1. Introduction

Why is the Song of Birds Important?

The songs of birds have inspired great musicians, poets, writers and ordinary mortals alike (Scholes 1970; Campbell & Lack 1985). The prospect of a "Silent Spring" has urged many people to take a profound look at the importance of the natural world, ecology and the destructive powers of *Homo sapiens* (Carson 1965; Terborgh 1990). In the words of Rachel Carson: "There was a strange stillness. The birds, for example – where had they gone? Many people spoke of them, puzzled and disturbed. The feeding stations in the backyards were deserted. The few birds seen anywhere were moribund; they trembled violently and could not fly. It was a spring without voices. On the mornings that had once throbbed with the dawn chorus of robins, catbirds, doves, jays, wrens, and scores of other bird voices there was now no sound; only silence lay over the fields and woods and marsh."

The singing and songs of birds have inspired and captured the attention of many naturalists and scientists. Darwin (1871), for example, wrote, "Naturalists are much divided with respect to the object of the singing of birds." One of the reasons for this inspiration and captivation is due to great variation in the acoustic structure of song, in combination with questions like those posed by Tinbergen (1963) about causation, survival value, ontogeny and evolution. Songs, the signal units usually comprising these performances (Smith 1991), vary from simple, stereotyped and discrete forms found in some species through a spectrum of variation to extremes of complexity, variability and continuity found in others. This broad spectrum has provided fertile ground for scientific research, which has developed into a popular and active field of science (Kroodsma & Miller 1982, 1996; Catchpole & Slater 1995). Enormous progress has been made in this field since early pioneering days (Witchell 1896), and impressive advances have occurred since the works of Thorpe (1961), Hinde (1969), Armstrong (1973), Hartshorne (1973) and Thielcke (1976). These advances include the testing of hypotheses about the functions of song in birds. Evidence generally favours the dual hypothesis of territorial defence or rival male repulsion and mate attraction, but there is also support for conspecific recognition, individual recognition, the signalling of male and territory quality, female mate choice, male-male competition, female stimulation, pair-bonding and keeping contact (Catchpole & Slater 1995). Research on the singing and song of birds continues to generate and test hypotheses, which address questions within the broader science of animal communication (Sebeok 1977; Smith 1977; Halliday & Slater 1983; Hauser 1996; Bradbury & Vehrencamp 1998), animal behaviour (Hind 1982; Dawkins 1986; Alcock 1993), behavioural ecology (Krebs & Davies 1993, 1997) and sociobiology (Wilson 1975). It is clear from these major reference works that research on the singing and song of birds has provided, and continues to provide insight into unravelling the complexities of learning, development, sound production and reception, physiology, anatomy, neurobiology and evolution.

There is no question that the study of the singing and song of birds is science, and hence it must satisfy the basic characteristics and criteria of science, which are explained by Chalmers (1982) and referred to by distinguished scientists, like Schrödinger (1992). No field of scientific endeavour is perfect however, and this field of study is no exception. My thesis draws attention to some fundamental aspects that are poorly defined and described, and provides suggestions for solving some of these deficiencies.

What is Bird "Song"?

Throughout my thesis, "a song" or "songs" is used when referring to the signal units that usually comprise singing performances, whereas "singing" is used when referring to the activity of performing songs. This makes the important operational distinction between songs and singing, which are two different classes of signalling (Smith 1991).

The great variety of vocalisations produced by birds usually are divided into calls and songs by researchers studying bird acoustic communication (Thorpe 1961; Catchpole & Slater 1995; Kroodsma & Miller 1996). In a review of over 80 definitions of "bird song" however, Spector (1994) found the application of a wide range of defining criteria and an absence of consensus between and clarity within authors about what defines "song" or differentiates it from calls. Furthermore, calls given in a particular context in some species are identical to songs, e.g., the contact call in the Black-cheecked Waxbill, *Estrilda erythronotos* (Goodwin 1982), while in other species, songs are produced that are inconsistently distinct from calls, e.g., natural hybrids of the humming bird species, *Calypte anna* and *Selasphorus sasin* (Wells & Baptista 1979). There also are exceptions to standard definitions of "song", e.g., *Poecile* tit vocalisations, which vary considerably in structure and are similar to calls, songs and mixtures of the two (Ficken, Ficken & Witkin 1978; Gaddis 1985; Hailman 1989; Hailman & Ficken 1996). The latter two studies have cautioned against pre-conceived ideas of dividing bird vocalisations into calls and songs and against a unitary view of song, which they argue, obscure evidence for different types of vocalisation and their specific uses and functions in these tits. If call-song terminology inhibits our understanding of bird vocalisations and confuses underlying concepts, e.g., evolutionary function (*sensu* Williams 1966), then the issue should continue to be addressed in a constructive way (Spector 1994), using ideas proposed by, for example, Tinbergen (1963) and Beer (1982).

A description of songs and their differentiation from calls are necessary as a working hypothesis because part of my thesis deals with song structure. They are best given specifically in terms of the subject of my thesis, the European Sedge Warbler (*Acrocephalus schoenobaenus*), which hereafter will be referred to as the Sedge Warbler.

It is evident from field observations (Wilson, unpublished data) and other studies (Géroudet 1963; Sauer & Sauer 1960; Bergmann & Helb 1982; Simms 1985), that the calls of male and female Sedge Warblers are simple and short, are composed of one to a few discrete units called syllables, vary up to several seconds in duration, and are given in particular contexts, e.g., contact, alarm and copulation. In contrast, male advertising songs of this species have been described as some of the longest and most variable and complex among the oscines, and are thought to be performed for mate attraction (Catchpole 1973, 1976, 1980, 1982, 2000; Catchpole & Slater 1995). Catchpole has described singing in the Sedge Warbler as spontaneous, performed with a characteristic diurnal rhythm, and as some of the most sustained vocalising in the oscines. Male advertising songs are reported to be composed of sequences of many syllables of different types that are combined in an almost infinite number of different ways (Catchpole 1976). My study is based on advertising songs (Tinbergen 1939) produced by males at high intensity in their territories during the breeding season, prior to pairing with a female (Catchpole 1973, 1976). Advertising songs include songs delivered from song posts and during special display flights called song flights (Catchpole 1973, 1976; Heuwinkel 1985; Buchanan & Catchpole 1997; Wilson, unpublished data). These songs have been described as audibly distinct from lower intensity songs such as "sub-song" (Thorpe & Pilcher 1958; Catchpole 1973; Marler & Peters 1982), short songs given by males to their mates (Brown & Davies 1949; Catchpole 1973), spring migrant songs (Géroudet 1963), winter quarter songs (Sauer & Sauer 1960; Ruwet 1965; but see Immelmann & Sossinka 1973/4) and short female songs (Howard 1907-14). It is not known how advertising songs differ in acoustic structure from these putative kinds of songs, besides general qualitative accounts of differences in variation, duration and intensity.

Why the European Sedge Warbler?

Most of the fascinating work on songs and singing in birds has been on species with discrete songs comprised of one or more types, e.g., the White-crowned Sparrow (*Zonotrichia leucophrys*) with one type (Baptista 1975), the Great Tit (*Parus major*) with two to eight types (Krebs, Ashcroft & Webber 1978) and the legendary Common Nightingale (*Luscinia megarhynchos*) with around 200 types (Todt 1971; Todt & Hultsch 1996). Song types performed by an individual bird make up its song type repertoire (Shiovitz 1975; Kroodsma 1982; Thompson, LeDoux & Moody 1994). The use of song types as units of vocal organisation depends on the presence of discrete songs with an apparent finite number of syllable type combinations, although there is important ongoing work and debate about intra- versus inter-song type variation (Kroodsma 1982; Table 1). There has been a growing emphasis on determining how the birds themselves categorise conspecific songs compared to classifications developed by researchers (Table 2). Recently, male response was tested to differing degrees of song type similarity in Song Sparrow (*Melospiza melodia*) vocal repertoires, and song types were reported to function as fundamental units (Searcy, Nowicki & Peters 1999). The way in which song types are organised in singing performances varies within and among species, but there are three broad categories of organisation. Some species usually sing with eventual variety, repeating the same song type before switching to another one, e.g., the Song Sparrow (Saunders 1951) and Chaffinch (*Fringilla coelebs*, Slater 1983), while others usually sing with immediate variety, performing a succession of different song types, e.g., the Common Nightingale (Hultsch & Todt 1989 a, b) and Long-billed Marsh Wren (*Cistothorus palustris*, Verner 1976). Some species sing with eventual or immediate variety depending on certain factors, e.g., Grace's Warbler (*Dendroica graciae*, Staicer 1989), the Yellow Warbler (*Dendroica petechia*, Spector 1991) and Banded Wren (*Thryothorus pleurostictus*, Molles & Vehrencamp 1999).

In sharp contrast to species with definite, discrete songs, much less work has been conducted on more continuous singers, like the Sedge Warbler and Brown Thrasher, *Toxostoma rufum* (Table 3). These two species, and others like the Sedge Warbler's close relatives which have been described as lacking discrete songs (Catchpole 1980), namely the Marsh Warbler (*Acrocephalus palustris*), Reed Warbler (*A. scirpaceus*) and Moustached Warbler (*A. melanopogon*), pose difficult problems of song classification (Kroodsma 1982).

Table 1. Studies examining variation within and/or between song types in various species of songbirds.

Species	Author
Great Tit (*Parus major*)	Lambrechts & Dhondt 1990.
Black-capped Chickadee (*Parus atricapillus*)	Ratcliffe & Weisman 1985.
	Nowicki & Nelson 1990.
	Horn, Leonard, Ratcliffe, Shackleton & Weisman 1992.
Mountain Chickadee (*Parus gambeli*)	Gaddis 1985.
Winter Wren (*Troglodytes troglodytes*)	Van Horne 1995.
Marsh Wren (*Cistothorus palustris*)	Kroodsma & Verner 1978.
Banded Wren (*Thryothorus pleurostictus*)	Molles & Vehrencamp 1999.
Varied Thrush (*Zoothera naevia*)	Whitney 1981.
European Starling (*Sturnus vulgaris*)	Adret-Hausberger 1989.
	Adret-Hausberger & Jenkins 1988.
Solitary Vireo (*Vireo solitarius*)	Martindale 1982.
Yellow Warbler (*Dendroica petechia*)	Spector 1991.
Grace's Warbler (*Dendroica graciae*)	Staicer 1989.
Chestnut-sided Warbler (*Dendroica pennsylvanica*)	Lein 1978.
Bobolink (*Dolichonyx oryzivorus*)	Bakker, Withrow & Thompson 1983.
	Trainer & Peltz 1996.
Western Meadowlark (*Sturnella neglecta*)	Horn & Falls 1988.
Yellow-headed Blackbird (*Xanthocephalus xanthocephalus*)	Cosens & Falls 1984.
Dark-eyed Junco (*Junco hyemalis*)	Williams & MacRoberts 1977.
Field Sparrow (*Spizella pusilla*)	Nelson 1989.
	Nelson & Croner 1991.
Harris' Sparrow (*Zonotrichia querula*)	Shackleton, Ratcliffe, Horn & Naugler 1991
Nuttall's White-crowned Sparrow (*Zonotrichia leucophrys nuttalli*)	Cunningham & Baker 1989.
Fox Sparrow (*Passerella iliaca*)	Martin 1977.
Swamp Sparrow (*Melospiza georgiana*)	Marler & Pickert 1984.
	Clark, Marler & Beeman 1987.
	Nelson & Marler 1989.
Song Sparrow (*Melospiza melodia*)	Podos, Peters, Rudnicky, Marler & Nowicki 1992.
	Nowicki, Podos & Valdes 1994.
	Hughes, Nowicki, Searcy & Peters 1998.
Zebra Finch (*Taeniopygia guttata castanotis*)	Zann 1993.
Darwin's Finches (*Geospiza conirostris*)	Grant 1984.
Village Indigobird (*Vidua chalybeata*)	Payne 1979.
Corn Bunting (*Miliaria calandra*)	Holland, McGregor & Rowe 1996.
Snow Bunting (*Plectrophenax nivalis*)	Espmark 1995.

Table 2. Studies investigating how songbirds categorise conspecific songs or their parts or components compared to classifications developed by researchers.

Species	**Author**
Great Tit (*Parus major*)	Falls, Krebs & McGregor 1982.
	Weary 1989, 1990, 1991.
	Weary, Falls & McGregor 1990.
	Weary, Norris & Falls 1990.
	Weary & Krebs 1992.
Black-capped Chickadee (*Parus atricapillus*)	Weisman & Ratcliffe 1989.
	Horn, Leonard, Ratcliffe, Shackleton & Weisman 1992.
European Blackbird (*Turdus merula*)	Wolffgramm & Todt 1982.
	Dabelsteen & Pedersen 1988a, 1992, 1993.
Veery (*Catharus fuscescens*)	Weary, Lemon & Date 1986.
Golden-winged Warbler (*Vermivora chrysoptera*)	Ficken & Ficken 1973.
Common Yellowthroat (*Geothlypis trichas*)	Wunderle 1979.
Kentucky Warbler (*Oporornis formosus*)	Morton & Young 1986.
American Redstart (*Setophaga ruticilla*)	Date, Lemon, Weary & Richter 1991.
Willow Warbler (*Phylloscopus trochilus*)	Järvi, Rådesäter & Jakobsson 1980.
Great Reed Warbler (*Acrocephalus arundinaceus*)	Catchpole 1983.
Aquatic Warbler (*Acrocephalus paludicola*)	Catchpole & Leisler 1989, 1996.
	Schmidt, Schaefer & Leisler 1999.
Western Meadowlark (*Sturnella neglecta*)	Falls 1985.
	Falls, Horn & Dickinson 1988.
	Horn & Falls 1988; 1991.
Eastern Meadowlark (*Sturnella magna*)	Falls, Dickinson & Krebs 1990.
Red-winged Blackbird (*Agelaius phoeniceus*)	Sinnott 1980.
	Brenowitz 1982, 1983.
	Searcy & Brenowitz 1988.
	Searcy, Coffman & Raikow 1994.
Cowbird (*Molothrus ater*)	Sinnott 1980.
	West, King & Eastzer 1981.
	Dufty & Hanson 1999.
Indigo Bunting (*Passerina cyanea*)	Shiovitz 1975.
Field Sparrow (*Spizella pusilla*)	Nelson 1988, 1989.
	Nelson & Croner 1991.
Harris' Sparrow (*Zonotrichia querula*)	Shackleton, Ratcliffe, Horn & Naugler 1991.
Swamp Sparrow (*Melospiza georgiana*)	Nelson & Marler 1989.
Song Sparrow (*Melospiza melodia*)	Searcy & Marler 1981.
	Searcy 1984.
	McArthur 1986.
	Stoddard, Beecher & Willis 1988.
	Stoddard, Beecher, Campbell & Horning 1992.
	Stoddard, Beecher, Loesche & Campbell 1992.
	Horning, Beecher, Stoddard & Campbell 1993.
	Beecher, Campbell & Burt 1994.
	Searcy, Podos, Peters & Nowicki 1995.
	Searcy, Nowicki & Peters 1999.
Canary (*Serinus canaria*)	Vallet & Kreutzer 1995.
	Vallet, Beme & Kreutzer 1998.
	Kreutzer, Beme, Vallet & Kiosseva 1999.

In the Sedge Warbler, syllable types and not song types have been used as vocal units comprising a male's repertoire, as songs are thought to be composed of a sustained succession of syllables produced with an open-ended number of type combinations (Catchpole 1976). In discussing similarities and differences between particular songs in one male, Catchpole (1976) remarked: "This is the closest to repetition of a song type that searching by audition has yet revealed. Indeed, if all the known variations in song duration, syllable repertoire, numbers, intervals, repetition, alternation and sequencing are considered, the number of permutations for song types become astronomical, and the probability of repetition remote." It is this variability that has led to the Sedge Warbler's song being perceived as extremely elaborate and complex and being referred to as an acoustic analogue of the peacock's tail (Catchpole 1986, 1987, 1988, 2000).

The Sedge Warbler is a suitable candidate to test the hypothesis that females prefer males with more elaborate and complex song (Darwin 1871). Field studies have reported a significant correlation between syllable repertoire size, used as a measure of song complexity, and pairing date, used as a measure of female choice (Catchpole 1980; Buchanan & Catchpole 1997). In the latter study, no significant correlation was found between three measures of song complexity and structure, and pairing date. Recently, Buchanan and Catchpole (2000) reported a significant correlation between syllable repertoire size and male provisioning rate, which was used as a measure of male parental effort. In the laboratory, a significant correlation was reported between copulation solicitation display behaviour of estradiol primed female Sedge Warblers and a series of artificially constructed songs differing in number of syllable types from one male (Catchpole, Dittami & Leisler 1984). These findings were used by the authors as support for Darwin's theory of sexual selection by female choice. Furthermore, a study of the *Acrocephalus* and closely related *Locustella* genera reported that sexual selection may have resulted in the evolution of syllable repertoire size by increasing the size of the higher vocal centre, a controlling area of the brain (Székely, Catchpole, De Voogd, Marchl & De Voogd 1996). Syllable repertoire size, song complexity and song length were used to test Sheldon's phenotype-linked fertility insurance hypothesis (Birkhead & Fletcher 1995) and Møller's (1994) directional asymmetry hypothesis in the Sedge Warbler, but no support for these hypotheses was found (Birkhead, Buchanan, Devoogd, Pellatt, Székely & Catchpole 1997).

All these studies on the Sedge Warbler involving syllable repertoire size and song complexity and structure depend on a song being a discrete, reliable and readily identifiable unit as described by Catchpole (1976). The problems around this dependency, which is dealt with in

Table 3. Studies investigating aspects of acoustic communication in species of songbird that sing more continuously than species with definite, discrete songs.

Species	**Author**
Sedge Warbler	Catchpole 1976, 1977, 1980 part 1, 1982, 1986, 1987, 1988, 1989.
	Brackenbury 1978, 1979.
	Catchpole, Dittami & Leisler 1984.
	Heuwinkel 1985.
	Buchanan & Catchpole 1997, 2000.
	Birkhead, Buchanan, De Voogd, Pellatt, Szekely & Catchpole 1997.
	Buchanan, Catchpole, Lewis & Lodge 1999.
Sedge and Reed Warblers	Catchpole 1973, 1982.
	Jilka & Leisler 1974.
Sedge, Reed & Marsh Warblers	Catchpole 1978.
	Heuwinkel 1982.
Marsh Warbler	Dowsett-Lemaire 1979.
Moustached Warbler	Feßl & Hoi 2000.
Genus *Acrocephalus*	Catchpole 1980, 1981, 1995, 1996, 2000.
	Székely, Catchpole, De Voogd, Marchl & De Voogd 1996.
Brown Thrasher	Kroodsma & Parker 1977.
	Boughey & Thompson 1981.

Chapters 2 and 3, started to become clear during a preceding three year study of singing behaviour and mate choice in a breeding population of Sedge Warblers at Wraysbury Lakes in the Thames Valley, south-east England, United Kingdom (51°27'N, 0°32'W). Visual and auditory examination of the spectrograms of 15 minutes of natural, spontaneous song of each of 70 males, using a Uniscan real-time spectrum analyser and fibre-optic printer, revealed that songs differed from Catchpole's (1976) description. Subsequently, a qualitative and quantitative study was undertaken to investigate this issue and other aspects of song structure, described in Chapter 2.

The different types of syllables comprising the repertoires of male Sedge Warblers have never been described or defined, not least in terms of criteria for type assignment, yet they have been used to test Darwin's theory of sexual selection by female choice (Catchpole 1980; Catchpole et al. 1984; Buchanan & Catchpole 1997). Fundamental questions naturally arise about the nature of these units of vocal organisation not only within individual males, but also among males, especially with respect to intra- and inter-type variation (Kroodsma 1982; Table 1). These questions are addressed qualitatively and quantitatively within individual males in Chapter 3 and among males in Chapter 4. This presents a formidable challenge, as Kroodsma (1982) has noted: "It is a simple matter to recognize the existence of differences in vocal behaviors, yet it is very difficult to quantify those differences adequately".

Fundamental problems of definition and description need to be resolved before hypotheses about the biological relevance of song can be formulated and tested (Horn & Falls 1996; Macdougall-Shackleton 1997). In reviewing methods of assessing how birds themselves perceive their vocalisations, Horn and Falls (1996) emphasise the crucial importance of song structure analyses: "detailed analyses of song structure are essential if these response gradients are to be compared with structural variation in songs. Without such comparisons, categorization cannot be rigorously demonstrated". This is an important theme running through my thesis and it has been emphasised before by Kroodsma (1982): "Description and correlation are not enough, of course, but description is not a second-class approach to that of experimentation; rather, it is a fundamental prerequisite to any experimental approach." Thus, more work is needed on more continuous and continuous singers, and in my thesis, some of the difficult classification problems are addressed qualitatively and quantitatively.

Aims

1. To define the temporal, sequential and spectral structure of Sedge Warbler song quantitatively.
2. To provide this definition in a transparent, reliable and verifiable way.
3. To consider the implications of this definition in terms of:
    a) Published studies involving the acoustic structure of Sedge Warbler song, and
    b) The communication system of this species.

The attainment of these aims and the following objectives will help by, first, enabling valid, dependent and repeatable empirical studies dealing with the acoustic structure of Sedge Warbler song, and second, the formulation and testing of hypotheses concerning the biological relevance of the extremely variable song found in this species.

Objectives

1. To clarify why more definition of the temporal, sequential and spectral structure of Sedge Warbler song is needed.
2. To test the following hypotheses:

A. Chapter 2

i) The element, syllable and song are three kinds of basic, discrete vocal units of different and increasing temporal magnitude comprising Sedge Warbler song.

ii) A Sedge Warbler song is a discrete and readily identifiable unit in terms of the intervals between syllables and the temporal and sequential organisation of syllables.

iii) There are differences between males in element, inter-element, syllable and inter-syllable durations and syllable rate that provide a basis for individual recognition and female mate choice.

iv) There are differences between males in syllable type rate that provide a basis for female mate choice.

B. Chapter 3

i) The syllable repertoire of a male Sedge Warbler is composed of a large number of types that differ qualitatively and quantitatively from one another in temporal and acoustic frequency characteristics.

The separate analysis of each male's entire estimated syllable type repertoire distinguishes Chapter 3 from Chapter 4, where the focus changes to syllable type variation and composition in a population of Sedge Warblers.

C. Chapter 4

i) At the population level, the syllable repertoires of males are composed of syllable types that are qualitatively and quantitatively distinct in temporal and acoustic frequency characteristics and thus form the basis for a communication system in this species.

ii) There is structural constancy of syllable types in the population that provides a basis for more reliable communication.

iii) There are differences in the levels of abundance of syllable types among males and within males at different times of the same season and in different years.

iv) A proportion of syllable types is shared in a population of male Sedge Warblers, while other syllable types are peculiar to individual males. Sharing of syllable types may provide the basis for a specific-mate recognition system as defined by Paterson (1985, 1993), while peculiar types may provide a basis for individual recognition and female choice.

3. To develop methods for analysing temporal, sequential and acoustic frequency structure of Sedge Warbler song.
4. To consider limitations of the methods used and if necessary to propose modifications or better methods.
5. To discuss the interpretation and implications of my results in terms of relevant studies.

2. What's in a Song? - The Case of the European Sedge Warbler, *Acrocephalus schoenobaenus*.

"The analysis of song patterning, as of any aspect of behaviour, depends upon the definition of the elements that are the basis for description, and upon the ease of recognizing these elements and of extracting pertinent correlative data." (Isaac & Marler 1963).

Abstract

The purpose of my study is to examine the structure of Sedge Warbler song quantitatively, define the basic units of vocal organisation, explore relationships between important structural variables and consider the implications of the new findings. Temporal analysis using two independent methods and single spectrum (Fourier) and cross-spectrum analysis of 10-min. natural sequences of song of four males do not support a song as a discrete and reliable temporal unit in this species. This evidence has implications for the interpretation of Sedge Warbler communication and for studies that depend on this vocal unit. Sedge Warbler song is composed of two fundamental acoustic units, syllables and their component elements. Element and inter-element durations are within the limits of auditory discrimination in songbirds. The considerable variability in the cyclical behaviour of syllable, syllable type and mean inter-syllable duration rates and out of phase variation between them in each male indicate higher levels of variability and complexity in the structure of Sedge Warbler song than originally described. This variability and complexity may promote attention levels in prospecting conspecifics and may reduce specific neuro-muscular activity and fatigue that will be important in sustaining the almost continuous singing over long periods, characteristic of male Sedge Warblers. The out of phase variation and absence of a clear pattern of co-variation between syllable and syllable type rate cycles together with significant differences between males in both these variables provide a basis for them to function as independent multiple cues in female choice. Analysis of the relationships between inter-syllable duration, syllable rate and syllable type rate indicate that there are constraints on the possible sexually selected traits of syllable and syllable type rates such as the approach of the limits to inter-syllable duration, a ceiling on syllable type rate and trade-off between syllable and syllable type rates. Significant differences were found between 10-min. song samples of males in

element, inter-element, syllable and inter-syllable durations and syllable and syllable type rates that may reflect differences between males.

Introduction

The hierarchical temporal structure of bird song is well known (Kroodsma & Miller 1982, 1996; Catchpole & Slater 1995), although there has been great variation and inconsistency in the naming of vocal units (Shiovitz 1975; Spector 1994), despite efforts at creating a uniform nomenclature (Shiovitz 1975; Thompson, Ledoux & Moody 1994). Songs and their component syllables are two basic units that form part of this hierarchy and that have become fundamental to studies investigating many aspects of songbird vocal communication (Kroodsma & Miller 1982, 1996; Searcy & Andersson 1986; Kroodsma & Byers 1991; Andersson 1994; Catchpole & Slater 1995).

Despite the fundamental nature and importance of these two vocal units, very few studies have defined them quantitatively (Isaac & Marler 1963; Lemon 1971; Lemon & Chatfield 1971, 1973). In contrast, many studies have provided qualitative descriptions of songs and syllables that have proved too vague and difficult to understand (Spector 1994; Thompson et al. 1994).

In the Sedge Warbler, the syllable has been described briefly as a basic temporal unit of vocal organisation (Catchpole 1976, 2000) that sometimes can be divided into smaller temporal units called sub-syllables (Catchpole 1976) or elements (Catchpole & Slater 1995). The syllable is crucial to the description, definition and use of syllable types as important vocal units in the communication system of this species (Wilson, Chapters 3, 4). Furthermore, syllable rate has been implicated in the only laboratory study of female choice for greater syllable repertoire size in this species (Catchpole, Dittami & Leisler 1984), and in a test of the phenotype-linked fertility hypothesis (Birkhead, Buchanan, De Voogd, Pellatt, Székely & Catchpole 1997).

Catchpole (1976, 2000) described a Sedge Warbler song as a temporally discrete, readily identifiable unit and further in terms of the temporal and sequential organisation of syllables. This unit has been used extensively in studies on this species to derive estimates of syllable repertoire size that have been used to test Darwin's theory of sexual selection by female choice (Catchpole 1980; Székely, Catchpole, De Voogd, Marchl & De Voogd 1996; Buchanan & Catchpole 1997). Recently, song complexity, song length and repertoire size were used to test the phenotype-linked fertility hypothesis and the directional asymmetry hypothesis in this species (Birkhead et al. 1997).

Since a rigorous quantitative analysis has not been conducted on the structure of Sedge Warbler song, my study aims to do this in four males, define the basic units of vocal organisation, explore relationships between important structural variables and consider implications of the new findings, by testing the following four hypotheses.

The first hypothesis focuses on the intervals between vocal units and states that elements, syllables and songs are three kinds of basic, discrete vocal units of different and increasing temporal magnitude comprising Sedge Warbler song. The first prediction tested is that if song in this species is composed of these units, then they will be separated by three distinct peaks in a frequency distribution of the intervals between vocal units in a long sequence of song of each of several males. Catchpole (1976) described the intervals between songs as being ca. 1 s and the intervals between syllables as being ca. 0.1 s. Thus, in a frequency distribution of intervals between vocal units, songs should be separated by a peak around 1 s, while syllables should be separated by a peak around 0.1 s. Catchpole provided a qualitative description of sub-syllables or elements, but gave no indication about the approximate magnitude of such a unit. In a frequency distribution of intervals between vocal units, elements must be separated by intervals at a lower order of temporal magnitude than are syllables. The second prediction tested is that if songs are temporally discrete units, which are separated from one another by intervals around 1 s, then their presence should be defined by a single, distinct plateau at around 1 s, when number of songs is plotted against intervals between a continuous sequence of syllables, comprising many bouts of song. This plateau should be distinct from the first part of the graph, which shows a rapid decrease when intervals are in the range of inter-syllable durations within songs at around 0.1 s. Similarly, the plateau should be distinct from the third part of the graph, which rapidly decreases to a value of one when the interval between songs becomes longer than normal, e.g., when a male stops singing after a bout of songs.

The second hypothesis focuses on the rhythmic or cyclical behaviour of syllable and syllable type rates and intervals between syllables and songs described by Catchpole (1976, 2000), who reported that a Sedge Warbler song is composed of three sections: (1) An initial section consisting of long complex patterns of repetition and alternation, usually of two syllable types. (2) A middle section characterised by a sudden and rapid increase in the number of syllables and syllable types. (3) A terminal section, similar to the initial section, but with two syllable types selected from the middle section. If Sedge Warbler songs can be defined temporally and rhythmically in this way, then the following predictions should hold: (i) Inter-syllable duration defining the end of one song and the beginning of the next should be distinct and large after and only after each cycle, comprising an increase and then a decrease in syllable and syllable type rates. Thus, cycles of mean inter-syllable duration/s should have the same period as syllable and syllable type rate cycles in each male. (ii) Inter-syllable duration should cycle out of phase with syllable and syllable type rates, i.e., as the number of syllables and syllable types increase and then decrease per unit time, the duration between syllables should decrease and then increase. (iii) Syllable and syllable type rates should be in phase, with syllable rate being tracked closely by syllable type rate in each rate cycle. Thus, syllable and syllable type rate cycles should have the same period within each male and should co-vary.

The third hypothesis addressed is that males differ in element, inter-element, syllable and inter-syllable durations and syllable rate. These differences are likely to result from various proximate factors that influence sound production in individuals such as neurobiology (Brenowits & Kroodsma 1996), physiology and morphology of the sound production system and energetic condition (Lambrechts 1996). These differences may provide a basis for individual recognition in the Sedge Warbler (Falls 1982; Stoddard 1996). Furthermore, syllable rate may be a sexually selected trait as a result of female preference for males with higher syllable rates (Andersson 1994). Syllable rate has been mentioned as a possible cue for female choice in the Sedge Warbler (Catchpole et al. 1984), although no published accounts have investigated its function in this species to date. If it is a cue, then mate choice theory predicts that males should differ significantly in this trait (Searcy 1979). Whatever the proximate and ultimate factors, the prediction tested is that males differ significantly in element, inter-element, syllable and inter-syllable durations and syllable rate over a 10-min. sample period.

The fourth hypothesis addressed is that males differ in syllable type rate. Repertoire size has been widely reported to be an important cue in female choice in several species of songbird (Searcy & Andersson 1986; Andersson 1994; Searcy & Yasukawa 1996; Macdougall-Shackleton 1997), including the Sedge Warbler (Catchpole 1980, 2000; Catchpole et al. 1984; Buchanan & Catchpole 1997). However, it takes approximately 12.5 minutes for a male Sedge Warbler to cycle through his repertoire of over 100 syllable types (Wilson, Chapter 3). Syllable type rate would provide a female with more of an immediate (Parker 1974; Sullivan 1994) and easily remembered measure (Kroodsma 1976; Krebs & Kroodsma 1980; Falls 1982) of repertoire size as she surveyed singing males in the population. According to mate choice theory (Searcy 1979), the prediction tested is that males differ significantly in syllable type rate over a 10-min. sample period.

Materials and Methods

Acquisition of Sound Recordings

In the field: A breeding population of Sedge Warblers was studied from 1988 to 1990 at Wraysbury Lakes, Surrey, UK (51°27'N, 0°32'W). Males arrived at the study area in small groups in a staggered pattern in April and May of each year and were uniquely marked with colour and aluminium rings. After taking up territory and a day of settling in, males sang in their characteristic advertising manner (Catchpole 1973, 1976).

Fifteen min. continuous sound recordings were taken for each male during the time of peak daily singing from sunrise until 10.00 am until pairing occurred, after which singing stopped. On average, males paired after 12 days of singing ($\bar{x} \pm SE=12.8 \pm 0.8, N=70$) during the three field seasons. A Uher 4000 Report-L tape recorder (tape speed: 19 cm/s) and a DP4/M Grampian omni-directional microphone attached to a parabolic sound reflector (diameter: 45.72 cm) were used. The high output pressure type dynamic microphone had a uniform, wide range frequency response (50 Hz-15 kHz), which was flat in the 800 Hz to 15 kHz frequency range. This covered the known frequency range of the Sedge Warbler (1.778-14.120 kHz, Heuwinkel 1982).

In the Laboratory: Sound recordings of four males were chosen from the 1990 field season (N=18 males) by random number generator for spectrographic analysis. Environmental variables such as temperature, humidity and windspeed differed by <10% when the recordings were made in the field. One 15 min. song sample for each male was recorded onto a Macintosh Quadra 840 AV using Canary (Charif, Mitchell & Clark 1995). A 22.05 kHz sampling rate exceeding twice the maximum signal frequency, and 16 bit sample size were selected to prevent aliasing and minimise quantization noise respectively (Charif et al. 1995). Each song sample contained perch song predominantly and occasional song flights (Catchpole 1976).

Quantitative Analysis

Durations of successive putative syllables and inter-syllable intervals were measured from spectrograms of 10-min. of continuous song of each of the four males (Fig. 1), using optimal temporal analysis settings (resolution=5.75 ms, Appendix 1). For finer scale temporal analysis, a large number of syllables (Fig. 3d) comprised of putative elements in each male's 10-min. sample were chosen from different phrases (Fig. 1; Brackenbury 1978) by random number generator. In this analysis, an interval, called an inter-element duration, was measured between the first two elements in each syllable from spectrograms (Fig. 1), using optimal temporal analysis settings (resolution=0.719 ms, Appendix 4). The duration of the second element in each syllable also was measured.

Syllable types were determined by multi- and univariate analysis (Wilson, Chapters 3, 4). Syllable and syllable type rates were obtained by counting respectively the number of syllables and syllable types per second in each male's 10-min. song sample.

Statistical Methods

General: Uni- and multivariate analyses were conducted using Unistat (Toker 1994). The confidence level for rejecting the Null Hypothesis was 0.05 and all tests were 2-tailed unless otherwise stated. Element, inter-element, syllable and inter-syllable duration were continuous variables, whereas syllable and syllable type rates were meristic variables (Sokal & Rohlf 1981). Data were checked for normality with the Kolmogorov-Smirnov one sample test and for homogeneity of variances with Bartlett-Box F- and Hartley's F tests (Sokal & Rohlf 1981). None of the variables satisfied the assumption of normality and most transformations (Sokal & Rohlf 1981) were not successful either, thus requiring the use of non-parametric tests (Siegel & Castellan 1988).

Hypothesis 1 - Elements, syllables and songs are basic, discrete vocal units: Frequency distributions of inter-syllable duration were generated for each male at 0.1 s and 0.01 s resolution. Frequency distributions of inter-element duration were generated for each male at 1 ms resolution. Mann-Whitney U tests determined whether syllable and inter-syllable durations were significantly greater than element and inter-element durations respectively (1-tailed tests).

The frequency distributions of successive inter-syllable durations in 10-min. of continuous song of the four males that I studied were log-normal around modal values (Figs. 2 a, b). Expected successive inter-syllable durations were generated for Catchpole's (1976) males 2 and 3 in 10-min. of song from two simulations based on, first, the log-normal distribution found for my four males and second, his reported means of the following variables for males 2 and 3: Syllable duration (90 ms), inter-syllable duration (120 ms, 110 ms), song duration (14.51 s, 29.31 s), inter-song duration (1.35 s, 1.88 s) and number of syllables per song (74.65, 145.35). From the inter-syllable durations of each of the six males, minimum intervals between syllables that defined songs, starting at 0.1 s and increasing at 0.1 s intervals, determined the number of resulting songs for each male. The upper limit to these intervals for each of the males was set by the particular male's maximum inter-syllable duration, e.g., 45.1 s for male 9 (Figs. 3c, 4). The numbers of songs were plotted for each 0.1 s increase in interval (e.g., 0.1, 0.2, 0.4,….,45.1 s) for each male.

Hypothesis 2 - Cycles of inter-syllable duration and syllable and syllable type rates define songs: The cyclical nature of these variables and the time taken for one complete cycle (i.e., period) were investigated quantitatively using single spectrum (Fourier) analysis of each male's whole 10-min. song sample. The problem of leakage was addressed by padding and tapering each time series and smoothing each periodogram. The cyclical nature of each time series was tested statistically using the Kolmogorov-Smirnov one-sample D statistic (Siegel & Castellan 1988). This tested the distribution of observed periodogram values against the exponential distribution, which would be expected if the time series represented white noise. Co-variation in cyclical behaviour between syllable and syllable type rates in each male was investigated using cross-spectrum analysis. In order to study the real time relationships between inter-syllable duration and syllables/s and syllable types/s, mean inter-syllable duration/s was calculated for each male's 10-min. song sample. As mean inter-syllable durations were calculated per second, all means greater than 1 s had to be removed from the datasets. These large means accounted for <0.01% of the data in each of the four males. Time series plots of the three variables were made for the 10-min. continuous song samples of each male.

Relationships between variables: The relationship The relationships between mean inter-syllable duration/s and syllable and syllable type rates were determined for the four males using pooled data. A non-linear regression equation, coefficient of multiple determination (R^2), F-statistic and significance level were obtained for each relationship.

A graphical technique was developed to study the relationship between syllable and syllable type rates as linear regression could not be used because the variables were not normally distributed even after all possible transformations. A frequency distribution of syllable rate was plotted for each syllable type rate from 1 to a maximum of 5 syllable types/s. The statistical relationship between syllable and syllable type rates was determined by Kendall's rank correlation test (Siegel & Castellan 1988), a non-parametric version of linear regression (Sokal & Rohlf 1981). Kruskal-Wallis one-way ANOVA tested whether median syllable rate for the four males differed significantly at syllable type rates 1 to 5 syllable types/s. A non-parametric multiple comparison test determined which pairs of syllable rates differed significantly in their median values (Siegel & Castellan 1988).

Hypotheses 3 and 4 - Males differ in element, inter-element, syllable and inter-syllable durations and syllable and syllable type rates: Kruskal-Wallis one-way ANOVA tested whether males differed significantly in each type of duration and rate in the particular 10-min. song samples that were analysed. A non-parametric multiple comparison test determined the pairs of males among males 1, 4, 5 and 9 that differed significantly in their median values (Siegel & Castellan 1988).

Results

General

Sedge Warbler song was composed of long, continuous sequences of syllables (Fig. 1; Appendix 2). Syllables were discrete units of sound, had complex and variable temporal and frequency structure with amplitude and frequency modulation, were generally composed of smaller temporal units called elements and usually were repeated in varying numbers of the same syllable type before changing to another type (Fig. 1; Appendix 2).

Hypothesis 1 - Elements, syllables and songs are basic, discrete vocal units

The first prediction that songs, syllables and elements are separated by three distinct peaks in a frequency distribution of the intervals between vocal units in a long natural sequence of song was not supported for males 1, 4, 5 and 9. Examination of the frequency distributions of consecutive inter-syllable durations in the song of four males at 0.1 s resolution revealed a log-normal distribution with one distinct peak at <0.1 s that separated syllables, but no peak at ca. 1 s that separated songs (Fig. 2a). Furthermore, this figure indicates that there is no clear interval defining a song in these males in the range of durations shown. A finer scale analysis at 0.01 s resolution also revealed one distinct peak that separated syllables with modes for males 1 and 5 at 0.03 s, and for males 4 and 9 at 0.04 s (Fig. 2b). The considerable range of almost continuous variation of inter-syllable duration, as well as the irregular nature of this variation as occurring in a 10-min. natural sequence of song, was clearly evident for each of the four males (Figs. 1-4 in Appendix 3). Examination of the frequency distributions of inter-element durations of the four males at

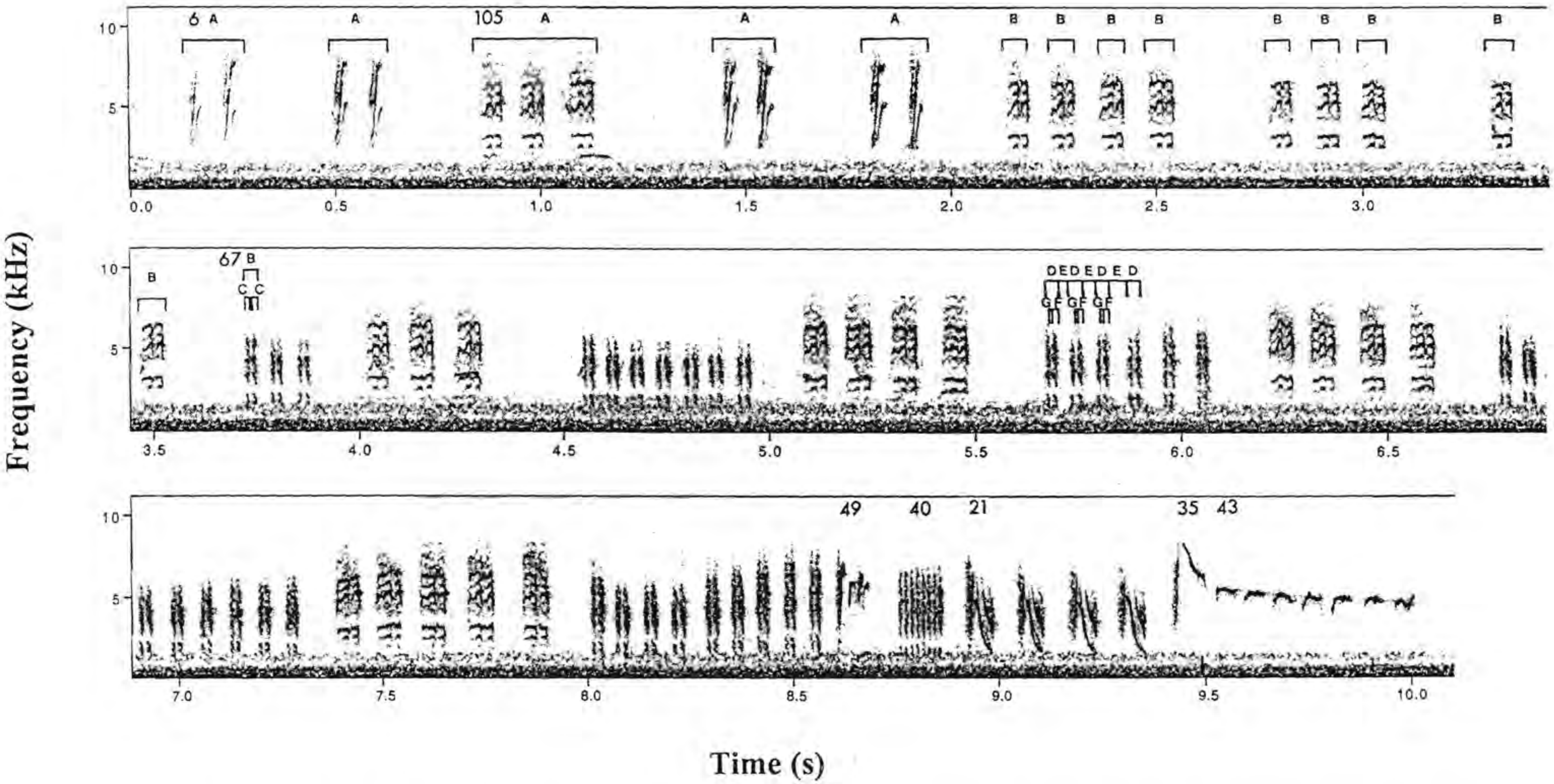


Figure 1. Spectrogram of a natural sequence of phrases (A), syllables (B) and their component elements (C), and syllable types (numbered when they first appear) comprising the song of male 1. Examples of measurements of syllable duration (D), inter-syllable duration (E), element duration (F) and inter-element duration (G) are shown that formed the basis of the quantitative analysis of the continuous song of males 1, 4, 5 and 9.

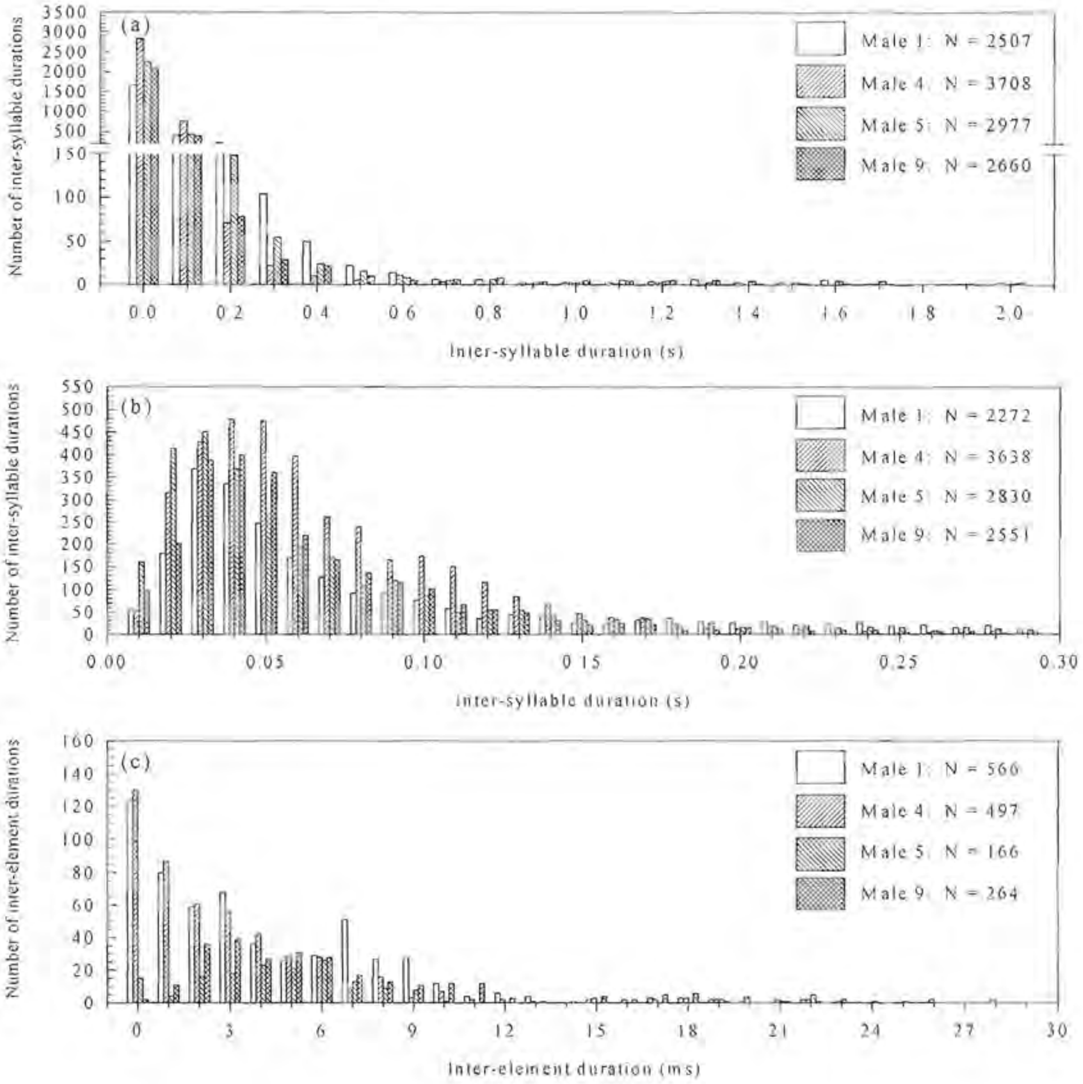


Figure 2. Syllables (a, b) and elements (c), but not songs are revealed as basic discrete vocal units in frequency distributions of male 1's, 4's, 5's and 9's inter-syllable and inter-element durations. (a) At 0.1 s intervals, one peak at <0.1 s separates syllables, but no other peak is evident that separates songs. (b) The distinct peak of inter-syllable durations that separates syllables can be seen clearly at 0.01 s intervals. (c) At 1 ms intervals, a distinct peak that separates elements is evident.

i 15951728
b15367368

1 ms resolution indicated the presence of a distinct peak that separated elements with modes for males 1 and 4 at <1 ms, for male 5 at 5 ms and for male 9 at 3 ms (Fig. 2c).

In each of the four males, syllable duration was significantly greater than element duration (Mann-Whitney U test: Adjusted Z>-21.6, P<0.0001; Figs. 3a, b) and inter-syllable duration was significantly greater than inter-element duration (Mann-Whitney U test: Adjusted Z>-21.4, P<0.0001; Figs. 3c, d). Mean element and especially mean inter-element duration were of small magnitude (Figs. 3b, d). Considerable variation in each type of duration resulted in large standard deviations and in overlap between minimum syllable duration and maximum element duration, and minimum inter-syllable duration and maximum inter-element duration (Figs. 3a-d).

The second prediction that songs are temporally discrete units defined by the formation of a single, distinct plateau at ca. 1 s when number of songs is plotted against minimum intervals between syllables that define a song, was not supported for males 1, 4, 5 and 9 (Fig. 4). More importantly this figure indicates that there is no clear interval defining a song in these males. Two graphs, however, that were generated from simulations derived from summary data in Catchpole's (1976) study, in which Sedge Warbler songs were described statistically and graphically, supported the prediction (Fig. 4). The plateau of stability around 1 s that was evident in both these graphs was formed as the intervals between syllables equalled the range of observed intervals between songs. The plateau was distinct from the preceding rapid decrease in song number when intervals between syllables were in the range of observed inter-syllable durations within songs. The plateau also was distinct from the ensuing part of the graph, which showed a rapid decrease in song number alternating with minor plateaus when the intervals between songs became longer than normal. Males 1, 5 and 9 showed a rapid decline in song number up to ca. 2 s after which many plateaus that alternated with rapid decreases were evident (Fig. 4). Male 4 showed a rapid decline in song number up to 0.8 s, after which there was a brief levelling to 0.9 s followed by alternating rapid decreases and further levelling, which occurred from 1.3 to 1.7 s, and from 2 to 3.5 s (Fig. 4). Kolmogorov-Smirnov two sample test results supported the differences in shape between the distributions of males 1 and 9 and Catchpole's (1976) two males, and male 4 and Catchpole's male 2 (Fig. 4). Catchpole's two males did not differ significantly, but neither did they differ significantly from male 5 (Fig. 4).

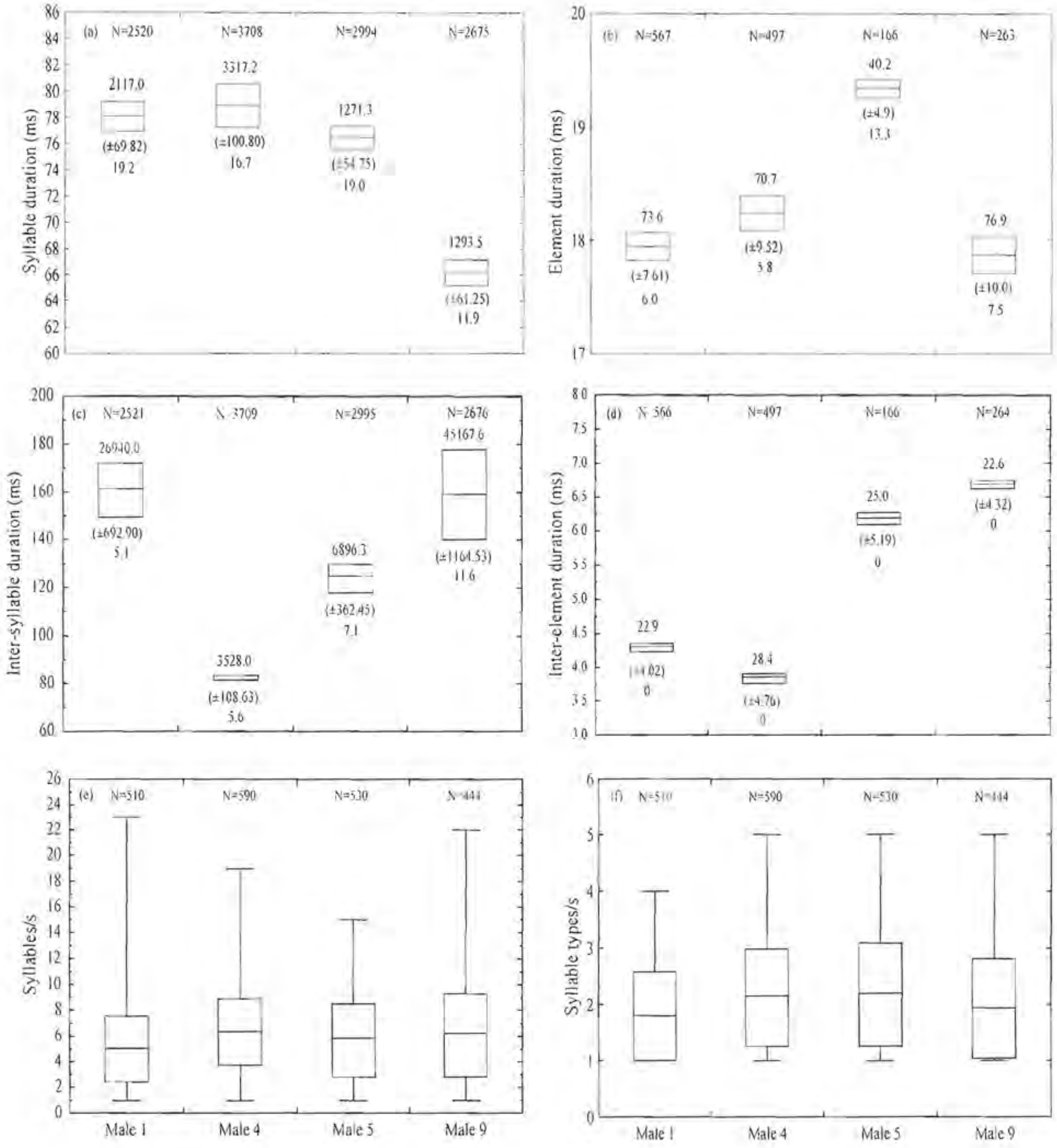
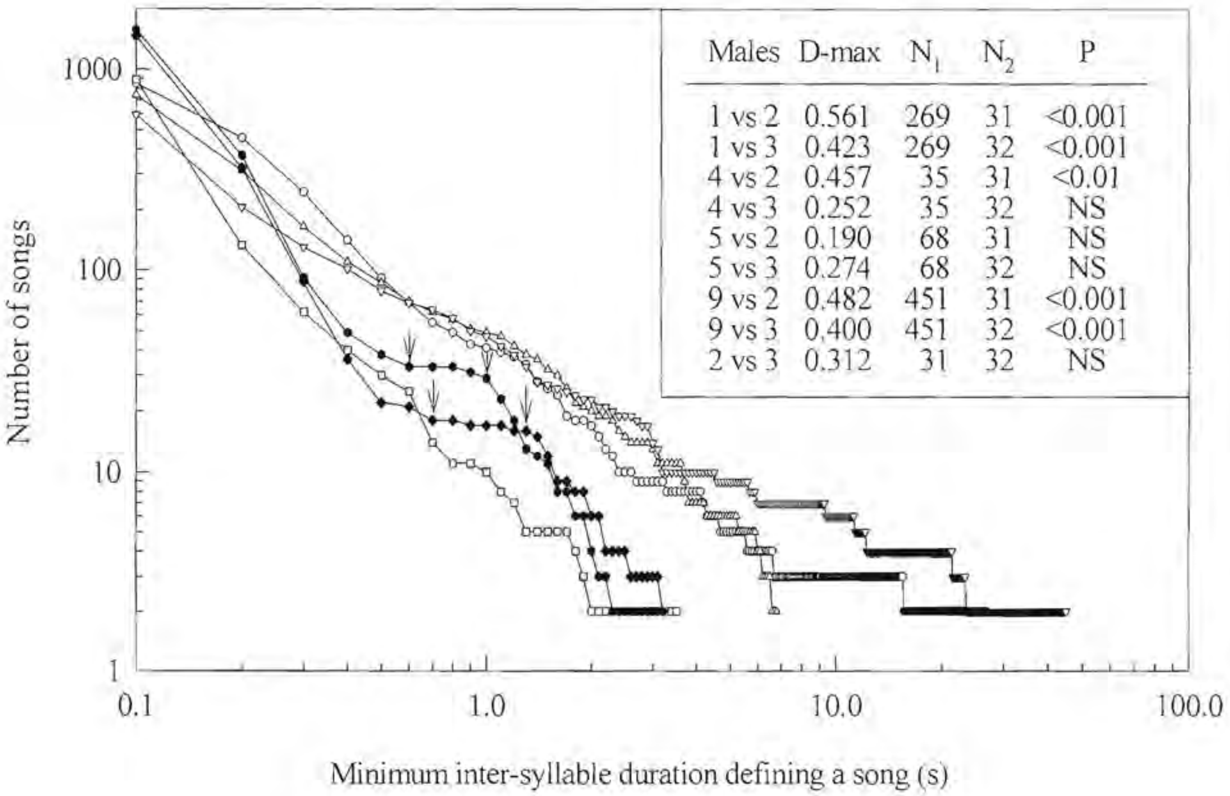


Figure 3. Descriptive statistics of (a) syllable duration, (b) element duration, (c) inter-syllable duration, (d) inter-element duration, (e) syllable rate and (f) syllable type rate for males 1, 4, 5 and 9. In (a)-(d), boxes show mean±1SE, brackets contain ±1SD and maximum and minimum values appear above and below each box respectively. In (e)-(f), boxes show mean±1SD, while whiskers show min. and max. rates. Sample size (N) is indicated above each plot.



Males	D-max	N_1	N_2	P
1 vs 2	0.561	269	31	<0.001
1 vs 3	0.423	269	32	<0.001
4 vs 2	0.457	35	31	<0.01
4 vs 3	0.252	35	32	NS
5 vs 2	0.190	68	31	NS
5 vs 3	0.274	68	32	NS
9 vs 2	0.482	451	31	<0.001
9 vs 3	0.400	451	32	<0.001
2 vs 3	0.312	31	32	NS

Figure 4. Plots of males 2 (–●–) and 3 (–◆–), derived from simulations based on summary data from two of Catchpole's (1976) males with discrete songs, show areas of distinct stability (↓ to ↓) at around 1 s that indicate the ranges of intervals between songs. These areas contrast with less stable areas that indicate intervals between syllables within songs (before 1st arrows) and longer than normal intervals between songs (after 2nd arrows). Songs as distinct vocal units however, are not evident when minimum inter-syllable duration defining a song is plotted against the resulting number of songs for the entire ranges of inter-syllable duration in males 1 (–O–), 4 (– –), 5 (–Δ–) and 9 (–∇–). Kolmogorov-Smirnov two sample test results show differences (D-max) between the distributions of males 1, 4, 5 and 9 and Catchpole's (1976) males 2 and 3. N_1 and N_2 indicate sample sizes for the distributions, while P values are reported after sequential Bonferroni correction.

Hypothesis 2 - Cycles of inter-syllable duration and syllable and syllable type rates define songs

Single spectrum (Fourier) analysis and Kolmogorov-Smirnov one sample tests revealed that although mean inter-syllable durations/s and syllable and syllable type rates showed cyclical behaviour, strong periodicity identified by a single dominant period was not present in each male's time series variable. Rather, a range of relatively prominent periods was found (Table 1). Single spectrum analysis and time series plots for each of the four males showing real-time variation in mean inter-syllable duration/s (ISD/s) and syllable and syllable type rates, revealed no support for the three predictions that were tested: (i) Mean ISD/s was not distinct and large, i.e., did not form a major peak, after each cycle of syllable and syllable type rates. Every major peak in mean ISD/s occurred after an irregular number of syllable and syllable type rate cycles (Fig. 5). These observations were corroborated by single spectrum analysis, which showed that cycles of mean inter-syllable duration/s had very different prominent periods compared to syllable and syllable type rate cycles in each male (Table 1). (ii) Mean ISD/s did not cycle out of phase with syllable and syllable type rates. When syllable or syllable type rates reached a peak, mean ISD/s usually was not at an absolute low, but on the decline, or increase or at a minor peak (Fig. 5). (iii) Syllable and syllable type rates were not in phase, i.e., syllable rate was not tracked closely by syllable type rate in each cycle. A variable, irregular and complex behaviour was evident for all four males with the cycles often being out of phase and with syllable type rate cycles usually containing an irregular number of syllable rate cycles (Fig. 5). These observations were confirmed by single and cross-spectrum analyses. Single spectrum analysis showed that syllable and syllable type rate cycles had very different prominent periods in each male, except for male 9 where the periods were the same (Table 1). Cross-spectrum analysis showed that syllable and syllable type rates co-varied over a wide range of periods with no clear pattern within and among males (Table 2).

Relationships Between Variables

The relationship between syllable rate and mean ISD/s was exponential and significant with 42% of the variation in syllable rate being explained by mean ISD/s, indicating that higher syllable rates resulted in shorter intervals between syllables (Fig. 6a). As mean ISD/s decreased to ca.

Table 1. Summary results of single spectrum (Fourier) analysis of mean inter-syllable duration/s, syllable rate and syllable type rate time series for males 1, 4, 5 and 9 over 10 min. of continuous, natural song, showing cyclical behaviour for each time series (D-max*, P<0.01), but divergent periods within and among time series variables. A significant Kolmogorov-Smirnov one-sample D statistic (after sequential Bonferroni correction) indicates that the distribution of observed periodogram values differed significantly from the expected exponential distribution associated with white noise and that accordingly, the time series displayed cyclical behaviour. Sample size of each time series is shown (N). The prominent period corresponding to the largest periodogram value and the range of periods corresponding to the 10 largest periodogram values are shown for each time series.

Time series variable	Male	D-max	N	Period (s)	Range of periods (s)
Mean inter-syllable duration/s	1	0.3422*	492	21.39	10.04 - 164
	4	0.2638*	586	15.42	4.22 - 83.71
	5	0.2872*	510	11.59	8.36 - 18.89
	9	0.1825*	420	10.77	3 - 105
Syllable rate	1	0.4207*	600	100	15.38 - 600
	4	0.4175*	600	85.71	13.64 - 200
	5	0.4648*	600	16.22	10.17 - 150
	9	0.4949*	600	100	12.5 - 600
Syllable type rate	1	0.4502*	600	85.71	15.38 - 150
	4	0.3672*	600	46.15	17.65 - 300
	5	0.3829*	600	75	10.17 - 150
	9	0.5068*	600	100	14.63 - 600

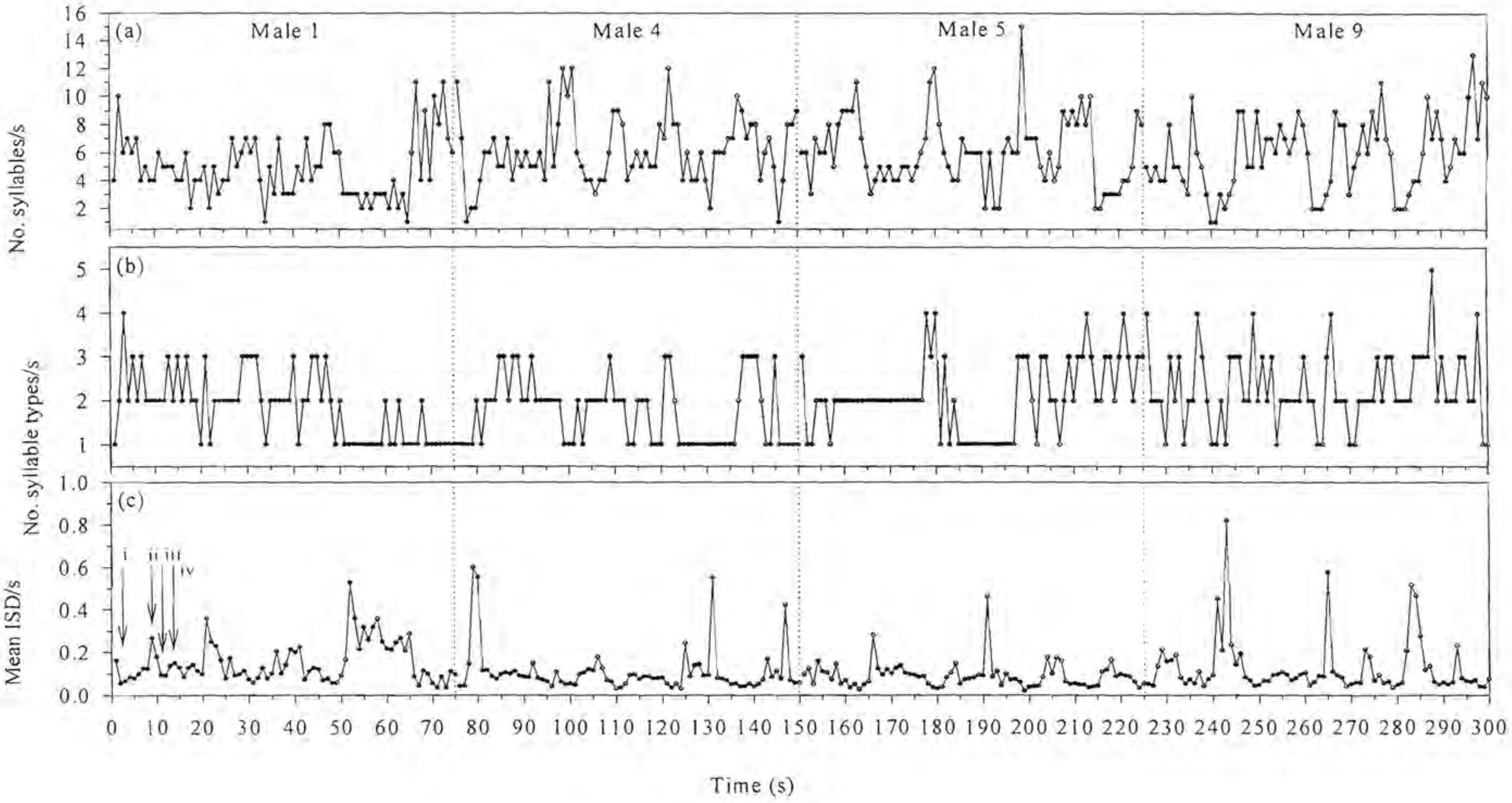


Figure 5. Representative snap-shots (75 s) of continuous song in each of the four males showing, (a) number of syllables/s, (b) number of syllable types/s, and (c) mean inter-syllable duration/s (mean ISD/s). In each variable, a cycle is composed of a trough (i), peak (ii) and trough (iii). In mean ISD/s, major peaks (ii) are differentiated from minor peaks (iv).

Table 2. Summary results of the cross-spectrum analysis of syllable rate and syllable type rate time series for males 1, 4, 5 and 9 over 10 min. of continuous, natural song, showing covariance over a wide range of periods, with no clear pattern within and among males. The 10 highest ranked periods, for which covariance in cyclical behaviour between syllable and syllable type rates was found, are reported for each male, together with the range of cross-amplitude (C-A) values, which indicates the strength of the covariance between the periods in the two time series. Cross-amplitude values are greatest for highest ranked periods and decrease as rank of period decreases. Each time series has a sample size of 600 observations.

	Male 1		Male 4		Male 5		Male 9	
Rank	Period (s)	C-A range	Period (s)	C-A range	Period (s)	C-A range	Period (s)	C-A range
1	85.3	35.7	78.8	24.3	16.0	56.0	102.4	95.8
2	78.8	↑	46.5	↑	73.1	↑	93.1	↑
3	128.0	|	113.8	|	16.3	|	37.9	|
4	20.9	|	73.1	|	23.3	|	256.0	|
5	20.5	|	39.4	|	128.0	|	512.0	|
6	23.3	|	85.3	|	146.3	|	341.3	|
7	68.3	|	19.7	|	78.8	|	113.8	|
8	256.0	|	24.4	|	15.3	|	146.3	|
9	15.5	↓	17.4	↓	28.4	↓	14.0	↓
10	15.3	12.0	15.8	5.4	35.3	14.9	12.5	16.9

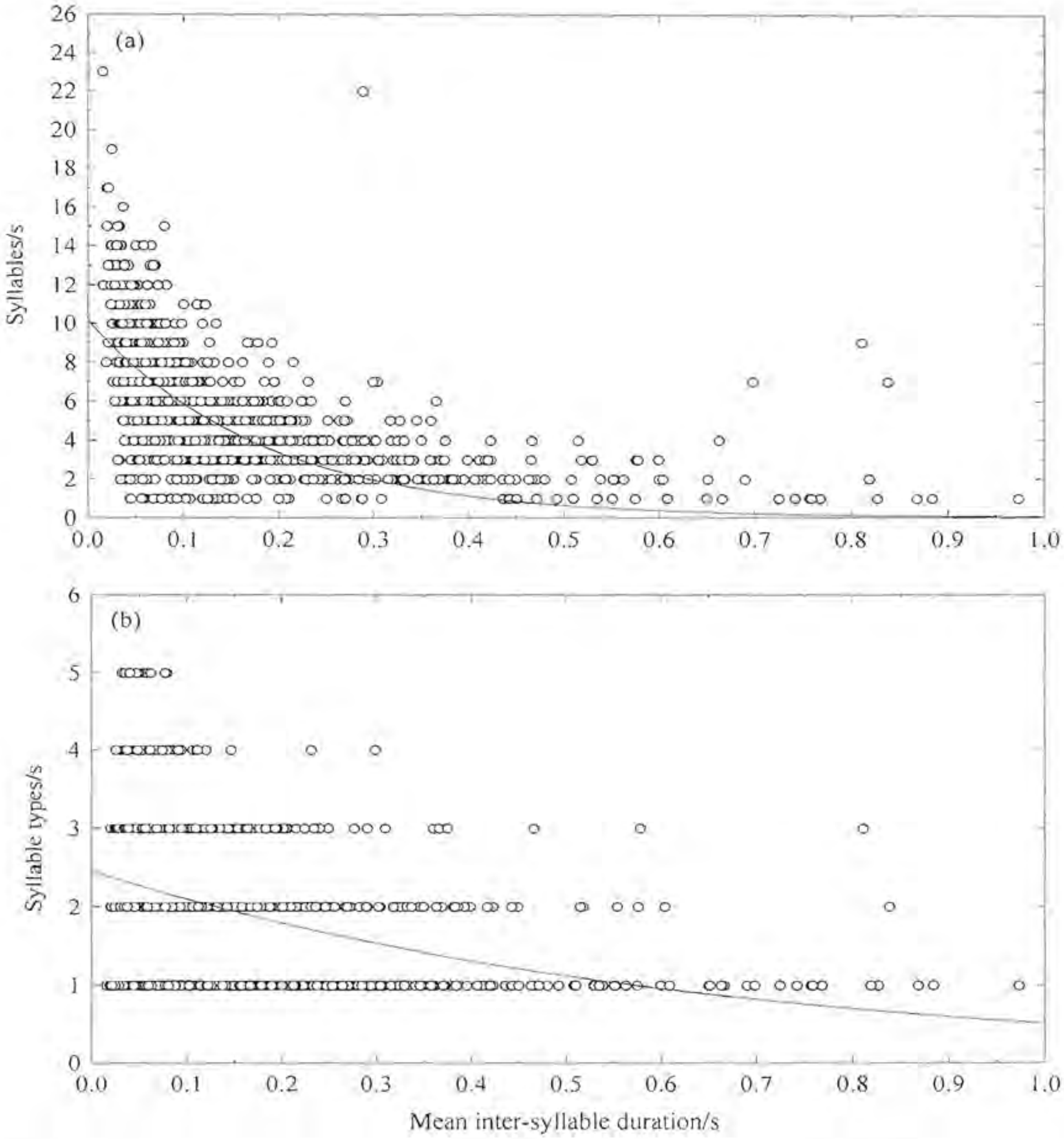


Figure 6. Relationship between mean inter-syllable duration/s and (a) syllable rate, and (b) syllable type rate, for males 1, 4, 5 and 9's pooled data, showing that higher syllable and syllable type rates result in shorter intervals between syllables. The regression equation and statistics for (a) are $y=10.17*\exp(-5.49*x)$, $R^2=0.42$, $F_{1,3914}=1416$, $P<0.001$, $N=1958$, and for (b) are $y=2.46*\exp(-1.57*x)$, $R^2=0.10$, $F_{1,3914}=217$, $P<0.001$, $N=1958$.

0.4 s, syllable rate increased gradually. Further decreases in mean ISD/s were associated with more and more rapid increases in syllable rate. Increases in syllable rate reached high levels in each of the males (Fig. 3e) and were constrained as mean ISD/s approached zero. Extremely small inter-syllable durations were found in each of the males (Fig. 3c). The relationship between syllable rate and mean ISD/s followed an irregular cyclical pattern in real time (Figs. 5a, c).

The relationship between syllable type rate and mean ISD/s was weakly exponential and significant, indicating that higher syllable type rates resulted in shorter intervals between syllables, although only 10% of the variation in syllable type rate was explained by mean ISD/s (Fig. 6b). Syllable type rate did not show any marked increase as mean ISD/s decreased. The relationship followed an irregular cyclical pattern in real time (Figs. 5b, c).

The relationship between syllable and syllable type rates was significant (Kendall's rank correlation: T=0.238, N=1958, P<0.0001; Fig. 7). The four males contributed approximately equally to each of the five frequency distributions (Fig. 7), except for male 1, which had a maximum syllable type rate of 4 (Fig. 3f). Variation in syllable rate in each of the males differed markedly from syllable type rate, which reached a ceiling at 5 (Figs. 3e, f). Median syllable rate differed significantly at syllable type rates 1 to 5 (Kruskal-Wallis test: H_4=166.78, P<0.0001; Fig. 7). Median syllable rates at syllable type rates 1 and 2 differed significantly from one another and from other median syllable rates, while median syllable rates at syllable type rates 3, 4 and 5 were not significantly different (Fig. 7). Thus there was a significant increase in syllable rate as syllable type rate increased from 1 to 3, but as syllable type rate increased above 3 there was no longer a significant increase in syllable rate (Fig. 7). There was a sharp decrease in the number of cases from syllable type rates 2 to 5, and high syllable rate decreased with each increase in syllable type rate (Fig. 7).

Hypotheses 3 and 4 - Males differ in element, inter-element, syllable and inter-syllable durations and syllable and syllable type rates

There was support for hypotheses 3 and 4 in terms of each male's 10-min. song sample for the following reasons. A significant difference was found between the 10-min. song samples of males in the median values of all six variables (Table 3). There was however, considerable variation within males in element, inter-element, syllable and inter-syllable durations and syllable rate (Figs. 3a-e). Three of the six pairs of males differed significantly in median element duration, while five of the six pairs differed significantly in median inter-element duration (Table 3). All

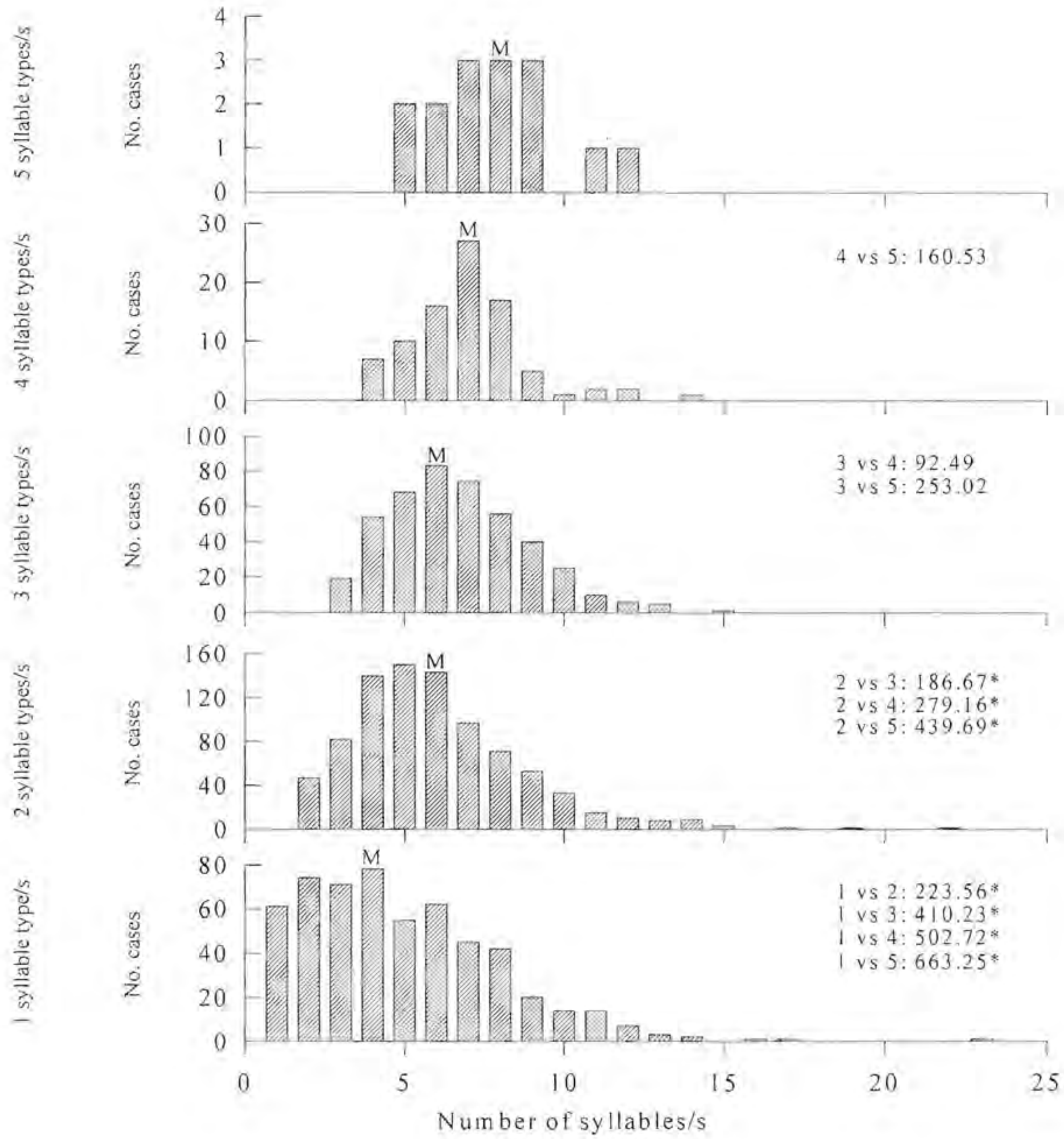


Figure 7. Syllable rate frequency distributions at syllable type rates 1 to 5 for the pooled data of males 1, 4, 5 and 9. Median (M) syllable rates are indicated. Non-parametric multiple comparisons between syllable rates at syllable type rates 1 to 5 are shown. Significant differences between pairs of median syllable rates were found (*P<0.05) if $|R_u-R_v| \geq$ critical value, where $|R_u-R_v|$ is the difference between the mean ranks in each comparison, and critical value=$z_{\alpha/k(k-1)}\sqrt{N(N+1)/12(1/n_u+1/n_v)}$ (Siegel & Castellan 1988). For 10 comparisons, $z_{\alpha/k(k-1)}=2.807$. $N=1958$, $n_1=550$, $n_2=864$, $n_3=441$, $n_4=88$, $n_5=15$.

Table 3. Kruskal-Wallis one-way ANOVA results showing a significant difference (**$P<0.0001$) between the 10 min. song samples in males 1, 4, 5 and 9 in the medians of four different types of duration, and in syllable and syllable type rates (D.F.=3). Non-parametric multiple comparison test results also are reported for the males. Significant differences between pairs of median values were found (*$P<0.05$) if $|R_u-R_v| \geq$ critical value, where $|R_u-R_v|$ is the difference between the mean ranks in each comparison, and critical value=$z_{\alpha/k(k-1)}\sqrt{N(N+1)/12(1/n_u+1/n_v)}$ (Siegel & Castellan 1988). For 6 comparisons, $z_{\alpha/k(k-1)}=2.638$. For syllable duration, $N=11897$, $n_1=2520$, $n_4=3708$, $n_5=2994$, $n_9=2675$. For inter-syllable duration, $N=11901$, $n_1=2521$, $n_4=3709$, $n_5=2995$, $n_9=2676$. For element duration, $N=1493$, $n_1=567$, $n_4=497$, $n_5=166$, $n_9=263$. For inter-element duration, $N=1493$, $n_1=566$, $n_4=497$, $n_5=166$, $n_9=264$. For syllable and syllable type rates, $N=2074$, $n_1=510$, $n_4=590$, $n_5=530$, $n_9=444$.

		$\lvert R_u-R_v \rvert$ values for male comparisons					
Variable	H	1 vs 4	1 vs 5	1 vs 9	4 vs 5	4 vs 9	5 vs 9
Syllable duration	161.0**	326.86*	9.69	1446.03*	336.55*	1119.17*	1455.72*
Inter-syllable duration	105.21**	431.47*	968.13*	675.28*	536.66*	243.81*	292.85*
Element duration	32.97**	14.86	181.37*	48.71	196.23*	33.85	230.08*
Inter-element duration	155.75**	95.71*	200.81*	274.52*	296.52*	370.23*	73.71
Syllable rate	77.12**	307.80*	166.93*	228.84*	140.87*	78.96	61.91
Syllable type rate	67.97**	221.71*	250.45*	87.52	28.74	134.19*	162.93*

males differed significantly in median inter-syllable duration and five of the six pairs differed significantly in median syllable duration (Table 3). Four of the six pairs of males differed significantly in median syllable and syllable type rates (Table 3).

Discussion

Songs as basic, discrete vocal units: The two predictions of hypothesis 1 and the three predictions of hypothesis 2 that addressed songs as discrete and reliable temporal units in the Sedge Warbler were not supported by the results. In the frequency distributions of inter-syllable duration (Fig. 2a), the absence of a peak at ca. 1 s that separated songs was contrary to the first prediction of hypothesis 1. This may be contested by the argument that the song sample time of 10-min. allowed too few songs and hence too few inter-song intervals to form a distinct peak. According to Catchpole's (1976) study, however, mean song length was 19.49 s (N=60), which on average should yield around 31 songs per male and 124 songs for the four males in my study. This should be a sufficient number of songs to form a distinct peak of inter-song intervals at ca. 1 s ($\bar{x}$=1.31 s, N=58) as described by Catchpole (1976). The frequency distribution method has been used to show songs as discrete temporal units in the Mistle Thrush (Isaac & Marler 1963), Red-eyed Vireo (Lemon 1971) and Cardinal (Lemon & Chatfield 1971).

Results for the second prediction of hypothesis 1 were clear (Fig. 4). The graphs based on two of Catchpole's (1976) males which were described in his study as having discrete songs had three distinct parts, the middle one of which was a stable plateau at ca. 1 s that defined the presence of songs. In contrast, the graphs of the four males in my study neither revealed these distinct parts nor a stable plateau at ca. 1 s. Kolmogorov-Smirnov two sample test results supported the differences in shape between the distributions of the two groups of males overall.

Apart from the predictions of hypothesis 1, Figs. 2a and 4 reveal that there is no clear interval defining a song in males 1, 4, 5 and 9. However, an alternative interpretation of these figures is that inter-song interval varies to such an extent within and among males that it cannot be clearly defined quantitatively. But if this is the case, then the question about Sedge Warbler song definition remains.

With respect to the three predictions of hypothesis 2, there was weak cyclical behaviour of syllable and syllable type rates and mean inter-syllable duration/s (ISD/s; Table 1), and relatively large inter-syllable durations did occur as major peaks of mean ISD/s (Fig. 5), but none of these occurrences was as predicted. All the evidence against hypotheses 1 and 2 indicates that a distinction between inter-syllable and inter-song interval cannot be made and is artificial, and that the rhythmic or cyclical behaviour of syllable and syllable type rates and mean ISD/s do not reveal the presence of distinct songs as predicted in the Sedge Warbler.

This evidence has implications for studies that depend on the presence of songs in the Sedge Warbler. Estimated repertoire size in this species has been determined by visually identifying and counting new syllable types from spectrograms of a sample of continuous song and plotting the new syllable types against the cumulative number of songs in each male (Catchpole 1980, 2000; Buchanan & Catchpole 1997). However, if a song as a discrete and reliable vocal unit is artificial in this species, then estimates of repertoire size based on this unit are likely to be misleading. Furthermore, a song has been used as a quantitative unit in several other ways in this species. The number of syllable types per song, which was used as a measure of inter-song complexity, and song length formed part of a study of multiple female choice cues (Buchanan & Catchpole 1997). Both these variables also were involved in a test of the phenotype-linked fertility hypothesis (Birkhead et al. 1997).

Syllables and elements as basic, discrete vocal units: The evidence against songs as discrete temporal units in the Sedge Warbler has important implications for communication in this species, which is unlikely to involve songs, but which is likely to involve syllables and their component elements that both were supported clearly by the results (Figs. 2b, c). In a quantitative study of the structure of Sedge Warbler song, Brackenbury (1978) also identified syllables and elements as basic vocal units, although he named them chirps and pulses respectively. Much greater variation in syllable and element durations were shown in my study compared to Brackenbury's analysis of one male (Figs. 3a, b), although his study recorded lower minimum values for element duration. Catchpole (1976) described syllables as a basic unit of vocal organisation in the Sedge Warbler, although he reported much lower levels of variation in syllable duration than were found in my study. The importance of syllables as a basic vocal unit in the Sedge Warbler and other songbird species has been emphasised in at least two comparative oscine neurobiological studies, first in terms of learning and complexity of song (DeVoogd, Krebs, Healy & Purvis 1993), and second in terms of complexity and evolution of song (Székely et al. 1996).

The small magnitude of element and inter-element durations shown in my study is well within the limits of auditory discrimination in songbirds (Figs. 3b, d). Temporal resolving power in the order of two milliseconds has been demonstrated for the auditory systems of several species of songbird for narrow band (Wilkinson & Howse 1975) and wide band signals (Klump & Maier 1989; Klump & Gleich 1991). This high degree of resolution not only enables the detection of elements and inter-element intervals, but also the detection of small changes in duration. Thus, the difference in duration and number of elements between two successive syllables differing in one or more similar elements is likely to be detected within the active space of the signal (Brenowitz 1982; Wilson, Chapter 3).

Cyclical behaviour of syllable, syllable type and mean inter-syllable duration rates: The considerable variability in the cyclical behaviour of these rates, out of phase variation between them in each male (Table 1, Fig. 5) and absence of a pattern of co-variation in syllable and syllable type rates within and among males (Table 2) indicate higher levels of variability and complexity in Sedge Warbler song than originally described by Catchpole (1976). This variability and complexity may help maintain attention levels in prospecting conspecific females and males (Hartshorne 1973; Kroodsma 1982), and together with the large repertoire size of males (Wilson, Chapter 3), may reduce specific neuro-muscular activity and fatigue, as different sounds are generated by the action of different neurons and muscles (Suthers & Hector 1985; Vicario 1991). Reduction in neuro-muscular activity and fatigue may be an important factor in the sustained, almost continuous singing over long periods, characteristic of male Sedge Warblers (Catchpole 1973, 1976).

Differences between males in element, inter-element, syllable and inter-syllable durations and syllable rate: If differences between 10-min. song samples of males in these variables reflect differences between males (Table 3), then the differences may be due to various proximate factors that influence sound production in individuals such as neurobiology (Brenowits & Kroodsma 1996), physiology and morphology of the sound production system and energetic condition (Lambrechts 1996). Differences between males nevertheless provide a possible basis for individual recognition of potential mates and rival males (Falls 1982; Stoddard 1996). Syllable rate, however, has been mentioned as a possible cue for female choice in the Sedge Warbler (Catchpole et al. 1984). If significant differences between 10-min. song samples of males in syllable rate reflect differences between males (Table 3), a possible basis for female choice arises. There is support for this idea from the following studies on songbirds with discrete songs where song rate has been measured. A correlation between song rate and food abundance has been found in several songbird species, suggesting that song rate is energy-limited and provides reliable information about the energetic condition of males (Searcy 1979; Gottlander 1987; Reid 1987; Alatalo, Glynn & Lundberg 1990; Cuthill & Macdonald 1990). Female preference for males with high song rates has been reported in various songbird species (Payne & Payne 1977; Gottlander 1987; Radesäter, Jakobsson, Andbjer, Bylin & Nyström 1987; Reid & Weatherhead 1990). Song rate also has been found to be important in female stimulation (Wasserman & Cigliano 1991) and a reliable indicator of paternal care (Greig-Smith 1982).

Although a large number of element, inter-element, syllable and inter-syllable durations and syllables comprised the analysis of differences between male song samples in these durations and syllable rate (Figs. 3a-e), the differences found may not reflect differences between males, but rather may indicate a male's state of motivation or fatigue (Lambrechts 1996) over the particular 10-min. sampling period. This may have some support as there was considerable variation within males in the different types of duration and syllable rate (Figs. 3a-e). More song samples spread over the season and over several years will have to be taken for each male, to determine whether consistent differences are found between males. This argument also applies to syllable type rate discussed below (Fig. 3f).

Differences between males in syllable type rate: Syllable type rate differed significantly between 10-min. song samples of males in my study and also may provide females with a cue for choosing particular males, if differences shown reflect differences between males (Table 3). Although syllable repertoire size has been reported as an important cue for female choice in the Sedge Warbler (Catchpole 1980, 2000; Catchpole et al. 1984; Buchanan & Catchpole 1997), it takes approximately 12.5 minutes for a male to cycle through most of his repertoire of syllable types (Wilson, Chapter 3). Syllable type rate would provide a female with more of an immediate measure of song complexity as she surveyed singing males in the population (Parker 1974; Sullivan 1994). This measure would also ease the burden on a female's memory (Kroodsma 1976; Krebs & Kroodsma 1980; Falls 1982), especially in the Sedge Warbler, which has a repertoire size exceeding 100 syllable types (Wilson, Chapter 3).

Syllable and syllable type rates as multiple cues in female choice: It is possible that both syllable and syllable type rates are involved as multiple cues in female choice, providing females with more information about particular males to make a more accurate and reliable choice in an economical time (Iwasa & Pomiankowski 1994, 1995; Sullivan 1994; Møller & Pomiankowski 1993; Johnstone 1996). Although there was a significant correlation between syllable and syllable type rates, there was no significant increase in syllable rate as syllable type rate increased above 3 syllable types/s (Fig. 7). Together with the absence of in phase variation between syllable and syllable type rate cycles in males (Table 1), there is the possibility that each rate delivers a different message to receivers about the quality of the sender, rather than each rate being a backup signal of the other (Johnstone 1996).

Structural constraints on syllable and syllable type rates: If either or both syllable and syllable type rates are sexually selected traits, the question arises whether there are any aspects of song structure that may constrain this selection (Halliday 1987; Lambrechts 1996). The following aspects were revealed in my study. First, the high maximum syllable rates (Fig. 3e), low minimum inter-syllable durations that approached zero (Fig. 3c) and strong exponential relationship between mean inter-syllable duration/s and syllable rate (Fig. 6a) indicated constraints on syllable rate. Second, there were uniform maximum syllable type rates among males that indicated a ceiling on syllable type rate (Fig. 3f). Third, as syllable type rate increased from 2 to a maximum of 5 syllable types/s, there was a drastic concomitant decrease in frequency of occurrence of this rate and decrease in high syllable rate that indicated a trade-off between syllable and syllable type rates (Fig. 7). Further evidence of a trade-off between the two variables was seen by the absence of a significant increase in syllable rate as syllable type rate increased above 3 syllable types/s (Fig. 7).

Conclusion: My study has provided quantitative evidence that syllables and their component elements are the basic vocal units in Sedge Warbler communication and that songs are artificial units. This means that songs cannot be used reliably as quantitative units to derive any measures such as syllable repertoire size in the Sedge Warbler. Studies that depend on any such derived measures will have to be reassessed. Future studies on Sedge Warbler communication should focus on syllables and elements.

My study has shown that there may be a basis for element, inter-element, syllable and inter-syllable durations and syllable rate to function in individual recognition and for syllable and syllable type rates to function in female choice in the Sedge Warbler, but more samples are required to test differences between males, after which these hypotheses remain to be tested in the field and laboratory.

3. Syllable Repertoires in the European Sedge Warbler, *Acrocephalus schoenobaenus*: Size, Composition and Biological Significance.

"To experiment first is human, to describe first, divine." (Kroodsma & Byers 1991)

Abstract

In the past, syllable repertoire size in the Sedge Warbler has been determined in the usual qualitative way without describing or defining the syllable types that comprise each male's repertoire. This gives rise to fundamental questions about syllable type definition, especially when hypotheses about the function of repertoire size are tested. The purpose of my study is to determine syllable repertoire composition and size objectively in two male Sedge Warblers using qualitative and quantitative methods so as to provide a reference for future studies involving syllable types and repertoire size in this species. In the qualitative analysis, syllable assignment to putative syllable types (PSTs) often was difficult due to complexity, variability and similarity of syllable frequency and temporal structure and involved extensive comparison of syllables from the large library of PSTs. In the quantitative analysis, most PSTs differed significantly in temporal and acoustic frequency measures in multi- and univariate analyses and were supported well by multivariate analyses. The quantitative analysis largely supports the qualitative analysis of repertoire composition and size in both males. Repertoire size is appreciably higher than previous qualitative estimates in both males and this may be attributed to differences in syllable type definition and sample time. The influence of the latter two factors on previous estimates of syllable repertoire size in the Sedge Warbler may have implications for the studies that depend on these estimates. The temporal and acoustic frequency differences between syllable types are well within songbird auditory system resolving power. These differences form a basis for the syllable types to function as fundamental units of syllable variation and for categorical perception of the syllable types in the Sedge Warbler.

Introduction

Repertoire size has received considerable attention as a possible cue in male contest and female choice studies testing Darwin's theory of sexual selection (Searcy & Andersson 1986; Andersson 1994; Searcy & Yasukawa 1996; Macdougall-Shackleton 1997). Repertoires usually are divided into different types of songs or syllables (Searcy 1992). The use of song types as units of vocal organisation depends on the presence of discrete songs with an apparent finite number of syllable type combinations, while the syllable type is favoured when a sustained succession of syllables is produced with an open-ended number of type combinations, as in the Sedge Warbler (Catchpole 1976).

Sedge Warbler song is some of the longest, most complex and variable known to science (Catchpole 1976, 2000). Syllable repertoire size has been used as a measure of song complexity in this species and has been reported as an important cue in female choice (Catchpole 1980; Catchpole, Dittami & Leisler 1984; Buchanan & Catchpole 1997), and as a reliable indicator of parasitism (Buchanan, Catchpole, Lewis & Lodge 1999) and paternal effort (Buchanan & Catchpole 2000). Syllable repertoire size has been used in a test of the phenotype-linked fertility insurance hypothesis and the directional asymmetry hypothesis (Birkhead, Buchanan, Devoogd, Pellatt, Székely & Catchpole 1997). The response of territorial male Sedge Warblers to playback of different repertoire sizes also has been tested (Catchpole 1989). A study of the *Acrocephalus* and closely related *Locustella* genera indicates that sexual selection may have resulted in the evolution of syllable repertoire size by increasing the size of the higher vocal centre, a controlling area of the brain (Székely, Catchpole, De Voogd, Marchl & De Voogd 1996).

However, the different types of syllables in Sedge Warbler song have not been described or defined, not least in terms of criteria for type assignment. This is a weakness shared by other studies investigating the function of song repertoires (Podos, Peters, Rudnicky, Marler & Nowicki 1992; Tchernichovski, Nottebohm, Ho, Pesaran & Mitra 2000). A fundamental question naturally arises about the nature of these units of vocal organisation which are used to test Darwin's theory of sexual selection, and to address other functions of song (Kroodsma & Byers 1991).

Estimated repertoire size in the Sedge Warbler has been determined by visual inspection of spectrograms (Catchpole 1976, 1980; Székely et al. 1996; Buchanan & Catchpole 1997). Syllable types in a sample of continuous song are identified, counted and plotted against the cumulative number of songs, with the asymptote of this function being taken as an estimate of repertoire size

(Catchpole 1976; Kroodsma 1982). Estimates of syllable repertoire size range in the Sedge Warbler have varied considerably (Table 1). This may be due to an increase in the number of males sampled (Catchpole 2000), songs being used to derive syllable repertoires (Wilson, Chapter 2), and insufficient sampling time considering that inter-observer variability was minimized (Catchpole 1976, 1980; Székely et al. 1996; Buchanan & Catchpole 1997).

It is fundamentally important that syllable types and estimated repertoire size be determined in an accurate and reliable way for a particular species, to enable repeatable empirical studies (Isaac & Marler 1963; Tinbergen 1963; Kroodsma 1982; Hauser 1996; Macdougall-Shackleton 1997; Jones, Ten Cate & Bijleveld 2001). Furthermore, a biologically relevant determination of repertoire size can be undertaken effectively only after repertoire size has been determined accurately and reliably (Horn & Falls 1996; Macdougall-Shackleton 1997). The units of sound thought to comprise a male's repertoire must be known before females and males can be assayed in the field or in the laboratory.

The aim of my study is to determine Sedge Warbler repertoire size and composition accurately, reliably and transparently using qualitative and quantitative methods. The objectives are to: 1) Define the syllable types used by two male Sedge Warblers. 2) Compare the results of the qualitative and quantitative analyses. 3) Explore the implications of the findings. Although this is the first quantitative determination of repertoire size and composition in a songbird with complex song structure and a very large syllable type repertoire, a similar quantitative method of summary descriptive measurement was used successfully to classify Solitary Vireo syllables (Martindale 1982) and to examine microgeographic variation in Nuttall's White-crowned Sparrow song (Cunningham & Baker 1989). A similar qualitative and quantitative approach was used to examine the microstructure of Swamp Sparrow song (Marler & Pickert 1984), to define natural categories among syllables in Black-capped Chickadee calls (Nowicki & Nelson 1990), and to define song categories in Field Sparrows (Nelson & Croner 1991).

Materials and Methods

Digital Acquisition of Sound Recordings

Recordings of male song were made during the breeding season at Wraysbury Lakes, Surrey, UK (51°27'N, 0°32'W). A 15-min. sample of continuous song was obtained for each of two males

Table 1. History of reported syllable repertoire size ranges in the Sedge Warbler based on estimated syllable repertoire size in individual males. Syllable types that comprised the repertoires of individual males were determined by visual inspection. The number of males involved in each study and number of songs used for each male are shown.

Syllable repertoire size range	No. of males	Sample size (No. of songs)	References
35-55	3	20	Catchpole 1976
14-41	10	10	Catchpole 1980 (part one)
35-55	≥3	Not given	Catchpole 1980 (part two)
Maximum of 50	Not given	Not given	Catchpole 1987
35-75	80	20	Székely et al. 1996; Buchanan & Catchpole 1997; Catchpole 2000

(males 1 and 4) and recorded onto a Macintosh Quadra 840 AV using Canary (Charif, Mitchell & Clark 1995). A 22.05 kHz sampling rate exceeding twice the maximum signal frequency, and 16 bit sample size were selected to prevent aliasing and minimise quantization noise respectively (Charif et al. 1995). Spectrogram settings were determined for separate frequency (resolution=21.57 Hz) and temporal analyses (resolution=0.719 ms) of syllables because of the trade-off between temporal and frequency resolution (Charif et al. 1995) and to enable optimal levels of accuracy in the quantitative spectrographic analysis (Appendix 4). Both song samples contained perch song predominantly and occasional song flights.

Qualitative Analysis

Spectrograms of male 1's (N=3946) and 4's (N=3420) syllables were examined and sorted visually into putative types according to shape, acoustic frequency and temporal pattern, maximum and minimum acoustic frequency and number of elements (Shiovitz 1975; Marler & Pickert 1984; Thompson et al. 1994). The general criterion of intra- versus inter-type variation was used to define putative types (Kroodsma 1982), whereby variation within types was judged to be less than variation between types. Comparison and verification of similar putative syllable types were aided by examining their acoustic frequency and temporal structure at different screen resolutions together with playback at normal and slow speeds to integrate visual and auditory assessments.

Putative syllable types (PSTs) were placed in one of six groups according to similarity in acoustic frequency and temporal structure (Appendix 6). This facilitated assigning some syllables, which were more difficult to categorise and testing the difference between similar PSTs in uni- and multivariate analyses. Group 1 syllables had positive frequency modulation rates and were generally based on the structure of syllable type 1 (Appendix 6). Group 2 syllables had negative frequency modulation rates and were based on the structure of syllable type 14 (Appendix 6). Group 3 syllables were composed of harmonics generally, with positive and negative frequency modulation rates, and were based on the structure of syllable type 24 (Appendix 6). Group 4 syllables generally had positive and negative frequency modulation rates and were based on the structure of syllable type 38 (Appendix 6). Group 5 syllables were composed of two to many elements which were usually made up of harmonics and had wide frequency band structure (Appendix 6). Group 6 syllables were composed of elements with a negative frequency modulation rate and relatively narrow frequency band structure (Appendix 6). The repeatability of syllable assignments to PSTs in the song samples of both males was checked after 30 days.

A modification of Catchpole's (1976) method for determining syllable repertoire size in the Sedge Warbler was used by plotting the number of new PSTs against the duration of continuous song. An asymptote was formed at approximately 12.5 min. of continuous song and hence this duration was used as a representative sample for determining estimated syllable repertoire size and composition in each male (Fig. 2).

Quantitative Analysis

PSTs comprising the estimated repertoires of the two males were identified in the qualitative analysis and 10 representative syllables of each PST were analysed spectrographically. Syllables of a particular PST (Appendix 5) were randomly selected for measurement from different phrases (Brackenbury 1978) in the 12.5 min. of song of each male, to satisfy the condition of independence of cases necessary for uni- and multivariate analyses (Sokal & Rohlf 1981; Morrison 1990; Hair, Anderson, Tatham & Black 1995). Seven accurate measurements that could be applied to all Sedge Warbler syllables were identified (Fig. 1) and the entire set of 1980 syllables was measured. Accuracy of the seven syllable measurements was checked by repeating each one 30 times for one syllable from each of the six PST groups. No significant differences were found for the different syllable measurements for each of the six groups (ANOVA: $P>0.05$), indicating a low measurement error.

Statistical Methods

General: Uni- and multivariate analyses were performed on each of the six and five groups of PSTs in males 1 and 4 respectively, using Unistat (Toker 1994). The confidence level for rejecting the Null Hypothesis was set at 0.05 and all univariate tests were 2-tailed unless otherwise stated. Infrequently or solitary occurring PSTs were not included in these analyses because they did not satisfy sample size requirements, which left 73 and 64 PSTs to be analysed for males 1 and 4 respectively (Table 2; Appendix 5). The PST groups were well represented in these analyses except male 4's group 6 which was omitted due to sample size criteria (Table 2; Appendix 5). Elements, the smaller temporal units comprising many syllables (Fig. 1), occurred in relatively few PSTs in groups 1 to 4 (Appendix 6) and consequently element duration and inter-element interval were not included in multivariate analyses for these groups. Sample sizes of PSTs, especially for the multivariate analyses, met the requirements for analysis (Sokal & Rohlf 1981; Hair et al. 1995). Data were checked for univariate normality using the Kolmogorov-Smirnov one sample test and for homogeneity of variances using Bartlett-Box F- and Hartley's F tests (Sokal & Rohlf 1981). Multivariate normality was checked according to Morrison (1990). Logarithmic, reciprocal, reciprocal square root, square root and arcsine transformations were applied to non-parametric variables (Sokal & Rohlf 1981).

Differences between PSTs were examined in a complimentary multivariate way using principal components analysis (PCA), canonical multiple discriminant function analysis (MDA) and multivariate analysis of variance (MANOVA). MDA, which maximised the differences between the pre-specified PSTs possibly masking continuous variation among them, was evaluated by MANOVA of principal component (PC) coordinates generated by PCA. The number of principal components (PCs) used in MANOVA was selected according to latent root (>1) and scree test criteria (Jackson 1991; Hair et al. 1995). The table of MANOVA results for each syllable type group was subjected to a sequential Bonferroni test to allow simultaneous inference and prevent spurious significant results (Rice 1989).

PCA, based on the correlation matrix (Jackson 1991), also allowed the identification of temporal and acoustic frequency variables that had significant eigenvector loadings, using criteria for significance (Hair et al. 1995). Duncan's multiple range tests (DMRTs) were applied to the PC coordinates to determine whether the means of the same PCs differed significantly between PSTs. These tests were used in combination with frequency distribution plots to assess whether pairs of types had bimodal distributions for each principal component. Although DMRTs examined differences between the means of types in terms of individual PCs, the inference was that types came from two different statistical populations if significant differences were found and if the frequency distribution was bimodal (Sokal & Rohlf 1981; Spector 1994).

A t test was used to determine whether PSTs 13 and 14 differed in their frequency modulation rate, which was calculated by dividing the frequency range by the duration of each syllable.

Internal and external validity in the MDAs were ensured by dividing each data set randomly into analysis and holdout samples and validating the discriminant functions (Hair et al. 1995).

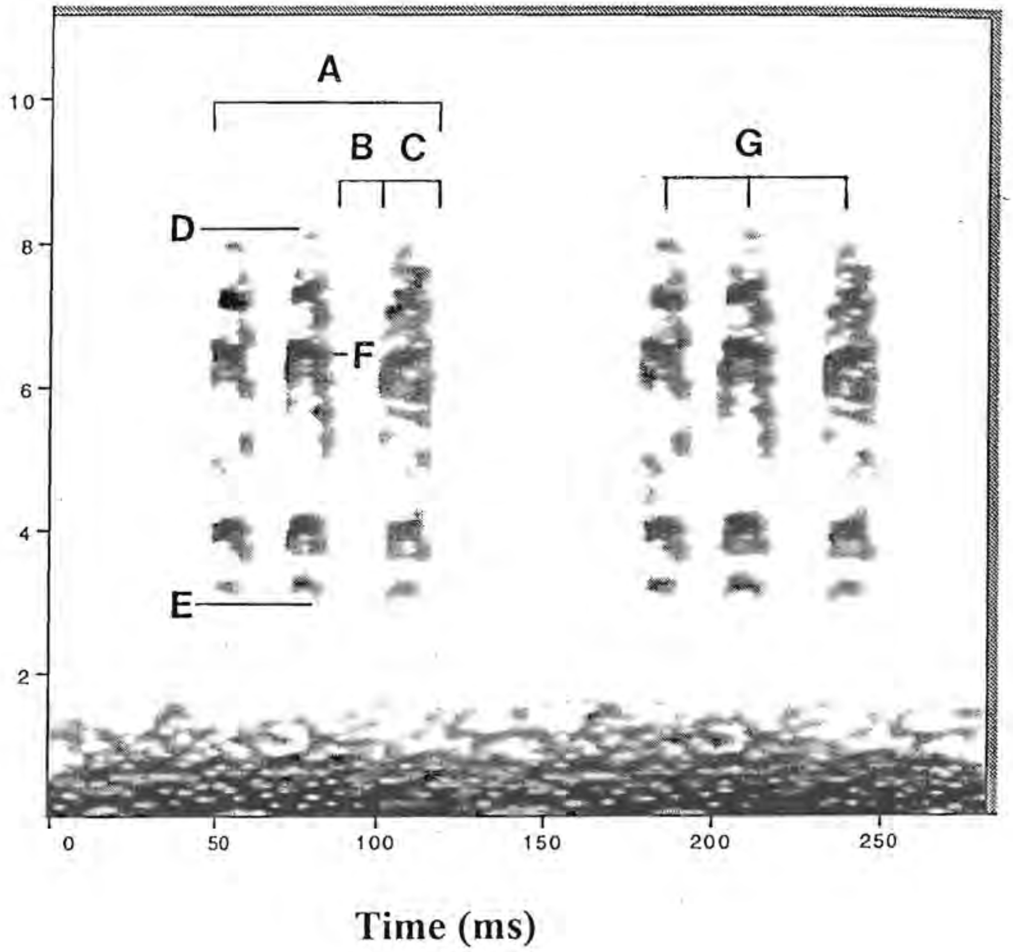


Figure 1. Spectrogram of two Sedge Warbler syllables of putative syllable type 103 and their component elements, illustrating examples of the seven temporal and acoustic frequency measurements, which were used in the quantitative analysis: Syllable duration (A), inter-element duration (B), element duration (C), maximum syllable frequency (D), minimum syllable frequency (E), peak syllable frequency (F) and the number of elements comprising a syllable (G). Peak syllable frequency, which is the frequency at which the highest amplitude in the syllable occurs, is not easily shown.

Syllable types were considered to be strongly supported in the MDA if they each received ≥80% classification accuracy. Significant Wilk's lambda values and high structure matrix loadings were obtained for all variables in the MDAs for both males.

Splitting of PSTs: There was a need for re-assigning or possible splitting if: 1) MDA revealed a low classification accuracy (<50%) for a particular PST and that type was misclassified as another or several other PSTs. 2) MANOVA found no significant differences between the particular PST and the other types with which it was misclassified. 3) DMRTs showed no significant differences between these types or a significant difference in a PC that explained the least variance. 4) Examination of spectrograms found no type assignment errors. Then the following steps were implemented: 1) Affected syllables were re-assigned according to the MDA results, and MDA, MANOVA and DMRTs were re-run on the adjusted data set. 2) Re-assignments were accepted if they were supported by the multi- and univariate analyses. 3) If the re-assignments were not supported by these analyses, the affected syllables were assigned to a new PST, and the analyses were re-run. 4) The new PST was accepted if there was multi- and univariate analytical support.

PSTs kept separate: PSTs became analytically determined syllable types (ADSTs) if MANOVA and DMRTs showed significant differences and MDA revealed strong support (≥80%). If MANOVA indicated no significant difference between two or more PSTs, they still became ADSTs if MDA revealed ≥80% classification accuracies and/or DMRTs showed significant differences in PCs that explained the most variance and that had significant eigen-vector loadings. If DMRT results were the main criterion, classification accuracies would have to be >50% and mutual misclassification <50% in MDA.

Merging of PSTs: If PSTs were not supported by the multi- and univariate analyses, spectrograms of their syllables were re-examined to ensure that type assignment errors had not been made. Two or more PSTs were merged to form one ADST if the following conditions held: 1) MANOVA revealed no significant differences. 2) MDA indicated low classification accuracies (<50%) and high mutual misclassification (≥50%). 3) DMRTs showed no significant differences or a significant difference in a PC that explained the least variance.

Results

General: In the qualitative analysis, 111 and 114 PSTs were identified in male 1's and

4'srepertoires respectively (Fig. 2; Appendix 5). Male 1 produced new PSTs at a higher rate than male 4 for the first 57 types after which there was convergence and similarity in rate, with male 4 ultimately overtaking male 1 (Fig. 2). Syllable assignments to the wide range of PSTs were found to be consistent and repeatable for most of the syllables examined in each male's song sample. Repeatability of 100% was obtained for PSTs in syllable type groups 1 to 4 and 6 and 98% for PSTs in syllable type group 5. However, extensive comparison of syllables from the large library of PSTs often was necessary due to complexity, variability and similarity of syllable frequency and temporal structure, especially for group 5 syllables which constituted the biggest group (Appendices 5, 6). In both males, 79% of PSTs in groups 1 to 4 were composed of one temporal unit, whereas all PSTs in groups 5 and 6 were composed of two or more elements (Appendix 6). Most PSTs covered a broad acoustic frequency range and showed amplitude and frequency modulation, harmonics and repetition of elements (Appendix 6).

In the quantitative analysis, the following points were evident from each male's PST groups. PCs used in MANOVA explained a high percentage of the variance (Table 2), and eigenvector loadings were significant. MANOVA showed significant differences in 98% (N=745) and 95% (N=503) of the pairwise comparisons of PSTs in males 1 and 4 respectively. MDA showed high mean classification accuracies, only two PSTs with low classification accuracies (<50%, Fig. 3) and no PSTs with high mutual misclassification (>50%). DMRTs revealed significant differences between the means of PC coordinates in 99.6% (N=760) and all (N=528) of the pairwise comparisons of PSTs in males 1 and 4 respectively (Table 2). PSTs that differed mainly in the number of elements in the qualitative analysis (Appendix 6) were well supported by multi- and univariate analyses except for those that did not differ significantly in MANOVA (Table 2).
PSTs not split: Male 4's type 2 (group 1) received a low classification accuracy (Fig. 3) and was misclassified as type 10 by 41%. Misclassified type 2 syllables were not re-assigned to type 10 because significant differences were found (MANOVA: $P<0.05$; DMRTs: PCs 2, 3, $P<0.05$). These two types were very different in acoustic frequency and temporal structure (Appendix 6).

Male 4's type 40 (group 4) was classified with 43% accuracy (Fig. 3) and was misclassified as type 42 by 29% and as types 39 and 41 by 14%. Misclassified type 40 syllables were not re-assigned because DMRTs showed significant differences between the types in two PCs that had significant loadings for syllable duration and maximum acoustic frequency (Table 2, Appendix 6).

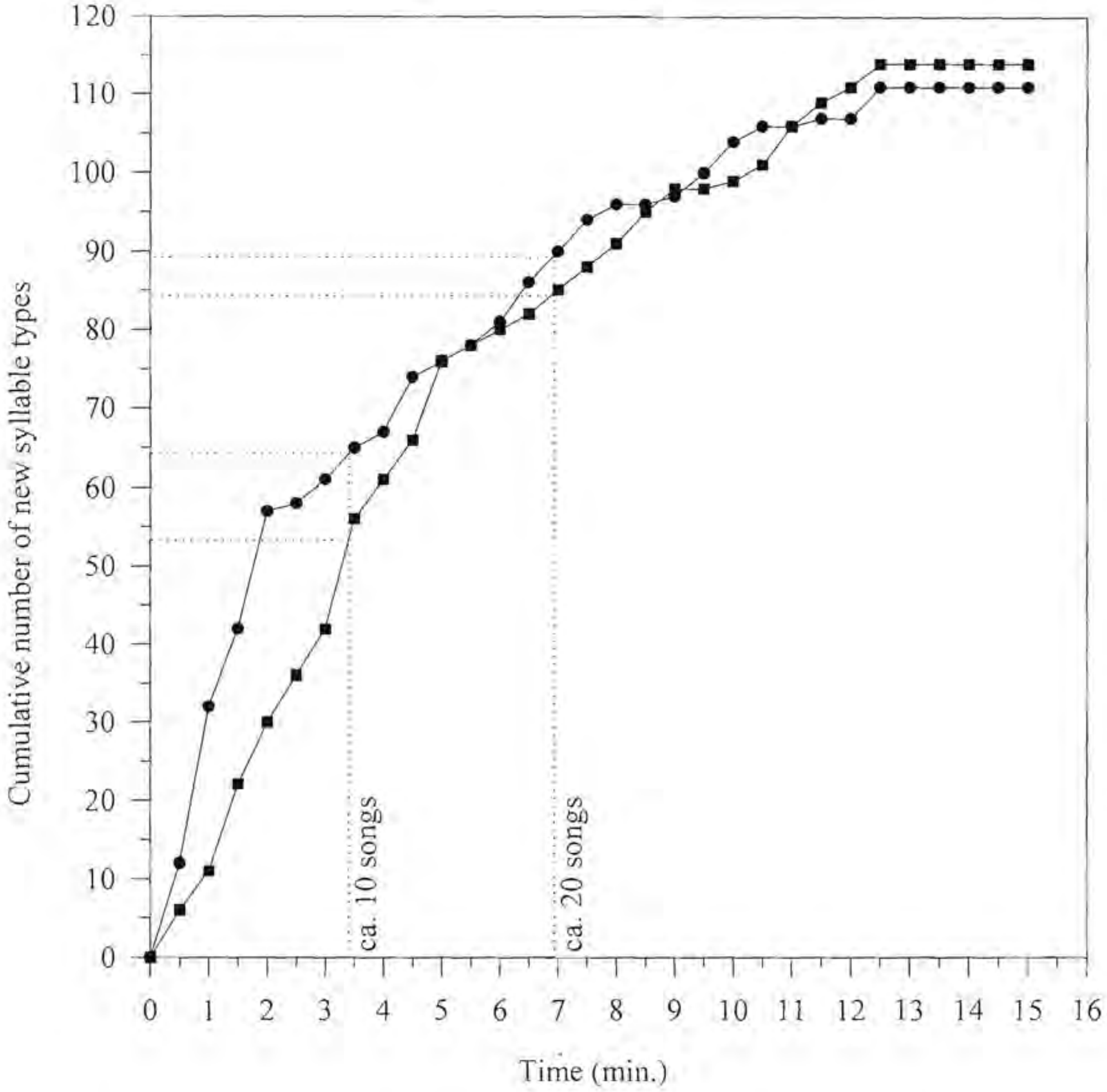


Figure 2. Determination of estimated syllable repertoire size and song sample duration in males 1 (—●—) and 4 (—■—). Ten and 20 songs were used in previous estimates of repertoire size in the sedge warbler (see Table 1) and these sample sizes are approximately equal to 3.44 and 6.91 min. of song respectively (.....) according to summary data provided by Catchpole (1976). Both sample sizes produce estimated syllable repertoire sizes that are appreciably less (.....) than those determined by the formation of asymptotes for males 1 and 4. Asymptotes are formed at approximately 12.5 min. of continuous song which is used as a representative sample for determining estimated syllable repertoire size and composition in each male.

Table 2. Summary PCA, MANOVA and D[illegible] yllable type groups in two male Sedge Warblers. Numbers of syllable types are shown that [illegible] the qualitative analysis (Qualitative), b) included in (Incl.) and determined by (Det.) the quantitative analysis, and c) determined by synthesis of both analyses (Final). The following also are shown: (i) The number of variables (No. vars.) included in each PCA. (ii) Principal components (PC) with their cumulative variance (cum. var.) used in each MANOVA. (iii) PSTs not differing significantly in MANOVA after sequential Bonferroni correction (P≥0.05). (iv) Principal components whose means differed significantly between types in DMRTs (P<0.05). (v) PSTs that were merged (M) and kept separate (S). (vi) PSTs (*) in each pairwise comparison that differed mainly in the number of elements in the qualitative analysis are indicated.

Syllable Type Group	Male	No. of Syllable Types Qual-itative	Quantitative Incl.	 Det.	 Final	No. vars.	PC (cum. var.)	MANOVA PSTs		DMRT PC
1	1	9	7	7	9	5	1-3 (92%)	6, 7	S	2, 3
								6, 8	S	1, 3
								7, 8	S	3
1	4	9	7	7	9	5	1-3 (89%)	2, 7	S	1, 2
								7, 8	S	3
2	1	7	6	6	7	5	1-3 (93%)	13, 14	S	2
2	4	7	5	5	7	5	1-3 (95%)	16, 33	S	2, 3
3	1	10	9	9	10	5	1-4 (95%)	21, 26	S	1, 2, 4
								21, 32	S	3, 4
3	4	12	11	11	12	5	1-4 (95%)	21, 26	S	1-4
								21, 32	S	1, 2, 4
								25, 28	S	1, 4
4	1	12	11	11	12	5	1-4 (94%)	39, 40	S	1, 3, 4
4	4	16	14	14	16	5	1-4 (98%)	39, 40	S	2, 4
								39, 42	S	2, 3
								40, 41	S	2, 3
								40, 42	S	2, 3
								41, 42	S	2, 3
5	1	65	36	34	63	7	1-4 (86%)	64, 101	S	1, 2
								64, 112	S*	1
								78, 121	S	1, 3, 4
								87, 89	M*	0
								87, 91	M*	0
								89, 91	M*	0
								101, 109	S	1-4
								106, 131	S	1-3
								130, 135	S*	2
								140, 144	S*	2
5	4	67	27	27	67	7	1-4 (89%)	64, 101	S	1, 2, 4
								64, 112	S*	1, 2, 4
								67, 68	S	2, 3
								67, 101	S	1-3
								71, 72	S	1-3
								73, 74	S	2, 4
								74, 104	S*	2, 3
								74, 118	S	1-3
								80, 101	S	1, 2
								81, 103	S	1-2
								92, 96	S	2-4
								96, 112	S	2
								103, 135	S	1, 2, 4
								104, 107	S	1, 2
6	1	8	4	4	8	5	1-2 (89%)	164, 166	S	2

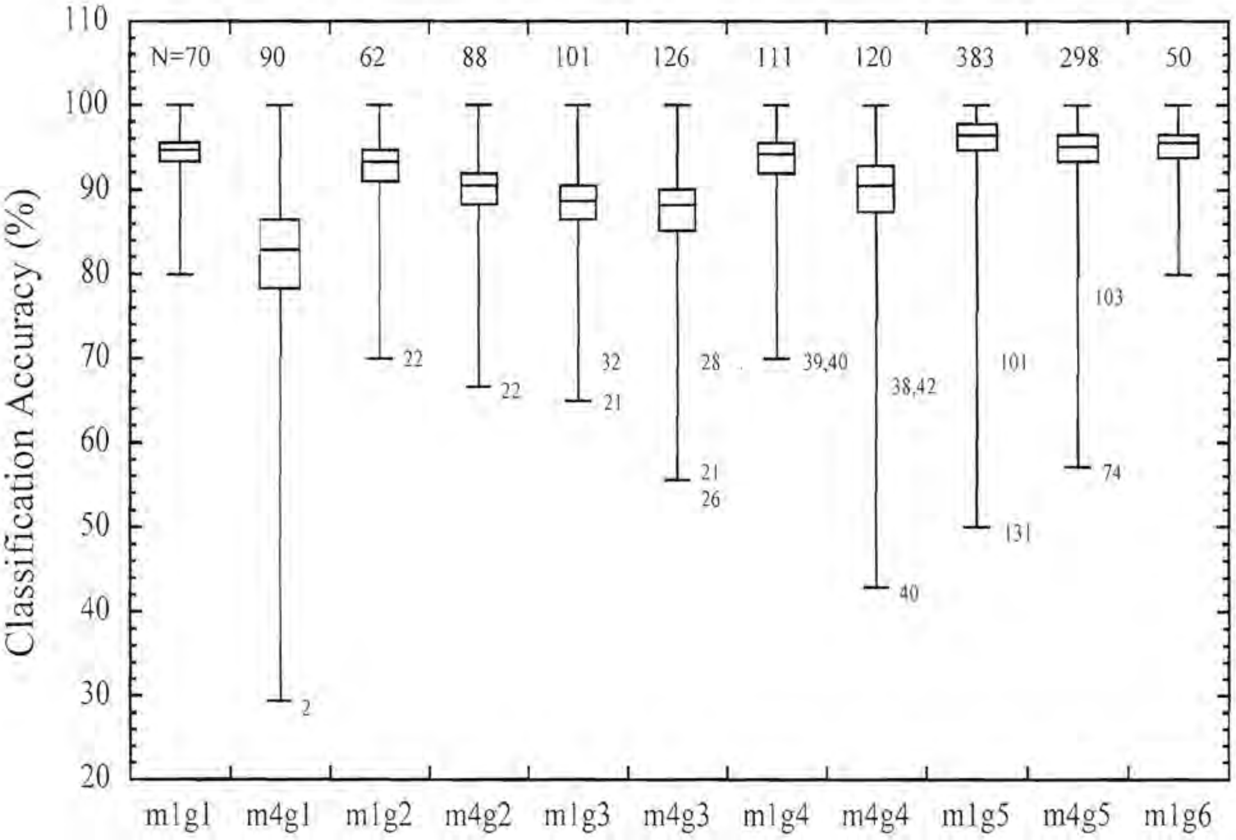


Figure 3. Descriptive statistics of male 1's (m1) and 4's (m4) syllable type group (g1-g6) classification accuracies obtained by canonical multiple discriminant function analysis, showing a high mean classification accuracy (>80%) for each syllable type group. Boxes show mean±1SE and whiskers indicate maxima and minima. Syllable types with an accuracy <80% and sample sizes (N) are indicated.

PSTs kept separate although not differing significantly in MANOVA: These PSTs were kept separate for the following reasons: For most of them (Table 2), MDA showed strong support (≥80%, Fig. 3) and mutual misclassification was low (<15%). For all of them, DMRTs revealed significant differences between the means of the coordinates of PCs that had significant loadings for temporal and/or acoustic frequency variables (Table 2). In addition to these reasons, male 1's types 13 and 14 differed significantly in frequency modulation rate (t test: $t_{6.5}$=-11.2, P<0.00001; Appendix 6).

PSTs merged: Although male 1's types 87, 89 and 91 received strong support in the MDA (Fig. 3), they were merged because MANOVA and DMRTs found no significant differences (Table 2). These three types were very similar in acoustic frequency and temporal structure, but differed in the number of elements (Appendix 6).

Repertoire size: The 111 PSTs identified in male 1's repertoire by qualitative analysis were reduced by quantitative analysis to 109 syllable types. Quantitative analysis indicated that no changes should be made to the 114 PSTs that were identified in male 4's repertoire by qualitative analysis.

Discussion

The size and composition of the syllable repertoires of two male Sedge Warblers were determined by qualitative and quantitative analyses, which produced very similar results. Most syllable types differed significantly in temporal and acoustic frequency measures in multi- and univariate analyses and were strongly supported by MDA. Repertoire size in both males was appreciably higher than previous qualitative estimates.

Qualitative analysis and the influence of subjectivity: In this analysis, syllables occurring in long natural sequences of song were assigned to types within each male according to shape, acoustic frequency and temporal pattern, maximum and minimum acoustic frequency and number of elements using a rigorous comparative method. Assignment, however, often was difficult due to complexity, variability and similarity of syllable frequency and temporal structure and involved extensive comparison of syllables from the large library of PSTs (Appendix 6). There are problems associated with subjective assessments of similarity that distort the real relationships among syllables in unpredictable ways, and assessments can vary according to stringency of criteria (Marler & Pickert 1984; Martindale 1982; Podos et al. 1992; Jones et al. 2001). The aspects of syllable structure that complicated assignment in my study may explain some of the variation in repertoire size range in the past (Table 1) and differences between previous and present estimates of repertoire size. A recent study of song complexity and song structure in the closely related Moustached Warbler, *Acrocephalus melanopogon* (Feßl & Hoi 2000), revealed that repertoire size after only 2 min. of song was twice as high as that calculated by Catchpole (1980). A possible explanation for the discrepancy was given as a difference in the stringency of syllable type definition between the two studies (Feßl & Hoi 2000). The influence of subjectivity in the qualitative determination of syllable types in my study was checked to a large degree by the quantitative analysis.

Quantitative analysis - minimising the influence of subjectivity: In this analysis, most PSTs differed significantly in temporal and acoustic frequency measures (Table 2), and the high classification accuracies obtained in MDA indicated limited overlap between PSTs (Fig. 3). In the numerous multi- and univariate analyses that were conducted, only one MDA yielded anomalous results involving male 4's types 2 and 10. The complimentary MANOVA and DMRT approach detected this anomaly. Multi- and univariate analyses were used to determine whether PSTs should be split, merged or kept separate. Most PSTs were kept separate because the analyses indicated consistent differences. Only for a minority of PSTs did MANOVA show no significant differences (Table 2), but for most, MDA and DMRTs gave consistent support. All PSTs that differed mainly in the number of elements in the qualitative analysis, were well supported by multi- and univariate analyses, except for male 1's types 87, 89 and 91, which failed to show significant differences in MANOVA and DMRTs and which consequently were merged as one syllable type (Table 2; Appendix 6). Multivariate analysis of acoustic measurements has been used successfully to define song categories in Field Sparrows (Nelson & Croner 1991) and natural categories among syllables in Solitary Vireo Songs (Martindale 1982) and in the chick-a-dee calls of Black-capped Chickadees (Nowicki & Nelson 1990). In the latter study, as with my study, multivariate analysis yielded very similar results to those produced by visual inspection.

Although the method of summary descriptive measurement was suitable for the quantitative analysis of Sedge Warbler syllable types, it has limitations. First, there is a level of subjectivity as it is based on pre-specified variables (Fig. 1). Second, these variables were not able to capture the complexity of Sedge Warbler acoustic frequency and temporal structure, notably harmonics which are common (Appendix 6). Some PSTs found to be similar in the quantitative analysis, differed in aspects of acoustic frequency and temporal pattern not captured by the standard set of measurements, e.g., types 7 and 8 (Table 2; Appendix 6). Third, due to the labour intensive and time-consuming nature of the work, only two males were analysed comprehensively. Recently, a procedure was developed for the automated measurement of acoustic similarity that is likely to solve these three limitations (Tchernichovski et al. 2000). This procedure probably also will be useful for assessing rare syllable types that could not be included in the uni- and multivariate analyses and that comprised an appreciable proportion of the repertoires of the two males.

Song sample duration and the determination of estimated syllable repertoire size: Apart from the possible influence of subjectivity in explaining differences between previous and present estimates of repertoire size in male Sedge Warblers (Table 1), my study has drawn attention to song sample duration as another possible factor. Previous estimates have been based on 10 and 20 songs, which equal ca. 3.44 and 6.91 min. respectively, using Catchpole's (1976) mean song duration of 19.49 s and mean song interval of 1.31 s. Much longer samples of 12.5 min. were necessary to capture most of the syllable types in each male's repertoire (Fig. 2). Thus, repertoire size in individual males and repertoire size range in the Sedge Warbler probably have been underestimated in the past. Figure 2 also shows that individual differences in repertoire size that were evident early on in the sample provided an inaccurate indication of later differences. This is contrary to the finding of Catchpole (2000) who suggested that Sedge Warbler song structure was designed to reveal a male's repertoire as quickly as possible to attract a mate before rival males.

Implications of subjectivity and insufficient song sample duration: The influence of subjectivity and insufficient song sample duration on previous estimates of syllable repertoire size in the Sedge Warbler may have adverse implications for the studies that depend on these estimates. The implications are likely to be compounded if songs are used to derive syllable repertoires (Wilson, Chapter 2). All three factors probably will result in inaccurate estimates of syllable repertoire size that have the following outcomes. The inaccurate estimates may be affected in different, unpredictable ways (Martindale 1982; Podos et al. 1992) changing the significant relationships found between repertoire size and pairing date (Catchpole 1980; Buchanan & Catchpole 1997), female copulation solicitation display (Catchpole et al. 1984), age (Birkhead et al. 1997), parasite load (Buchanan et al. 1999) and paternal effort (Buchanan & Catchpole 2000). Alternatively, the error may be systematic and may affect all males in the same way, thus not changing the significant relationships found between repertoire size and the other variables mentioned.

Temporal and acoustic frequency differences between syllable types - biological significance: Although the differential behavioural response of female Sedge Warblers to the playback of different repertoire sizes indicates that Sedge Warbler syllable types have some meaning to females (Catchpole et al. 1984), how likely is it that Sedge Warblers are able to detect the temporal and acoustic frequency differences found between syllable types in my study? A close match has been shown between variability in vocalisations and auditory system resolving power in birds (Dooling 1982; Okanoya & Dooling 1988). Temporal resolution of 2 ms has been found for the auditory systems of several species of songbird for narrow band (Wilkinson & Howse 1975) and wide band signals (Klump & Maier 1989; Klump & Gleich 1991), while frequency resolution of 20 Hz at 2 kHz has been demonstrated for birds (Dooling 1982), and songbird ability to detect small changes in acoustic frequency is well developed (Klump 1996). Close correspondence also has been found between hearing sensitivity and the acoustic frequency characteristics of vocalisations in songbirds (Dooling 1982). Thus it is highly likely that Sedge Warblers are able to detect temporal and acoustic frequency differences found between syllable types, including types that differed mainly in the number of elements. These differences are likely to facilitate the ability of Sedge Warbler receivers to detect (Brenowitz 1982; Dooling 1982; Wiley & Richards 1982) discriminate, identify and remember syllable types (Horn & Falls 1996) that will result in more effective communication (Johnstone 1997). In this way, these differences form a basis for the syllable types to function as fundamental units of syllable variation (Searcy, Nowicki & Peters 1999) and for categorical perception of the syllable types in the Sedge Warbler (Horn & Falls 1996). Syllable types would have to function in this way and to be perceived categorically if female Sedge Warblers are able to use syllable repertoire size in their assessment of mate quality as argued (Catchpole 2000).

It may be argued that categorical perception may not be possible for syllable types that were very similar in acoustic frequency and temporal structure, but that differed in the number of elements or duration (Appendix 6). Nelson and Marler (1989), however, demonstrated that territorial male Swamp Sparrows were able to divide a natural continuum of note duration into two categories that have different roles in song organisation. Thus structural differences can have large consequences, as similar signals on either side of a perceptual category boundary would appear very different to receivers (Horn & Falls 1996).

Receiver memory may place a significant constraint on the number of perceivable syllable or song types in songbird repertoires, especially if identification of types is important. Although the ability of songbirds to recognise individuals, neighbours and strangers using acoustic cues in their songs is impressive (Falls 1982; Stoddard 1996; O'Loghlen & Beecher 1999), it remains to be established whether female Sedge Warblers can use syllable repertoire size of the magnitude found in my study to assess mate quality in a population as argued (Catchpole 2000).

Conclusion: The size and composition of the complete estimated syllable repertoires of two male Sedge Warblers were determined by quantitative and qualitative analyses which yielded very similar results, but appreciably higher estimates of size than previous qualitative studies. This discrepancy may be due to subjectivity of syllable type determination and insufficient song sample duration of the previous studies that are likely to have produced inaccurate estimates of syllable repertoire size. This error may affect the significant relationships found between repertoire size and other variables in previous studies. Significant quantitative differences and qualitative differences in acoustic frequency and temporal structure found between syllable types form a basis for their function as fundamental units of syllable variation and for their categorical perception in the Sedge Warbler.

4. Syllable Repertoire and Syllable Type Differences in the European Sedge Warbler, *Acrocephalus schoenobaenus*.

Abstract

The structure of syllable types and composition of syllable type repertoires of 11 males in a breeding population of Sedge Warblers were studied. Male repertoires were composed of syllable types, which were qualitatively and quantitatively distinct from one another in temporal and acoustic frequency characteristics. In the qualitative analysis, there was ≥95% repeatability of syllable assignments to the wide range of putative syllable types (PSTs). In the quantitative analysis, multi- and univariate analysis of temporal and acoustic frequency variables revealed significant differences between PSTs, which also were supported well by canonical multiple discriminant function analysis (MDA). Structural distinctness of syllable types provides a quantitative basis for them to function as fundamental units of Sedge Warbler vocal repertoires and for categorical perception and effective communication in this species. MDA and MANOVA of temporal and acoustic frequency variables showed that there was structural constancy within syllable types among males, and within males at different times of the breeding season and in consecutive years. In MDA, syllables were classified correctly regardless of which males sang them or whether they were sung at different times of the season or in different years. In MANOVA, no significant differences were found within syllable types between males, and within males at different times of the season and in consecutive years, in a large proportion of syllable types and in the majority of males. Non-metric multidimensional scaling and histogram analysis showed that there were differences in the frequency of occurrence of syllable types between males in song samples taken in the same season, and within males in song samples taken at different times of the season and in consecutive years. Log-linear analysis indicated that the differences within males between samples taken at different times of the season were significant. Approximately 50% of the syllable types were shared between the complete repertoires of two males and a similar proportion was peculiar to each of these males. Out of the 30 syllable types that were sampled, 17% were shared by all 11 males, with a high proportion (47%) being shared by 9 of the 11 males, while none were peculiar to any of these males. Structural distinctness, constancy and sharing of syllable types provide a basis for shared types to function as part of the Sedge Warbler specific mate recognition system. The findings are discussed in terms of the Recognition Concept, individual recognition and sexual selection and provide a basis for further research in these areas in the Sedge Warbler.

Introduction

Vocal complexity and differences between individuals are central issues in studies testing Darwin's theory of sexual selection (Andersson 1994; Macdougall-Shackleton 1997). In contrast, widespread stability or constancy and minimal variability of acoustic features are important predictions of the Recognition Concept (Paterson 1985, 1993; Villet 1995), while constancy and presence of acoustic signature features in individuals are essential for individual vocal recognition (Falls 1982; Beecher 1989; Weary & Krebs 1992; Stoddard 1996).

Vocal complexity usually is measured simply in terms of the number of song or syllable types an individual sings and is expressed as repertoire size (Kroodsma 1982; Catchpole & Slater 1995). Song and syllable types are thought to be important functional units in songbird communication (Kroodsma & Miller 1982, 1996; Kroodsma & Byers 1991; Andersson 1994; Catchpole & Slater 1995; Macdougall-Shackleton 1997).

There are two factors that lessen the importance given to song and syllable types. First, they usually are determined subjectively by visual inspection of spectrograms without any description or definition (Catchpole 1980; Buchanan & Catchpole 1997; Macdougall-Shackleton 1997). In species with large, complex repertoires like the Sedge Warbler and the closely related Moustached Warbler, *Acrocephalus melanopogon*, this method has yielded considerable variation in repertoire size range (Feßl & Hoi 2000; Wilson, Chapter 3). Very few studies have used quantitative methods to determine repertoire size as emphasised by Podos, Peters, Rudnicky, Marler and Nowicki (1992), and to date there have been no quantitative studies determining repertoire composition and size in species with large, complex repertoires. Second, structural differences within and between types have not been described well within and among conspecific males (Podos et al. 1992; Catchpole & Slater 1995; Kroodsma & Miller 1996), despite the fact that repertoire differences based on qualitative studies have received much attention in a wide range of species in the following ways. Differences in repertoire size between conspecific males have been well documented (Searcy & Andersson 1986; Andersson 1994; Catchpole & Slater 1995; Searcy & Yasukawa 1996; Macdougall-Shackleton 1997). Song type differences within the same season and differences and constancy between years have been reported in individual males (Avery & Oring 1977; Payne 1985; Sorjonen 1987; Trainer 1989). Since there is evidence that intra-type differences may be functionally as important as inter-type differences (Stoddard, Beecher & Willis 1988), both kinds of differences need to be described well.

There is evidence of differences in repertoire composition among males in the occurrence of syllable types (Catchpole 1976). Rarity and commonness of types also have received attention, e.g., the presence of unique types in individual males has been reported (Slater, Ince & Colgan 1980; Slater 1986), while in many species, sharing of song types (Kroodsma & Verner 1978; Morton 1987; Ewert & Kroodsma 1994; Beecher 1996; Kroodsma 1996; Hughes, Nowicki, Searcy & Peters 1998; Molles & Vehrencamp 1999) and syllable types (Catchpole 1976) has been found. However, these studies are based on the qualitative determination of repertoire composition, and few have explored differences within males.

Structural differences within and between syllable types and differences in syllable repertoire composition were studied in a breeding population of Sedge Warblers using qualitative and quantitative methods by testing the following hypotheses: i) At the population level, syllable repertoires of males are composed of syllable types, which are qualitatively and quantitatively distinct in temporal and acoustic frequency characteristics and thus form the basis for a communication system in this species. ii) There is structural constancy of syllable types in the population that provides a basis for more reliable communication, which has the following two predictions: a) Syllables of a particular type are structurally similar among males and b) in individual males when sung at different times of the breeding season and in different seasons. iii) There are differences in the levels of abundance of syllable types among males and within males at different times of the season and in different seasons. iv) A proportion of syllable types is shared in a population, while other syllable types are peculiar to individual males. Sharing of syllable types may provide the basis for a specific-mate recognition system as defined by Paterson (1985, 1993), while peculiar types may provide a basis for individual recognition and female choice.

The testing of these hypotheses will help establish a quantitative basis for determining the functional significance of syllable types and for understanding vocal complexity in terms of differences in and constancy of syllable type structure and repertoire composition in the Sedge Warbler.

Materials and Methods

Acquisition of sound recordings: A breeding population of Sedge Warblers was studied from 1988 to 1990 at Wraysbury Lakes, Surrey, UK (51°27'N, 0°32'W) and sound recordings were made as described by Wilson (Chapter 2). The song of two males from 1990 was analysed and the composition and size of their estimated syllable repertoires were determined (Wilson, Chapter 3). A second song sample of both males and two song samples of a third male from 1990 were used to study syllable type differences within the same season. The first and second song samples were recorded near the beginning and end of each male's advertising period, which lasted for 16, 8 and 26 days. Song samples of two males from both 1988 and 1989 were used to study syllable type differences between years. In addition to the five males already mentioned, a further six males were chosen randomly for spectrographic analysis from 1990's 18 males for the study of syllable type differences between males. The 14 continuous song samples of the 11 males, excluding the two samples already analysed (Wilson, Chapter 3), were recorded onto a Macintosh Quadra 840 AV using Canary (Charif, Mitchell & Clark 1995) as described by Wilson (Chapter 3).

Qualitative analysis of putative syllable types (PSTs): Spectrograms of each sample's syllables were sorted by visual inspection into PSTs, each of which was placed in one of six syllable type groups as described by Wilson (Chapter 3). The repeatability of syllable assignments to PSTs in each of the 14 song samples was checked after 30 days. A representative duration of 12.5 min. that captured most of the PSTs in each song sample was used as described by Wilson (Chapter 3).

Quantitative analysis of PSTs: Ten syllables of each of 30 randomly chosen PSTs from syllable type groups 1 to 5 (Fig. 4) were analysed spectrographically. In this analysis, syllables of a particular PST were randomly selected for measurement from different phrases (Brackenbury 1978) in each of the 14 12.5 min. song samples of the 11 males. Seven accurate measurements that could be applied to all the Sedge Warbler syllables were identified and the entire set of 4104 syllables was measured (Fig. 2c), using the same methods as for two males studied previously (Wilson, Chapter 3).

Structural differences between PSTs among males: Uni- and multivariate analyses were performed on each of the five syllable type groups using Unistat (Toker 1994) to determine whether PSTs should be split, kept separate or merged to form analytically determined syllable types (ADSTs). The same uni- and multivariate methods were used as for the study on two males

(Wilson, Chapter 3), except that in the present study the PSTs of all males in each syllable type group were analysed together, whereas in the previous study the PSTs of the two males in each syllable type group were analysed separately. The confidence level for rejecting the Null Hypothesis was 0.05 and all univariate tests were two-tailed unless otherwise stated. The 67 syllable types of male 1 and 64 syllable types of male 4 that were determined by quantitative analysis of syllable type groups 1 to 5 (Wilson, Chapter 3) were included with the 30 PSTs for uni- and multivariate analyses. In total, 98 syllable types were analysed. Elements, the smaller components of many syllables (Fig. 2c), occurred in relatively few syllable types in syllable type groups 1 to 4 (Appendix 6) and consequently element duration and inter-element interval could not be included in the multivariate analyses that were conducted in each of these groups. PSTs were split, kept separate or merged to form ADSTs according to the same criteria that were used by Wilson (Chapter 3).

Structural differences within syllable types: The same acoustic frequency and temporal measurements, which were used to define ADSTs, were used to study differences within 49 syllable types between males that sang in the same season, and in particular males that sang at different times of the season and in consecutive years. Multivariate analysis of variance (MANOVA) of principal component (PC) coordinates that were generated by principal components analysis (PCA), was conducted on ADSTs in each of the five syllable type groups. The number of principal components (PCs) used in MANOVA was selected according to latent root (>1) and scree test criteria (Jackson 1991; Hair et al. 1995). The table of MANOVA results for each syllable type group was subjected to a sequential Bonferroni test to allow simultaneous inference and prevent spurious significant results (Rice 1989). A one-tailed Fisher exact test was used to determine whether the number of significant differences found within syllable types in males that sang at different times of the season was significantly less than the number found between males that sang in the same season.

Differences in syllable type occurrence: ADSTs of the 30 PSTs that formed part of the uni- and multivariate analyses were ranked according to whether they occurred frequently, rarely (<5 syllables) or not at all in each of the 12.5 min. song samples. The data matrix was analysed using non-metric multidimensional scaling (NMDS, Kruskal & Wish 1978). Log-linear analysis determined whether the NMDS differences found were significant within males that sang at different times of the season and in consecutive years. The occurrence of each of the ADSTs in terms of presence or absence among the 11 males was analysed using a histogram. In this analysis, the median number of males that sang syllable types in each of the five syllable type groups was determined and differences between groups were tested using a Kruskal-Wallis one-way ANOVA.

Differences in common and peculiar syllable types among males: The number of syllable types shared by males out of the 30 types that were sampled, was determined by histogram analysis. Common and peculiar syllable types in the complete repertoires of males 1 and 4 were counted in each syllable type group and overall (Appendix 5) and expressed as percentages.

Results

Structural Differences Between Syllable Types Among Males

Qualitative Analysis

Syllables of a particular type were recognised as that type regardless of where they occurred in a male's song or which males sang them, as there was repeatability of 100% (syllable type groups 1-4, 6) and 95% (syllable type group 5) in the thousands of syllable assignments to the wide range of PSTs in the repertoires of the 11 males. Assignment, however, often was difficult and involved extensive comparison of syllables from the large library of PSTs due to complexity and variability of syllable acoustic frequency and temporal structure, especially group 5 syllables (Appendix 6). Most of the PSTs in syllable type groups 1 to 4 were composed of one temporal unit, while all PSTs in syllable type groups 5 and 6 were composed of two or more elements (Appendix 6). Most PSTs covered a broad acoustic frequency range and showed amplitude and frequency modulation, harmonics and repetition of elements (Appendix 6).

Quantitative Analysis

General: The following points were evident from the five syllable type groups and demonstrated in a complimentary way that Sedge Warbler syllables can be designated as types with distinct temporal and acoustic frequency characteristics within and among males: First, principal components used in MANOVA explained a high percentage of the variance (Table 1), and eigen-vector loadings were all significant. Second, MANOVA with sequential Bonferroni adjustments, showed significant differences in 1564 pairwise comparisons of PSTs (95%, N=1646). Third, MDA indicated high mean classification accuracies for syllable type groups 1, 2, 4 and 5, moderately high levels for syllable type group 3 (Fig. 1), and no PSTs with high levels of misclassification and mutual misclassification (>50%). Fourth, DMRTs revealed significant differences between the means of principal component coordinates in all pairwise comparisons of PSTs (Table 1).

PSTs not split: Although eight PSTs received low classification accuracies (<50%, Fig. 1), none were split for the following reasons. Misclassification in MDA was appreciably less than 50% (Max.=37%). Misclassification of any one PST was spread among several PSTs. DMRTs revealed significant differences between the means of principal component coordinates in all pairwise comparisons of poorly classified types and types with which they were misclassified (Table 1), and acoustic frequency and temporal variables loaded significantly on the principal components.

PSTs kept separate although not differing significantly in MANOVA: In syllable type groups 1 to 5, MANOVA found no significant differences in a minority of pairwise comparisons between PSTs (Table 1). These PSTs were kept separate for the following reasons. First, DMRTs found significant differences between the PSTs (Table 1), and there were significant eigen-vector loadings for the variables on the principal components. Second, mutual misclassification between the PSTs in MDA was low generally (<20%) with maximum values being substantially less than 50% (max.=25%). Third, MDA showed strong support for most of the types (≥80%) and moderately strong support for others (≥50%), except for eight types that were classified poorly (<50%, Fig. 1).

PSTs defined as ADSTs: All 98 PSTs that were included in the uni- and multivariate analyses of 11 males were established as ADSTs (Table 1). These ADSTs included the 30 PSTs that were sampled among the 11 males and the 67 and 64 syllable types of males 1 and 4 respectively.

Structural Constancy Within Syllable Types

Generally, syllable types were classified correctly in MDA regardless of which males sang them and whether they occurred in samples taken at different times of the season or in consecutive years (Fig. 1).

Between males in the same season: There was structural constancy in a large proportion of syllable types and in the majority of males for the following reasons. MANOVA showed no significant differences between samples of different males among 48% of the syllable types

Table 1. Summary PCA, MANOVA and DMRT results for five syllable type groups from 11 males in a population of Sedge Warblers, showing that the number of PSTs determined by visual inspection of syllables was verified by quantitative and uni- and multivariate analyses. The following are indicated for each syllable type group: Number of putative syllable types (No. PSTs) and number of syllables included in the quantitative analysis. The number of variables (No. vars.) included in each PCA. Principal components (PC) and their cumulative variance (cum. var.) used in each MANOVA. Pairwise comparisons conducted in MANOVA between putative syllable types (PST Comparisons) showing, a) number of pairwise comparisons that were not significantly different after sequential Bonferroni adjustment (No. NS, $P \geq 0.05$), and b) total number of pairwise comparisons conducted. Duncan's multiple range test results (DMRTs) showing numbers of principal components (No. PCs) whose means differed significantly ($P<0.05$) between pairwise comparisons that were not significantly different in MANOVA. Number of analytically determined syllable types (No. ADSTs).

Syllable Type Group	No. Males	No. PSTs	No. syllables	No. vars.	PC (cum. var.)	PST Comparisons (MANOVA)		DMRTs:	
						No. NS	Total no.	No. PCs	No. ADSTs
1	11	10	530	5	1-3 (86%)	4	45	2	10
2	11	7	481	5	1-4 (98%)	3	21	3	7
3	10	11	634	5	1-4 (89%)	7	66	3	11
4	11	17	516	5	1-2 (67%)	10	136	1	17
5	11	53	1943	7	1-3 (72%)	58	1378	2	53

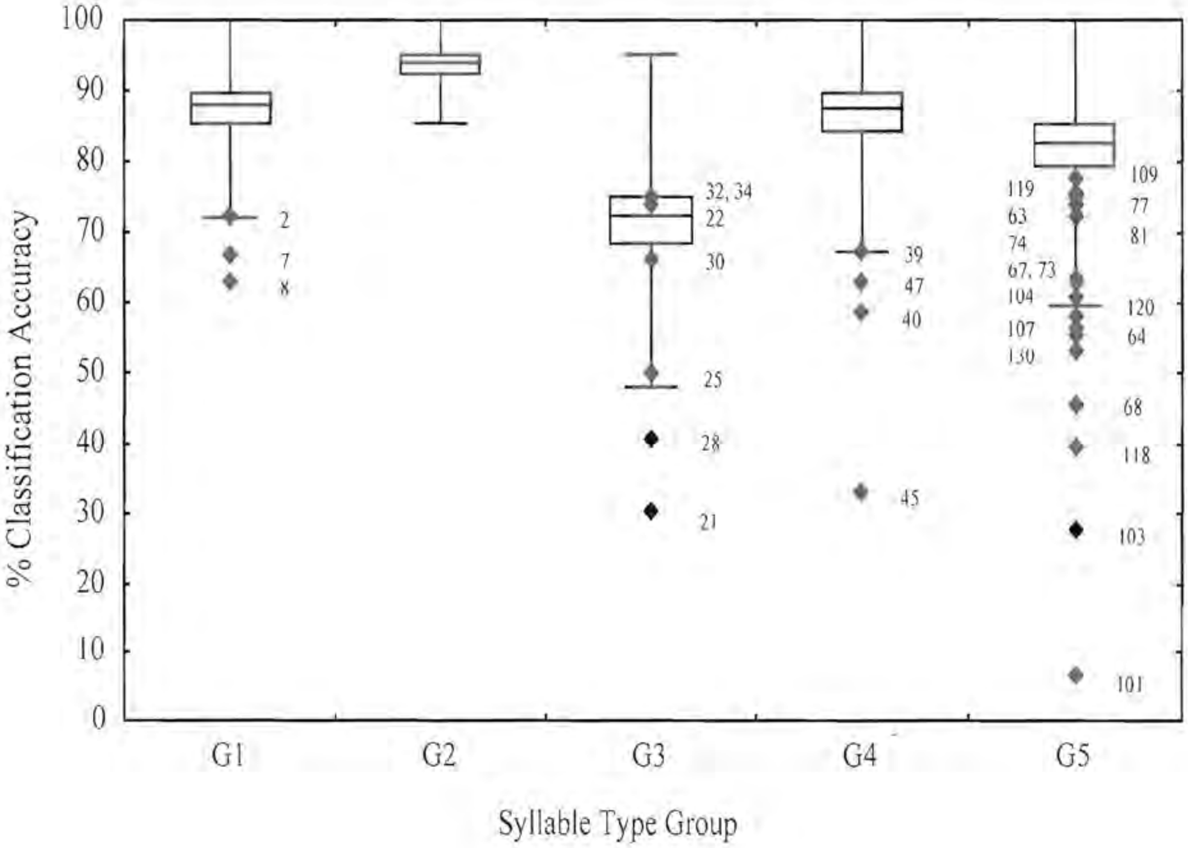


Figure 1. Descriptive statistics of classification accuracies of putative syllable types (PSTs) in syllable type groups 1 to 5 (G1-5), showing high mean classification accuracy for each syllable type group. Classification accuracies were obtained by canonical multiple discriminant function analysis (MDA) of acoustic frequency and temporal parameters (Fig. 2c) of the syllables of males that sang in the same season, at different times of the season and in different years. Boxes show mean accuracy (±1SE), while whiskers indicate ±1SD. PSTs with classification accuracies of <80% are indicated. The numbers of males, PSTs and syllables involved in the MDA of each syllable type group are reported in Table 1.

analysed (Table 2, Fig. 2a). A mean of only 38% of male pairs showed significant differences, which occurred in syllable types in each of the syllable type groups (Table 2).

Within males at different times of the season: There was structural constancy in a large proportion of syllable types as 70% of the syllable types from all five syllable type groups showed no significant differences in particular males when sung at different times of the season (Table 2). Syllable type 107 provides an example of significant differences found in the remaining 30% of the syllable types (Table 2, Fig. 2b). Furthermore, fewer significant differences were found within syllable types in males that sang at different times of the season than between males that sang in the same season (Fisher exact test: N=26, P=0.001).

Within males between years: There was structural constancy in a large proportion of syllable types as 75% of the syllable types from all five syllable type groups showed no significant differences between samples taken in consecutive years in particular males (Table 2). Syllable types 30 and 67 provide examples of significant differences found in the remaining 25% of the syllable types (Table 2, Fig. 2c). The same number of non-significant (N=14) and significant differences (N=4) were found within syllable types in males that sang in different years as in males that sang at different times of the season, with non-significant differences being predominant (78%).

Differences in Syllable Type Occurrence

Among males: There were considerable differences in the occurrence of the 30 syllable types sung by different males (Fig. 3). Males that sang most of the syllable types frequently and that had few rare and absent types appear in quadrant B (Fig. 3). The greater the distance that separates other males from males in quadrant B, the fewer the syllable types that were sung frequently and the greater the number of rare and absent syllable types.

There also were considerable differences in the syllable types sung by males overall and in each syllable type group (Fig. 4). Most of the types were sung by the majority of males whose median numbers did not differ significantly between syllable type groups. Uncommon syllable types were sung by at least four males, e.g., syllable type 104.

Within males between samples taken at different times of the same season: Song samples of males 1, 4 and 15 taken later in the season appear in quadrant B, which contained samples with the frequent occurrence of most of the 30 syllable types and few rare and absent types (Fig. 3).

Table 2. Structural differences within syllable types in syllable type groups 1 to 5 between 11 males that sang in the same season (Male), and in particular males that sang at different times of the season (Season) and in different years (Year). MANOVAs were conducted on principal component coordinates of syllable types that were generated by PCA (See Table 1 for summary PCA results). For differences in a particular syllable type between samples of different males, the number of pairwise comparisons conducted between males and differing significantly are shown. For differences in a particular syllable type between samples taken at different times of the season and in consecutive years in particular males, the number of males tested and significant differences found are reported. MANOVA results include sequential Bonferroni adjustments.

		Male		Season		Year	
Syllable Type Group	Syllable type	Total no. male pairs	No. male pairs MANOVA: $P<0.05$	No. males	MANOVA $P<0.05$	No. males	MANOVA $P<0.05$
1	1	1	0	-	-	-	-
1	2	45	14	2	1	2	0
1	3	1	0	-	-	-	-
1	4	5	0	1	1	-	-
1	5	1	0	-	-	-	-
1	7	1	0	-	-	-	-
1	8	28	0	2	0	2	0
1	9	3	0	1	0	-	-
2	12	15	6	1	0	-	-
2	14	21	7	2	0	1	0
2	16	45	16	2	1	1	0
2	19	10	0	-	-	1	0
3	21	10	0	2	0	1	0
3	22	1	0	-	-	-	-
3	24	28	4	3	0	2	1
3	25	1	1	-	-	-	-
3	27	1	0	-	-	-	-
3	28	21	14	2	0	-	-
3	30	36	17	3	1	2	1
3	32	1	0	-	-	-	-
4	38	10	2	1	1	-	-
4	39	15	0	1	1	-	-
4	40	1	0	-	-	-	-
4	43	1	0	-	-	-	-
4	45	6	2	-	-	-	-
4	46	21	9	1	0	-	-
4	47	15	7	3	0	1	0
4	48	-	-	1	0	-	-
4	50	1	0	-	-	-	-
5	63	21	1	-	-	2	1
5	64	10	3	2	0	2	0
5	67	15	6	2	0	2	1
5	68	21	11	1	0	2	0
5	73	15	1	2	0	1	0

5	74	1	0	-	-	-	-
5	81	10	0	1	0	1	0
5	82	1	0	-	-	-	-
5	87	1	0	-	-	-	-
5	101	28	10	2	0	2	2
5	103	28	12	2	0	2	0
5	104	3	1	-	-	-	-
5	107	15	3	2	1	1	0
5	108	1	1	-	-	-	-
5	118	3	2	2	0	1	0
5	119	3	1	1	0	1	0
5	120	21	3	2	1	-	-
5	123	1	0	-	-	-	-
5	170	1	0	-	-	-	-
5	172	1	0	-	-	-	-

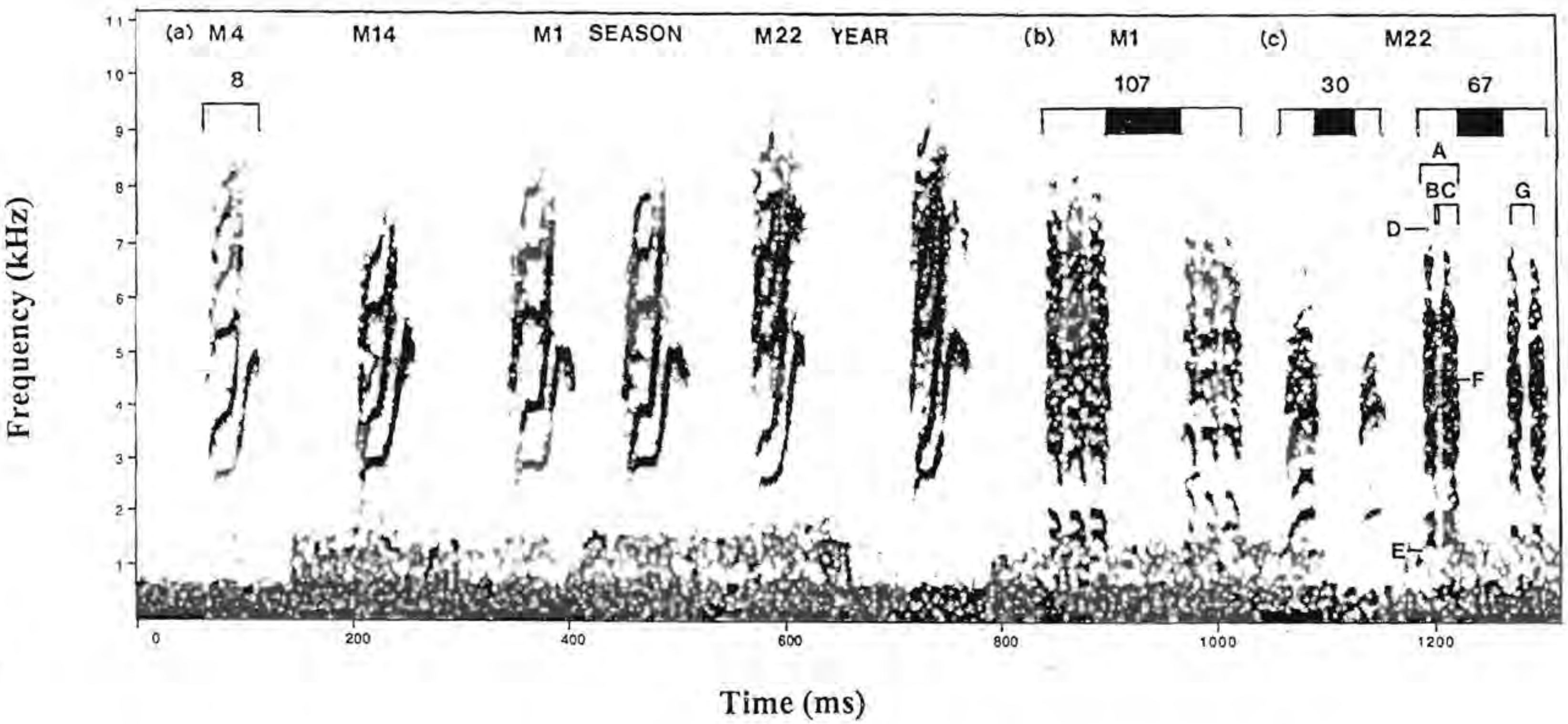


Figure 2. Spectrogram showing examples of: (a) Structural constancy in syllable type 8 among song samples of males 4, 14, 1 and 22; in male 1's song samples taken at different times of the season and in male 22's song samples taken in consecutive years. (b) Structural differences in syllable type 107 in male 1's song samples taken at different times of the season. (c) Structural differences in syllable types 30 and 67 in male 22's song samples taken in consecutive years. Structural constancy was based on an absence of significant differences obtained by MANOVA of principal component coordinates generated by PCA. Structural differences were based on the presence of significant differences obtained by MANOVA of principal component coordinates generated by PCA. These multivariate analyses were based on seven temporal and acoustic frequency measurements, examples of which are shown (c): Syllable duration (A), inter-element duration (B), element duration (C), maximum syllable frequency (D), minimum syllable frequency (E), peak syllable frequency (F) and the number of elements comprising a syllable (G). Peak syllable frequency, which is the frequency at which the highest amplitude in the syllable occurs, is not easily shown.

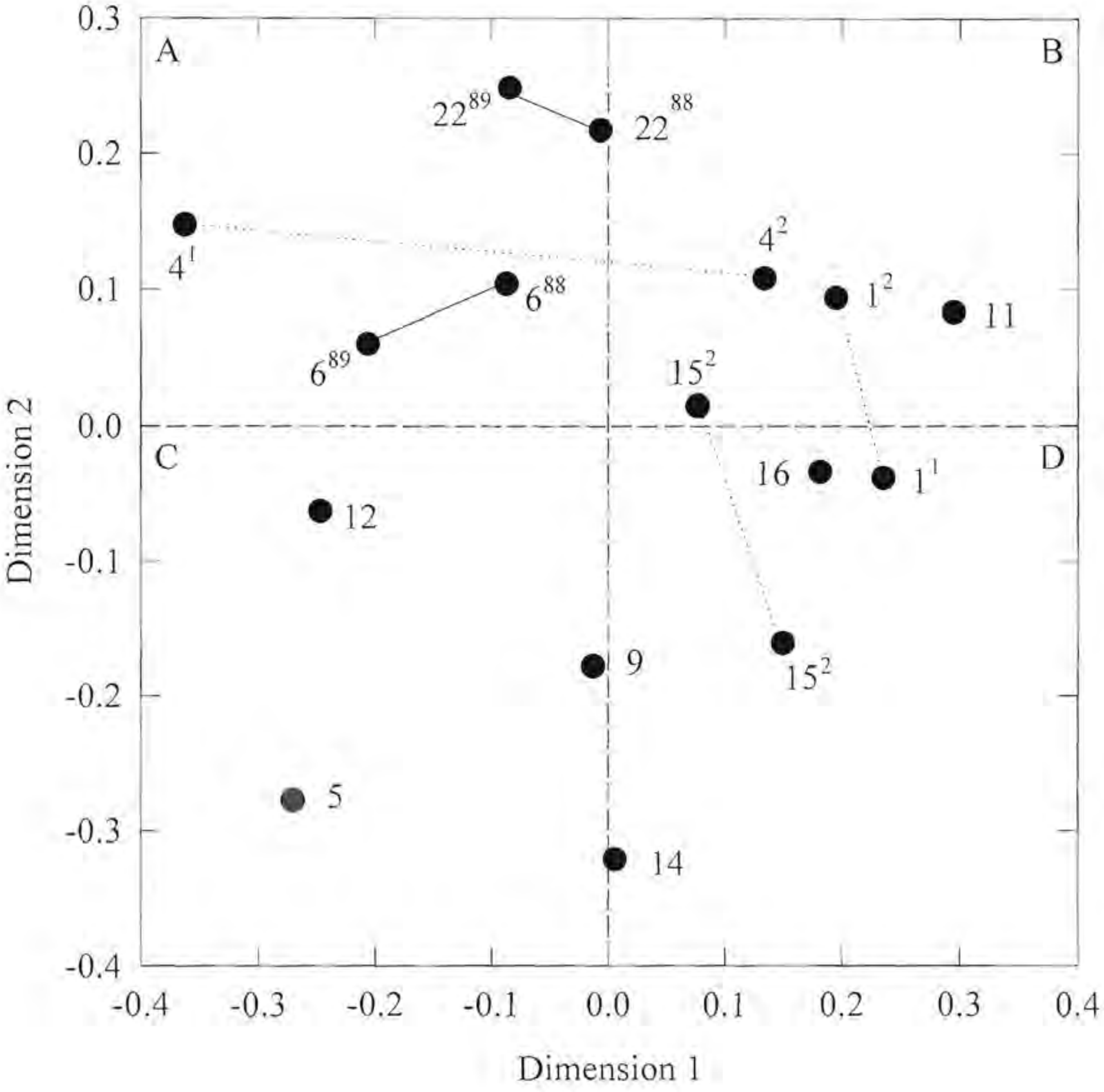


Figure 3. Ordination using non-metric multidimensional scaling showing differences in the levels of abundance of 30 syllable types among 11 males in a population of Sedge Warblers. Dotted lines show differences in samples of males 1, 4 and 15 between the beginning (1) and end (2) of each male's advertising period in 1990's breeding season. Solid lines show differences in samples of males 6 and 22 between the years of 1988 (88) and 1989 (89). Quadrant B indicates song samples with the frequent occurrence of most of the 30 syllable types and few rare and absent types. The higher the incidence of rare and absent syllable types, the greater the distance between quadrant B data points and other data points in quadrants A, C and D.

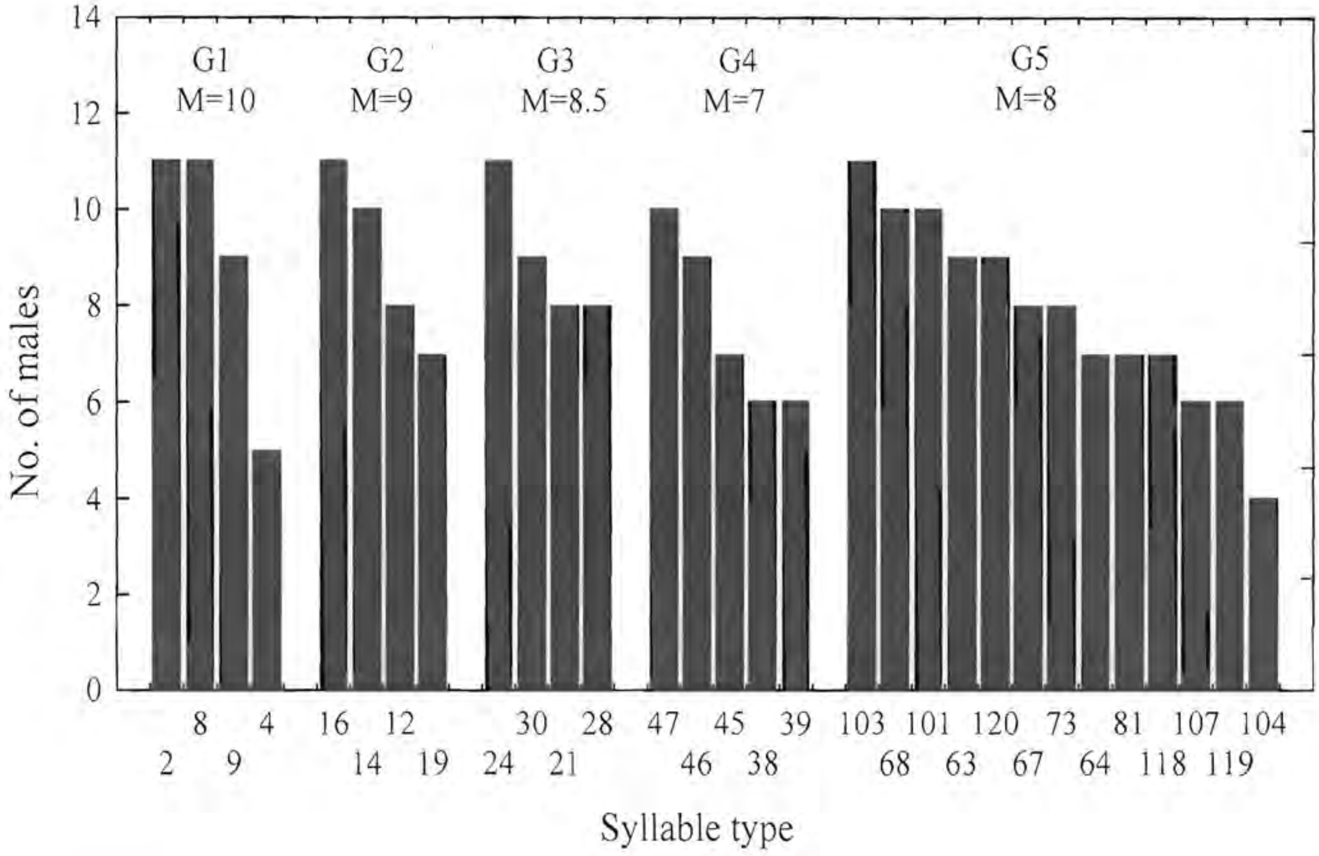


Figure 4. Differences in the composition of syllable type repertoires among 11 males in a population of Sedge Warblers with respect to 30 syllable types that were sampled. Syllable types from syllable type groups 1 to 5 (G1-G5) are shown. The median (M) number of males that sang the syllable types in each syllable type group also is indicated. The median number of males did not differ significantly among syllable type groups (Kruskal-Wallis test: H_4=3.14, NS).

Song samples of these males taken earlier in the season appear in quadrants A and D, which contained samples with a greater rarity and absence of syllable types than quadrant A (Fig. 3). Differences in occurrence of the 30 syllable types in terms of absence, rarity and commonness were significant with respect to samples taken at different times of the season for males 1, 4 and 15 (Max. likelihood Chi-square test, male 1: χ^2_{28} =74.37, P<0.00001; male 4: χ^2_{28} =65.22, P=0.00008; male 15: χ^2_{28}=52.57, P=0.0033).

Within males between samples taken in consecutive years: There were differences in the occurrence of the 30 syllable types sung by males 6 and 22 between samples taken in consecutive years (Fig. 3). More rare types were sung and fewer types were sung frequently by both males in samples taken in 1989 compared to 1988. Male 6 also had more types with zero occurrence in 1989's sample compared to 1988's sample. These differences in occurrence between samples taken in consecutive years were significant for male 22 (Max. likelihood Chi-square test: χ^2_{28}=60.02, P=0.0004), but not for male 6 (Max. likelihood Chi-square test: χ^2_{28}=40.64, NS).

Differences in Common and Peculiar Syllable Types Among Males

In the analysis of 11 males, five (17%) and 14 (47%) of the 30 syllable types were shared by 11 and nine males respectively, with all syllable type groups being represented, while no syllable types were peculiar to individual males (Fig. 4). Approximately half of male 1's and 4's repertoires were composed of shared syllable types, the other half being composed of peculiar types (Table 3). All six syllable type groups were represented, although they contributed differently to the proportion of common and peculiar types in both these males (Table 3).

Discussion

Hypothesis 1 - At the population level, syllable repertoires of male Sedge Warblers are composed of syllable types, which differ qualitatively and quantitatively from one another in temporal and acoustic frequency characteristics: There was strong support for this hypothesis in the qualitative and quantitative analysis. In the qualitative analysis, repeatability of syllable assignments to the wide range of types in the repertoires of 11 males was high, indicating that syllables of a particular type were recognised as that type regardless of where they occurred in a male's song or which males sang them, despite variability and complexity of syllable acoustic frequency and

Table 3. The high and similar total proportions of syllable types shared by and peculiar to males 1 and 4. Common and peculiar syllable types also are shown according to syllable type group. This analysis is based on the complete syllable type repertoire of each male.

Syllable type group	Male	No. of syllable types	% peculiar types	% common types
1	1	9	22	78
	4	9	22	78
2	1	7	43	57
	4	7	43	57
3	1	10	10	90
	4	12	25	75
4	1	12	33	67
	4	16	50	50
5	1	63	57	43
	4	67	60	40
6	1	8	62	38
	4	3	0	100
Total	1	109	47	53
	4	114	49	51

temporal structure. In the quantitative analysis, MANOVA showed significant differences between most types, and DMRTs found significant differences between all types in each syllable type group (Table 1). MDA revealed high mean classification accuracies for types in groups 1, 2, 4 and 5 and moderately strong support for types in group 3, while only a small number of types received weak support (Fig. 1). Misclassification and mutual misclassification of types occurred at a low level generally. These complementary results provide evidence that Sedge Warbler syllables can be designated as types with distinct temporal and acoustic frequency characteristics within and among males as was done with Black-capped Chickadee syllables (Nowicki & Nelson 1990) and Swamp Sparrow notes (Marler & Pickert 1984; Clark, Marler & Beeman 1987). More importantly these results are consistent with syllable types being fundamental units of Sedge Warbler vocal repertoires (Catchpole 1976, 2000) and provide a quantitative basis for categorical perception in this species (Searcy, Nowicki & Peters 1999). Structural distinctness of syllable types increases the efficacy of signal transmission, detection, reception (Wiley & Richards 1982) and perception (Horn & Falls 1996), and provides the basis for effective communication in the Sedge Warbler (Johnstone 1997).

The differences within and similarity between some syllable types shown in the multivariate analyses, notably in groups 3 (Fig. 1) and 5 (Table 1), are not unexpected, especially in a population study of a species with a very large repertoire. But what are the implications of syllable type similarity in Sedge Warbler communication? If receivers are using the temporal and acoustic frequency features that were used in my study to discriminate between and identify syllable types, then the smaller the multivariate range of variation among types, the more difficult discrimination and identification will be (Horn & Falls 1996). Significant differences however, between the means of similar syllable types shown by DMRTs in my study (Table 1) still may indicate sufficient structural distinctness between these types that would facilitate categorical perception (Spector 1994; Horn & Falls 1996). In the Song Sparrow, which has discrete songs, greater numbers of song types have been associated with increasing similarity of song structure (Podos et al. 1992). This however, has not compromised categorical perception of song types that has been shown recently in this species (Searcy et al. 1999). Furthermore, there is evidence in songbirds of categorical perception of signals that vary continuously (Nelson & Marler 1989) and recognition of subtle differences in the acoustic properties of songs (Falls, Dickinson & Krebs 1990), both within and among song types (Stoddard et al. 1988).

Hypothesis 2 - Structural constancy of syllable types among males and within males over time: MDA results supported this hypothesis as syllable types received high mean classification accuracies in groups 1, 2, 4 and 5 (Fig. 1), moderately strong support in group 3 (Fig. 1), and low levels of misclassification and mutual misclassification generally. Thus, syllable types were classified correctly regardless of which males sang them and whether they occurred in samples taken at different times of the season or in consecutive years. The first prediction that syllables of a particular type are structurally constant among males was supported largely by MANOVA, as no significant differences were found between males among 48% of the syllable types that were analysed, and a mean of only 38% of male pairs showed significant differences in the remaining 52% of the syllable types (Table 2; Fig. 2a). The second prediction that syllables of a particular type are structurally constant within males when sung at different times of the breeding season and in different years also received support from MANOVA (Table 2). First, 70% and 75% of the syllable types respectively revealed no significant differences in particular males. Second, significantly fewer significant differences occurred within syllable types in males at different times of the season than between males in the same season. Third, the same numbers of non-significant (N=14) and significant differences (N=4) were found within syllable types in males in consecutive years as in males at different times of the season, with the numbers of non-significant differences being predominant (78%). These results provide evidence for syllable type constancy in a large proportion of syllable types in a breeding population of Sedge Warblers that forms a basis for reliable communication in this species. Signals involved in territorial proclamation, mate attraction and rival male repulsion need stability of components for effective functioning (Paterson 1985, 1993).

The constancy shown in my study is striking in two ways. First, in the context of a large syllable repertoire size in the Sedge Warbler as large repertoires are likely to make stereotypy of learned syllables more difficult (Kroodsma 1974). Second, compared to variability in other aspects of Sedge Warbler display behaviour involving syllables, e.g., the variability in syllable type occurrence shown in my study (Figs. 3, 4) and by Catchpole (1976). Some syllable types did show structural differences within and between males (Table 2; Fig. 2b, c). It remains to be established whether these differences affect the perception of the particular syllable types by receiving Sedge Warblers. Both constancy and variability of note types were shown in the Swamp Sparrow (Clark et al. 1987).

Hypothesis 3 - Differences in syllable type occurrence among males and within males over time: This hypothesis was supported by NMDS (Fig. 3), histogram analysis (Fig. 4) and log-linear analysis. Catchpole (1976) also recorded considerable differences in the numbers of syllable types within and among three Sedge Warblers. It remains to be established whether the differences in syllable type occurrence among and within males have any biological significance, e.g., result from song development, or functional significance, e.g., in female choice (Andersson 1994).

Some syllable types were rare and some were absent from males in my study (Fig. 4). Marler and Pickert (1984) also found rare note types in a population of Swamp Sparrows. The significance of rare and absent syllable types is not known in the Sedge Warbler or more generally, but may reduce a male's effective and total repertoire size respectively which may affect female choice (Andersson 1994; Macdougall-Shackleton 1997).

Syllable repertoire composition changed significantly in three males in song samples taken later in the season compared to those taken earlier in the season. More syllable types were sung frequently and fewer rare and absent types formed part of each male's repertoire later than earlier in the season (Fig. 3). If syllable repertoire size (Catchpole 1980, 2000; Buchanan & Catchpole 1997) and composition are important in female choice, this finding may explain when an individual male Sedge Warbler pairs in the breeding season. There also may be implications for determining syllable repertoire size in individual males, as a song sample taken earlier in the season is likely to yield a smaller repertoire size than one taken later in the season for a particular male. Buchanan and Catchpole (1997) reported that syllable repertoire size remained constant at different times of the breeding season, but provided no data to support this. My finding may not have any functional significance, e.g., in female choice, but may be the result of song development that may occur each season after the males return from migration (Beecher, Campbell & Stoddard 1994).

Syllable repertoire composition changed significantly in samples taken in consecutive years in male 22, but not in male 6 (Fig. 3). The significant occurrence of more rare types and fewer types that were sung frequently in 1989 compared to a year earlier may just be peculiar to male 22, but more than two males need to be analysed to determine whether a trend is present in males.

Hypothesis 4 - A proportion of syllable types is shared in a population, while other syllable types are peculiar to individual males: This hypothesis was partially supported, as approximately 50% of the syllable types were shared between the complete repertoires of two males (Table 3), while 17% of the 30 syllable types that were sampled among 11 males were shared, with a high proportion (47%) being shared by nine of the 11 males (Fig. 4). Although approximately 50% of the syllable types of the complete repertoires of the two males were peculiar to each of these males (Table 3), none of the 30 syllable types that were sampled were peculiar to any of the 11 males (Fig. 4). Catchpole (1976) also reported a high proportion of shared syllable types among three Sedge Warblers from a different study population. The sharing of syllable types together with structural type distinctness (Table 1, Fig. 1) and constancy (Table 2) in a breeding population of Sedge Warblers provide a possible basis for shared types to form part of the specific mate recognition system (SMRS) of this species (Paterson 1985, 1993). The SMRS is composed of adaptations that are involved in signalling between potential mates, forming a co-adapted signal-response reaction chain that allows the recognition and bringing together of conspecific male and female mating partners. Shared, stable signals throughout the range of a species are important characteristics of its SMRS, although signal variability will be determined by receiver specificity (Paterson 1993). Further work will be needed to establish whether the syllable types, which were common to all 11 males, were sung frequently on a regular basis, and whether more than 17% of syllable types in the complete repertoires of a population of Sedge Warblers are shared and sung frequently. Affirmative answers to these questions will make the operation of a SMRS based on shared syllable types more likely. It also remains to be established whether other acoustic features of Sedge Warbler song form part of its SMRS, such as shared elements (Marler & Pickert 1984; Clark et al. 1987), syllable structure (Searcy & Marler 1981) or syllable acoustic frequency (Nelson 1989).

Further work also will be necessary to establish whether a proportion of syllable types in the complete repertoires of a population of Sedge Warblers are peculiar to individual males. The presence of peculiar syllable types together with syllable type constancy (Table 2) provide a basis for individual recognition in the Sedge Warbler, as males possibly can be distinguished from one another by these types by conspecific females and males (Stoddard 1996; O'Loghlen & Beecher 1999). Syllable types that are peculiar to individual males also may be important in attracting a mate. Repertoire size may be less important in female choice, since females might be stimulated by the number of peculiar types in a male's repertoire. A male with a small repertoire, but high proportion of peculiar syllable types may be preferred to a male with a large repertoire of largely shared types. The constancy of a large proportion of syllable types within and among males makes this female choice scenario possible (Table 2).

Conclusion: The present study has provided quantitative and qualitative evidence for structural distinctness of syllable types in a population of Sedge Warblers. This evidence is consistent with syllable types functioning as fundamental units of Sedge Warbler vocal repertoires and provides a quantitative basis for categorical perception and effective communication in this species. Structural constancy in a large proportion of syllable types within and between breeding seasons, together with syllable type distinctness and the sharing of an appreciable proportion of syllable types in the population, provide a basis for the shared, constant types to function as part of the Sedge Warbler's specific mate recognition system. Differences in syllable type occurrence and repertoire composition found within males in the same breeding season may explain when they pair and may have implications for determining syllable repertoire size in individual males. Differences in syllable type occurrence and repertoire composition found between males in the same season and within males in successive seasons provide scope for further work to gain a better idea about the biological significance of these differences.

5. Synthesis

Achievement of Aims and Objectives

Why was more structural definition of Sedge Warbler song needed?

It was evident from Chapter 2 that although previous studies had described elements, syllables (Catchpole 1976; Brackenbury 1978) and songs (Catchpole 1976) as basic units of temporal organisation in the Sedge Warbler, these units needed to be defined more quantitatively for the following reasons. First, prior to my study, they had not been defined in a rigorous, quantitative way for several males. Second, syllable rate had been suggested as a possible cue in female choice (Catchpole et al. 1984), and had been used in a test of the phenotype-linked fertility hypothesis (Birkhead et al. 1997). Third, syllables were crucial to the description, definition and use of syllable types as important vocal units in the Sedge Warbler communication system as discussed in Chapters 3 and 4. Fourth, songs had been used extensively to derive estimates of syllable repertoire size that in turn had been used to test Darwin's theory of sexual selection by female choice (Catchpole 1980; Catchpole et al. 1984; Székely et al. 1996; Buchanan & Catchpole 1997). Fifth, song dependent song complexity and song length had been used to test female choice (Buchanan & Catchpole 1997), the phenotype-linked fertility hypothesis and the directional asymmetry hypothesis in the Sedge Warbler (Birkhead et al. 1997).

It was clear from Chapter 3 that Sedge Warbler syllable types needed to be defined as objectively as possible for the following reasons. First, this had not been achieved previously, not least in terms of qualitative or quantitative criteria for type assignment. Second, the danger of too much subjectivity influencing the visual determination of syllable and song types had been emphasised in recent studies (Podos et al. 1992; Feßl & Hoi 2000; Tchernichovski et al. 2000). The variable and complex acoustic frequency and temporal structure of Sedge Warbler syllable types increased this danger. Third, estimates of syllable type-dependent syllable repertoire size range had varied over time (Catchpole 1976, 1980, 1987, 2000; Székely et al. 1996; Buchanan & Catchpole 1997). Fourth, despite not being described or defined, syllable types had been used quantitatively to test the following ideas in the Sedge Warbler: (i) Darwin's theory of sexual selection by female choice (Catchpole 1980; Catchpole et al. 1984; Buchanan & Catchpole 1997). (ii) Syllable repertoire size as a reliable indicator of parasitism (Buchanan et al. 1999) and paternal effort (Buchanan & Catchpole 2000). (iii) Male response to different syllable repertoire sizes (Catchpole 1989). (iv) The phenotype-linked fertility insurance hypothesis and the directional asymmetry hypothesis (Birkhead et al. 1997).

It was evident from Chapter 4 that structural variation in syllable types and variation in syllable repertoire composition in a population of Sedge Warblers needed to be determined quantitatively for the following reasons. First, to be able to compare syllable types and syllable type composition within males over time and among males in an objective way. Second, the study was novel for the Sedge Warbler and apparently for other songbird species, and was a logical sequel to the quantitative determination of syllable repertoire size and composition in two males as dealt with in Chapter 3. Third, syllable or song types had not been described well quantitatively in other songbird species in terms of structural variation within and between types and males (Podos et al. 1992; Catchpole & Slater 1995; Kroodsma & Miller 1996). Fourth, there had been important ongoing work and debate about intra- versus inter-song type variation (Kroodsma 1982; Wilson, Chapter 1, Table 1) and its functional importance (Stoddard et al. 1988) that had implications for syllable types. Fifth, studies on repertoire composition had been based on the qualitative determination of this variable and had concentrated on comparisons between males, neglecting the situation within males over time.

Generally it was clear that the above aspects of song structure needed to be determined in an accurate, transparent and verifiable way for two important reasons. First, to enable valid, dependent and repeatable empirical studies (Isaac & Marler 1963; Kroodsma & Byers 1991; Hauser 1996). Second, to enable a biologically relevant determination of the vocal units themselves and any derived measures, such as repertoire size (Horn & Falls 1996; Macdougall-Shackleton 1997).

Methods for the analysis of Sedge Warbler song structure: Achievements and limitations.

Qualitative and quantitative methods were developed and applied, and uni- and multivariate analytical methods were used for testing the hypotheses and predictions in an accurate, transparent and verifiable way. In all cases, quantitative analysis, including either or both uni- and multivariate analyses, were used in the final testing of the hypotheses, except in Chapter 3 where rare syllable types were determined qualitatively. These types, however, were defined as true types because they differed in aspects of temporal and acoustic frequency structure that were consistent with the differences found in the quantitative analysis that separated types.

Although the method of summary descriptive measurement was successful for the analysis of Sedge Warbler syllable types in Chapters 3 and 4, it had certain limitations. First, there was a level of subjectivity as it was based on pre-specified measurement categories. Second, these categories were not able to capture the complexity of Sedge Warbler acoustic frequency and temporal structure, notably harmonics that were common. Some syllable types that were found to be similar in the quantitative analysis, differed in complex aspects of acoustic frequency and temporal pattern not captured by the standard set of measurements. Third, the syllable type repertoires of only two males were analysed comprehensively due to the labour intensive and time-consuming nature of the work. Recently, a procedure was described for the automated measurement of acoustic similarity that may overcome these three limitations (Tchernichovski et al. 2000). This procedure also would be useful for assessing rare syllable types that comprised an appreciable proportion of the repertoires of the two males that were analysed in detail.

Implications of findings

The quantitative evidence against songs and for syllables and their component elements as discrete and reliable temporal units in the Sedge Warbler has several important implications. First, songs as they have been described (Catchpole 1976, 2000), cannot be used reliably as quantitative units to derive any measures such as syllable repertoire size. Studies that depend on any such derived measures will have to be reassessed. These studies include testing Darwin's (1871) theory of sexual selection by female choice (Catchpole 1980; Catchpole et al. 1984; Székely et al. 1996; Buchanan & Catchpole 1997), the phenotype-linked fertility hypothesis and the directional asymmetry hypothesis (Birkhead et al. 1997), and the relationship between syllable repertoire size and age (Birkhead et al. 1997), parasitism (Buchanan et al. 1999) and paternal effort (Buchanan & Catchpole 2000). Second, communication in the Sedge Warbler is unlikely to involve songs, but is likely to involve syllables and their component elements. Syllables and elements have been identified quantitatively before as basic vocal units in the Sedge Warbler (Brackenbury 1978) and are well within the limits of auditory discrimination in songbirds (Wilkinson & Howse 1975; Klump & Maier 1989; Klump & Gleich 1991). Neurobiological studies have emphasised syllables as basic vocal units in the Sedge Warbler and other songbird species (DeVoogd et al. 1993; Székely et al. 1996). Third, syllables and their component elements provide several possible channels of communication as basic vocal units in the Sedge Warbler. The significant differences shown between song samples of different males in element, inter-element, syllable and inter-syllable durations and syllable rate, provide a possible basis for individual recognition of potential mates and rival males (Falls 1982; Stoddard 1996), and in the case of syllable rate, a possible basis for female choice (Andersson 1994; Catchpole & Slater 1995), if differences shown reflect differences between males. The idea that different types of syllables may provide a channel of communication became a real possibility once syllables were defined quantitatively as basic vocal units.

Significant quantitative differences and qualitative differences in acoustic frequency and temporal structure that were found between syllable types in individual males provide a basis for such a channel of communication in several possible ways. Syllable type rate differed significantly between song samples of different males. If these differences between song samples reflect differences between males, syllable type rate may be more of an immediate (Parker 1974; Sullivan 1994) and more easily remembered (Kroodsma 1976; Krebs & Kroodsma 1980; Falls 1982) cue for female choice than syllable repertoire size. After all, syllable repertoire size was shown to be appreciably greater than the latest estimates (Catchpole 2000) in this species.

The importance of syllable types as a possible channel of communication increased once it was shown that the syllable repertoires of males in a population of Sedge Warblers were composed of different syllable types, which differed consistently in acoustic frequency and temporal structure regardless of which males used them. This enables syllable types to be fundamental units in Sedge Warbler vocal repertoires and provides a quantitative basis for categorical perception in this species (Searcy et al. 1999). Structural distinctness of syllable types also increases the efficacy of signal transmission, detection, reception (Wiley & Richards 1982) and perception (Horn & Falls 1996), and provides the basis for a communication system in this species. Furthermore, evidence for the structural stability of syllable types within males over time and between males enables reliable communication, with particular types in one male being the same in other males at different times of the breeding season and in successive seasons. Thus, the scope for the function of Sedge Warbler syllable types had increased from mere quantity in terms of syllable type rate and repertoire size, to quality in terms of syllable type composition. Syllable type composition was analysed in terms of types shared and peculiar, present and absent, and common and rare.

A proportion of syllable types was shared among males in the population, and together with structural type stability, provide a possible basis for a specific mate recognition system (SMRS) in the Sedge Warbler (Paterson 1985, 1993). This system would enable female Sedge Warblers to recognise males as appropriate (i.e. conspecific) mating partners and male Sedge Warblers to recognise other conspecific males. The appreciable variation in syllable type composition that was found between males singing in the same season and particularly within males singing at different times of the season and in successive seasons may have implications for female choice. Some syllable types may be more effective in attracting females than others and individual males thus may vary their use of syllable types to increase their chances of attracting a mate. Males used a wider range of syllable types and reduced the number of infrequently sung types later in the season, suggesting that maximum use of the types was then being made to attract a mate. This was not consistent with the assertion that a male's repertoire size remains constant from day to day (Buchanan & Catchpole 1997, 2000; Buchanan et al. 1999). Caution should thus be exercised when making inferences about the relationship between syllable repertoire size and other important variables such as age (Birkhead et al. 1997), parasite load (Buchanan et al. 1999), and paternal effort and the size of the higher vocal centre (HVC, Buchanan & Catchpole 2000).

Syllables and Syllable Types in Sedge Warbler Communication: The Bigger Picture

In the palaearctic migratory Sedge Warbler, it is essential that males that have obtained a breeding territory repel other males and attract a mate. It is important for newly arrived males at the breeding grounds to know which territories are occupied so they too can establish themselves and start advertising for a mate. It is essential that newly arrived female Sedge Warblers are able to detect available males in the population for pairing. As Sedge Warblers are small, cryptically coloured, monomorphic songbirds (Cramp 1992), the striking singing behaviour of males provides a likely way to communicate this necessary information. However, Sedge Warblers would need to be able to recognise Sedge Warbler acoustic features, which would have to be common to all males and stable among and within males over time. Amidst all the variability and complexity of acoustic structure shown in my study, shared syllable types satisfied these recognition criteria. Thus they were identified as possibly fulfilling a recognition function and forming part of the Sedge Warbler SMRS (Paterson 1985, 1993). Several studies on other songbird species have identified and investigated acoustic features, which are used by females for recognising and responding to conspecific males (West et al. 1979, 1981; Searcy & Marler 1981; King & West 1983; Catchpole 1986; Dabelsteen & Pedersen 1988a, b, 1993; Searcy & Brenowitz 1988; Catchpole & Leisler 1989, 1996; Feßl & Hoi 1996; Schmidt et al. 1999). Likewise, several studies have identified and investigated acoustic features that are used by males for recognising and responding to conspecific males (Ficken & Ficken 1973; Shiovits 1975; Brémond 1976; Wunderle 1979; Becker 1982; Brenowits 1982, 1983; Catchpole 1983; Nelson 1988; Catchpole & Leisler 1989, 1996; Feßl & Hoi 1996; Schmidt et al. 1999).

It is in the interests of an unpaired female Sedge Warbler to choose a healthy mate that will provide direct benefits to her in terms of territory quality, paternal care and protection from predators, and benefits to her offspring in terms of resistance to disease and parasites, and because of the high levels of predation (Wilson, unpublished data), the ability to attract a mate as early in the breeding season as possible. There is a well established theoretical basis for female choice and its evolutionary explanation in terms of sexual selection that have considerable support in songbirds (Bateson 1983; Andersson 1994; Catchpole & Slater 1995; Searcy & Yasukawa 1996). Although a female Sedge Warbler possibly can assess territory quality directly (Buchanan & Catchpole 1997), she has no direct measure of the qualities of her prospective mate, and thus she would have to use a reliable indicator or indicators. The use of several indicators may improve the accuracy, reliability and timing of her choice (Parker 1974; Iwasa & Pomiankowski 1994, 1995; Sullivan 1994; Møller & Pomiankowski 1993; Johnstone 1996). One possibility is syllable rate which has been highlighted in my study, but which has not been tested in the Sedge Warbler to date. There is considerable circumstantial evidence suggesting that syllable rate may be involved in female choice. Several studies suggest that song rate provides reliable information about the energetic condition of males (Searcy 1979; East 1982; Davies & Lundberg 1984; Gottlander 1987; Reid 1987; Alatalo et al. 1990; Cuthill & Macdonald 1990). Female preference for males with high song rates has been reported in various songbird species (Payne & Payne 1977; Gottlander 1987; Radesäter, Jakobsson, Andbjer, Bylin & Nyström 1987; Reid & Weatherhead 1990). Song rate also has been found to be important in female stimulation (Wasserman & Cigliano 1991) and a reliable indicator of paternal care (Greig-Smith 1982).

Another possible reliable indicator of male quality is syllable repertoire size, which in the Sedge Warbler was reported to be significantly correlated with pairing date (Catchpole 1980; Buchanan & Catchpole 1997), female copulation solicitation display (Catchpole et al. 1984), male age (Birkhead et al. 1997), paternal care, chick weight and higher vocal centre (HVC) size (Buchanan & Catchpole 2000), and significantly lower in parasitised individuals (Buchanan et al. 1999). Furthermore, there is considerable support for syllable or song repertoire size to be involved in female choice in a wide range of other songbird species (Table 1). Active female choice, which is very difficult to show in the field, has been demonstrated for the first time in songbirds in the Great Reed Warbler, a close relative of the

Table 1. Songbird species in which female choice or preference for male syllable repertoire size or song repertoire size has been reported.

***Acrocephalus* and related species**		
1. *A. schoenobaenus*	Sedge Warbler	Catchpole 1980; Catchpole et al. 1984; Buchanan & Catchpole 1997.
2. *A. arundinaceus*	Great Reed Warbler	Catchpole 1986; Hasselquist et al. 1996.
3. *A. paludicola*	Aquatic Warbler	Catchpole & Leisler 1996.
4. *Phylloscopus trochilus*	Willow Warbler	Järvi 1983; Radesäter & Jakobsson 1988.
Other species		
5. *Mimus polyglottus*	Mockingbird	Howard 1974.
6. *Serinus canarius*	Canary	Kroodsma 1976.
7. *Melospiza melodia*	Song Sparrow	Searcy & Marler 1981; Searcy 1984.
8. *Melospiza georgiana*	Swamp Sparrow	Searcy et al. 1982.
9. *Parus major*	Great Tit	Baker et al. 1986.
10. *Emberiza citrinella*	Yellowhammer	Baker et al. 1987.
11. *Agelaius phoeniceus*	Red-winged Blackbird	Searcy 1988.
12. *Sturnus vulgaris*	European Starling	Eens et al. 1991, 1993.
13. *Ficedula hypoleuca*	Pied Flycatcher	Lampe & Saetre 1995.

Sedge Warbler (Bensch & Hasselquist 1992). A significant correlation also has been reported between repertoire size and offspring survival in the Great Reed Warbler (Hasselquist, Bensch & Von Schantz 1996). My study, however, has shown that syllable repertoire size cannot be measured as described in the above-mentioned Sedge Warbler studies, thus casting doubt on the results described and inferences made about female choice and sexual selection in these studies. My study also found that repertoire size was appreciably greater than the latest estimates for the Sedge Warbler and has raised questions about a female's ability to use a cue of this magnitude. Syllable type rate was suggested as possibly a more realistic cue. Furthermore, my study has drawn attention to the possible importance of repertoire composition in female choice rather than just repertoire size, such as a proportion of peculiar types in a male's repertoire. Finally, I showed that repertoire size may change in individual males during the breeding season, rather than remaining constant as has been reported (Buchanan & Catchpole 1997, 2000; Buchanan et al. 1999). This may have implications for the reliability of repertoire size as a measure of male quality.

The benefits of individual recognition are large to the philopatric Sedge Warbler and the singing behaviour of males again provides a likely way to communicate this information. Time and energy are probably major constraints to this long distance migrant, as territorial proclamation, mate attraction, feeding and rearing of offspring to independence, and restarting the whole process in the likely event of clutch or nestling predation (Wilson, unpublished data) all have to occur in approximately five months before the birds return to Africa. Less time and energy would need to be spent in establishing and defending territorial boundaries between familiar neighbours during the same breeding season and in subsequent seasons, leaving more time for mate attraction. Females could identify their mates from the previous season as well. There is a considerable body of evidence showing that song is used in neighbour-stranger discrimination in a wide range of songbird species (Falls 1982; Stoddard 1996). A study on the migratory Hooded Warbler has demonstrated long-term neighbour-stranger and neighbour-neighbour discrimination ability in successive breeding seasons (Godard 1991). The use of peculiar songs (McGregor & Avery 1986), signature syllables (Weary, Norris & Falls 1990) and acoustic features of syllables (Brooks & Falls 1975; Nelson 1989) in individual discrimination also has been shown. Pairing with the same male in successive seasons has been reported in the migratory Marsh Warbler, a close relative of the Sedge Warbler (Kelsey 1987). My study has shown that song samples of different male Sedge Warblers differ significantly in element, inter-element, syllable and inter-syllable durations and syllable rate. If differences between song samples reflect differences between males then any one or a combination of the duration and rate variables may provide a basis for individual recognition in this species.

Syllables, their component elements, syllable types and the sequential structure of Sedge Warbler song have been determined quantitatively in an accurate, transparent and verifiable way in my study, which also has emphasised their fundamental importance in the Sedge Warbler communication system. Further research will have to be conducted to determine whether these features of song structure function as suggested in the SMRS, female choice and individual recognition in the Sedge Warbler.