium cynomolgi***	**AF069616**	**AB444126**	**AB471873**	—
P. falciparum	Humans	***Plasmodium falciparum***	**DQ642845**	**M76611**	**DQ642846**	**AB023579**
P. fieldi	Old World Monkeys	*Plasmodium fieldi*	AB354574	AB354574	AB471874	—
P. gaboni	*Pan troglodytes*	*Plasmodium gaboni*	FJ895307	FJ895307	HQ842630	—
P. gonderi	Old World Monkeys	*Plasmodium gonderi*	AF069622	AB434918	AB471877	—
P. hylobati	*Hylobates moloch*	*Plasmodium hylobati*	AB354573	AB354573	AB471878	—
P. inui	Old World Monkeys	*Plasmodium inui*	AF069617	AB354572	AB471879	—
P. knowlesi	Old World Monkeys	***Plasmodium knowlesi***	**AF069621**	**AY598141**	**AF348341**	**XM_002260326**
P. malariae	Humans	***Plasmodium malariae***	**AF069624**	**AB489193**	**AF348342**	—
P. ovale	Humans	***Plasmodium ovale***	**AF069625**	**JF894415**	**AY634623**	—
P. reichenowi	*Pan troglodytes*	***Plasmodium reichenowi***	**AJ251941**	**AJ251941**	**EU560464**	—
P. simiovale	Old World Monkeys	*Plasmodium simiovale*	AF069614	—	AB471881	—
P. simium	*Alouatta fuscus*	*Plasmodium simium*	AF069620	—	—	—
P. vivax	Humans	***Plasmodium vivax***	**AF069619**	**AAY26841**	**AF348344**	**XM_001615828**
P. sp.	*Pan troglodytes*	Ex. Chimp_C1	HM235391	HM235391	HM235147	—
P. sp.	*Pan troglodytes*	Ex. Chimp_C2	HM235404	HM235400	HM235145	—
P. sp.	*Pan troglodytes*	Ex. Chimp_C3	HM235360	HM235360	HM235151	—
P. sp.	*Gorilla gorilla*	Ex. Gorilla_G1	HM235288	HM235308	HM235163	—
P. sp.	*Gorilla gorilla*	Ex. Gorilla_G2	HM234984	HM235383	HM235148	—
P. sp.	*Gorilla gorilla*	Ex. Gorilla_G3	HM234998	HM235290	HM235154	—
P. sp.	*Propithecus verreauxi*	**Ex. *Propithecus verreauxi* (Sifaka)**	**AY762079**	—	—	—
Rodent *Plasmodium*						
P. atheruri	*Atherurus africans*	P. atheruri	AY099051	DQ414588	DQ417611	—

Table S4. Cont.

Parasite	Host species	Abbreviation in Fig. 2*A* (and Fig. S2)	GenBank accession nos.			
			cytb	*cox1*	*clpc*	*ef2a*
P. berghei (Anka)	*Grammomys surdaster**	***Plasmodium berghei* (Anka)**	**DQ414645**	**DQ414589**	**DQ417612**	—
P. berghei (NK65)	*G. surdaster**	*P. berghei* NK65	DQ414646	DQ414590	DQ417613	—
P. chabaudi adami (408XZ)	*Thamnomys rutilans*[†]	*P. chabaudi adami* 408XZ	DQ414647	DQ414591	DQ417614	—
P. chabaudi adami (556KA)	*T. rutilans*[†]	*P. chabaudi adami* 556KA	DQ414648	DQ414592	DQ417615	—
P. chabaudi chabaudi (AS)	*T. rutilans*[†]	***Plasmodium chabaudi* (AS)**	**DQ414649**	**DQ414593**	**DQ417616**	**XM_736543**
P. chabaudi chabaudi (CB)	*T. rutilans*[†]	*P. chabaudi chabaudi* CB	AY099050	DQ414594	DQ417617	—
P. vinckei brucechwatti	*Praomys tullbergi*	*P. vinckei brucechwatti*	DQ414652	DQ414597	DQ417620	—
P. vinckei lentum (194ZZ)	*T. rutilans*[†]	*P. vinckei lentum* 194ZZ	DQ414653	DQ414598	DQ417621	—
P. vinckei lentum (408XZ)	*T. rutilans*[†]	*P. vinckei lentum* 408XZ	DQ414654	DQ414599	DQ417622	—
P. vinckei petteri (BS)	*T. rutilans*[†]	*P. vinckei petteri* BS	DQ414655	DQ414600	DQ417623	—
P. vinckei petteri (CR)	*T. rutilans*[†]	*P. vinckei petteri* CR	DQ414656	DQ414601	DQ417624	—
P. vinckei vinckei (VIBA CyPl)	*G. surdaster**	***Plasmodium vinckei* (VIBA CyPl)**	**DQ414651**	**DQ414596**	**DQ417619**	—
P. vinckei ("EL")	*Thamnomys sp.*	*P. vinckei* EL	DQ414650	DQ414595	DQ417618	—
P. yoelii yoelii (17X)	*T. rutilans*[†]	*P. yoelii yoelii* 17X	AY099051	DQ414605	DQ417628	—
P. yoelii yoelii (33x)	*T. rutilans*[†]	***Plasmodium yoelii* (33x)**	**DQ414660**	**DQ414606**	**DQ417630**	—
P. yoelii (EL)	*T. rutilans*[†]	*P. yoelii* EL	DQ414657	DQ414602	DQ417625	—
P. yoelii killicki	*T. rutilans*[†]	*P. yoelii Killicki* 193L	DQ414658	DQ414603	DQ417626	—
P. yoelii nigeriensis	*T. rutilans*[†]	P. yoelii nigeriensis N67	DQ414659	DQ414604	DQ417627	—
Bat *Plasmodium*						
P. cyclopsi	*Hipposideros cyclops*	**Ex. *Hipposideros cyclops* (L-1-1)**	**KF159710**	**KF159788**	**KF159635**	**KF159729**
P. cyclopsi	*Hipposideros cyclops*	Ex. *Hipposideros cyclops* (L-4-1)	KF159674	KF159789	KF159630	KF159728
P. cyclopsi	*Hipposideros cyclops*	Ex. *Hipposideros cyclops* (L-4-2)	KF159716	KF159791	KF159637	F
P. voltaicum	*Myonycteris angolensis*	**Ex. *Myonycteris angolensis* (G-1-1)**	**KF159671**	**KF159792**	**KF159648**	**F**
P. voltaicum	*Myonycteris angolensis*	Ex. *Myonycteris angolensis* (G-3-1)	F	F	KF159634	F
P. voltaicum	*Myonycteris angolensis*	Ex. *Myonycteris angolensis* (G-5-1)	KF159692	KF159793	F	F
Hepatocystis						
Primate *Hepatocystis*						
Hep. sp.	*Cercopithecus cephus*	**Ex. *Cercopithecus cephus* (Gabon/GAB_1)**	**JF923760**	—	—	—
Hep. sp.	*Cercopithecus cephus*	Ex. *Cercopithecus cephus* (Gabon/GAB_2)	JF923758	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (Cambodia)__1	JQ070884	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_2)	JQ070893	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_3)	JQ070872	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_4)	JQ070903	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_5)	JQ070940	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_6)	JQ070922	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_7)	JQ070892	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_8)	JQ070926	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_9)	JQ070897	—	—	—
Hep. sp.	*Cercopithecus nictitans*	Ex. *Cercopithecus nictitans* (CAM_10)	JQ070925	—	—	—

Table S4. Cont.

Parasite	Host species	Abbreviation in Fig. 2*A* (and Fig. S2)	GenBank accession nos.			
			cytb	*cox1*	*clpc*	*ef2a*
Hep. sp.	*Macaca sp.*	Ex. *Macaca* sp. (MYA_1)	GU930064	—	—	—
Hep. sp.	*Macaca sp.*	Ex. *Macaca* sp. (THA_1)	GU930051	—	—	—
Hep. sp.	*Macaca sp.*	Ex. *Macaca* sp. (THA_2)	EU400409	—	—	—
Hep. sp.	*Macaca sp.*	Ex. *Macaca* sp. (THA_3)	GU930042	—	—	—
Hep. sp.	*Mandrillus sphinx*	**Ex. *Mandrillus sphinx* (Gabon/GAB_1)**	**JF923759**	—	—	—
Hep. sp.	*Miopithecus talapoin*	Ex. *Miopithecus talapoin* (GAB_2)	JF923757	—	—	—
Hep. sp.	*Papio nubensis*	**Ex. *Papio nubensis* (Ethiopia/ETH_1)**	**AF069626**	—	—	—
Bat *Hepatocystis*						
Hep. sp.	*Cynopterus brachyotis*	**Ex. *Cynopterus brachyotis* (Singapore/SIN_1)**	**EU254526**	**EU254569**	**EU254616**	—
Hep. sp.	*Cynopterus brachyotis*	Ex. *Cynopterus brachyotis* (Singapore/SIN_2)	AY099030	—	—	—
Hep. sp.	*Cynopterus horsfieldii*	**Ex. *Cynopterus horsfieldii* (Borneo/BOR_1)**	**DQ396527**	—	—	—
Hep. sp.	*Cynopterus horsfieldii*	Ex. *Cynopterus horsfieldii* (Borneo/BOR_2)	DQ396528	—	—	—
Hep. sp.	*Epomophorus gambianus*	**Ex. *Epomophorus gambianus* (G-4-1)**	**KF159695**	**F**	**F**	**F**
Hep. sp.	*Epomops buettikoferi*	Ex. *Epomops buettikoferi* (G-4-1)	KF159701	KF159768	KF159636	F
Hep. sp.	*Epomops buettikoferi*	Ex. *Epomops buettikoferi* (G-4-2)	KF159706	KF159779	KF159612	KF159757
Hep. sp.	*Epomops buettikoferi*	Ex. *Epomops buettikoferi* (G-4-3)	KF159703	KF159790	F	F
Hep. sp.	*Epomops buettikoferi*	**Ex. *Epomops buettikoferi* (C-13-1)**	**KF188067**	**F**	**F**	**F**
Hep. sp.	*Epomops buettikoferi*	**Ex. *Epomops buettikoferi* (L-2-1)**	**KF159694**	**KF159798**	**F**	**F**
Hep. sp.	*Epomops buettikoferi*	Ex. *Epomops buettikoferi* (L-4-1)	F	F	F	KF159741
Hep. sp.	*Hipposideros larvatus*	Ex. *Hipposideros larvatus* (CAM_1)	EF179356	—	—	—
Hep. sp.	*Hypsignathus monstrosus*	Ex. *Hypsignathus monstrosus* (L-2-1)	KF159689	KF159799	F	KF159734
Hep. sp.	*Hypsignathus monstrosus*	**Ex. *Hypsignathus monstrosus* (L-4-1)**	**KF159712**	**F**	**KF159647**	**F**
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-1)	KF159691	KF159800	KF159614	F
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-2)	KF159688	KF159758	KF159621	F
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-3)	KF188068	KF188070	F	KF188072
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-4)	KF159713	KF159759	KF159622	KF159738
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-5)	KF159677	KF159760	KF159620	KF159736
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-6)	KF159718	KF159761	KF159613	KF159722
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-7)	KF159719	KF159762	F	KF159725
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-8)	KF159686	KF159763	KF159646	KF159727
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-9)	KF159702	KF159764	KF159643	KF159745
Hep. sp.	*Micropteropus pusillus*	**Ex. *Micropteropus pusillus* (G-1-10)**	**KF159683**	**KF159801**	**KF159623**	**KF159744**
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-11)	KF159682	KF159802	KF159632	KF159732
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-1-12)	KF159711	KF159803	KF159624	KF159743
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-2-1)	KF159670	F	KF159619	KF159752
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-2-2)	KF159709	KF159765	KF159644	F
Hep. sp.	*Micropteropus pusillus*	**Ex. *Micropteropus pusillus* (G-2-3)**	**KF159685**	**KF159766**	**KF159627**	**KF159747**
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-2-4)	KF159708	KF159767	KF159638	KF159739
Hep. sp.	*Micropteropus pusillus*	**Ex. *Micropteropus pusillus* (G-3-1)**	**KF159696**	**KF159769**	**KF159640**	**KF159724**
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-3-2)	KF159669	KF159770	KF159617	KF159726
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-3-3)	KF159697	KF159771	KF159629	KF159737
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-4-1)	KF159679	KF159772	KF159615	F
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-4-2)	KF159693	KF159773	KF159645	KF159746
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-4-3)	KF159672	KF159774	KF159633	KF159730
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-4-4)	F	F	KF159628	F
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-4-5)	KF159704	KF159775	KF159618	KF159753
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-4-6)	F	F	F	KF159735
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-5-1)	KF159717	F	F	KF159748
Hep. sp.	*Micropteropus pusillus*	**Ex. *Micropteropus pusillus* (G-5-2)**	**KF159676**	**KF159776**	**KF159626**	**F**
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-5-3)	F	KF159777	KF159625	KF159721
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-5-4)	KF159680	KF159778	KF159641	KF159723
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-5-5)	KF159673	KF159780	F	KF159733
Hep. sp.	*Micropteropus pusillus*	Ex. *Micropteropus pusillus* (G-5-6)	KF159687	KF159781	KF159649	KF159756

Table S4. Cont.

Parasite	Host species	Abbreviation in Fig. 2*A* (and Fig. S2)	GenBank accession nos.			
			cytb	*cox1*	*clpc*	*ef2a*
Hep. sp.	*Myonycteris leptodon*	**Ex. *Myonycteris leptodon* (C-7-1)**	**KF188066**	**KF188069**	**F**	**KF188071**
Hep. sp.	*Myonycteris leptodon*	**Ex. *Myonycteris leptodon* (C-7-2)**	**F**	**KF159782**	**F**	**F**
Hep. sp.	*Myonycteris leptodon*	**Ex. *Myonycteris leptodon* (L-2-1)**	**KF159707**	**KF159783**	**F**	**KF159755**
Hep. sp.	*Myonycteris leptodon*	**Ex. *Myonycteris leptodon* (L-3-1)**	**KF159705**	**N**	**F**	**KF159754**
Hep. sp.	*Myonycteris leptodon*	**Ex. *Myonycteris leptodon* (L-4-1)**	**KF159678**	**KF159784**	**F**	**KF159750**
Hep. sp.	*Nanonycteris veldkampii*	**Ex. *Nanonycteris veldkampii* (C-8-1)**	**KF159684**	**KF159785**	**F**	**F**
Hep. sp.	*Nanonycteris veldkampii*	Ex. *Nanonycteris veldkampii* (C-8-2)	KF159715	F	F	F
Hep. sp.	*Nanonycteris veldkampii*	**Ex. *Nanonycteris veldkampii* (L-1-1)**	**KF159698**	**KF159786**	**KF159631**	**KF159749**
Hep. sp.	*Nanonycteris veldkampii*	**Ex. *Nanonycteris veldkampii* (Guinea/GUI_1)**	**EU254528**	**EU254571**	**EU254618**	—
Hep. sp.	*Nanonycteris veldkampii*	Ex. *Nanonycteris veldkampii* (Guinea/GUI_2)	EU254527	EU254570	EU254617	—
Hep. sp.	*Pteropus hypomelanus*	**Ex. *Pteropus hypomelanus* (Malaysia/MAL_1)**	**FJ168565**	**FJ168565**	—	—
Hep. sp.	*Pteropus vampyrus*	Ex. *Pteropus vampyrus* (Borneo/BOR_1)	DQ396530	—	—	—
Hep. sp.	*Pteropus vampyrus*	**Ex. *Pteropus vampyrus* (Borneo/BOR_2)**	**DQ396531**	—	—	—
Polychromophilus						
Poly. melanipherus	*Miniopterus schreibersii*	*Polychromophilus melanipherus* (_3)	JN990708	JN990714	JN990720	—
Poly. melanipherus	*Miniopterus schreibersii*	***Polychromophilus melanipherus* (_4)**	**JN990709**	**JN990715**	**JN990721**	—
Poly. melanipherus	*Miniopterus schreibersii*	*Polychromophilus melanipherus* (_5)	JN990710	JN990716	JN990722	—
Poly. melanipherus	*Miniopterus schreibersii*	*Polychromophilus melanipherus* (_6)	JN990711	JN990717	—	—
Poly. murinus	*Myotis daubentonii*	***Polychromophilus murinus* (_1)**	**HM05583**	**JN990718**	**JN990723**	—
Poly. murinus	*Myotis daubentonii*	*Polychromophilus murinus* (_2)	HM05584	JN990719	JN990724	—
Poly. murinus	*Myotis daubentonii*	*Polychromophilus murinus* (_3)	HM05585	—	—	—
Poly. murinus	*Myotis daubentonii*	*Polychromophilus murinus* (_4)	HM05586	—	—	—
Poly. murinus	*Myotis daubentonii*	*Polychromophilus murinus* (_5)	HM05587	—	—	—
Poly. murinus	*Myotis daubentonii*	*Polychromophilus murinus* (_6)	HM05588	—	—	—
Poly. murinus	*Myotis daubentonii*	Ex. *Myotis daubentoni_7*	HM05589	—	—	—
Poly. sp.	*Myotis goudoti*	Ex. *Myotis goudoti* (Madagascar)	AY762072	—	—	—
Poly. sp.	*Miniopterus manavi*	Ex. Min_manavi (Madagascar) _MAD1	AY762070	—	—	—
Poly. sp.	*Miniopterus manavi*	Ex. Min_manavi (Madagascar) _MAD2	AY762071	—	—	—
Poly. sp.	*Miniopterus manavi*	Ex. Min_manavi (Madagascar) _MAD3	AY762074	—	—	—
Poly. sp.	*Kerivoula hardwickii*	Ex. *Kerivoula hardwickii* (Cambodia)_CAM1	EF179354	—	—	—
Poly. sp.	*Miniopterus villiersi*	Ex. *Miniopterus villiersi* (G-3-1)	KF159675	KF159794	F	F
Poly. sp.	*Miniopterus villiersi*	**Ex. *Miniopterus villiersi* (G-3-2)**	**KF159699**	**KF159795**	**KF159616**	**KF159740**
Poly. sp.	*Miniopterus villiersi*	Ex. *Miniopterus villiersi* (G-3-3)	KF159681	KF159796	KF159642	KF159731
Poly. sp.	*Neoromicia capensis*	**Ex. *Neoromicia capensis* (G-3-1)**	**KF159700**	**F**	**F**	**F**
Poly. sp.	*Pipistrellus* aff. *grandidieri*	**Ex. *Pipistrellus grandidieri* (G-3-1)**	**KF159714**	**KF159797**	**KF159639**	**KF159742**
Nycteria						
N. sp.	*Rhinolophus alcyone*	**Ex. *Rhinolophus alcyone* (C-9-1)**	**KF159720**	**F**	**F**	**KF159751**
N. sp.	*Rhinolophus landeri*	**Ex. *Rhinolophus landeri* (G-3-1)**	**KF159690**	**KF159787**	**F**	**F**
Haemosporida sp.	*Megaderma spasma*	**Ex. *Megaderma spasma* (Cambodia)**	**EF179355**	—	—	—

Samples/accession numbers in bold have been included in the analysis for both Fig. 2*A* and Fig. S2. F, failed to amplify. Sequences from this study are highlighted in red. A dash signifies the sequence is not available/not attempted to amplify.

*Grammomys surdaster = Grammomys dolichurus.

†Thamnomys rutilans = Grammomys poensis.

Table S5. GenBank accession numbers for additional genes used for bat and rodent *Plasmodium* taxa only

Plasmodium species	Host species	Abbreviation in Fig. 2*B* and Fig. S4	GenBank accession numbers*								
			Actin-1	*Actin-2*	*asl*	*cyspro*	*dhfr*	*H2A*	*InMPDH*	*P25*	*UBI*
P. falciparum	**Humans**	***Plasmodium falciparum***	**XM_001350811**	**M22718**	**XM_001349541**	**XM_001348691**	**XM_001351443**	**M86865**	**XM_001352043**	**XM_001347551**	**XM_001350490**
P. knowlesi	**Old World Monkeys**	***Plasmodium knowlesi***	**XM_002262498**	**XM_002260836**	**XM_002257931**	**XM_002260255**	**XM_002258192**	**XM_002261462**	**XM_002258443**	**XM_002261792**	**XM_002260588**
P. vivax	**Humans**	***Plasmodium vivax***	**XM_001613936**	**XM_001616779**	**XM_001612942**	**XM_001615757**	**XM_001615032**	**XM_001616141**	**XM_001614690**	**XM_001608410**	**XM_001616541**
P. atheruri	*Atherurus africans*	*P. atheruri*	—	—	—	DQ414607	DQ414626	—	—		
P. berghei	***Grammomys surdaster***†	***Plasmodium berghei (P. berghei Anka)***	**PBANKA_145930**‡	**XM_675072**	**AF262049**	**DQ414608**	**DQ414627**	**PBANKA_111700**§	**XM_673207**	**PBANKA_051500**§	**PBANKA_061030**§
P. berghei (NK65)	*G. surdaster*†	*P. berghei* NK65	—	—	—	DQ414609	DQ414628	—	—	—	—
P. chabaudi adami (408XZ)	*T. rutilans*‡	*P. chabaudi adami* 408XZ	—	—	—	DQ414610	DQ414629	—	—	—	—
P. chabaudi adami (556KA)	*T. rutilans*‡	*P. chabaudi adami* 556KA	—	—	—	DQ414611	DQ414630	—	—	—	—
***P. chabaudi chabaudi* (AS)**	***Thamnomys rutilans***‡	***Plasmodium chabaudi* (AS)**	**XM_730543**	**EU254668**	**DQ414612**	**M30834**	**DQ414631**	**XM_740624**	**XM_739934**	—	—
P. chabaudi chabaudi (CB)	*T. rutilans*‡	*P. chabaudi chabaudi* CB	—	—	—	DQ414613	DQ414632	—	—	—	—
P. vinckei brucechwatti	*Praomys tullbergi*	*P. vinckei brucechwatti*	—	—	—	DQ414616	DQ414635	—	—	—	—
P. vinckei lentum (194ZZ)	*T. rutilans*‡	*P. vinckei lentum* 194ZZ	—	—	—	DQ414617	DQ414636	—	—	—	—
P. vinckei lentum (408XZ)	*T. rutilans*‡	*P. vinckei lentum* 408XZ	—	—	—	DQ414618	DQ414637	—	—	—	—
P. vinckei petteri (BS)	*T. rutilans*‡	*P. vinckei petteri* BS	—	—	—	DQ414619	DQ414638	—	—	—	—
P. vinckei petteri (CR)	*T. rutilans*‡	*P. vinckei petteri* CR	—	—	—	DQ414620	DQ414639	—	—	—	—
***P. vinckei vinckei* (VIBA CyPl)**	***G. surdaster***†	***P. vinckei* (VIBA CyPl)**	—	—	**EU254667**	**DQ414615**	**DQ414634**	—	—	—	—
P. vinckei ("EL")	*Thamnomys sp.*	*P. vinckei* EL	—	—	—	DQ414621	DQ414633	—	—	—	—
P. yoelii yoelii (17X)	*T. rutilans*†	*P. yoelii yoelii* 17X	—	—	—	DQ414624	DQ414643	—	—	—	—
***P. yoelii yoelii* (33x)**	***T. rutilans***‡	***Plasmodium yoelii* (33x)**	**XM_725017**	**XM_724231**	**AF262050**	**DQ414625**	**DQ414644**	**XM_720384**	**XM_718689**	**XM_720005**	**PYYM_0611900**
P. yoelii ("EL")	*T. rutilans*‡	*P. yoelii* EL	—	—	—	DQ411462	DQ414640	—	—	—	—
P. yoelii killicki	*T. rutilans*‡	*P. yoelii Killicki* 193L	—	—	—	DQ421462	DQ414641	—	—	—	—

Table S5. Cont.

Plasmodium species	Hosts pecies	Abbreviation in Fig. 2*B* and Fig. S4	GenBank accession numbers*								
			Actin-1	*Actin-2*	*asl*	*cyspro*	*dhfr*	*H2A*	*InMPDH*	*P25*	*UBI*
P. yoelii nigeriensis	*T. rutilans*‡	*P. yoelii nigeriensis* N67	—	—	—	DQ414623	DQ414642	—	—	—	—
P. cyclopsi	***Hipposideros cyclops***	**Ex. *Hipposideros cyclops* (L1-1)**	**KF159609**	**KF159650**	**F**	**F**	**F**	**KF159658**	**F**	**KF159663**	**KF159664**
P. cyclopsi	***Hipposideros cyclops***	**Ex. *Hipposideros cyclops* (L-4-1)**	**KF159610**	**KF159651**	**KF159653**	**KF159655**	**F**	**KF159659**	**KF159661**	**F**	**KF159665**
P. cyclopsi	***Hipposideros cyclops***	**Ex. *Hipposideros cyclops* (L-4-2)**	**KF159611**	**KF159652**	**F**	**F**	**F**	**KF159660**	**F**	**F**	**KF159666**
P. voltaicum	***Myonycteris angolensis***	**Ex. *Myonycteris angolensis* (G-1-1)**	**F**	**F**	**KF159654**	**KF159656**	**KF159657**	**F**	**KF159662**	**F**	**F**

Samples/accession numbers in bold have been included in the analysis for both Fig. 2*B* and Fig. S4.

*Additional nine genes inlcuded in the analysis for Fig. 2*B* and Fig. S4 (accession numbers for cytochrome *b*; cytochrome oxidase I; caseinolytic protease and elongation factor 2 are given in Table S4). *ACT1*, Actin-1; *ACT2*, Actin-2; *asl*, adenylosuccinate lyase; *cyspro* , cysteine proteinase; *dhfr*/*ts*, dihydrofolate reductase/thymidylate synthase; F, failed to amplify; H2A, histone H2A; *InMPDH,* Inosinmonophosphat-Dehydrogenase; *P25*, ookinete surface protein; *UBI*, poly-ubiquitin. Sequences from this study are highlighted in red. A dash represents sequence not available/not attempted.

†*Grammomys surdaster* = *Grammomys dolichurus*.

‡*Thamnomys rutilans* = *Grammomys poensis*.

§PlasmoD.