Species limits in Northern Eurasian taxa of the common stonechats, *Saxicola torquatus* complex (Aves: Passeriformes, Muscicapidae)

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Abstract

The common stonechat *Saxicola torquatus* traditionally was considered as a polytypic species widely distributed in Africa, Europe and Asia. Recently, several authors have suggested that this formerly single species needs to be split into several distinct species composing the *Saxicola torquatus* complex based on mitochondrial markers. However, mitochondrial DNA alone is not sufficient for the evaluation of species status. In this paper, we reviewed the taxonomy of Northern Eurasian taxa from the complex based on morphometrics, plumage, song and alarm calls. The morphological and vocal data clearly matched the phylogroups reconstructed from mitochondrial DNA sequences, and separated Northern Eurasian taxa of the *Saxicola torquatus* complex into three groups: *rubicola*, *maurus* and *stejnegeri*. We proposed the species status for these three groups: European stonechat *Saxicola rubicola*, eastern stonechat *Saxicola maurus* and Japanese stonechat *Saxicola stejnegeri*. Among them, *S. stejnegeri* is a cryptic species as it cannot be distinguished by morphometrics and by worn spring plumage from *S. maurus*, but differs noticeably by male song.

Key words

Common stonechat, *Saxicola torquatus* complex, geographic variation, cryptic species, song divergence.

Introduction

In the last decades, it was revealed that many bird species actually are not a single species but rather complexes of several species, which are similar in morphology, but distinct in behavior, ecology, physiology etc (Bickford et al., 2006; Pfenninger & Schwenk, 2007). It is especially true for South Asia thanks to many recent advances in the taxonomy of Sino-Himalayan and Southeast Asian birds (Johansson et al., 2007; Martens et al., 2011; Päckert et al., 2012; Alström et al., 2013).

At same time, the revaluation of cryptic species in Northern Asia had received insufficient attention. Several recent studies however reveal large genetic and/or biocoustic and/or morphological differentiation between western and eastern population groups of a species across Northern Eurasia. In several cases it was found that a taxon that was treated as a single species with discontinuous distribution in Northern Asia would in fact better be classified as two distinct species: white stork *Ciconia ciconia* – oriental stork *C. boyciana* (Archibald & Schmitt, 1991; Pavlova & Panov, 2005), and azure-winged magpie (*Cyanopica cyanus*) – Iberian magpie (*C. cooki*) (Fok et al., 2002; Kryukov et al., 2004; Zhang et al., 2012). In several other examples newly evaluated species lack an apparent distributional gap: red-breasted flycatcher *Ficedula parva* – taiga flycatcher *F. albicilla* (Svensson et al., 2005), western marsh harrier *Circus aeruginosus* –
eastern marsh harrier *C. spilonotus* (FEFLOV, 2001), great grey shrike *Lanius excubitor* – northern grey shrike (*Lanius borealis*) (OLSSON et al., 2010; TÁKÓVA & RED’KIN, 2014), common buzzard *Buteo buteo* – eastern buzzard *B. japonicus* (KRUCKENHAUSER et al., 2004), great tit *Parus major* – Japanese tit *Parus minor* (KVIST et al., 2003; PACKERT et al., 2005), western yellow wagtail *Motacilla flava* – eastern yellow wagtail *M. tschutschensis* (PAVLONA et al., 2003), and arctic warbler *Ph. borealis* – Kamchatka leaf warbler *Ph. eximianus* (SAITOH et al., 2010; ALSTRÖM et al., 2011; but the taxonomy is still debated: RED’KIN, 2013; RED’KIN et al., 2016).

More or less deep east-west divergence in Northern Eurasia was also found between different subspecies of a species. Examples include marsh tit *Poecile palustris* (TRITSCHE et al., 2017), coal tit *Periparus ater* (PENTZOLD et al., 2013), goldcrest *Regulus regulus* (PACKERT et al., 2003), Eurasian nuthatch *Sitta europaea* (ZINK et al., 2006), barn swallow (*Hirundo rustica*) (SCORDATO & SAFRAN, 2014), black-tailed godwit *Limosa limosa* (HÖGLUND et al., 2009), great spotted woodpecker (ZINK et al., 2002), winter wren *Troglodytes troglodytes* (DROVETSKI et al., 2004), and several corvids (HÅRING et al., 2007). The willow tit *Poecile montanus* differs from the aforementioned examples because it does not display a continental east-west split (PAVLONA et al., 2006; TRITSCHE et al., 2017).

The most plausible interpretations for east–west divergence within a species and/or a species group in Northern Eurasia would be a wide distribution before the Pleistocene followed by the geographic isolation because of climatic changes during the ice age (HEWITT, 2000, 2004; SCHMIDT, 2007). The exact evolutionary scenario could differ across species and species complexes (ZINK et al., 2008). Other theories, e.g. emergence of east-west divides by speciation from a spatially in a circular overlap (IRWIN et al., 2005) have received cautious but steady criticism (PACKERT et al., 2005; MARTENS & PACKERT, 2007; KOVYLOV et al., 2012; ALCAIDE et al., 2014).

The majority of the aforementioned studies analyzed mitochondrial DNA (mtDNA) and/or though rarely nuclear DNA. The taxonomic implications from mitochondrial DNA studies are somewhat limited, because several authors have concluded that mtDNA alone is not sufficient for the evaluation of species status (EDWARDS et al., 2005; TOBIAS et al., 2010). The species diversity of Northern Eurasian birds could be thus underestimated. The plausible solution is to use integrative taxonomy that takes into account also nuclear genetic markers, morphology, vocalization, ecology and behavior (PADIAL et al., 2010). Integrative taxonomy is especially helpful in the evaluation of cryptic species. Studies on the magpie *Pica pica* for example, have shown a clear east-west divergence in the Northern Palearctic based on DNA analysis (KRYUKOV et al., 2004; ZHANG et al., 2012; SONG et al., 2018). It was also found that bioacoustic data based on chatter call reflect DNA lineages. Taking together, these results allowed splitting Northern Eurasian magpies into the two taxa: *Pica pica* and *P. serica* (KRYUKOV et al., 2017).

The common stonechat *Saxicola torquatus* traditionally was considered as a single species with many subspecies widely distributed in Europe, Asia and Africa (ROBERTSON, 1977; CRAMP, 1988; ECK, 1996). In a view of some recent taxonomic advances, based on mtDNA sequence information this formerly single species needs to be split into several distinct species composing the *Saxicola torquatus* complex (WITTMAN et al., 1995; WINK et al., 2002; ILLERA et al., 2008; ZINK et al., 2009). However, as was mentioned above, mitochondrial markers alone are not sufficient for the evaluation of species status. Therefore, the taxonomy is still debated (URQUHART, 2002; CLEMENT & ROSE, 2015; DEL HOYO & COLLAR, 2016).

Moreover, two currently accepted species (DEL HOYO & COLLAR, 2016) were nested within the *Saxicola torquatus* complex in mitochondrial DNA study(-ies): Fuerteventura stonechat *S. dacotiae* (ILLERA et al., 2008; ZINK et al., 2009) from the Canary Islands, and Reunion stonechat *S. tectes* from Reunion Island (ZINK et al., 2009).

Seven taxa from the *Saxicola torquatus* complex breed in Eurasia (Fig. 1): *rubicola* and *hibernans* in Europe, *stejnegeri* in the eastern Palearctic, *maurus* in the central Palearctic, *variegatus* and *armenicus* in the Caucasus and adjacent areas, and *indicus* and *przewalskii* in the Himalayas and adjacent China. Other names (e.g. *hempichii*) should be in synonymy. In our opinion, the name “*S. m. hempichii* EIHENBERG, 1833” was mistakenly applied by SVENSSON et al., (2012) to the west Caspian populations. Actually, *hempichii* is in the synonymy of *variegatus*, the latter having priority. The reason is as follows. According to SVENSSON et al., (2012), a large mountain subspecies was firstly described under the name “*Parus variegatus* S. G. GMELIN, 1774” (terra typica – Shamakhi, east of Azerbaijan). Later, according to SVENSSON et al., (2012), this large subspecies was renamed as “*Saxicola torquatus armenica* STEGMANN, 1935”. If the point of view of SVENSSON et al., (2012) was correct, West Caspian populations had to be named *S. m. hemprichii*, and populations from the mountains of Iran, Turkey and the Transcaucasia had to be named *S. m. variegatus*. However, the point of view of SVENSSON et al., (2012) based (1) on the mistake of STEGMANN (1935) who incorrectly placed the breeding specimen from Shamakhi in “*armenica*”, and (2) on the incorrect interpretation of the characters of “*Parus variegatus* S. G. GMELIN, 1774”, represented on the image in the original description. Actually, (1) the breeding area of “*armenica*” does not reaches East Transcaucasia including Shamakhi and (2) the coloration of specimen depicted in the original description of “*variegatus*” clearly fitted the variability limits of West Caspian populations (own data). That is why we place *S. m. hemprichii* in the synonymy of *S. m. variegatus*. Besides, the Sicilian population was given the name archimedes (CLANCHEY, 1949). However, it remains questionable, whether the Sicilian birds are confirmed as a separate race, or archimedes should be in synonymy with *rubicola* (CORSO, 2001).

Recently it was found that *rubicola* (including *hibernans*), *maurus* (including *variegatus*) and *stejnegeri*
could be classified as separate species based on the variation of mitochondrial ND2 gene sequences. It was also shown that stejnegeri, although very similar in appearance, had separated from rubicola-maurus well before differentiation occurred among the latter two taxa (Zink et al., 2009). However, in the study by Zink et al., (2009), haplotypes from the central Siberian clade (maurus) were found in a sample from Rostov-na-Donu area, just east of the Sea of Azov, in Russian Federation, and haplotypes from the east Siberian clade (stejnegeri) in a sample from Astrakhan, west of the Caspian Sea. Both findings may suggest that any firm conclusions about the phylogenetic relationships of maurus and stejnegeri are premature.

In this paper we analyzed morphometrics, plumage, song and alarm calls from throughout the range of three Northern Eurasian taxa of the Saxicola torquatus complex: rubicola, maurus and stejnegeri. We proposed that S. rubicola, S. maurus and S. stejnegeri could be recognized as separate species. We also shortly discussed the phylogenetic relationships of these taxa.

Materials and Methods

Field work

The two taxa (maurus and stejnegeri) were studied in the field (Fig. 1, Table S2–S3). Observations and sound recordings were carried out in the following locations: (1) stejnegeri: near Khasan, Primorsky Kray, Russian Federation in June 2016 by E.K., (2) stejnegeri: in Khingan State Nature Reserve, Amur Oblast, Russian Federation in May–June 2013 by A.O. and M.G., (3) stejnegeri: in Ivan-Arachleisk Nature Park, Chita Oblast, Russian Federation in June–July 2015 by E.K., and (4) maurus: near Irkutsk, Irkutsk Oblast, Russian Federation in May–June 2015 by E.K.

Morphology

We measured and described the plumage of adult males and females of most specimens of three taxa of the Saxicola torquatus complex in several Russian collections (listed in Table S1). We described plumage under daylight condition using Naturalist’s Color Guide (Smith, 1975). In the morphometry analysis, we used male specimens only. We measured the specimens of rubicola (n=14), maurus (n=73) and stejnegeri (n=63) from throughout their ranges (Table S1). Measurements taken and used in the analysis were (in mm): (1) wing length (flattened and stretched), (2) tail length (measured from the base of central rectrices), (3) tarsus length, (4) bill length from skull to distal nares, (5) maximum bill depth, (6) bill width from bill base, (7) distance between wing tip and the tip of 2nd primaries, and (8) length of primaries projection.

Song

Recordings of 57 individuals of rubicola (n=10), maurus (n=20) and stejnegeri (n=27), comprising 567 song strophes, were analyzed (Table S2). Most of the recordings were made by the authors (A.O. and E.K.). Additionally, we used song and calls (see below) recordings either taken from B. N. Veprintsev’s collection of animal voices (Severtsov Institute of Ecology and Evolution of
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Russian Academy of Sciences, Moscow, Russian Federation or downloaded from Xeno-canto (www.xeno-canto.org). Besides, Alexander Rubtsov (Darwin State Museum, Moscow, Russian Federation) kindly provided recordings of two *maurus* males. Sonograms were produced and analyzed using Syrinx PC v. 2.6 (John Burt, www.syrinxpc.com) with an FFT size = 256, and a window type = Hanning.

Territorial song of each stonechat male consists of song strophes. Each song strophe consists of a number of syllables (Fig. 1, a). We define a syllable as either continuous line on a sonogram (i.e. element) or a combination of two or more elements separated by the pauses less than 10 ms or by a rapid alternation in frequency. Two or more elements in a syllable could be either different or similar. We thus introduced the term ‘trill’ that is the syllable consisted of several identical elements (Fig. 1, a). We chose 10 ms cut-off point because the distribution of pauses between elements was clearly bimodal, with most pauses either less than 10 ms (i.e. intra-syllable pauses) or more than 10 ms (between-syllable pauses) (Fig. S1, b).

In most cases we analyzed 10 consecutive song strophes per individual (mean 9.9).

1. Duration of strophe,
2. number of syllables,
3. number of syllable types,
4. median syllable length,
5. median between-syllable pause,
6. minimum frequency,
7. maximum frequency,
8. median frequency range.

We chose 10 ms cut-off point because the distribution of pauses between elements was clearly bimodal, with most pauses either less than 10 ms (i.e. intra-syllable pauses) or more than 10 ms (between-syllable pauses) (Fig. S1, b).

### Alarm call

All species on the approach of nest predators give two types of alarm calls, whit and chack (Johnsong, 1971; Greig-Smith, 1980). They are commonly given in mixed sequences, although both occur singly. Using one-way ANOVA we found that males and female did not differ in time and frequency parameters of whits and chacks in the two taxa for which we had enough data (*rubicola*: $F = 3.03, p = 0.07$, 9 males and 6 females; *stejnegeri*: $F = 2.38, p = 0.08$, 10 males and 11 females). These findings were in accordance with the fact, that in bird species that have particular calls used by both sexes, there are minimal or no sex differences in those shared calls (Vicario et al., 2001). Therefore, in the analysis the data were pooled across both sexes, and individuals of unknown sex were added.

For the alarm call analysis we used recordings of 53 individuals, belonging to three taxa of the *Saxicola torquatus* complex: *rubicola* ($n = 16$), *maurus* ($n = 15$) and *stejnegeri* ($n = 22$) (Table S3). All recordings had both whits and chacks. Usually we analyzed 10 whits and 10 chacks per individual (mean 9.9, for both alarm call types).

1. Duration,
2. minimum frequency,
3. maximum frequency and
4. frequency range were measured for each whit call. We measured duration only for each chack call. Median values of measurements of both types of alarm calls of an individual were calculated and used in the analysis.

### Statistical analysis

The analysis was performed in Statsoft Statistica 6.0 (StatSoft Inc [Internet] 2001) and in R 3.3.2 (R Core Team, 2016). We analyzed morphometrics, song and two types of alarm calls separately.

Firstly, we used linear discriminant analysis (LDA) in the ‘MASS’ package in R (Ripley et al., 2017). LDA classify a given sample of predictors with highest posterior probability using Bayes’ rule and tries to find a linear combination of predictors that gives maximum separation between the centers of the data. We then ran a principal component analysis (PCA) in Statistica 6.0. Bonferroni-adjusted two-sample Student t-tests were used to test differences between groups.

### Results

#### Morphometrics

With respect to the morphometrics, the LDA of *rubicola*, *maurus*, and *stejnegeri* males resulted in 90.0% correct classification of these three groups (Wilks’ $\lambda = 0.108$, $F = 13.44, n = 150$ males). The majority (11 out of 15) of misclassification cases were *maurus* males that were predicted to belong to *stejnegeri* (5 males), and vice versa (6 males).

The first three principal components had eigenvalues greater than one and PC1 explained 30.0% of the total variation. PC2 explained a cumulative 50.0% and PC3 a cumulative 63.8% of the total variation. Parameters of body size showed the highest absolute correlation with Factor 1: wing length, tail length and distance between wing tip and the tip of 2nd primaries. By contrast, bill size showed the strongest correlation with Factor 2: bill width and bill height. PC1 vs. PC2 showed individual measurements clustering according to the taxa (Fig. 2, a).

Among the three species, *rubicola* is characterized by comparatively short wing and tail, and had thus comparatively small body size (Table 1). The bill of *rubicola* is somewhat longer than that of both *maurus* and *stejnegeri*. By contrast, *stejnegeri* had relatively wide and deep bill.

It seemed impossible, however, to distinguish between these three taxa based on morphometrics only, because the measurements overlapped broadly (Table 1).

#### Plumage

The three taxa from the *Saxicola torquatus* complex could be distinguished based on the color of the fringes of the

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feathers of upperparts in fresh autumn plumage (Fig. 3), and on the amount of white on the sides of the neck, on the inner upperwing and on the rump in worn spring plumage.

In worn spring plumage the male of \textit{rubicola} is distinguishable from the male of the other taxa by its comparatively small patches of white on the sides of the neck, on the inner upperwing and on the rump. There are prominent darkish centers of the feathers on the upper tail coverts, which appear as dark longitudinal streaks. The rectrices are completely black showing no white at their base. The adult male in fresh autumn plumage is somewhat duller than in spring (fringes of the feathers of the upperparts – Mikado Brown C. 121C, according to \textit{Smithie}, 1975) and has chestnut (Raw Sienna C. 136) fringes of the uppertail coverts. The female of \textit{rubicola} is readily distinguishable by its darkish olive-brown upperparts and head, both having broad black stripes. It is due to broad white tips on the underwing coverts, that both sexes have whitish underwing.

The male of \textit{maurus} in worn spring plumage is distinguishable by its broad patches of white on the sides of the neck, on the inner upperwing and on the rump. The rectrices have a white base in the majority of \textit{maurus} individuals. The underwing is blackish and much darker than that of \textit{rubicola}. In fresh autumn plumage the male of \textit{maurus} has ochreous fringes (Ground Cinnamon C. 239) of the feathers of the upperparts. The feathers of the rump

Table 1. Univariate statistics for measurements of the males of three taxa of the \textit{Saxicola torquata} complex. The values given are means ± SE, and min – max given in parentheses. Significance levels (Student t-test): **\( p < 0.006 \) (Bonferroni-adjusted \( p \) value); *\( p < 0.01 \); all others not significant.

<table>
<thead>
<tr>
<th></th>
<th>( S. \textit{r. rubicola} ) ((n=14))</th>
<th>( S. \textit{m. maurus} ) ((n=73))</th>
<th>( S. \textit{(m.) stejnegeri} ) ((n=63))</th>
<th>( S. \textit{r. rubicola} ) versus ( S. \textit{m. maurus} )</th>
<th>( S. \textit{r. rubicola} ) versus ( S. \textit{(m.) stejnegeri} )</th>
<th>( S. \textit{m. maurus} ) versus ( S. \textit{(m.) stejnegeri} )</th>
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<tbody>
<tr>
<td>Wing length (mm)</td>
<td>65.3 ± 1.2 ((59.0 – 68.0))</td>
<td>68.9 ± 1.7 ((63.5 – 73.0))</td>
<td>68.0 ± 1.8 ((63.0 – 72.0))</td>
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<tr>
<td>Tail length (mm)</td>
<td>45.5 ± 1.5 ((42.5 – 48.0))</td>
<td>47.5 ± 2.1 ((43.0 – 52.9))</td>
<td>48.0 ± 2.0 ((42.0 – 52.7))</td>
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<tr>
<td>Tarsus length (mm)</td>
<td>21.7 ± 0.7 ((20.5 – 23.2))</td>
<td>20.9 ± 0.7 ((18.5 – 23.0))</td>
<td>21.0 ± 0.7 ((19.0 – 22.6))</td>
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<tr>
<td>Bill length (mm)</td>
<td>7.9 ± 0.3 ((7.4 – 8.7))</td>
<td>7.4 ± 0.4 ((6.4 – 8.4))</td>
<td>7.6 ± 0.4 ((6.4 – 8.6))</td>
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<tr>
<td>Bill depth (mm)</td>
<td>3.6 ± 0.2 ((3.3 – 4.3))</td>
<td>3.7 ± 0.2 ((3.2 – 4.4))</td>
<td>3.9 ± 0.2 ((3.3 – 4.4))</td>
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<tr>
<td>Bill width (mm)</td>
<td>6.7 ± 0.4 ((5.7 – 7.3))</td>
<td>6.5 ± 0.3 ((5.5 – 7.2))</td>
<td>7.2 ± 0.3 ((6.2 – 8.0))</td>
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<tr>
<td>Distance between wing tip and 2nd primaries (mm)</td>
<td>14.6 ± 1.3 ((12.3 – 17.5))</td>
<td>15.5 ± 1.2 ((12.1 – 18.7))</td>
<td>15.5 ± 1.3 ((11.7 – 18.1))</td>
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<tr>
<td>Primaries projection (mm)</td>
<td>5.4 ± 0.7 ((3.7 – 6.5))</td>
<td>6.0 ± 1.1 ((3.3 – 11.0))</td>
<td>5.4 ± 1.0 ((2.8 – 8.3))</td>
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Fig. 2. PCA of eight external measurements of whole skin specimens (a) and PCA of eight acoustic parameters of song strophes (b) of males of three taxa of the \textit{Saxicola torquatus} complex.
Fig. 3. Dorsal and lateral views of males of *rubicola*, *maurus* and *stejnegeri* in worn spring plumage (*a*, *b*) and fresh autumn plumage (*c*).
Table 2. Univariate statistics for measurements of the song strophes of the males of three taxa of the Saxicola torquata complex. The values given are means ± SE, and min – max given in parentheses. Significance levels (Student t-test): **p < 0.006 (Bonferroni-adjusted p value); *p < 0.01; all others not significant.

<table>
<thead>
<tr>
<th></th>
<th>S. r. rubicola (n=10)</th>
<th>S. m. maurus (n=20)</th>
<th>S. (m.) stejnegeri (n=27)</th>
<th>S. r. rubicola versus S. m. maurus</th>
<th>S. r. rubicola versus S. (m.) stejnegeri</th>
<th>S. m. maurus versus S. (m.) stejnegeri</th>
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<tr>
<td>Duration of strophe (s)</td>
<td>1.36 ± 0.10 (1.23 – 1.53)</td>
<td>1.34 ± 0.25 (1.13 – 2.11)</td>
<td>1.01 ± 0.21 (0.73 – 1.49)</td>
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<tr>
<td>Number of syllables</td>
<td>13.0 ± 2.6 (8.5 – 16.5)</td>
<td>15.8 ± 3.7 (11.0 – 26.0)</td>
<td>10.5 ± 2.2 (7.0 – 15.0)</td>
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<tr>
<td>Number of syllable types</td>
<td>12.1 ± 2.3 (8.5 – 15.0)</td>
<td>13.8 ± 2.8 (10.5 – 22.5)</td>
<td>9.8 ± 2.1 (6.0 – 14.5)</td>
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<tr>
<td>Median syllable length (s)</td>
<td>0.07 ± 0.02 (0.05 – 0.10)</td>
<td>0.05 ± 0.01 (0.04 – 0.07)</td>
<td>0.07 ± 0.01 (0.05 – 0.10)</td>
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<tr>
<td>Median between-syllable pause (s)</td>
<td>0.03 ± 0.01 (0.01 – 0.04)</td>
<td>0.02 ± 0.01 (0.008 – 0.03)</td>
<td>0.02 ± 0.01 (0.008 – 0.03)</td>
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<tr>
<td>Minimum frequency of a strophe (kHz)</td>
<td>2.3 ± 0.2 (2.1 – 2.7)</td>
<td>2.3 ± 0.4 (1.4 – 2.8)</td>
<td>2.4 ± 0.2 (2.0 – 2.8)</td>
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<tr>
<td>Maximum frequency of a strophe (kHz)</td>
<td>7.5 ± 0.4 (7.0 – 8.1)</td>
<td>7.3 ± 0.3 (6.9 – 7.9)</td>
<td>6.4 ± 0.3 (5.8 – 6.9)</td>
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<tr>
<td>Median frequency range of a syllable (kHz)</td>
<td>2.2 ± 0.2 (1.7 – 2.9)</td>
<td>2.0 ± 0.3 (1.2 – 4.1)</td>
<td>1.4 ± 0.2 (1.0 – 2.2)</td>
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and the uppertail coverts have pale sandy (Cinnamon C. 39) or rusty-brown (Antique Brown C. 37) fringes. Compared to female rubicola, the maurus female shows upperparts having the more expressed black streaks.

We have detected no stable differences in worn plumage between maurus and stejnegeri. We have found however only some weak differences in worn plumage between these two taxa, but these are evident only in a series of individuals and thus were not useful for species distinction. There was a little less white on the sides of the neck, on the inner upperwing, on the rump and on the base of rectrices in males of stejnegeri. Compared to female maurus, the females of stejnegeri have little more dark upperparts, which somewhat obscured black streaks.

The prominent differences between maurus and stejnegeri appear in fresh autumn plumage only. It is the color of the fringes of the feathers of the rump and the uppertail, that is useful for identification (Fig. 3): stejnegeri has chestnut-red fringes (Amber C. 36) and maurus has much more pale sandy (Cinnamon C. 39) or rusty-brown fringes (Antique Brown C. 37). Besides, stejnegeri has chestnut (Raw Sienna C. 136) fringes of the feathers of the upperparts that appear darker than that of maurus.

Song

The songs of maurus, rubicola and stejnegeri were generally similar. Each song strophe lasted 1–1.5 sec and built up of 10–20 short syllables interspersed with one or several single-element trills(s) (Fig. 4). The trills were very characteristic for the songs of maurus and rubicola, but were more rarely observed in stejnegeri. Because of a rarity of trills, singing of stejnegeri audibly seemed to be more melodious.

The LDA of songs in maurus, rubicola and stejnegeri males resulted in 86.0% correct classification of these three groups (Wilks’ λ = 0.090, F = 13.67, n = 57 males). The majority (5 out of 8) of misclassification cases were rubicola males that were predicted to belong to maurus. In addition to the misclassified ones above, there were also two maurus predicted to belong to rubicola and one stejnegeri predicted to belong to rubicola.

The first three principal components had eigenvalues greater than one. PC1 explained 45.1% of the total variation; PC2 explained a cumulative 68.1% and PC3 a cumulative 81.2% of the total variation. The following parameters showed the highest absolute correlations with Factor 1: duration of strophe, number of syllables, number of syllable types, and maximum frequency. By contrast, both median syllable length and median between-syllable pause showed the strongest correlation with Factor 2. PC1 vs. PC2 showed individual measurements clustering according to the taxa (Fig. 2, b).

Therefore, the LDA and PCA analysis clearly separated stejnegeri from both maurus and rubicola, which were closer to each other. Moreover, it was impossible to separate songs of maurus and rubicola in several cases. Song strophes of stejnegeri were the shortest, and had the lowest maximum frequency and the narrowest frequency range (Table 2).

Alarm call

All species on the approach of nest predators give two types of alarm calls, whits and chack. Whits are often modulated notes with energy limited to a narrow frequency range. Chacks are usually shorter than whits, and cover a much wider frequency range. Both whits and
chacks are generally similar in all species analyzed, although differed in detail (Fig. 5).

The LDA of whits in maurosi, rubicola and stejnegeri individuals resulted in 94.3% correct classification (Wilks’ $\lambda = 0.111$, $F = 23.37$, $n = 53$ individuals). The first two principal components had eigenvalues greater than one. PC1 explained 45.6%, and PC2 explained a cumulative 80.6% of the total variation. Frequency range and
maximum frequency showed the highest absolute correlations with Factor 1, and whit’s duration and minimum frequency showed the strongest correlation with Factor 2. Fig. 6, a shows the ‘PC1 vs. PC2’ scatterplot (see also Table 3).

Besides, we have found one more stable differences between the structure of whits of rubicola and that of the other two taxa. The shape of frequency modulation of whits of rubicola was always ‘ascending’ (frequency ascends from the begging to the end of a whit), while whits of the others had ‘descending’ modulation (Fig. 5).

The duration of chacks in rubicola and stejnegeri did not significantly differ one from another, but differed significantly from that of maurus (Fig. 6, b; Table 3).

**Discussion**

The morphological (both plumage and measurements) and vocal (both song and calls) data clearly matched the phylogroups reconstructed from mtDNA sequences (Zink et al., 2009), and separated Northern Eurasian taxa of the Saxicola torquatus complex into the three groups: rubicola, maurus and stejnegeri. Besides, variegatus was thought to be a closely relative to maurus based on mitochondrial DNA study (Zink et al., 2009). The taxonomy of Southern Eurasian’s armenicus, indicus and przewalskii remains to be studied.

With respect to vocalization, maurus and rubicola had the most similar songs, although they clearly differed in alarm calls. These two species also differed subtly, but noticeably, in external morphology. Our result confirmed previously published information concerning differences between the taxa in plumage and morphometrics (Robertsion, 1977; Urquhart, 2002; Hellström & Wern, 2011). In worn spring plumage male of rubicola is distinguishable by its comparatively small patches of white on the sides of the neck, by pale (not blackish) under-wing-coverts and by the prominent darkish centers of the feathers on the uppertail coverts, which appear as dark longitudinal streaks on the rump.

The two Asian taxa (maurus and stejnegeri) morphologically were the most similar. These two taxa cannot
be reliably distinguished by morphometrics and by worn spring plumage. The only feature useful for identification appears in fresh autumn plumage: the color of the fringes of the feathers of the rump and the uppertail. Besides, HELLSTROM & NOREVIK (2014) revealed the presence of dark spotting on the uppertail-covert of ca. 60% of birds in fresh autumn plumage, whereas maurus generally but not always (own data) shows unmarked rump. In contrast to morphology, song of stejnegeri was the most distinctive among the three taxa analyzed. Bioacoustic data are thus in agreement with mitochondrial lineages in so far as stejnegeri has apparently split from maurus and rubicola before differentiation occurred among the latter two taxa. S. stejnegeri thus appeared to be a cryptic species that is extremely similar in the suite of external characters to maurus, but differs in song and calls (this study) and mtDNA (ZINK et al., 2009).

The zones of sympathy between different taxa from the Saxicola torquatus complex in Eurasia are poorly known. The only exception is the sympathy zone between rubicola and variegatus in Rostov Oblast, Russian Federation (Fig. 1). Here, these two taxa bred in different although overlapping habitats without any signs of intergradations (KAZAKOV & BAKHTADZE, 1999; BAKHTADZE, 2002).

Little is known about the distributional ranges of maurus and stejnegeri in Transbaikalia, where a sympathy zone might exist. Although several authors stated a wide range of intergradation between maurus and stejnegeri in Siberia (Cramp, 1988; HELLSTROM & WERN, 2011), this statement is apparently wrong (HELLSTROM & NOREVIK, 2014). Interestingly, stonechats are more or less absent (except on migration) from the area just south and east of Lake Baikal, including the delta of the Selenga River (Fefelov et al., 2001). Therefore, the transition from maurus to stejnegeri in southern Siberia may be abrupt. According to our analysis of museum collections, as well as on published data (ZINK et al., 2009, 2010), there were only two points, where both taxa have been collected (Fig. 1). First point located near Chikoy urban-type settlement in the Republic of Buryatia, Russian Federation, where stejnegeri is the commonest stonechat, but several maurus specimens were collected during the breeding season (ZINK et al., 2010). The second point located in the eastern part of the Khangai Mountains (Central Mongolia), where one specimen of stejnegeri was collected along with several specimens of maurus. In the second point, however, the specimen of stejnegeri was collected on 21.08.1926 (deposited in the collection of Zoological Institute of the Russian Academy of Sciences), and could be thus on migration. Therefore, new studies on distribution ranges of maurus and stejnegeri in Eastern Siberia and Mongolia are extremely needed.

What happens where rubicola and armenicus meet in Turkey is unclear (Kirwan et al., 2008). URQUHART (2002), however, stated that the two taxa show different habitat preferences in that region, with armenicus present in mountain habitats and rubicola in the lowlands. In the Himalayas, przewalskii was found to be parapatric with indicus being separated by elevation (MARTENS & ECK, 1995).

The comparative studies of breeding biology, ecology and behavior of Eurasian stonechats are almost completely absent. Recently, however, GOLOVINA & OPAEV (2016) reviewed some original and published data (FUJIMAKI & SHIBBEY, 1991; FUJIMAKI et al., 1994) on the breeding biology and social organization of stejnegeri. Comparing biology and behavior of stejnegeri, with those of well-studied rubicola and hibernans (Parrinder & Parrinder, 1945; Johnson, 1971; Fuller & Glue, 1977; Greg-Smith, 1979; Urquhart, 2002; Banik, 2003) showed that most differences between them concern territory size and breeding density. S. stejnegeri usually had smaller territories and bred in higher densities, than both rubicola and hibernans. By contrast, habitat and breeding biology (e. g. nest and egg) appeared to be rather similar among the two taxa.

Summarizing, our study presented data that is valuable for the evaluation of the taxonomy of the Saxicola torquatus complex. Based on our and earlier genetic findings, we proposed the species status for the following three taxa:

1. European stonechat Saxicola rubicola (LINNAEUS, 1766), including S. r. rubicola and S. r. hibernans.
2. Eastern stonechat Saxicola maurus (PALLAS, 1773), including at least three subspecies: S. m. maurus, S. m. variegatus and S. m. armenicus (the taxonomic affinities of indicus and przewalskii that usually merge into S. maurus remain to be studied).
3. Japanese stonechat Saxicola stejnegeri (PARROT, 1908), monotypic.

Our song data are also in agreement with previous phylogenetic reconstructions, because we revealed that the song of S. stejnegeri is the most distinct among the three taxa analyzed.

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Electronic Supplement Files

at http://www.senckenberg.de/vertebrate-zoology

File 1. Supplementary_Table_S1.pdf.
File 2. Supplementary_Table_S2.pdf.
File 3. Supplementary_Table_S3.pdf.
File 4. Supplementary_Fig_S1.pdf.