Taxonomic revision of the Red-fronted Tinkerbird

_Pogoniulus pusillus_ (Dumont, 1816) based on molecular and phenotypic analyses

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Summary.—Red-fronted Tinkerbird _Pogoniulus pusillus_ (Dumont, 1816) presently comprises three recognised subspecies, of which two are found in East Africa and one occurs disjunctly in southern Africa. Based on their respective distributions and phenotypic differences, a taxonomic reassessment of the species is warranted. We performed a phylogenetic reconstruction using the mitochondrial genes ATPase 6/8 based on 33 samples from across the distribution of Red-fronted Tinkerbird and four outgroup samples, and then determined correspondence between genetic distances and differences in song and morphology among clades using the Tobias _et al._ criteria. Our phylogenetic analyses revealed 4.4% sequence divergence in mtDNA between northern and southern populations, with plumage, morphometric and song differences of a similar magnitude to those between _P. pusillus_ and Yellow-fronted Tinkerbird _P. chrysoconus_, and above species-level thresholds according to the Tobias _et al._ criteria. Furthermore, the molecular phylogeny supports recognition of a synonymised taxon (_P. p. eupterus_) as a distinct, but phenotypically cryptic, subspecies in East Africa, with c.1.5% sequence divergence from _P. p. affinis_ and _P. p. uropygialis_, which in turn differ less (1%) from each other. We propose that northern and southern Red-fronted Tinkerbirds are treated as separate species, and that the subspecies _eupterus_ is resurrected. Our findings suggest that _P. chrysoconus_ as presently constituted may also merit taxonomic revision.

Red-fronted Tinkerbird _Pogoniulus pusillus_ (Dumont, 1816) has a disjunct distribution (Fig. 1), with two subspecies, _P. p. affinis_ (Reichenow, 1879) and _P. p. uropygialis_ (von Heuglin, 1862) from East Africa currently recognised by the IOC world bird list, and which are separated from the third, the nominate southern African race, by at least 1,500 km (Table 1). _P. p. pusillus_ (including _niethammeri_ Clancey, 1952), ranging from Eastern Cape, South Africa, to Sul do Save, Mozambique, differs in body size and plumage from the taxa in East Africa (Short & Horne 2001), but these differences have not hitherto been quantified to determine if they meet species-level criteria (e.g. Tobias _et al._ 2010). Furthermore, the transition zone between the two currently recognised subspecies in East Africa, _P. p. affinis_ ranging from southern Ethiopia and Somalia south to the Rufiji River in Tanzania and west to Oldeani (Tanzania) and north Uganda, and _P. p. uropygialis_ in northern and central Ethiopia to Eritrea and northern Somalia (White 1965), has remained unclear (Ash & Atkins 2009). These subspecies differ primarily in the yellow dorsal streaking of _P. p. uropygialis_, vs. white in _affinis_. Two additional subspecies have been recognised but since synonymised: subtle morphological differences led Grote (1928) to describe _eupterus_ (type locality
Ukerewe Island; see Fig. 1, Table 1), from inland Tanzania as distinct from *P. p. affinis*, which he considered a coastal race (Grote 1939). Grant & Mackworth-Praed (1942), finding overlap in wing length between coastal and inland forms, disagreed and synonymised it within *affinis* but instead recognised *lollesheid* (van Someren, 1931, 1932) from the Juba
River, Somalia, based on the latter’s shorter wing. Subsequently, White (1965) synonymised lollesheid (sic, possibly an unjustified emendation) within affinis, citing the variation in size as clinal. Thus, with several forms described from East Africa, a taxonomic reappraisal is warranted, ideally incorporating molecular phylogenetics.

Red-fronted Tinkerbird had been distinguished from its presumed sister taxon, Yellow-fronted Tinkerbird *P. chrysoconus*, primarily using plumage differences, most notably forecrown colour. It is most parsimonious for a derived trait, such as the red forecrown colour, to have evolved once, but with the red forecrown feathers in this species explained by a gene that converts yellow dietary carotenoids to red ketocarotenoids (Kirschel et al. 2020a), it is possible that homoplasy, wherein different mutations lead to a convergent phenotype, explains such parallel patterns. It is therefore possible that disjunct populations of tinkerbirds with red forecrowns have resulted from independent mutations among genes functioning in the conversion of carotenoids. A taxonomic reappraisal of Red-fronted Tinkerbird should thus incorporate all currently recognised subspecies of Yellow-fronted Tinkerbird (*P. c. chrysoconus, P. c. extoni* and *P. c. xanthostictus*) to determine whether they might actually be closer relatives of any of the subspecies of Red-fronted Tinkerbird.

We re-examine the relationships between populations of *P. pusillus* using molecular genetics and relate our findings to morphological variation according to Tobias et al. (2010) criteria. We then suggest a taxonomic reappraisal based on our findings. Our results suggest southern and eastern Africa forms have long been diverging, reflected by phenotypic differences, whilst there are genetic breaks in regions of East Africa that have seldom been identified as playing a role in the avian biogeography of the region.

**Methods**

**Field work and sampling.**—Field work was performed in Ethiopia in January and April 2019, and in Eswatini (formerly Swaziland) in July 2019. Blood samples were collected from the brachial vein of tinkerbirds. Of the samples collected by the University of Cyprus (UCY; see Table 2), seven *P. p. uropygialis*, five *P. p. affinis* and one *P. c. xanthostictus* from Ethiopia, along with one *P. p. pusillus* from Eswatini were then pooled with five samples of presumed *P. p. affinis* and one of *P. c. extoni* collected during field work in Kenya in 2011–14 and Tanzania in 2013 (described in Nwankwo et al. 2018), and three of *P. p. pusillus* collected in South Africa in 2015 (see Nwankwo et al. 2019). Two additional samples were provided by museum collections (a blood sample of *P. p. pusillus* from Morgan’s Bay, South Africa, provided by the Museum of Vertebrate Zoology of the University of California Berkeley...

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**TABLE 1**
Currently recognised taxa (by the IOC world bird list) and synonyms, years of description, authors, and type localites.

| Scientific name | Original name | Year | Author | Type locality |
|-----------------|---------------|------|--------|---------------|
| *Pogoniulus pusillus pusillus* | *Bucco pusillus* | 1816 | Dumont | Sunday River, East Cape Province |
| * junior synonym | *Pogoniulus pusillus niethammeri* | 1952 | Clancey | Makane’s Point, Pongola River, north-east Zululand |
| *Pogoniulus pusillus uropygialis* | *Barbatula uropygialis* | 1862 | von Heuglin | Ain Saba, Eritrea |
| *Pogoniulus pusillus affinis* | *Barbatula affinis* | 1879 | Reichenow | Kipini, Kenya |
| * junior synonym | *Pogoniulus pusillus eupterus* | 1928 | Grote | Ukerewe Island, Tanzania |
| * junior synonym | *Pogoniulus pusillus lollesheid* | 1931 | van Someren | Serenli, Juba River, Somalia |
| Source | Sample | Accession no. | Species | Subspecies | Locality | Country          |
|--------|--------|---------------|---------|------------|----------|------------------|
| KU     | 8528   | OK649847      | scolopaceus | flavisquamatus | Centro Sur, Rio Lobo at Asoc | Equatorial Guinea |
| UCY    | J3146  | OK649849      | chrysoconus | extoni     | Mikumi  | Tanzania          |
| UCY    | J3130  | MH364276      | chrysoconus | chrysoconus | Queen Elizabeth | Uganda           |
| UCY    | AS32040| OK649848      | chrysoconus | xanthostictus | Harena forest | Ethiopia         |
| UCY    | AS39214| OK649850      | pusillus    | pusillus   | Mlawula  | Eswatini          |
| UCY    | AR93131| MK492415      | pusillus    | pusillus   | Kube Yini | South Africa     |
| UCY    | AR93132| OK649871      | pusillus    | pusillus   | Kube Yini | South Africa     |
| UCY    | AR93133| OK649872      | pusillus    | pusillus   | Vernon Crookes | South Africa |
| UCY    | AR93138| OK649869      | pusillus    | pusillus   | Lake Eland | South Africa     |
| UCY    | AR93139| MK492417      | pusillus    | pusillus   | Lake Eland | South Africa     |
| MVZ    | JF551  | MH364289      | pusillus    | pusillus   | Table Farm | South Africa     |
| MVZ    | JF734  | OK649870      | pusillus    | pusillus   | Morgan’s Bay | South Africa   |
| MVZ    | JF789  | MK492418      | pusillus    | pusillus   | Morgan’s Bay | South Africa     |
| MVZ    | JF790  | MH364292      | pusillus    | pusillus   | Morgan’s Bay | South Africa     |
| UCY    | AS32033| OK649854      | pusillus    | uropygialis | Bishangari | Ethiopia         |
| UCY    | AS32034| OK649852      | pusillus    | uropygialis | Bishangari | Ethiopia         |
| UCY    | AS32046| OK649851      | pusillus    | uropygialis | Wondo Genet | Ethiopia         |
| UCY    | AS32055| OK649853      | pusillus    | uropygialis | Wondo Genet | Ethiopia         |
| UCY    | AS32071| OK649859      | pusillus    | uropygialis | Blue Nile Falls | Ethiopia |
| UCY    | AS32086| OK649858      | pusillus    | uropygialis | Checheho   | Ethiopia         |
| UCY    | AS32087| OK649860      | pusillus    | uropygialis | Lalibela   | Ethiopia         |
| UCY    | AS32041| OK649855      | affinis     | affinis    | Dolo Mena | Ethiopia         |
| UCY    | AS32042| OK649861      | affinis     | affinis    | Dolo Mena | Ethiopia         |
| UCY    | AS32043| OK649862      | affinis     | affinis    | Dolo Mena | Ethiopia         |
| UCY    | AS32045| OK649856      | affinis     | affinis    | Dolo Mena | Ethiopia         |
| UCY    | AS32052| OK649863      | affinis     | affinis    | Harena forest | Ethiopia |
| UCY    | AS32056| OK649857      | affinis     | affinis    | Harena forest | Ethiopia |
| UCY    | K69359 | MH364298      | affinis     | affinis    | Watamu     | Kenya            |
| UCY    | K69360 | MH364299      | affinis     | affinis    | Lamu       | Kenya            |
| FMNH   | T37337 | MH364303      | affinis     | affinis    | Wenge East | Kenya            |
| UCY    | J31303 | OK649865      | pusillus    | eupterus   | Kongelai   | Kenya            |
| UCY    | J31306 | OK649866      | pusillus    | eupterus   | Kongelai   | Kenya            |
| UCY    | J31312 | MH364279      | pusillus    | eupterus   | Awasi      | Kenya            |
| UCY    | J31315 | MH364281      | pusillus    | eupterus   | Diani      | Kenya            |
| UCY    | K69301 | OK649867      | pusillus    | eupterus   | Mbeya      | Tanzania         |
| UCY    | K69340 | MH364297      | pusillus    | eupterus   | Diani      | Kenya            |
| UCY    | K69357 | OK649864      | pusillus    | eupterus   | Tiwi       | Kenya            |
| UCY    | K69358 | OK649868      | pusillus    | eupterus   | Vipingo    | Kenya            |
Lab work.—DNA was extracted from blood and tissue samples using a Qiagen DNeasy blood and tissue kit, following the manufacturer's protocols (Qiagen, Valencia, CA, USA). Polymerase chain reaction (PCR) was then performed to amplify DNA of ATPase 6/8 genes in 10 μL reactions containing: 1× reaction buffer, 25 mM MgCl₂, 10 mM dNTP, 1 U/μL Taq polymerase (Kapa Biosystems), and 1 μM of each primer. The primers ATPC02GQL (5'-GGACAATGCTCAGAAATCTGC-3') and ATPC03HMH (5'-CATGGGCTGGGGTCRACTATG-3') (Eberhard & Bermingham 2004) were used, and PCR amplification was performed using an Applied Biosystems 2720 Thermocycler. The thermal protocol involved an initial denaturation step at 95°C for five minutes, followed by 35 cycles (98°C / 40 seconds, 52°C / 45 seconds, 72°C / 40 seconds) and a final extension step at 72°C for five minutes. Resulting bands from gel electrophoresis of PCR products were visualised on a 1% agarose gel and extracted using a Macherey-Nagel gel extraction kit for Sanger sequencing. Resultant sequences were pooled, with 12 further sequences obtained from previous studies (see Nwankwo et al. 2019, Kirschel et al. 2020b), resulting in a total of 38 sequences (Table 2).

Phylogenetic reconstruction.—Sequences were aligned using MUSCLE in MEGA v.7 (Kumar et al. 2016) and imported as Fasta into Geneious, where they were converted to phylip format for maximum likelihood phylogenetic analysis using IQtree (Minh et al. 2020). Blocks were partitioned by codon position (Chernomor et al. 2016), then model selection was performed (Kalyaanamoorthy et al. 2017) based on best Bayesian Information Criterion (BIC) score, and maximum likelihood estimation was performed using 1,000 ultrafast bootstraps (Hoang et al. 2017).

Bayesian analysis was performed using MrBayes (Huelsenbeck & Ronquist 2001) in Geneious (Kearse et al. 2012), running four heated chains for one million generations, sampling every 1,000 generations with a burn-in of 250,000 generations. We also calculated uncorrected pairwise sequence divergence using distance estimates provided in Geneious.

Morphometrics.—We measured wing chord, tarsus, tail, bill (maxilla and mandible) length, and bill width of 128 P. pusillus and 148 P. chrysoconus specimens (Table 3) from the Natural History Museum, Tring (NHMUK); Natural History Museum of Los Angeles County, Los Angeles (LACM); Field Museum of Natural History, Chicago (FMNH); National Museum of Natural History, Smithsonian Institution, Washington, DC (USNM); American Museum of Natural History, New York (AMNH); Museum of Natural Science of Louisiana State University, Baton Rouge, LA (LSUMZ); Peabody Museum of Yale University, New Haven, CT (YPM); and the University of California Los Angeles Dickey Collection, Los Angeles (UCLA). One of us (ANGK) took all morphometric measurements. In accord with Tobias et al. (2010), we used Cohen’s d statistic to quantify the effect size of morphological differences and assigned magnitude scores to plumage data and behavioural and ecological differences, to determine if different populations might qualify for species status according to the yardstick approach (Tobias et al. 2010). Specimens of both sexes were included in analyses after first running tests for differences between the sexes; in these monomorphic species, sex was determined by collectors based on gonad identification, and comprised 171 males and 105 females. Comparisons were made between P. chrysoconus and P. pusillus to establish the yardstick, then between P. p. pusillus (n = 23) and the East African populations (n = 125), and finally between three sets of East African populations that accorded with clades identified from the phylogenetic reconstruction.

Song analysis.—Songs of Pogoniulus tinkerbirds are very simple in structure, typically comprising ‘pulses’ (short tones, see Fig. 2) repeated at a constant frequency and rate.
(Kirschel et al. 2009, 2020b, Nwankwo et al. 2018, Sebastianelli et al. 2021), and are innate (Lukhele et al. unpubl.). The songs of *P. chrysoconus* and *P. pusillus* are the simplest in the genus, with the pulses repeated continuously without separation into discrete songs or bouts (Monadjem et al. 1994). They can thus be characterised simply by their pace and pitch. We analysed peak frequency and the temporal measurement of inter-onset interval (IOI), a measure of pace, calculated as the time between the onset of two consecutive ‘pulses’, from 386 recordings of *P. chrysoconus* and 187 *P. pusillus*, collated for a parallel study (Kirschel et al. unpubl.). To detect the large number of tinkerbird ‘pulses’ present in each recording (typically 100+), we used Raven Pro (K. Lisa Yang Center for Conservation Bioacoustics 2019) to detect tinkerbird ‘pulses’ automatically using the built-in band-limited energy detector (BLED) and extracted separately high-resolution spectral (DFT size = 65,536) and temporal measurements (DFT size = 512). The BLED detects sound signals when their amplitude in a specified band of frequency exceeds a specific signal-to-noise ratio against background noise (in dB). However, given the non-selectivity of the BLED, both

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Figure 2. Spectrograms showing song structure of Yellow-fronted Tinkerbird *Pogoniulus chrysoconus* and the four subspecies of Red-fronted Tinkerbird *P. pusillus*. 

![Spectrograms showing song structure of Yellow-fronted Tinkerbird *Pogoniulus chrysoconus* and the four subspecies of Red-fronted Tinkerbird *P. pusillus*.](image-url)
natural and anthropogenic sounds with frequency similar to tinkerbird ‘pulses’ might be detected erroneously or might lead to modification of the time duration of the signal detected, if overlapping with tinkerbird calls. Therefore, signal detection results were visually inspected for false positives and modified where necessary to match the actual start and end times of ‘pulses’. We then used Cohen’s $d$ statistic to quantify the magnitude of differences between first the two species, then between the 40 recordings of $P.\ p.\ pusillus$ and 147
recordings of East African populations (35 \textit{P. p. affinis}, 87 presumed \textit{P. p. eupterus} and 25 \textit{P. p. uropygialis}), and lastly between the three East African populations corresponding to the clades identified by phylogenetic reconstruction.

\textbf{Quantitative analysis.}—Cohen’s \(d\) statistics were calculated in R 4.0.1 (R Core Team 2020) using the ‘effsize’ (v.0.8.1) package (Torchiano 2016). The values were then converted into Tobias et al. (2010) levels of magnitude and plumage difference were scored according to the scales they suggest therein (Table 4). Ecology and behaviour, and geographical relationships, were also scored using Tobias et al. (2010) guidelines, based on differences evident from our field work but also described in field guides. We also compared morphology between groups using Principal Components Analysis (PCA), based on the correlation matrix of the six morphometric measurements using R 4.0.1 (R Core Team 2020), which allowed us to reduce the dimensionality of our dataset, as well as to discriminate the different clades in two-dimensional space. Ellipses (based on 95% confidence intervals) were drawn around the different clades identified in the phylogenetic reconstruction.

\textbf{Plumage coloration differences.}—These were scored qualitatively by ANGK based on experience from measuring the specimens, live birds in the field, and from quantitative analyses of all specimens as part of published (Nwankwo et al. 2019, Kirschel et al. 2020) and unpublished work.

\section*{Results}

\textbf{Maximum likelihood and Bayesian analyses.}—We resolved a concordant phylogeny using Bayesian and maximum likelihood approaches (Fig. 3). The \textit{P. pusillus} clade was nested within \textit{P. chrysoconus}, and southern populations of \textit{P. p. pusillus} were sister to northern populations. There were three distinct clades within East Africa, with ‘\textit{P. p. eupterus}’ sister to a clade including \textit{P. p. affinis} and \textit{P. p. uropygialis}. Mean pairwise distances between these three populations were of the same magnitude (1.0–1.6%, see Table 5) as between established Yellow-fronted Tinkerbird subspecies, \textit{P. c. chrysoconus} and \textit{P. c. xanthostictus}.

\textbf{Phenotypic differences.}—All morphometric comparisons between the sexes resulted in ‘small’ or ‘negligible’ Cohen’s \(d\) statistics (range 0.11 < \(d\) < 0.38) according to the ‘effsize’ package (Torchiano 2016), suggesting these species are sexually monomorphic, so all further tests were run on the entire specimen set with sexes pooled, although we provide means and standard deviations per sex (Table 3). We found greater differences in morphometrics and song between the larger and faster singing \textit{P. p. pusillus} and East African populations with smaller body size and slower songs, than between \textit{P. pusillus} and \textit{P. chrysoconus}, but differences between East African Red-fronted Tinkerbird populations were more subtle (Table 6; see also Fig. 4 for differences in IOI). Between \textit{P. pusillus} and \textit{P. chrysoconus} we

\begin{table}
\caption{Uncorrected mean pairwise genetic distances among populations of Red-fronted Tinkerbird \textit{Pogoniulus pusillus}, with pairwise distance from sample J31301 \textit{P. chrysoconus} (Yellow-fronted Tinkerbird) included as a reference. Values in lowest diagonal indicate intra-taxon variation (not estimated in \textit{chrysoconus}, represented here by a single individual).}
\centering
\begin{tabular}{lrrrrr}
 & \textit{chrysoconus} & \textit{pusillus} & \textit{uropygialis} & \textit{affinis} & \textit{eupterus} \\
\textit{chrysoconus} & - & 6.5\% & 6.2\% & 6.2\% & 6.2\% \\
\textit{pusillus} & - & - & 4.3\% & 4.4\% & 4.4\% \\
\textit{uropygialis} & - & - & - & 1.0\% & 1.5\% \\
\textit{affinis} & - & - & - & - & 1.6\% \\
\textit{eupterus} & - & - & - & - & 0.4\%
\end{tabular}
\end{table}
estimated a Tobias et al. (2010) score of 8, based on low effect sizes for morphometric (1) and song differences (1), 3 for plumage, based on the distinct forehead colour difference (Nwankwo et al. 2019, Kirschel et al. 2020a), 1 for ecology, based on differences in their preferred habitats, savanna woodland (P. chrysoconus) vs. forest (P. pusillus) (Short &

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**TABLE 6**

Cohen’s $d$ statistics for morphometric (wing chord, tarsus, tail, and exposed length of maxilla, mandible length, and bill width) and song characters (peak frequency and inter-onset interval = IOI) between populations of Red-fronted Tinkerbird Pogoniulus pusillus.

| Population 1          | Population 2          | Wing Length | Tarsus Length | Tail Length | Bill Length | Mandible Length | Bill Width | Peak Frequency | IOI |
|-----------------------|-----------------------|-------------|---------------|-------------|-------------|-----------------|------------|----------------|-----|
| P. pusillus nominate  | P. chrysoconus        | 1.02        | -0.12         | 0.09        | 0.47        | 0.43            | 0.87       | -0.23          | 1.34|
| pusillus              | uropygialis           | -1.92       | -3.81         | -2.75       | -1.38       | -1.05           | -0.45      | 1.25           | 2.1 |
| affinis               | uropygialis           | -2.59       | -1.37         | -1.76       | -1.22       | -1.08           | -0.2       | 0.42           | -0.85|
| eupterus              | uropygialis           | -1.9        | -1.2          | -1.0        | -1.13       | -1.16           | -0.44      | 0.27           | -1.14|
| affinis               | eupterus              | -0.54       | -0.11         | -0.65       | -0.08       | 0.11            | 0.24       | 0.15           | 0.26|

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Horne 2001) and 2 for their narrow hybrid zone (Nwankwo et al. 2019). The score for (southern) *P. p. pusillus* vs. East African (northern) populations was also 8, based on greater morphometric and song differences than between the recognised species (both scored 2); a plumage score of 3, based on marked facial differences in throat and eyebrow colour (white in the north, yellow in the south) and the golden-yellow wing patch in the south vs. yellow wingbar in the north (Fig. 5); and a score of 1 for ecology, with southern Red-fronted Tinkerbird primarily a tall-forest bird, whereas northern populations mostly are found in acacia woodland (Short & Horne 2001). The disjunct distribution of northern and southern Red-fronted Tinkerbirds means that no score is given for geographical relationship.
Within northern populations, scores were more difficult to assess because geographical relationships are not clear. We scored 5 between *P. p. affinis* vs. *P. p. uropygialis*, based mostly on wing length differences, and the latter’s yellower dorsal streaking (Fig. 5), but whose geographical relationship we expect involves either a broad (1 point) or narrow (2 points) hybrid zone in the Great Rift Valley; 4 for ‘*P. p. eupterus*’ vs. *P. p. uropygialis*, which again differ in wing length and dorsal streak colour, and we believe are allopatric; and 2–3 for *P. p. affinis* vs. ‘*P. p. eupterus*’, which we expect are either parapatric (3 points) or form a narrow hybrid zone (2 points), but with little or no differences in phenotypic traits (all Cohen’s $d < 1$). The first two principal components extracted explained 79.6% of morphometric variation (Table 7), and PCA supported the results of the Cohen’s $d$ analysis, with much overlap between the two species, but with *P. p. pusillus* almost completely distinct from all northern Red-fronted Tinkerbirds (Fig. 6).

### Figure 6

PCA plot of the first two principal components from morphometrics, explaining 79.6% of the variation. Ellipses are based on 95% CI. Yellow-fronted Tinkerbird *Pogoniulus chrysoconus* broadly overlaps with the various taxa of Red-fronted Tinkerbird (listed in the legend according to clades identified in the study), but there is almost no overlap between *pusillus* and other forms of Red-fronted Tinkerbird.

### Table 7

| Clade            | PC1  | PC2  |
|------------------|------|------|
| *affinis*        |      |      |
| *chrysoconus*    |      |      |
| *eupterus*       |      |      |
| *pusillus*       |      |      |
| *uropygialis*    |      |      |

Factor loadings, eigenvalues and proportions of variance explained by the first two principal components extracted using PCA from morphometrics of study skins of Red-fronted Tinkerbird *Pogoniulus pusillus*.

| Measure          | PC1  | PC2  |
|------------------|------|------|
| Wing             | 0.458| 0.284|
| Tarsus           | 0.444| 0.350|
| Tail             | 0.452| 0.436|
| Exposed          | 0.445| -0.528|
| Bill (lower)     | 0.434| -0.571|
| Eigenvalue       | 1.76 | 0.92 |
| Proportion of variance | 0.623 | 0.175 |
Discussion

*P. p. pusillus* is highly divergent from East African populations of Red-fronted Tinkerbird. Although our analysis of mtDNA suggests the *pusillus* clade is monophyletic, consistent with the findings in Nwankwo *et al.* (2019), by contrast, preliminary evidence from ongoing work (ANGK unpubl.) based on whole genomes is discordant with the topology found here and suggests it is polyphyletic. Indeed, whole genome data support a sister relationship between *P. p. pusillus* and *P. c. extoni*, and a sister relationship between a northern clade of *P. chrysoconus*, comprising *P. c. chrysoconus* and *P. c. xanthostictus*, and the northern clade of *P. pusillus*, rendering both *P. pusillus* and *P. chrysoconus* polyphyletic. If *P. p. pusillus* is not even sister to East African populations of Red-fronted Tinkerbird, this further supports our findings here demonstrating their genetic distance in mtDNA, and their phenotypic divergence. Using Tobias *et al.* (2010) criteria, we estimate a score of 8, the same as the difference assessed here between the presently recognised species *P. chrysoconus* and *P. pusillus*, and exceeding the Tobias *et al.* (2010) species-level threshold score of 7. Morphological differences, such as in the greater tarsal length of *P. p. pusillus*, may correspond to expectations under Bergmann’s Rule, but with no intermediate Red-fronted Tinkerbird populations occurring between their ranges, we suspect these size differences are biologically significant. Likewise, songs differ in pace between disjunct populations of Red-fronted Tinkerbird more than they do between Red-fronted and Yellow-fronted Tinkerbirds, with *P. p. pusillus* song much faster than those of *affinis*, *eupterus* and *uropygialis* in East Africa, and such differences might serve to differentiate these populations. We thus propose that Red-fronted Tinkerbird is split into two species, Southern Red-fronted Tinkerbird *Pogoniulus pusillus* (Dumont, 1816), and Northern Red-fronted Tinkerbird *P. uropygialis* (von Heuglin, 1862), including subspecies *P. u. affinis* (Reichenow, 1879) and *P. u. eupterus* Grote, 1928.

East African populations that span the range of *eupterus*, described by Grote (1928) from Ukerewe Island, Tanzania, as an inland race encompassing the Kilimanjaro area (Grote 1939), form a distinct group that is genetically more distant from *affinis* and *uropygialis* (c.1.5–1.6% sequence divergence) than the latter taxa are from each other (1%). Pairwise distance between *eupterus* and the other two is comparable to that between *P. c. chrysoconus* and *P. c. xanthostictus* (1.6%). Therefore, on the basis of equivalence and parsimony, *eupterus* should be resurrected as *P. u. eupterus*. We believe this status is merited based on the genetic divergence between *eupterus* and the clade including *affinis* and *uropygialis*. We appreciate, however, that based on our phenotypic yardstick analysis, morphometrics, plumage and songs differ less between *affinis* and *eupterus*, whereas *uropygialis* is more distinctive. Our analysis of morphometric data does not support Grote’s (1928) suggestion that *eupterus* and *affinis* differ in wing length.

It should be noted that the distribution of *eupterus* as understood here differs from the initial description by Grote (1928). Although this taxon occurs inland as far west as northwestern Uganda in the north, and to Mbeya in the south, we infer from data presented here and in parallel work (ANGK unpubl.) that it also extends east across much of Kenya and Tanzania to the Indian Ocean coast, from the south side of Kilifi Creek, Kenya, south to the Rufiji River, Tanzania. *P. u. affinis* occurs from north of Kilifi Creek to the type locality at Kipini, near the Tana River, and to Serenli along the Juba River in southern Somalia, from where *lollesheid* (van Someren, 1932) was described (Fig. 1). The distribution of *affinis* extends inland from Somalia to southern Ethiopia, to the edge of Harena forest on the south slope of the Bale Mountains, where its preferred habitat of acacia woodland meets montane forest. We were unable to sample along the southern Rift in Ethiopia for logistical reasons.
but samples of Red-fronted Tinkerbird from Wondo Genet, Bishangari and north across the Rift to the east side of Lake Tana and Lalibela were all *uropygialis*, which extends north into Eritrea and south-east along the Red Sea and Gulf of Aden coasts to northern Somalia.

The relative distributions of the three northern taxa of Red-fronted Tinkerbirds suggest that the Great Rift Valley may have played a role in their divergence. On the other hand, genetic divergence either side of Kilifi Creek is unexpected, and studies are ongoing to determine if there is gene flow between these forms (ANGK unpubl.). Tinkerbirds in this complex have been shown to hybridise, with *P. p. pusillus* and *P. c. extoni* interbreeding freely in their 50–100 km-wide contact zone in southern Africa (Nwankwo et al. 2019, Kirschel et al. 2020a), and introgressive hybridisation between the more closely related and phenotypically similar *eupterus* and *affinis* would not be unexpected if they do indeed come into contact.

Taxonomists are in much disagreement regarding how to define species limits, for example regarding the applicability of the Tobias et al. (2010) criteria (Rheindt & Ng 2021). Indeed, previous work on *Pogoniulus* tinkerbirds provides a relevant example in the case of ‘White-chested Tinkerbird *P. makawai’*. Considered a species by some based on its morphological distinctiveness (Collar & Fishpool 2006, del Hoyo & Collar 2014), but according to others it is an aberrantly plumaged Yellow-rumped Tinkerbird *P. bilineatus* (Dowsett & Dowsett-Lemaire 1980, 1993), a viewpoint supported by phylogenetic analysis (Kirschel et al. 2018), which demonstrated that it is nested within *P. b. mfumbiri*. In the present study, both a molecular phylogeny and phenotypic differences support species-level status for northern and southern forms of Red-fronted Tinkerbird according to the Phylogenetic and Evolutionary Species Concepts, and potentially even the Biological Species Concept, based on the phenotypic differences identified. Geographically parapatric cryptic taxa are more difficult to differentiate, yet such phylogenetic conservatism can result from vicariance and subsequent secondary contact, and we believe the genetic distance and even non-sister relationship of *eupterus* and *affinis* is sufficient to separate them subspecifically. The phylogeny also suggests Yellow-fronted Tinkerbird is paraphyletic with respect to the entire Red-fronted Tinkerbird clade, or polyphyletic according to unpublished work with whole genomes, meaning further research will be needed to elucidate species limits therein.

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