

## Green Warbler *Phylloscopus nitidus* ringed at Blåvand: molecular confirmation of a Danish first and European eighth vagrant record

*Kaukasisk lundsångare Phylloscopus nitidus ringmärkt i Blåvand: Danmarks första och Europas åttonde raritetsfynd bekräftas molekylärt*

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### Abstract

On 27 May 2015, a Green Warbler *Phylloscopus nitidus* was caught and ringed at Blåvand Bird Observatory, the westernmost point in Denmark. The species is challenging to identify on morphological basis alone, and the bird's identity was confirmed with phylogenetic analyses of mitochondrial DNA sequences. This constitutes the first record of Green Warbler in Denmark, the third record in Fennoscandia – following records on Öland, Sweden, on 29 May 2003, and Åland, Finland, on 20 May 2012 – and the eighth vagrant record in Europe outside the species' very restricted peripheral breeding range.

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Received 17 July 2017, Accepted 14 August 2017, Editor: Sören Svensson

### Introduction

Green Warbler *Phylloscopus nitidus* breeds from northern Turkey eastwards to Iran, and winters in India and southeast Asia (del Hoyo et al. 2006). It reaches marginally into Europe in southern Russia and northern Georgia (del Hoyo et al. 2006). East of its breeding distribution the Greenish Warbler *Ph. trochiloides* and the Two-barred Warbler *Ph. plumbeitarsus* display a continuous distribution along a broken ring around the Tibetan Plateau, with several discernible subspecies/populations (Irwin et al. 2001).

The relationship between Green Warbler, Greenish Warbler, and Two-barred Warbler is complex (Irwin et al. 2001) and the Green Warbler has traditionally been regarded as a subspecies of Greenish Warbler. It was raised to species status by the British Ornithologists' Union in 2008 (Knox et al. 2008, BOU 2009), and by the Clements Checklist of Birds of the World in 2009 (Clements et al. 2009), while the International Ornithological Union regarded it as a species already in its first edition (Gill & Wright 2006). The Two-barred Warbler was given species status later (Gill & Wright 2006, Clements et al. 2014).

Before this first Danish record of Green Warbler there were two records from Fennoscandia and another five from remaining Europe. Two new records were added in 2016, making the total until and including 2016 ten records outside the restricted European breeding range (Table 1). Furthermore, there are seven accepted records in Israel (Slack 2009, IRDC 2016).

### Description

On Wednesday 27 May 2015, the daily standardized ringing at Blåvand Bird Observatory could not be done because of strong wind (NW 12 m/s) and rain. Around noon, the wind decreased to 5 m/s the rain stopped, and the sun began to shine. Some of the mist nets were opened and after about one hour, one bird was caught. Surprisingly, this turned out to be a Greenish Warbler type. However, this bird clearly differed from the Greenish Warblers that are caught almost every year at Blåvand. The date was also quite early for Greenish Warbler at Blåvand, as most records of Greenish Warbler at Blåvand are from June. Because of the colouration, the bird was

Table 1. European records outside the peripheral breeding grounds of Green Warbler *Phylloscopus nitidus*.  
*Europeiska fynd utanför det perifera häckningsområdet av kaukasisk lundsångare Phylloscopus nitidus.*

\*Treatment/decision by national rarities committee. *Behandling/beslut av nationell raritetskommitté.*

\*\*Another record, made 2000-09-27 in Greece, was listed by Hudson (2010), but this has been rejected by the Hellenic Rarities Committee (Níkos Probonas *in litt.*). *Ytterligare ett fynd i Grekland 2000-09-27 rapporterades av Hudson (2010), men detta fynd har underkänts av den grekiska raritetskommittén (Níkos Probonas in litt.).*

Area, country	Site	Date (YMD)	RC treatment*	Sequenced	Reference
<i>Område, land</i>	<i>Lokal</i>	<i>Datum (ÅMD)</i>	<i>Rk-behandling*</i>	<i>Sekvenserad</i>	<i>Referens</i>
<b>Fennoscandia</b>					
Sweden	Ottenby, Öland	2003-05-29	accepted	yes	Irwin & Hellström 2007
Finland	Lågskär, Åland	2012-05-20	accepted	no	Väisänen et al. 2015
Denmark	Blåvand, Jylland	2015-05-27	pending	yes	this study
<b>Remaining NW Europe</b>					
Germany	Helgoland	1867-10-11	accepted	no	Gätkhe 1900
UK	St Mary, Isles of Scilly	1983-09-26	accepted	no	Hudson 2010
Faroe Islands	Nolsoy	1997-06-08	accepted	yes	Sørensen & Jensen 2001
UK	Foula, Shetland	2014-05-31	accepted	no	Pennington <i>in litt.</i>
UK	Unst, Shetland	2016-05-12	accepted	yes	Pennington 2016
<b>SE Europe</b>					
Greece**	Antikythira Island	1998-09-18	accepted	no	Hellenic Rarities Committee 2006 & 2009
Romania	Chituk	2016-10-05	pending	no	Milvus Group 2016

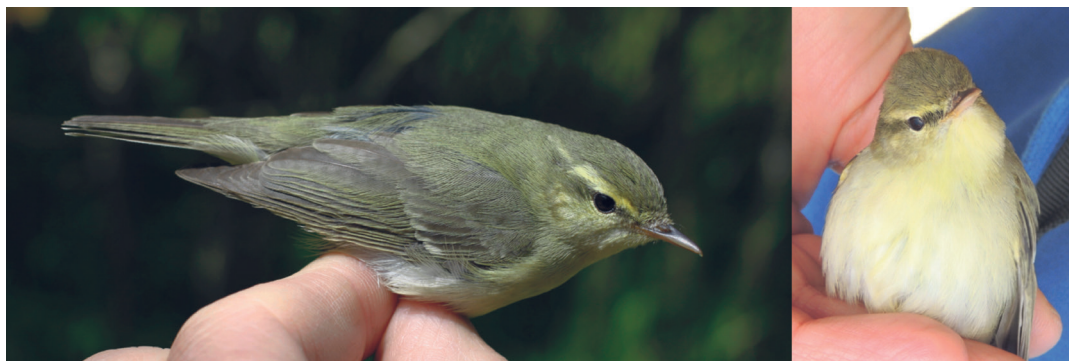


Figure 1. Green Warbler *Phylloscopus nitidus*, caught on 27 May 2015, in Blåvand, Denmark. Note rather green upperparts, one clearly visible wing-bar on the greater coverts, and yellowish breast, which are characters that distinguish it from Greenish Warbler *Ph. trochiloides* and Two-barred Warbler *Ph. plumbeitarsus*. Photos: Henrik Knudsen (left) and Morten Bentzon Hansen (right).

*Kaukasisk lundsångare Phylloscopus nitidus, fångad 27 maj 2015 i Blåvand, Danmark. Notera tämligen grön översida, endast ett synligt vingband längs de större armtäckarna samt gultonat bröst, vilka är karaktärer som skiljer den från lundsångare Ph. trochiloides och sibirisk lundsångare Ph. plumbeitarsus. Foton: Henrik Knudsen (vänster) och Morten Bentzon Hansen (höger).*

immediately suspected of being a probable Green Warbler.

The bird was measured and photographed (Figure 1). The wing length (maximum chord; Svensson 1992) was recorded at 61.5 mm, primary 3–4 formed the wing tip, and the tip of the second primary fell at the tip of primary 7. During the process, the bird shed two downs which were collected for DNA typing, as the species determination was not conclusive. After five hours, the bird was relocated and seen clearly in the field. The next day, however, the bird could not be found.

The colouration of the bird was quite different compared to a Greenish Warbler. The upperparts were bright green without the greyish wash, which is typical of Greenish Warbler. The supercilium was yellow, not white, as in Greenish Warbler. The throat and upper breast were yellow contrasting to the white lower breast and belly. The wing bar on the tip of the greater secondary coverts was prominent and broader compared to Greenish Warblers seen in the spring (Figure 1). In the hand, the size and shape of the bird was not perceived as different from Greenish Warbler, but on some photos the head looks larger and more robust than that of Greenish Warbler. The bird was not heard, but when a recording of Green Warbler song was played, the bird duly appeared. Regrettably, no recording of Greenish Warbler was played, so the potential response to that species is unknown.

## Methods

The bases of the collected downs were cut off and digested for three hours at 56 °C in 100 µl lysis buffer (0.1 M Tris, 0.005 EDTA, 0.2% SDS, 0.2 M NaCl, pH 8.5) with 1.5 µl proteinase K (10 mg/ml) and then precipitated with ethanol and eluted in 20 µl ddH<sub>2</sub>O. For typing of the mitochondrial cytochrome *b* (*cytb*) gene, we used the Qiagen Multiplex PCR Kit (Qiagen Inc.), with amplification reactions containing 5 µl Qiagen Multiplex PCR Master Mix, 0.2 µl each of 10 µM primers ND5-Syl (Stervander et al. 2015) and mtF-NP (Fregin et al. 2009), 2 µl template DNA (non-diluted elution), and 2.6 µl water. We ran the PCR reactions for activation at 95 °C for 15 min. Then followed 40 three-step cycles with denaturation at 94 °C for 30 s; annealing for 20 cycles in a touchdown profile decreasing by 0.5°C per cycle from 55°C, followed by 20 cycles at 45°C for 90 s; and extension at 72 °C for 90 s. Finally, we allowed extension at 72 °C for 10 min. PCR products were checked on a

1% agarose gel, precipitated with NH<sub>4</sub>Ac and ethanol, and then dissolved in 25 µl water. We used 2 µl for sequencing with the internal sequencing primer *Cytb\_seq\_H15541* (Stervander et al. 2015) with the BigDye sequencing kit (Applied Biosystems) in an ABI Prism 3100 capillary sequencer (Applied Biosystems).

The *cytb* sequence was manually inspected and edited in Geneious v. 10 (Biomatters). GenBank sequences of >900 bp were downloaded for all *Phylloscopus/Seicercus/Abrornis* (different genus names used for one monophyletic clade) and outgroups (*Cettia/Horornis*, *Aegithalos*, *Acrocephalus*, *Cisticola*, *Sylvia*), and aligned with the MAFFT v7.222 (Katoh et al. 2002) Geneious plugin. The 394 reference sequences included in this study are specified in Appendix 1.

Substitution models were evaluated with jModelTest v. 2.1.4 (Guindon & Gascuel 2003, Darriba et al. 2012), selecting from 88 available models allowing for rate heterogeneity according to four gamma categories and for a proportion of invariable sites. Model selection was performed per the Bayesian Information Criterion (BIC; Schwarz 1978).

*Cytb* gene trees were computed within a Bayesian inference (BI) framework with BEAST v. 2.4.4 (Bouckaert et al. 2014), using a Yule tree prior, and a strict molecular clock with a rate of 0.0105 substitution/site/lineage/million years, based on overall *cytb* substitution rates for a wide range of avian species (Weir & Schluter 2008). We applied four discrete categories over the gamma distribution, and estimated the frequency of invariant sites as well as the base frequencies. We performed two replicate runs, and sampled trees every 1,000 generations, over 30 million generations, of which the first 10% were discarded as burn-in. The results were inspected using Tracer v. 1.6 (Rambaut et al. 2013), ensuring stationarity and effective sample sizes (ESS) of >200, and ascertaining congruence between replicate runs.

The maximum clade credibility tree was calculated with TreeAnnotator (Bouckaert et al. 2014), using average node heights, and excluding 10% as burn-in. This tree was visualized and annotated in FigTree v.1.4.3 (Rambaut 2016) and Mega 7 (Kumar et al. 2016).

## Results

The trimmed sequence from the Blåvand bird was 681 bp, of which 633 bp covered parts of the *cytb* gene from its start, preceded by the last 40 bp of the ND5 gene and 8 intergenic bp. The *cytb* sequence

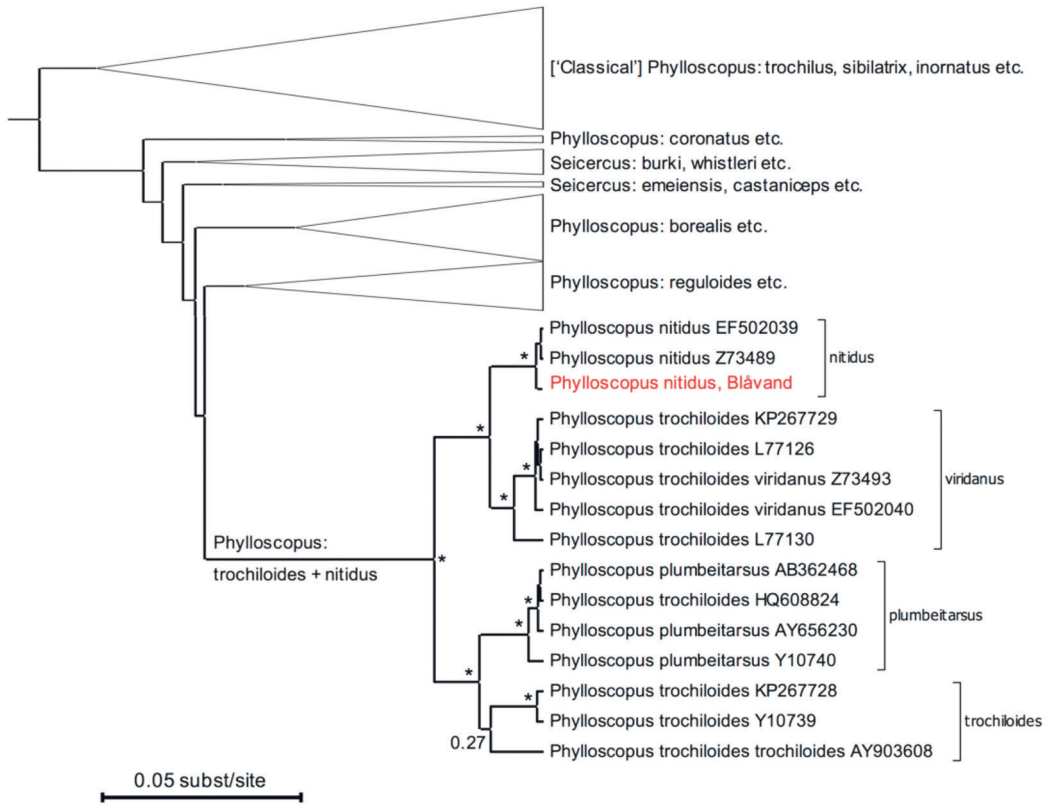


Figure 2. Phylogeny of *Seicercus* and *Phylloscopus* warblers, based on Bayesian inference of 394 cytochrome *b* sequences, which places the Blåvand bird (highlighted in red font) in the Green Warbler *Ph. nitidus* clade with maximal support. Sequences from the complex containing Green Warbler and Greenish Warbler *Ph. trochiloides* are shown with their taxon names as submitted to Genbank, with bracket annotations stating which clade they correspond to. Posterior probability (PP; 0.0–1.0) is indicated for nodes in this clade: \* indicates PP = 1.0, PP otherwise stated with a number at the node. All other clades have been collapsed, with the height of the triangle corresponding to number of sequences, and labels including taxa representative of the clade. For a full list of included taxa, and all Genbank accession numbers, see Appendix 1. Note that the traditional use of the genus names *Seicercus* and *Phylloscopus* does not reflect monophyletic groups. The outgroups have been removed for clarity.

Fylogeni över sångare inom släktena *Seicercus* och *Phylloscopus*, baserad på bayesiansk inferens av 394 cytokrom *b*-sekvenser, vilken placerar fågeln från Blåvand (markerad med röd text) i kladen med kaukasisk lundsångare *Ph. nitidus* med maximalt statistiskt stöd. Sekvenser från komplexet som omfattar kaukasisk lundsångare och lundsångare *Ph. trochiloides* visas namngivna såsom de skickats in till Genbank, med klamrar som indikerar vilken klad de tillhör. Posteriorisannolikhet (PP; 0,0–1,0) visas för noder inom denna klad: \* indikerar PP = 1,0, PP visas annars med en siffra vid noden. Alla andra klader har minimerats och triangelnas höjd motsvarar antalet sekvenser som ingår i kladen. Representerativa taxa har listats bredvid triangeln. För den fulla listan över inkluderade taxa, samt deras Genbank-nummer, se Appendix 1. Notera att den traditionella användningen av släktesnamnen *Seicercus* och *Phylloscopus* inte motsvarar monofyletiska grupper. Utgrupperna har uteläts från figuren för tydlighet.

aligned with no gaps to *cytb* sequences of related species, and is available at Genbank with the accession number MF188243.

The best estimated substitution model was HKY (Hasegawa et al. 1985) with rate variation following a discrete gamma distribution with four rate categories (G) and with an estimated fraction of invariant sites (I).

The computed *cytb* phylogeny groups the Blåvand bird with full statistical support (posterior probability 1.0) together with two sequences of Green Warblers in a *nitidus* clade, that is sister to the *viridanus* clade (Figure 2). Those two clades, in turn, make up as a sister clade to the clade containing the sisters *trochiloides* and *plumbeitarsus* (Figure 2).

## Discussion

Green Warbler may be an overlooked species in western and northern Europe because it is rather similar to Greenish Warbler. Even with good views the two species may be difficult to distinguish, and good views may be hard to achieve owing to the restless nature of the species and their habit of foraging high above the ground. However, very few Green Warblers have been caught in Europe compared to the number of Greenish Warblers, so the species is probably a quite rare vagrant in Europe.

The phylogenetic analysis clearly confirms that the Blåvand bird is a Green Warbler (Figure 2). It further highlights the special case that the Green/Greenish/Two-barred Warbler complex constitutes. Under a phylogenetic species concept, a species must be monophyletic, i.e. all of its subspecies/populations/individuals must be most closely related to each other, and together form a clade that contains no other taxa. However, the *cytb* phylogeny shows that the two Greenish Warbler clades (*trochiloides* and *viridanus*; Figure 2) are not each other's closest relatives, but instead make up sister clades with Two-barred Warbler (*plumbeitarsus*) and Green Warbler (*nitidus*), respectively. Thus, judging from the *cytb* phylogeny alone, Greenish Warbler is paraphyletic, and either all taxa within the complex represent one species, or all should be regarded separate species.

However, while mitochondrial markers such as *cytb* are appropriate for taxon identification, taxonomic conclusions should not rest solely on such information. Mitochondrial genetic markers differ from nuclear genetic markers because of a faster molecular substitution rate, and because of maternal inheritance. When populations diverge, mitochondrial and nuclear genetic markers may leave different phylogenetic signatures because they sort at different rates. Also, if diverged populations later come into secondary contact, mitochondrial and nuclear genetic markers will be differentially prone to introgression depending on the stage of divergence (Rheindt & Edwards 2011). For that reason, interpretations of phylogenetic relationship should best be based on multi-locus analyses including nuclear genetic markers (e.g. Rheindt & Edwards 2011, Stervander et al. 2015).

Further, the Greenish/Green/Two-barred Warbler clade is unusually well studied, and illustrates some of the problems highlighted above. The Greenish and Two-barred Warbler occur in a broken ring around the Tibetan plateau. The Two-barred Warbler occupies the north-eastern part of the ring,

and is isolated with a break in the ring to the south, while it meets the Greenish Warbler in a contact zone to the west. Despite the contact, the two species remain their integrity. The distribution of the Greenish warbler continues counter-clockwise around the Tibetan plateau until the gap the occurs east of the plateau, before reaching the distribution of the Two-barred Warbler. When Irwin et al. (2001) analysed divergence in song and mitochondrial DNA along the broken ring, they concluded that the complex had likely evolved along the ring. Neighbouring populations were only slightly differentiated, but the differentiation progressed on a gradient along the ring, and reproductive isolation was complete in the two end-points, represented by the Siberian populations of Greenish and Two-barred Warbler. However, when Alcaide et al. (2014) made use of “next generation sequencing” of massive amounts of nuclear DNA, they revealed that this interpretation was not correct. Instead they uncovered that there have been multiple historical breaks in gene flow, when populations have been isolated at several locations along the ring. Nevertheless, while this species complex may not represent a rare case of a “ring species”, it illustrates a complex evolutionary history resulting in varying levels of genetic differentiation between subspecies and species.

Finally, it is worth to mention that the two genera *Phylloscopus* and *Seicercus*, which presently constitute the family Phylloscopidae, are paraphyletic (Figure 2; Olsson et al. 2004). The family will therefore likely be split into multiple genera that better reflect their evolutionary relationships (P. Alström *pers. comm.*, Gill & Donsker 2017).

## Acknowledgements

Thanks to the late Martin Garner, Jochen Dierschke, Lars Svensson, Arnoud B. van den Berg, and Simon Sigaard Christiansen for initial comments about the identification of the bird. Sören Svensson and an anonymous referee gave valuable comments that improved the manuscript.

## Author contributions

H. K. trapped the bird and wrote the description; A. B. K. researched previous records and contributed to the manuscript; M. S. performed the lab work and the phylogenetic analyses, and wrote the manuscript.

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## Sammanfattning

Den 27 maj 2016 fångades och ringmärktes en lundsångarliknande fågel vid Blåvands fågelstation, som är belägen vid Danmarks västligaste punkt. Datumet är tidigare än tyngdpunkten för lundsångarfynd vid Blåvand, och fågeln avvek något i dräkt från lundsångare *Phylloscopus trochiloides*. Redan vid fångsten misstänktes att fågeln var en kaukasisk lundsångare *Ph. nitidus*, men dessa är

svåra att säkert bestämma enbart baserat på morfologi. Färgsättningen skilde sig jämfört med lundsångare på så sätt att den aktuella fågeln hade en tydligare grön ovalsida, utan gråton (Figur 1). Dessutom var ögonbrynsstrecket gult snarare än vitt och strupe samt övre bröst var gula, tydligt kontrasterande mot vitt nedre bröst och mage (Figur 1). Vingbandet, format av de större armtäckarnas ljusa spetsar, var bredare och tydligare jämfört med lundsångare (Figur 1).

Vid hanteringen tappade fågeln två dun, vilka användes för utvinning av DNA. Med hjälp av PCR mångfaldigades en DNA-sekvens från den maternellt nedärvda mitokondriegenen cytokrom *b*. En fylogenetisk analys av denna sekvens, tillsammans med alla publikt tillgängliga cytokrom *b*-sekvenser från fåglar inom släktena *Phylloscopus/Seicercus/Abrornis* (olika släktesnamn som används för en och samma monofyletiska klad) samt utgrupper (taxa från släktena *Cettia/Horornis*, *Aegithalos*, *Acrocephalus*, *Cisticola* och *Sylvia*) bekräftar att fågeln var en kaukasisk lundsångare (Figur 2).

Det fylogenetiska trädet (Figur 2) belyser ett par speciella förhållanden. *Phylloscopus* och *Seicercus* är båda parafyletiska släkten, dvs. de omfattar inte bara taxa som är närmast besläktade med varandra och bildar obrutna monofyletiska grupper. I stället förekommer arter som kallas *Phylloscopus* och *Seicercus* på olika ställen i fylogenin (Figur 2). Därmed är en taxonomisk revision att vänta för hela familjen Phylloscopidae. Dessutom uppvisar lundsångarkomplexet, med lundsångare, kaukasisk

lundsångare och sibirisk lundsångare *Ph. plumbeitarsus*, ett osedvanligt komplicerat inbördes förhållande. Medan den kaukasiska lundsångaren har ett isolerat utbredningsområde huvudsakligen i Kaukasus och norra Iran återfinns lundsångaren och den sibiriska lundsångaren öster därom i en bruten ring kring Tibetanska högplatån. De två arterna möts, utan att blandas, i östra Sibirien, medan urskiljbara populationer av lundsångaren förekommer med en kontinuerlig utbredning motsols till platåns östra sida. Tidigare, med stöd av analyser av mitokondriellt DNA och sång, har detta komplex tolkats som representanter för en s.k. ringart, där en art spridits och differentierats kontinuerligt längs en ring, så att ändarna, där de möts, är fullgoda arter, medan populationerna dem emellan endast skiljer sig gradvis. Nyare studier, baserade på analyser av stora delar av cellkärnans arvs massa hos flera av arterna, visar att det inte alls handlar om en ringart, utan att flera av populationerna runt ringen tidvis har varit isolerade. Hybridisering och introgression av genetiskt material mellan arterna har skapat ett komplext fylogenetiskt mönster, där det mitokondriella fylogenetiska trädet inte speglar arträdet (Figur 2).

Fyndet i Blåvand utgör Danmarks första fynd av kaukasisk lundsångare, Fennoskandias tredje – efter fynd på Öland den 29 maj 2003 och Åland den 20 maj 2012 – och Europas åttonde fynd (Tabell 1) utanför ett begränsat och perifert europeiskt häckningsområde i södra Ryssland och norra Georgien.

## Appendix 1

Taxa and GenBank accession number for cytochrome *b* sequences included in the phylogenetic analysis.  
*Taxa och GenBank-nummer för de cytokrom b-sekvenser som inkluderats i den fylogenetiska analysen.*

<b>Taxon</b>	<b>Accession no</b>	<b>Taxon</b>	<b>Accession no</b>
<b><i>Taxon</i></b>	<b><i>Genbank-nr</i></b>	<b><i>Taxon</i></b>	<b><i>Genbank-nr</i></b>
Abrornis chloronotus	KJ456391	Phylloscopus reguloides reguloides	AY656235
Abrornis chloronotus simlaensis	DQ008504	Phylloscopus reguloides reguloides	AY656236
Abrornis humei	KJ456394	Phylloscopus reguloides reguloides	AY656237
Abrornis humei humei	Z73488	Phylloscopus reguloides reguloides	AY656238
Abrornis inornata	AY635054	Phylloscopus reguloides ticehursti	AY656205
Abrornis inornata	DQ792799	Phylloscopus reguloides ticehursti	AY656206
Abrornis inornata	DQ792800	Phylloscopus reguloides ticehursti	AY656207
Abrornis inornata	KF742677	Phylloscopus reguloides ticehursti	AY656233
Abrornis inornata	L77135	Phylloscopus reguloides ticehursti	AY656234
Abrornis inornata	NC_024726	Phylloscopus reguloides ticehursti	AY656242
Abrornis inornata	Y10734	Phylloscopus reguloides ticehursti	AY656243
Abrornis maculipennis	HQ608828	Phylloscopus ricketti	AY656239
Abrornis maculipennis	KJ456395	Phylloscopus ricketti	GU045622
Abrornis maculipennis	Y10731	Phylloscopus ricketti	GU045623
Abrornis maculipennis maculipennis	AY635055	Phylloscopus ricketti	GU045624
Abrornis proregulus	AY635058	Phylloscopus ricketti	GU045625
Abrornis proregulus	DQ792796	Phylloscopus ruficapilla minullus	AY635060
Abrornis proregulus	DQ792797	Phylloscopus sarasinorum sarasinorum	AY656240
Abrornis proregulus	DQ792798	Phylloscopus schwarzi	AY635061
Abrornis proregulus	HQ608830	Phylloscopus schwarzi	HQ608825
Abrornis proregulus	L77134	Phylloscopus schwarzi	Y10728
Abrornis proregulus	Y10733	Phylloscopus sibilatrix	AY944178
Abrornis yunnanensis	HQ608833	Phylloscopus sibilatrix	L77123
Acrocephalus schoenobaenus	Z73475	Phylloscopus sibilatrix	Z73491
Acrocephalus scirpaceus scirpaceus	Z73483	Phylloscopus sibilatrix	L77136
Aegithalos concinnus	JX398849	Phylloscopus sindianus	L77136
Aegithalos concinnus concinnus	KF951091	Phylloscopus sindianus lorenzii	HQ706180
Cettia fortipes	L77122	Phylloscopus sindianus lorenzii	Z73478
Cisticola juncidis	Z73474	Phylloscopus subviridis	KJ456398
Horornis diphone	AB159194	Phylloscopus trivirgatus	L77145
Horornis diphone	AB159195	Phylloscopus trivirgatus trivirgatus	AY656244
Horornis diphone	AB159196	Phylloscopus trochiloides	HQ608824
Horornis diphone	AB159197	Phylloscopus trochiloides	KP267728
Horornis diphone	AB159198	Phylloscopus trochiloides	KP267729
Horornis diphone	AB694915	Phylloscopus trochiloides	L77126
Horornis diphone	AB694916	Phylloscopus trochiloides	L77130
Horornis diphone	AB694916	Phylloscopus trochiloides	Y10739
Horornis diphone	HQ608838	Phylloscopus trochiloides trochiloides	AY903608
Horornis seebohmi	AB281094	Phylloscopus trochiloides viridanus	EF502040
Horornis seebohmi	AB281095	Phylloscopus trochiloides viridanus	Z73493
Horornis seebohmi	AB281096	Phylloscopus trochilus	AJ004326
Phylloscopus affinis	DQ008503	Phylloscopus trochilus	AY944177
Phylloscopus affinis	KJ456389	Phylloscopus trochilus	DQ174582
Phylloscopus affinis	L77128	Phylloscopus trochilus	DQ174583
Phylloscopus affinis	Y10730	Phylloscopus trochilus	DQ174584
Phylloscopus amoenus	AY887676	Phylloscopus trochilus	DQ174585
Phylloscopus armandii	HQ608831	Phylloscopus trochilus	DQ174586
Phylloscopus bonelli	AY944179	Phylloscopus trochilus	DQ174587
Phylloscopus bonelli	L77127	Phylloscopus trochilus	DQ174588
Phylloscopus bonelli bonelli	Z73485	Phylloscopus trochilus	DQ174589
Phylloscopus bonelli bonelli	Z73486	Phylloscopus trochilus	DQ174590



*Phylloscopus borealoides* AB362459  
*Phylloscopus borealoides* AB362467  
*Phylloscopus borealoides* AY887677  
*Phylloscopus brehmi* AJ004325  
*Phylloscopus brehmi* Z73476  
*Phylloscopus calciatilis* GU045613  
*Phylloscopus calciatilis* GU045614  
*Phylloscopus calciatilis* GU045615  
*Phylloscopus calciatilis* GU045616  
*Phylloscopus calciatilis* GU045617  
*Phylloscopus calciatilis* GU045618  
*Phylloscopus calciatilis* GU045619  
*Phylloscopus calciatilis* GU045620  
*Phylloscopus calciatilis x ricketti* GU045621  
*Phylloscopus canariensis* Z73477  
*Phylloscopus cantator* KJ456390  
*Phylloscopus cantator cantator* AY656208  
*Phylloscopus cebuensis* JN827147  
*Phylloscopus cebuensis* L77144  
*Phylloscopus claudiae* AY656209  
*Phylloscopus claudiae* AY656210  
*Phylloscopus claudiae* AY656211  
*Phylloscopus claudiae* AY656212  
*Phylloscopus claudiae* AY656213  
*Phylloscopus collybita* AJ004324  
*Phylloscopus collybita* HQ608821  
*Phylloscopus collybita* L77125  
*Phylloscopus collybita abietinus* DQ174604  
*Phylloscopus collybita abietinus* DQ174605  
*Phylloscopus collybita abietinus* DQ174606  
*Phylloscopus collybita abietinus* DQ174607  
*Phylloscopus collybita abietinus* DQ174608  
*Phylloscopus collybita abietinus* HQ121526  
*Phylloscopus collybita abietinus* Z73479  
*Phylloscopus collybita brevirostris* Z73480  
*Phylloscopus collybita caucasicus* Z73481  
*Phylloscopus collybita collybita* Z73487  
*Phylloscopus collybita menzbieri* AF136374  
*Phylloscopus collybita tristis* HF562844  
*Phylloscopus collybita tristis* HF562845  
*Phylloscopus collybita tristis* HF562846  
*Phylloscopus collybita tristis* Z73482  
*Phylloscopus coronatus* AB362460  
*Phylloscopus coronatus* AY635053  
*Phylloscopus coronatus* HQ608834  
*Phylloscopus coronatus* L77139  
*Phylloscopus davisoni* AY656214  
*Phylloscopus davisoni disturbans* AY656217  
*Phylloscopus davisoni klossi* AY656215  
*Phylloscopus emeiensis* AY656218  
*Phylloscopus fulgiventis* KJ456392  
*Phylloscopus fuscatus* DQ119527

*Phylloscopus trochilus* DQ174591  
*Phylloscopus trochilus* DQ174592  
*Phylloscopus trochilus* DQ174593  
*Phylloscopus trochilus* DQ174594  
*Phylloscopus trochilus* DQ174595  
*Phylloscopus trochilus* DQ174596  
*Phylloscopus trochilus* DQ174597  
*Phylloscopus trochilus* DQ174598  
*Phylloscopus trochilus* JX869889  
*Phylloscopus trochilus* L77124  
*Phylloscopus trochilus trochilus* DQ673875  
*Phylloscopus trochilus trochilus* Z73492  
*Phylloscopus tytleri* AY887679  
*Phylloscopus tytleri* L77132  
*Seicercus affinis* KJ456460  
*Seicercus affinis intermedius* AY635066  
*Seicercus affinis intermedius* AY635069  
*Seicercus affinis intermedius* AY635070  
*Seicercus affinis intermedius* AY635071  
*Seicercus affinis ocularis* AY635063  
*Seicercus borealis* HQ608832  
*Seicercus borealis* L77138  
*Seicercus borealis* L77143  
*Seicercus borealis borealis* AB362438  
*Seicercus borealis borealis* AB362439  
*Seicercus borealis borealis* AB362440  
*Seicercus borealis borealis* AB362441  
*Seicercus borealis borealis* AB362442  
*Seicercus borealis borealis* AB362443  
*Seicercus borealis borealis* AB362444  
*Seicercus borealis borealis* AB362445  
*Seicercus borealis borealis* AB362446  
*Seicercus borealis borealis* AB362462  
*Seicercus borealis borealis* AB530997  
*Seicercus borealis borealis* AB530998  
*Seicercus borealis borealis* AB530999  
*Seicercus borealis borealis* AB531000  
*Seicercus borealis borealis* AB531001  
*Seicercus borealis borealis* AB531002  
*Seicercus borealis borealis* AB531003  
*Seicercus borealis borealis* AY635052  
*Seicercus borealis borealis* HQ243658  
*Seicercus borealis borealis* HQ243659  
*Seicercus borealis borealis* HQ243660  
*Seicercus borealis borealis* KP245888  
*Seicercus borealis borealis* Z73484  
*Seicercus borealis kennicotti* AB362435  
*Seicercus borealis kennicotti* AB362436  
*Seicercus borealis kennicotti* AB362437  
*Seicercus borealis kennicotti* AB362461  
*Seicercus burkii* AY635064  
*Seicercus burkii* AY635065

<i>Phylloscopus fuscatus</i>	DQ174599	<i>Seicercus burkii</i>	HQ608856
<i>Phylloscopus fuscatus</i>	DQ174600	<i>Seicercus castaniceps</i>	AY887680
<i>Phylloscopus fuscatus</i>	DQ174601	<i>Seicercus castaniceps</i>	HQ608857
<i>Phylloscopus fuscatus</i>	DQ174602	<i>Seicercus castaniceps</i>	KJ456461
<i>Phylloscopus fuscatus</i>	DQ174603	<i>Seicercus castaniceps castaniceps</i>	AY635067
<i>Phylloscopus fuscatus</i>	HQ608823	<i>Seicercus examinandus</i>	AB362424
<i>Phylloscopus fuscatus</i>	Y10729	<i>Seicercus examinandus</i>	AB362425
<i>Phylloscopus goodsoni fokiensis</i>	AY656219	<i>Seicercus examinandus</i>	AB362426
<i>Phylloscopus goodsoni fokiensis</i>	AY656220	<i>Seicercus examinandus</i>	AB362427
<i>Phylloscopus goodsoni fokiensis</i>	AY656221	<i>Seicercus examinandus</i>	AB362428
<i>Phylloscopus goodsoni fokiensis</i>	AY656222	<i>Seicercus examinandus</i>	AB362429
<i>Phylloscopus goodsoni fokiensis</i>	AY656223	<i>Seicercus examinandus</i>	AB362430
<i>Phylloscopus goodsoni goodsoni</i>	AY656225	<i>Seicercus examinandus</i>	AB362431
<i>Phylloscopus goodsoni goodsoni</i>	AY656226	<i>Seicercus examinandus</i>	AB362432
<i>Phylloscopus goodsoni goodsoni</i>	AY656227	<i>Seicercus examinandus</i>	AB362433
<i>Phylloscopus goodsoni goodsoni</i>	AY656228	<i>Seicercus examinandus</i>	AB362434
<i>Phylloscopus griseolus</i>	KJ456393	<i>Seicercus examinandus</i>	AB362463
<i>Phylloscopus hainanus</i>	AY656229	<i>Seicercus examinandus</i>	AB362464
<i>Phylloscopus magnirostris</i>	AY635056	<i>Seicercus examinandus</i>	KP245873
<i>Phylloscopus magnirostris</i>	AY887681	<i>Seicercus examinandus</i>	KP245874
<i>Phylloscopus magnirostris</i>	HQ608822	<i>Seicercus examinandus</i>	KP245875
<i>Phylloscopus magnirostris</i>	L77129	<i>Seicercus examinandus</i>	KP245876
<i>Phylloscopus magnirostris</i>	Y10737	<i>Seicercus examinandus</i>	KP245877
<i>Phylloscopus nigrorum benguensis</i>	AY656204	<i>Seicercus examinandus</i>	KP245878
<i>Phylloscopus nitidus</i>	EF502039	<i>Seicercus examinandus</i>	KP245879
<i>Phylloscopus nitidus</i>	Z73489	<i>Seicercus examinandus</i>	KP245880
<i>Phylloscopus occipitalis</i>	AY635057	<i>Seicercus examinandus</i>	KP245881
<i>Phylloscopus occipitalis</i>	EU372678	<i>Seicercus examinandus</i>	KP245882
<i>Phylloscopus occipitalis</i>	L77131	<i>Seicercus examinandus</i>	KP245883
<i>Phylloscopus occipitalis</i>	Y10735	<i>Seicercus examinandus</i>	KP245884
<i>Phylloscopus occisinensis</i>	HQ608829	<i>Seicercus examinandus</i>	KP245885
<i>Phylloscopus ogilviegranti ogilviegranti</i>	AY656216	<i>Seicercus examinandus</i>	KP245886
<i>Phylloscopus orientalis</i>	AY887678	<i>Seicercus examinandus</i>	KP245887
<i>Phylloscopus orientalis</i>	Z73490	<i>Seicercus examinandus</i>	KP245895
<i>Phylloscopus plumbeitarsus</i>	AB362468	<i>Seicercus examinandus</i>	KP245896
<i>Phylloscopus plumbeitarsus</i>	AY656230	<i>Seicercus examinandus</i>	KP245897
<i>Phylloscopus plumbeitarsus</i>	Y10740	<i>Seicercus examinandus</i>	KP245898
<i>Phylloscopus poliocephalus giulianetti</i>	AY656224	<i>Seicercus grammiceps grammiceps</i>	AY635068
<i>Phylloscopus pulcher</i>	HQ608826	<i>Seicercus ijimae</i>	L77141
<i>Phylloscopus pulcher</i>	KJ456396	<i>Seicercus ijimae</i>	Y10741
<i>Phylloscopus pulcher</i>	KJ567540	<i>Seicercus montis montis</i>	AY635073
<i>Phylloscopus pulcher</i>	KJ567541	<i>Seicercus omeiensis</i>	AY635078
<i>Phylloscopus pulcher</i>	KJ567542	<i>Seicercus poliogenys</i>	AY635079
<i>Phylloscopus pulcher</i>	KJ567543	<i>Seicercus poliogenys</i>	AY635080
<i>Phylloscopus pulcher</i>	KJ567544	<i>Seicercus poliogenys</i>	AY635081
<i>Phylloscopus pulcher</i>	KJ567545	<i>Seicercus poliogenys</i>	KJ456462
<i>Phylloscopus pulcher</i>	KJ567546	<i>Seicercus soror</i>	AY635082
<i>Phylloscopus pulcher</i>	KJ567547	<i>Seicercus soror</i>	AY635083
<i>Phylloscopus pulcher</i>	KJ567548	<i>Seicercus soror</i>	KM875497
<i>Phylloscopus pulcher</i>	KJ567549	<i>Seicercus tenellipes</i>	AY903607
<i>Phylloscopus pulcher</i>	KJ567550	<i>Seicercus tenellipes</i>	L77140
<i>Phylloscopus pulcher</i>	KJ567551	<i>Seicercus tenellipes</i>	Y10738

<i>Phylloscopus pulcher</i>	KJ567552	<i>Seicercus tephrocephalus</i>	AY635084
<i>Phylloscopus pulcher</i>	KJ567553	<i>Seicercus tephrocephalus</i>	AY635085
<i>Phylloscopus pulcher</i>	KJ567554	<i>Seicercus tephrocephalus</i>	HQ706182
<i>Phylloscopus pulcher</i>	KJ567555	<i>Seicercus umbrovirens mackenzianus</i>	AY635062
<i>Phylloscopus pulcher</i>	KJ567556	<i>Seicercus valentini latouchei</i>	AY635072
<i>Phylloscopus pulcher</i>	KJ567557	<i>Seicercus valentini latouchei</i>	AY635086
<i>Phylloscopus pulcher</i>	KJ567558	<i>Seicercus valentini latouchei</i>	AY635088
<i>Phylloscopus pulcher</i>	KJ567559	<i>Seicercus valentini valentini</i>	AY635087
<i>Phylloscopus pulcher</i>	KJ567560	<i>Seicercus valentini valentini</i>	AY635089
<i>Phylloscopus pulcher</i>	KJ567561	<i>Seicercus whistleri</i>	KJ456463
<i>Phylloscopus pulcher</i>	KJ567562	<i>Seicercus whistleri nemoralis</i>	AY635074
<i>Phylloscopus pulcher</i>	KJ567563	<i>Seicercus whistleri nemoralis</i>	AY635076
<i>Phylloscopus pulcher</i>	KJ567564	<i>Seicercus whistleri whistleri</i>	AY635075
<i>Phylloscopus pulcher</i>	KJ567565	<i>Seicercus whistleri whistleri</i>	AY635077
<i>Phylloscopus pulcher</i>	KJ567566	<i>Seicercus whistleri whistleri</i>	AY635090
<i>Phylloscopus pulcher</i>	KJ567567	<i>Seicercus xanthodryas</i>	AB362447
<i>Phylloscopus pulcher</i>	KJ567568	<i>Seicercus xanthodryas</i>	AB362448
<i>Phylloscopus pulcher</i>	KJ567569	<i>Seicercus xanthodryas</i>	AB362449
<i>Phylloscopus pulcher</i>	KJ567570	<i>Seicercus xanthodryas</i>	AB362450
<i>Phylloscopus pulcher</i>	KJ567571	<i>Seicercus xanthodryas</i>	AB362451
<i>Phylloscopus pulcher</i>	KJ567572	<i>Seicercus xanthodryas</i>	AB362452
<i>Phylloscopus pulcher</i>	KJ567573	<i>Seicercus xanthodryas</i>	AB362453
<i>Phylloscopus pulcher</i>	KJ567574	<i>Seicercus xanthodryas</i>	AB362454
<i>Phylloscopus pulcher</i>	KJ567575	<i>Seicercus xanthodryas</i>	AB362455
<i>Phylloscopus pulcher</i>	KJ567576	<i>Seicercus xanthodryas</i>	AB362456
<i>Phylloscopus pulcher</i>	KJ567577	<i>Seicercus xanthodryas</i>	AB362457
<i>Phylloscopus pulcher</i>	KJ567579	<i>Seicercus xanthodryas</i>	AB362458
<i>Phylloscopus pulcher</i>	KJ567580	<i>Seicercus xanthodryas</i>	AB362465
<i>Phylloscopus pulcher</i>	KJ567581	<i>Seicercus xanthodryas</i>	AB362466
<i>Phylloscopus pulcher</i>	KJ567582	<i>Seicercus xanthoschistos tephrodiras</i>	AY656246
<i>Phylloscopus pulcher</i>	KJ567583	<i>Seicercus xanthoschistos xanthoschistos</i>	AY635091
<i>Phylloscopus pulcher</i>	KJ567584	<i>Sylvia atricapilla atricapilla</i>	Z73494
<i>Phylloscopus pulcher</i>	KJ567585	<i>Sylvia melanocephala</i>	L77121
<i>Phylloscopus pulcher</i>	KJ567586		
<i>Phylloscopus pulcher</i>	KJ567587		
<i>Phylloscopus pulcher</i>	KJ567588		
<i>Phylloscopus pulcher</i>	KJ567589		
<i>Phylloscopus pulcher</i>	KJ567590		
<i>Phylloscopus pulcher</i>	KJ567592		
<i>Phylloscopus pulcher</i>	KJ567594		
<i>Phylloscopus pulcher</i>	KJ567595		
<i>Phylloscopus pulcher</i>	KJ567596		
<i>Phylloscopus pulcher</i>	L77133		
<i>Phylloscopus pulcher</i>	Y10732		
<i>Phylloscopus pulcher pulcher</i>	AY635059		
<i>Phylloscopus reguloides</i>	HQ608827		
<i>Phylloscopus reguloides</i>	KJ456397		
<i>Phylloscopus reguloides</i>	KM248527		
<i>Phylloscopus reguloides</i>	KM248529		
<i>Phylloscopus reguloides</i>	Y10736		
<i>Phylloscopus reguloides assamensis</i>	AY656231		
<i>Phylloscopus reguloides assamensis</i>	AY656232		