

UNIVERSITY OF SOUTH BOHEMIA, FACULTY OF SCIENCE

## **Song variation in the Chiffchaff**

*Phylloscopus collybita:*

**Song parameters suitable for individual coding**

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Supervisor: Mgr. Pavel Linhart

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**Anotation:**

My study deals with general individual characteristics in songs of a passerine bird species Chiffchaff (*Phylloscopus collybita*), especially I examine the potential of these characteristics for application in acoustic monitoring of bird individuals. I have found that Chiffchaffs have individually distinct songs but unfortunately, song features I measured were not stable in time and hence they would not be suitable for long-term monitoring of individuals. My study suggests that acoustic individual recognition might be challenging in songbird species due to the complexity and plasticity of their song.

**Declaration:**

I hereby declare that I have worked on my master thesis independently and used only the sources listed in the bibliography.

I hereby declare that, in accordance with Article 47b of Act No. 111/1998 in the valid wording, I agree with the publication of my master thesis, in shortened form resulting from deletion of indicated parts to be kept in the Faculty of Science archive, in electronic form in publicly accessible part of the STAG database operated by the University of South Bohemia accessible through its web pages.

Further, I agree to the electronic publication of the comments of my supervisor and thesis opponents and the record of the proceedings and results of the thesis defence in accordance with aforementioned Act No. 111/1998. I also agree to the comparison of the text of my thesis with the Theses.cz thesis database operated by the National Registry of University Theses and a plagiarism detection system.

In České Budějovice, 25<sup>th</sup> April 2014

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Bc. Alexandra Průchová

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# 1 Introduction

Recognition of individuals is widespread in animal kingdom. Both receivers and signallers could benefit from individual recognition in situations when they repeatedly communicate and interact between each other (Tibbetts and Dale 2007). Naturally, animals living in complex social environments (groups, colonies or flocks) are capable to recognise individuals. Distinguishing individuals is important for any group member; for example, it can facilitate understanding social structure, social hierarchy and the relationships between individuals in groups of primates (Bergman and Sheehan 2013). Individual recognition can also help animals reveal identity of reliable individuals from cheaters or unreliable individuals (Olendorf et al. 2004). Also, parent-offspring recognition (Jouventin et al. 1999; Charrier et al. 2003; Charrier and Harcourt 2006), sibling recognition (Wanker et al. 1998) or mate recognition represent recognition of individuals which can be challenging especially in colonially breeding species (Dale et al. 2001). Solitary animals may benefit from individual recognition as well, and can use it for recognition of neighbours ("dear enemies" - Wei et al. 2011) from strangers. For example, territorial American bullfrogs (*Rana catesbeina*) recognize voice of unfamiliar individual between familiar neighbours (Bee and Gerhardt 2002). Likewise, territorial birds can recognize neighbouring males from strangers (Stoddard et al. 1991) and even memorize them to the next year (Godard 1991; Draganoiu et al. 2014).

Individual recognition can be based on various mechanisms and use chemical, visual and acoustic cues (Tibbetts and Dale 2007). Olfactory signals inform about position in hierarchy, dominance and also show individual distinctiveness (Thom and Hurst 2004). Animals can also use visual cues, for example, Paper wasps (*Polistes fuscatus*) use uncial facial features to identify individual nest-mates (Tibbetts 2002). In comparison with olfactory and visual cues, vocalizations can carry current and up to date information even over long distances which receivers can react immediately.

The most basic individual vocal cues have origin in specific anatomy and morphology of vocal organs and of the whole vocal tract (Taylor and Reby 2010) of each individual. In birds, the voice is formed in syrinx, generally located at the area where trachea divides into bronchi. For example, birds with big syringes might have lower and stronger voices than birds with smaller syringes (Hardouin et al. 2007). Further, many birds can produce two voices simultaneously, each of them by different part of syrinx.

34 According to Aubin et al. (2000) this may lead to interactions of sound waves from both  
35 parts of syrinx reflecting tiny differences in vocal tract between individuals.

36 Given the information above, it is not surprising that many studies show  
37 individually variation in vocalization in birds (Thompson 1970; Lein 1978; Cicero and  
38 Benowitz - Fredericks 2000) and as I mention, this individual variation has many important  
39 implications for their lives. Scientists have focused their attention on individual variability  
40 in bird vocalizations for decades (Borror 1956; Thompson 1970; Williams and MacRoberts  
41 1977). Successively, visual comparison of individual spectrograms became standard  
42 method to demonstrate individual differences among individuals(Borror 1956), even  
43 though measuring of exact acoustic parameters that characterize individuals was still  
44 missing in the past but became common from late seventies (Güttinger et al. 1978).

45 Non-songbirds represent good study models because they have simple vocalizations  
46 with relatively stable acoustic structure (e.g. Mathevon et al. 2003). On the other hand,  
47 songbirds (and other avian vocal learners) are special regarding their vocal plasticity and  
48 vocal learning abilities. Therefore, they possess more complex identity coding and  
49 decoding mechanisms. For example, good imitation ability of many species could hinder  
50 individual recognition. However, identity can still be detected due to small individual  
51 modifications of the original imitated song (Catchpole and Slater 2008; Mennill 2011).  
52 Ortolan buntings (*Emberizia hortulana*) share syllables with similar shape but differ  
53 between males by slight shifts in frequency (Osiejuk et al. 2005). Song plasticity and song  
54 learning ability can also provide mechanisms to improve opportunity for individual  
55 recognition. Birds may develop individual repertoires (Gaddis 1985; Kroodsma 1996) or  
56 specific individual signatures (Wanker et al. 2005; Petrusková et al. 2010). Individual  
57 signatures can be located in the specific parts of songs (Linossier et al. 2013) only.

58 Recent technological progress in audio recordings and analysis make it possible  
59 that vocalizations will become a common monitoring tool in future (Blumstein et al. 2011).  
60 Nowadays different applications of acoustic monitoring are considered. For example, it  
61 might be possible to estimate species richness (Farina et al. 2011; Wimmer et al. 2013)  
62 based on recordings of soundscapes. One of the potential applications is also monitoring of  
63 individuals. This monitoring approach would be helpful for collecting information about  
64 species living in hardly accessible terrain, like tropical forests (Mennill and Vehrencamp  
65 2008), species with nocturnal activity (Lengagne 2001) or species that are difficult or  
66 problematic to catch (Marques et al. 2013). Individual variation in bird vocalizations has  
67 been widely documented and could be used in practical applications to identify individuals

68 in field. Birds could be recorded in the field, using special bioacoustics recording devices  
69 (Rempel et al. 2005) or acoustic sensors (Wimmer et al. 2013). These recordings should be  
70 subsequently analysed and assigned to correct individuals. The ideal method should be  
71 easily applicable between different species. Weary and Krebs (1992) suggested that Great  
72 tits (*Parus major*) can recognize familiar and unfamiliar individual by general voice  
73 characteristic independently on syllable content of the song. General voice characteristics  
74 could be therefore potentially used for acoustic monitoring of individuals.

75 In this study I would like to answer following questions; 1) Are general song  
76 characteristics individually distinct in Chiffchaffs? 2) Are these individually distinct song  
77 characteristics stable in time? Most studies focusing on individual vocal variation explore  
78 very short recordings. Consequently, my aim was to study individual variation on different  
79 time scales; I explored individual variation in songs recorded within single recording  
80 session; within single day; between two successive days, and between years.

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82

## 83 **2 Material and Methods**

84

### 85 **2.1 Study species**

86 According to (Cramp 1992), Chiffchaff (*Phylloscopus collybita*) is a small-size song bird  
87 (Passeriformes; Siilvidae) very common and wide spread in west Palearctic. Optimal  
88 habitats are lowland woodlands with sparse canopy level and rich undergrowth, often at  
89 moist areas. It is also common species in parks and gardens. Chiffchaffs are monomorphic  
90 species. Nevertheless, males and females differ in their behaviour. Females spend most of  
91 the time quietly deep and low in the shrubs. In contrast, males show strong and aggressive  
92 behaviour and defend their territory by singing and chasing out intruders. Chiffchaffs are  
93 monogamous, but the facultative polygyny (mostly bigamy) is also possible, especially, in  
94 the area with excessively high population density. They are migratory passerine birds  
95 wintering in Mediteranean and North Africa. First males arrive from mid March during the  
96 spring migration and they start to find and defend their territories immediately. Loud male  
97 songs, duets and antagonistic songs are very intensive at the beginning of season.  
98 Generally, females begin to arrive a week after males. Intensive chases as well as physical  
99 male fights are common during the territory establishment and pairing period. Pairs  
100 ordinary breed twice per season and first clutches (April, May) are usually slightly bigger  
101 (on average 5 eggs) than later clutches in June and early July. Although chiffchaffs are  
102 territorial, their territories may to some extent overlap at sites with high breeding density.

103

### 104 **2.2 Study area**

105 Males were recorded in former military training area on the outer boundary of České  
106 Budějovice, Southern Bohemia, the Czech Republic (48°59, 5′ N, 14°26, 5′ E). The area  
107 (ca 1 km<sup>2</sup>) is covered by habitat with small ponds, marshes and shrubs. Willows (*Salix* sp.),  
108 birches (*Betula* sp.) and poplars (*Populus* sp.) dominate in vegetation. Also large and old  
109 oaks (*Quercus* sp.) grow sporadically or in alleys on dams. We were studying Chiffchaffs  
110 at the locality since 2008 to 2012. The area hosts approximately 60 males every year and  
111 the breeding density is relatively high. Large majority of males was colour-banded during  
112 the years. About 16 - 25% banded males were detected again in at least one of the  
113 successive years.

114

115



### 116 **2.3 Recording**

117 Males were recorded for different purposes over the years 2008 – 2011. Recordings were  
118 made from April to June, due to the strong motivation of chiffchaff males to sing in this  
119 time. Recorded individuals were marked by combination of standard aluminium ring and  
120 up to three different colour rings for identification of individuals on locality. Recorded  
121 songs represent two different datasets: first dataset was focused on within and between day  
122 variation in song parameters (Days dataset; n = 13 males) and the second dataset was  
123 focused on between years song variation (Years dataset; n = 16 males). See below for  
124 detailed description. In total, songs from 29 different males were used in this study.  
125 Recording conditions were generally similar for both recording datasets. Recording were  
126 done from 5:30 to 11:00 in the morning. We used Marantz recorder and a directional  
127 microphone Sennheiser ME67. We tried to record birds as close as possible, usually within  
128 the distance of 5 - 15 meters from the singing bird and with no obstacles in between the  
129 microphone and the recorded male if possible.

130 Days dataset included recordings from the year 2011. Days dataset was recorded in  
131 two successive days to compare male songs from two days and find possible differences  
132 between them. During the first day recordings were taken from early morning from c.a.  
133 5:00 to c.a. 9:30 a.m. During second day, recordings were done only early in the morning  
134 (c.a 5:00 to 6:00). In detail, recordings from the first day (DAY1) represent on average 63  
135 min (minimum 28 min, maximum 115 min) long recordings (singing time) and recordings  
136 from the next day (DAY2) represent on average 14 min (minimum 5, maximum 61 min)  
137 long recordings. Overall, we recorded over 17 hours of recordings with 6216 songs in both  
138 days (Table 1).

139

140

**Table 1:** Number of songs recorded and analyzed for each male in dataset Days

Male	Number of songs	
	All	Analyzed
PC1101	456	212
PC1102	424	143
PC1103	354	150
PC1104	255	126
PC1105	237	48
PC1106	969	467
PC1107	793	365
PC1108	737	397
PC1109	848	314
PC1110	325	118
PC1111	103	77
PC1112	481	169
PC1113	234	184
Total	6216	2770

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Recordings for Years dataset were taken during years 2008 – 2011. Songs were recorded for each male in at least subsequent years in order to compare their songs between years. Years dataset contained on average 4 min long recordings (minimum 2 min, maximum 8 min). Altogether, 837 songs from 16 males were collected (Table 2). Initially, these data were collected for different studies, nevertheless, they also fit for this research.

**Table 2:** Number of songs recorded for each male in dataset Years

Male	Number of Songs	
	All	Analyzed
F72726	56	45
F91901	62	0
F91903	62	22
F91907	50	40
F91909	47	0
F91913	57	24
F91915	53	42
F91916	66	42
F91930	44	0
F91931	56	0
F91954	45	22
F91959	43	33
F91969	69	35
F91973	40	30
F91983	46	0
J23234	41	0
Total	837	335

149

## 150 **2.4 Song analysis**

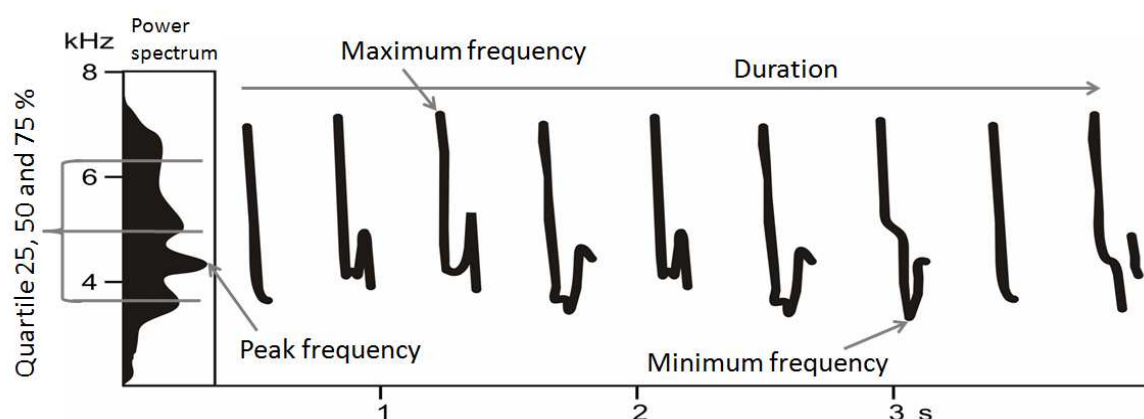
151 All songs were processed and analyzed in Avisoft SASLab Pro software (Raimund Spetch,  
152 Berlin). As a first step, I checked each recording to choose only the high quality songs for  
153 analysis. Any songs containing high background noise, other birds' vocalizations, and  
154 others disturbances, hindering song spectrogram analysis were rejected from further  
155 analyses. Subsequently, I applied a highpass filter (2500 Hz) on all preselected songs and  
156 down-sampled the songs to 22050 Hz sampling frequency.

157 Although many recordings were eliminated from further analysis, good sample of  
158 songs still remained for analysis. There were 2770 selected songs for Days dataset (from  
159 6216 original songs, see Table 1). Years dataset was much smaller and I analysed 335  
160 songs from 837 original songs. Furthermore, 6 birds were completely removed from

161 further analyses because of low number of song per recording (Table 2). I used the  
162 “Automatic parameter measurement” tool in Avisoft to measure song parameters.

163 I measured following basic song parameters: song length (s), syllable interval (s),  
164 minimum frequency (Hz), maximum frequency (Hz), peak frequency (Hz) and frequency  
165 quartiles (25, 50, 75 % of spectrum, all in Hz) (Figure 1). I determined these characteristic  
166 for their simplicity, easy measuring procedure, easy use across species and they give  
167 overall description of temporal and frequency song parameters.

168 .



169  
170 Figure 1. Spectrogram of chiffchaff song with marked measured characteristics, left side represents power  
171 spectrum

## 173 2.5 Statistical Analysis

174 Statistical analyses were calculated using statistical program R.

### 176 Potential of individual coding

177 To determine suitability of song parameters for individual coding I calculated Potential for  
178 individual coding scores (PIC) for each of the parameters. Following the formula:  $PIC =$   
179  $CV_b / CV_w$ , I counted PIC for each time-scale (see below) individually. The basic element  
180 of the formula is coefficient of variation ( $CV = SD / mean$ ) used in two different ways.  
181  $CV_b$  is coefficient of variation between males however  $CV_w$  is an average coefficient of  
182 variation within males (i.e. the CV is counted for each male and variable separately and the  
183 CV values for a particular variable are then averaged across males).

184 I divided data to four individual cases comparing the differences and changes in  
185 PIC values of each acoustic parameter on four different time-scales. “Recording session”  
186 PIC is based on the first 15 songs from each male during the recording at Day 1. To

187 compute “Within day” PICs, I used again the first 15 songs but included also the last 15  
188 songs on Day 1 from each male (so PICs were computed from 30 songs per male).  
189 “Between days” PICs were computed from the first 15 songs from Day1 and first 15 songs  
190 from Day2. In this case PIC for the males PC1105, PC1110 and PC1111 was not computed  
191 due to low number of suitable songs. In the last case “Between years” PICs were computed  
192 in similar way, but there were different numbers of songs per male. They varied from 9 to  
193 15 songs per male from each of the year (Year1 and Year2).

194

### 195 **Environmental conditions and song parameters**

196 To test whether song parameters systematically vary with environmental conditions, I  
197 compared averaged song parameters calculated from first successive 15 songs and last  
198 successive 15 songs of each male from the Day1 with nonparametric Wilcoxon matched-  
199 paired test. The analysis was done for 12 males because for the male PC1105, first and last  
200 15 songs were too close in time (recorded within one hour). First 15 songs were recorded  
201 between 5:00 and 7:00; last 15 songs between 7:30 and 9:30 depending on male. But for  
202 each male first and last 15 songs were at least 2 hours apart. I divided songs into these two  
203 groups to see whether the parameters change with daytime. Daytime likely correlates with  
204 changes in environmental circumstances at the initial and final part of recording. For  
205 example, early in the morning (first 15 songs) the noise level is low. On the other hand,  
206 later (last 15 songs) the environment is more noisy due to busy road near the locality (main  
207 traffic caused by people getting to work). Other conditions, possibly affecting song  
208 parameters, like temperature, humidity, or motivation of males could also change during  
209 the morning

210

### 211 **Repeatability of song parameters**

212 I calculated Spearman’s correlation coefficient ( $\rho$ ) to test parameter’s repeatability within  
213 day, between days and between years. Spearman’s correlation coefficient for Within day  
214 was calculated from same data as Wilcoxon matched-paired test and Within day PICs. The  
215 same data as in case Between days and Between years PICs were used to calculate  
216 Between days and Between years repeatability. First, I averaged song parameters for each  
217 bird and time interval (first and last 15 songs, first 15 songs from Day1 and Day2, songs  
218 from Year1 and Year2) and then I looked for correlations between those values. High and  
219 significant correlation between time intervals would indicate high repeatability of song  
220 parameters.

221 **Discrimination of individuals**

222 Finally, I conducted linear discriminant analysis (LDA) to assign songs to each bird  
223 according to measured song's characteristics and determine classification success achieved  
224 by the discriminant analysis (Figure 2). Again, four LDAs were conducted. The first LDA  
225 was conducted to see whether individuals can be discriminated within "Recording session"  
226 time-scale. It was based on 15 songs from Day 1. The second LDA on "Within day" time-  
227 scale involved all selected songs from Day 1. "Recording session" LDA and "Within Day"  
228 LDA used leave one out cross-validation. Data for LDAs on "Between day" and "Between  
229 years" time-scales used Jack-knife cross-validation, which means they were divided into  
230 training and test subdatasets. Training subdataset contained recordings from the Day 1 /  
231 Year 1 and test subdataset contained recordings from the Day 2 / Year 2. All acoustic  
232 variables were scaled to z-scores prior to LDA.

233

234 **3 Results**

235

236 **3.1 Potential of individual coding**

237 PIC values (Table 3) varied from 0.98 (peak frequency - between years) to 2.03 (syllable  
 238 interval - recording session). According to previous studies, the parameter has a potential  
 239 for identity coding if PIC reaches a value higher than 1. Only peak frequency between  
 240 years did not exceed 1. On the other hand, it is apparent that PIC values are generally low  
 241 so their potential for individual coding is weak. Syllable interval reach relatively high PIC  
 242 value (PIC = 2.03) during recording session. Generally, PICs showed declining trend with  
 243 increasing time-scale. CVb values for frequency parameters were very low, ranging  
 244 between 0.02 – 0.07. In contrast, duration showed the largest variation (CVb from 0.33 to  
 245 0.38).

246 **Table 3:** Coefficient of variation for each song characteristic between males (CVb), coefficient of variation  
 247 within males (Cv, mean) and PIC for datasets: Recording session, Within days, Between days, Between  
 248 years.

Song characteristic	Recording session			Within days			Between days			Between years		
	CVb	CVw, mean	PIC	CVb	CVw, mean	PIC	CVb	CVw, mean	PIC	CVb	CVw, mean	PIC
Duration (s)	0.38	0.26	<b>1.48</b>	0.38	0.28	<b>1.36</b>	0.37	0.29	<b>1.27</b>	0.33	0.27	<b>1.18</b>
Syllable interval (s)	0.04	0.02	<b>2.03</b>	0.04	0.03	<b>1.39</b>	0.04	0.03	<b>1.44</b>	0.31	0.26	<b>1.19</b>
Peak frequency (Hz)	0.07	0.05	<b>1.34</b>	0.06	0.05	<b>1.31</b>	0.07	0.05	<b>1.24</b>	0.06	0.06	<b>0.98</b>
Minimum frequency (Hz)	0.07	0.05	<b>1.35</b>	0.07	0.06	<b>1.23</b>	0.06	0.05	<b>1.22</b>	0.05	0.04	<b>1.39</b>
Maximum frequency (Hz)	0.07	0.06	<b>1.25</b>	0.08	0.07	<b>1.16</b>	0.07	0.06	<b>1.16</b>	0.03	0.03	<b>1.31</b>
Quartile 25 % (Hz)	0.02	0.01	<b>1.74</b>	0.03	0.02	<b>1.50</b>	0.03	0.02	<b>1.32</b>	0.04	0.03	<b>1.14</b>
Quartile 50 % (Hz)	0.02	0.02	<b>1.30</b>	0.03	0.02	<b>1.26</b>	0.02	0.02	<b>1.33</b>	0.04	0.03	<b>1.37</b>
Quartile 75 % (Hz)	0.02	0.02	<b>1.25</b>	0.03	0.02	<b>1.20</b>	0.02	0.02	<b>1.14</b>	0.03	0.02	<b>1.39</b>

249

250 **3.2 Environmental conditions and song parameters**

251 Wilcoxon matched-pairs test does not show strong changes in song parameters during the  
 252 day (Table 4; n=15) with the exception of syllable interval (p=0.034; V=66). Syllable  
 253 interval was shorter (chiffchaffs increased syllable rate) later in the day than early in the  
 254 morning.

255

256 **Table 4:** Wilcoxon matched-pairs test; first 15 and last 15 songs a day

Song characteristics	Median		Minimum		Maximum		p	V
	First	Last	First	Last	First	Last		
Duration (s)	4.54	4.45	2.65	2.54	9.62	8.29	0.97	40
Syllable interval (s)	0.38	0.36	0.34	0.34	0.39	0.38	<b>0.034</b>	66
Peak frequency (Hz)	4160.67	4184.83	3931.54	3913.33	4466.00	4459.33	0.569	47
Minimum frequency (Hz)	3473.67	3399.33	3294.67	2930.67	3740.00	3900.00	0.176	57
Maximum frequency (Hz)	6428.67	6408.00	5565.33	5528.00	7002.67	6864.00	0.733	44
Quartile 25 % (Hz)	4123.00	4051.00	3972.67	3930.67	4244.67	4302.67	0.266	54
Quartile 50 % (Hz)	4734.33	4724.00	4652.67	4617.78	4928.67	4962.67	0.97	40
Quartile 75 % (Hz)	5639.67	5651.76	5478.00	5348.00	5871.33	5922.67	0.301	25

257

### 258 **3.3 Repeatability of song parameters**

259 Spearman's correlation coefficients ( $\rho$ ) show relatively strong repeatability of most of the  
 260 song parameters. In total, average values of six parameters were significantly positively  
 261 correlated between first 15 and last 15 recorded songs ("Within day" time-scale) and  
 262 showed high correlation coefficients (e.g. duration, peak frequency, minimum frequency  $\rho$   
 263  $> 0.7$ ; see Table 5). Nevertheless, similar to PICs, repeatability of most song parameters  
 264 seemed to declines with increasing time-scale. On "Between days" time-scale, only  
 265 minimum frequency has still high and significant repeatability ( $\rho = 0.84$ ). Spearman's  
 266 correlation coefficients are also weak for most of the parameters on "Between years" time-  
 267 scale. Only quartile 50 % reaches  $\rho = 0.71$  and is significantly repeatable in songs from  
 268 Year 1 and Year 2. I do not consider peak frequency ( $\rho = - 0.65$ ) repeatable between years  
 269 due to the negative sign of the correlation which is in striking contrast with positive  
 270 correlation that was expected.

271

272

273 **Table 5:** Spearman's rank correlation coefficient  $\rho$  (significant correlations with  $p < 0.05$  are highlighted in  
 274 bold)

Song characteristics	Within day	Between days	Between years
Duration (s)	<b>0.76</b>	0.51	0.3
Syllable interval (s)	0.55	0.49	0.35
Peak frequency (Hz)	<b>0.73</b>	0.07	<b>-0.65</b>
Minimum frequency (Hz)	<b>0.71</b>	<b>0.84</b>	0.50
Maximum frequency (Hz)	0.39	0.47	0.18
Quartile 25 % (Hz)	<b>0.69</b>	0.55	0.31
Quartile 50 % (Hz)	<b>0.63</b>	0.3	<b>0.71</b>
Quartile 75 % (Hz)	<b>0.59</b>	-0.13	0.62

275

### 276 **3.4 Discrimination of individuals**

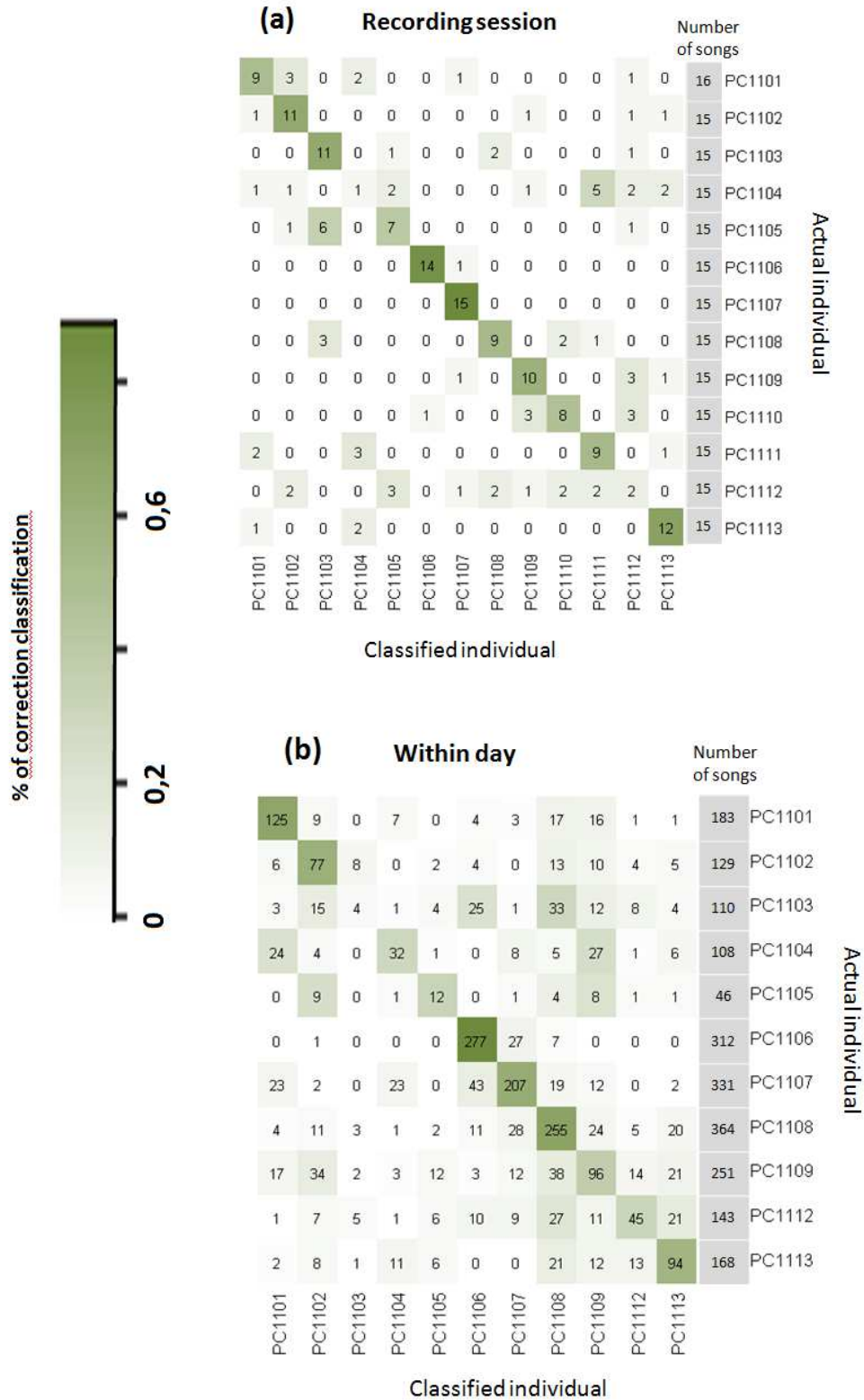
277 The highest number of correct classifications of songs to the right individuals is 60 % for  
 278 songs recorded during short time interval - within a single recording session. Within day  
 279 classification accuracy is 57 % and between day classification accuracy is 45 %. The  
 280 lowest overall accuracy is on between years time-scale - only 18 %. Even such a low  
 281 accuracy is still almost twice higher than accuracy expected by chance (correct  
 282 calssifications by chance = 9 for between years data) %. Classification accuracy also varied  
 283 markedly between males within each time-scale. For example, the accuracy within  
 284 "Recording session" for PC1107 was 100% in comparison with only 7 % of correct  
 285 classifications for PC1104.

286

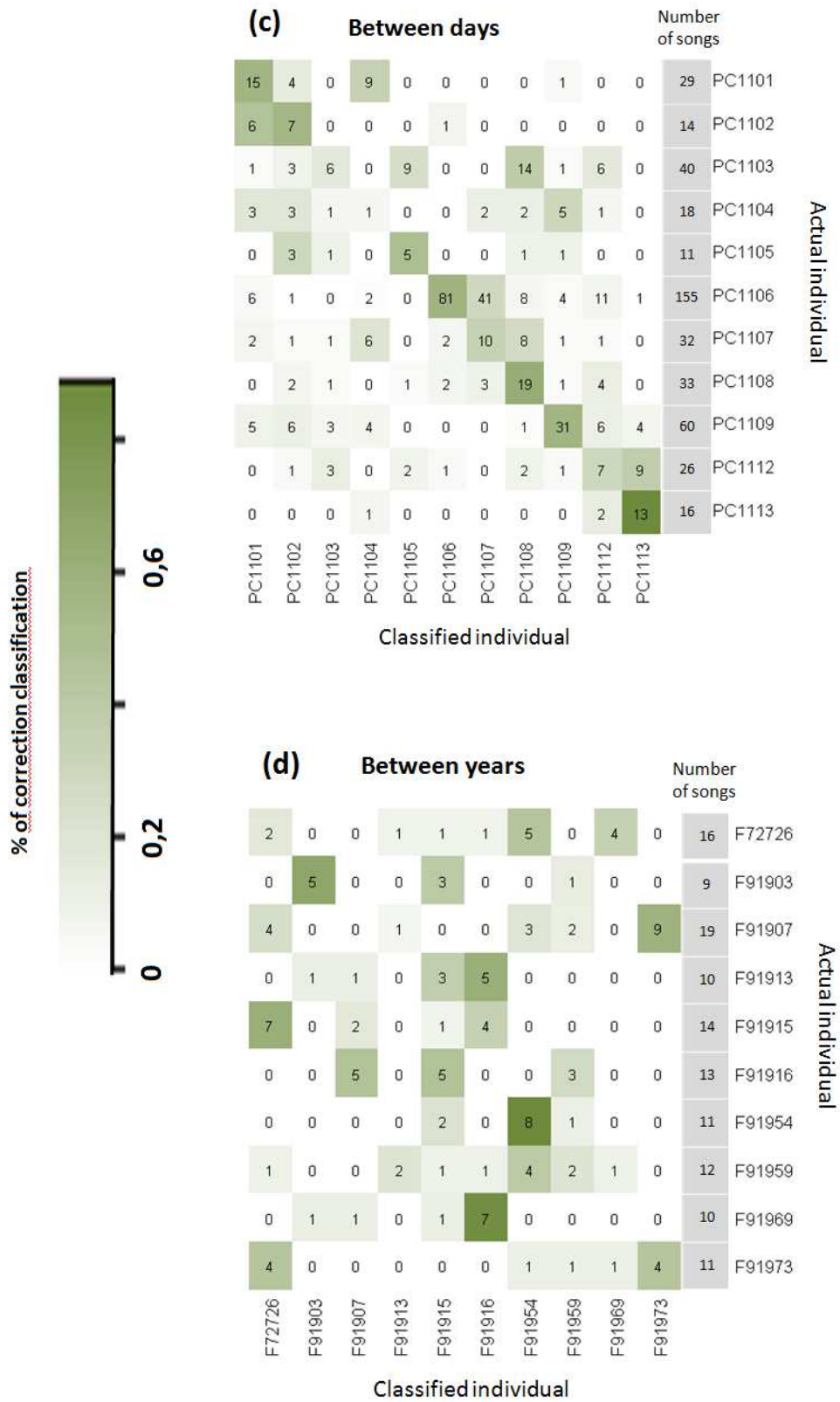
287



288 (Figure 2): Results of LDA (classification matrix) for songs recorded within Recording session (a), within  
 289 day (b), between days (c) and between years (d).  
 290  
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292



## 294 **4 Discussion**

295

296 This study is about individual variation in fundamental song parameters, their stability and  
297 their potential for individual coding in Chiffchaffs. My results suggest that general song  
298 parameters have a potential for individual coding but they are not stable in time. Therefore,  
299 identification of individuals based on these parameters could be possible only within a  
300 short time interval and their application for acoustic monitoring of individuals might be  
301 problematic in Chiffchaffs.

302 There is an evidence that some birds have individual syllable repertoires which  
303 even persist between years (Mamede and Mota 2012; Draganoiu et al. 2014). Other species  
304 may possess special individual signatures in their calls or within certain parts of a song  
305 (Wanker et al. 2005). Nevertheless, neither of the two mechanisms is appropriate for bird  
306 monitoring in general. Long recordings are required to get whole syllable repertoire in  
307 many species. Moreover, birds could use only part of their repertoire even during long  
308 recording sessions and possible results would thus be based on incomplete data.  
309 Chiffchaffs repertoire size is small, ranging from 7 to 13 syllables (Cramp 1992).  
310 However, Chiffchaffs probably use song types (only some combinations of syllables types  
311 are possible) and it may take tens or hundreds songs before they switch from one song type  
312 to another (Pavel Jaška pers. comm., pers. obs.). Moreover, using repertoires or individual  
313 signatures is not easily applicable across different species. Therefore, I decided to approach  
314 the song as a basic unit for analysis and analyse parameters which can easily describe basic  
315 parameters of whole song, without any special regard to syllable content.

316

### 317 **Potential of individual coding**

318 Potential of individual coding (PIC) is frequently used in studies dealing with  
319 individual recognition to determine parameters suitable for identity coding (Mathevon et  
320 al. 2003; Charrier et al. 2004; Sandoval et al. 2014). PIC values higher than 1 mean that  
321 parameter has a potential for individual coding (Lengagne et al. 1997). The higher the PIC  
322 value the greater is the potential of identity coding for a given parameter. My results show  
323 that all analyzed song parameters could possibly code identity, maybe with the exception  
324 of peak frequency (PIC=0.98 on between years time scale). On the other hand, PIC values  
325 of chiffchaff song parameters are relatively weak. Vocalization parameters of non-  
326 songbirds show considerably higher PIC values. For example calls of the black headed gull

327 (*Larus ridibundus*) and the slender-billed gull (*Larus genei*) show higher PIC not only in  
328 temporal parameters (PIC up to 2 or 3) but also for frequency parameters (PIC values  
329 ranging between 1.1 – 4.9.; Mathevon et al. 2003). Only two temporal parameters are used  
330 in this study (duration and syllable interval) both of them have relatively strong potential  
331 for individual coding especially on recording session time scale. Within songbirds, song  
332 duration PIC of chiffchaff has similar value like PIC for duration of specific song parts in  
333 White eared ground sparrows. But in contrast with my results, sparrows had also high PIC  
334 (from 1.1 to 4.9) for frequency parameters (Sandoval et al. 2014). To summarize, it seems  
335 that all of the measured song parameters could be used for distinguishing between  
336 individuals. Nevertheless, for practical applications in monitoring of individuals, song  
337 parameters should be also repeatable on the longer time scales and stable in different  
338 environmental and social circumstances.

339

#### 340 **Repeatability of song parameters**

341 Certainly, song parameters might be influenced by variation in environmental and social  
342 conditions (like temperature, air humidity, noise levels, motivation... etc.) during the day  
343 and between days (Wiley and Richards 1978; Slabbekoorn 2013). My results do not reveal  
344 any substantial impact of environmental and social conditions on song parameters taking  
345 the daytime as proxy for changing environmental and social condition during the day.  
346 Surprisingly, frequency parameters and song duration did not vary systematically during  
347 the morning although some studies might suggest changes of these parameters with  
348 daytime in chiffchaffs due to changes in e.g. noise levels (Verzijden et al. 2010) or  
349 motivation (Rodrigues 1996). Accordingly, repeatability of frequency parameters within  
350 day (Spearman's coefficient results) were high and significant in six parameters (duration,  
351 peak frequency, minimum frequency and frequency quartiles) but repeatability decreased  
352 on following time scales, possibly due to random variation in song parameters in time.

353

#### 354 **Discrimination of individuals**

355 Many studies dealing with individual variability in vocalizations use multivariate statistics  
356 like principal component analysis or discriminant analysis (Wanker and Fischer 2001;  
357 Charrier and Harcourt 2006; Digweed et al. 2012) to show that vocalizations can be  
358 assigned to correct individuals based on their acoustic parameters and hence, could be used  
359 to monitor individuals. These methods show relatively optimistic results and, of course,  
360 provide a good evidence that vocalizations are in principle individually distinct. These

361 findings are in accord with my results on discrimination of individuals within recording  
362 session. However, the problem of most of these studies is the limited time interval of  
363 recording individuals. In general, long-term individual recognition has been poorly  
364 investigated to date. One of the existing studies showed that owls might have hoots that are  
365 individually distinct and stable. Eagle owls (*Bubo bubo*) living in captivity did not change  
366 their vocalizations between two years (Lengagne 2001). It is apparent that long term  
367 recognition of individuals is important for animals. Birds are able to recognize songs of  
368 their neighbours from previous years (Godard 1991). Northern fur seal females  
369 (*Callorhinus ursinus*) were capable to recognize playback recording of their pups after four  
370 years of separation (Insley 2000). Therefore it seems likely that there should be identity  
371 cues in vocalizations stable over time. In conclusion, my results show that further studies  
372 considering vocalizations as monitoring tool should focus on parameters coding  
373 individuality on larger time scales.

374

375 Understanding identity coding in vocalizations is a basic knowledge for successful  
376 application of acoustic monitoring tools. Despite large interest in individual variation in  
377 vocalizations, we know very little about how song parameters change in time and which  
378 parameters could be suitable for acoustic monitoring of individuals. Song parameters that  
379 code individuality have to be stable and persist through time in order to be practically used  
380 for monitoring purposes. My results suggest that passerine birds represent challenge to  
381 methods of individual identification due to their complex song structure and vocal learning  
382 ability. However, songbird males can use songs to distinguish individuals even at longer  
383 time scales like days and seasons suggesting that some identity cues persist in  
384 vocalizations(Godard 1991). Classical methods of spectrogram song analysis might not be  
385 good enough to describe the long-term individual cues in songs. Therefore, future studies  
386 might try to determine new parameters based on the principles of song production and  
387 slight differences in song production organs between individuals.

388

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390

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