The Underwater Acoustic Repertoire of the Long-necked, Freshwater Turtle *Chelodina oblonga*

Chelodina oblonga Photo courtesy of Gerald Kuchling, 2005

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DECLARATION

Except where specifically acknowledged, this project is my own account of the research that I have undertaken and has not been submitted for any other awards from other institutions.

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With all the modern technology
hearing-aids of the 21st Century
listening into the quiet.

And lo – this is no longer a world of amber and silence.
This is a world full of voices and songs.
Ancient melodies modulating to a 200 million year old beat.

And now that we have heard the songs and the voices in the amber
what would they tell us ?
and are we going to listen….

.
J.C.G 2004

ABSTRACT

The major question addressed by this project was to determine if the long-necked, freshwater turtle *Chelodina oblonga,* vocalise underwater and whether their vocal activity could be related to behavioural or ecological aspects of their lives. These turtles often live in wetlands where visibility is restricted due to habitat complexity or light limitation caused by factors such as tannin-staining, or turbidity. For many aquatic animals, sound is a useful means of communication over distances beyond their visual acuity. This thesis gives the first detailed account of the underwater vocal repertoire of *C. oblonga*.

In total, over 230 days were spent in the field and more than 500 hours of tape recordings were made for this research. Initially, a number of recordings took place in three wetlands known to support turtle populations: Blue Gum Lake; Glen Brook Dam; and Lake Leschenaultia in Perth, Western Australia; in order to determine the nature of the freshwater sound field and place turtle vocalisations into the context in which they were vocalising. The wetlands differed in terms of degree of enrichment, substrate material, water depth and habitat complexity. Recordings were made over a four-week period in the last month of summer and the first week of autumn (Feb-Mar 2003). Invertebrate sweeps were also taken over a two-week period at each recording site to determine if invertebrate distributions were related to patterns of sonic activity. To determine the influence of wind on ambient noise; recordings were undertaken on winter mornings (June-August, 2003) at Blue Gum Lake and Glen Brook Dam at locations north, south, west and east for four different wind speeds – Beaufort Wind Scale (BWS) 0,1,2 & 3.

There were seven distinctive calls recognised in the recordings. The frequency bandwidth most utilised by organisms was between 3 kHz up to around 14 kHz, with the exception of

the ‘bird-like song’; which extended from 500 Hz up to around 10 kHz. Blue Gum Lake contained a more diverse and abundant assemblage of invertebrates than Lake Leschenaultia and Glen Brook Dam. Correspondingly, a greater diversity of calls was recorded at Blue Gum Lake, as well as the presence of chorus activity, which was not heard at the two less-enriched sites. The periods of greatest diversity and abundance of macroinvertebrates was synonymous with the increased sonic activity at dusk and midnight with noise levels greatest at dusk in particular, and to a lesser extent at midnight. There was no difference in ambient noise at Blue Gum Lake or Glen Brook Dam at wind speeds of Beaufort Wind Scale 0, 1 and 2.

Turtles from three populations were recorded in artificial environments: consisting of round, plastic, above-ground ponds (1.8m dia. x 0.65m depth), which were set up to re-create small wetlands. Recordings occurred from September to October, 2003 and from February to December, 2004 as well as January, 2005. Seven hatchling and five juvenile turtles (CL <10cm) were also recorded in order to ascertain whether very young turtles vocalised. Hatchlings were recorded in a glass aquarium (35.5cm length x 20cm width x 22.0cm depth) and juveniles were placed into a below-ground outdoor pond (1m length x 0.5m width x 0.4m depth). Recordings occurred from as early as 4.30am (dawn recordings) to as late as 1.30am (evening recordings).

The recordings revealed that turtles utilise an underwater acoustic communication system (calling at the water’s surface was also noted but these were not recorded or a part of this research) involving a repertoire of both complex and percussive sounds with short, medium and potentially long-range propagation characteristics. Complex structures included harmonically related elements (richly or sparsely) and different rates of frequency modulation. Frequency use extended beyond the in-air auditory sensitivity known for a single species of turtle studied from the family Chelidae; with calls ranging from around 100 Hz in some of the percussive displays, to as high as 3.5 kHz in some complex calls, with 'clicks' extending beyond the 20 kHz upper limit of the recording system. However, most of *C. oblonga's* vocalisations had dominant frequencies below 1 kHz. Turtles were intermittent callers with an extensive vocal repertoire of seventeen (17) vocal categories - highly suggestive of complex social organisation. Vocalisations included: a) clacks; b) clicks; c) squawks; d) hoots; e) short chirps; f) high short chirps; g) medium chirps; h) long chirps; i) high calls; j) cries or wails; k) cat whines; l) grunts; m) growls; n) blow bursts; o) staccatos; p) a wild howl; and q) drum rolling. Also, two sustained 'pulse-bouts' were recorded during the breeding months, hypothesised to function as acoustic advertisement displays – possibly 'calling songs'. Hatchling turtles were not heard to vocalise within the audible range. Only a single complex vocalisation was heard produced by the juvenile turtles, with a number of percussive calls.

Preliminary playback trials were conducted under free-field conditions and within an artificial environment, which consisted of a below ground rectangular tank (2.4m length x 0.8m width x 0.6m deep). A number of turtle calls recorded in the artificial ponds were selected for playback. A UW 30 speaker was used for broadcast of calls. The free-field playbacks occurred at Mabel Talbot Lake and Blue Gum Lake during the months of April and May, 2005. Playback using 14 seconds of an artificially constructed sequence from the sustained 'pulse-bout' occurred in the artificial channels. This sequence consisted of some of the first phase pulses followed by a section of the 'vibrato'.

The preliminary free-field playback trials indicated that turtles had some interest in the calls being played by responding with an ‘alert posture’. Turtles were shown to remain in the alert posture for a significantly longer time than when no sound was played or when white noise was played. The extensive repertoire and initial responses to the free-field playbacks indicated that sound has some biological importance for *C. oblonga*, although results of playbacks under artificial conditions were inconclusive.

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TABLE OF CONTENTS

Ch 1.0 Introduction

LIST OF FIGURES

LIST OF TABLES

LIST OF MAPS

Chapter 1.0 INTRODUCTION

1.1 General Introduction

The biological and natural contributions to sound underwater in freshwater environments, has been a little explored area. Most of the underwater sound research has been conducted within the ocean and on some of the marine life existing there. This research touched briefly on natural and anthropogenic contributions of sound into freshwater systems, but mainly investigated some of the biological contributions: particularly focusing on a long-necked, freshwater turtle species - the Oblong turtle (*Chelodina oblonga*) (Testudine; Pleurodira; Chelidae) (Lehrer, 1990). This turtle is only found in the south-west of Western Australia but it is more common around metropolitan Perth (Cann, 1998) where it inhabits wetland areas, including seasonal damplands (Burbidge, 1967).

Although *C. oblonga* is considered relatively common, our knowledge of this species is not extensive. Recent research conducted by Giles (2001) and Guyot and Kuchling (1998); and in the past Kuchling (1988; 1989); Porter (1987); Clay (1981); and Burbidge (1967); have filled some important gaps in the ecology and biology of *C. oblonga*. However, there are many aspects to their ecology, which are still unknown. One such aspect is the use of sound by these turtles. Although sound perception in reptiles is considered of less importance than the senses of vision and chemoreception (Bogert, 1960), transmission of chemical signals in water (as well as air) is inefficient as it is dependent on movement of the media (Pough *et al.,* 1998). In a flowing system such as streams or rivers, this would be a rapid and successful means of transmitting information. However in a lentic waterbody, such as those on the Swan Coastal Plain where *C. oblonga* exists; transmission of chemical signals would be slower as movement is dependent on the action of wind and some circulation occurring from the uneven heating of the water body.

It might be expected that visual observation would be a dominant sense in *C. oblonga* given the dorsolateral positioning of the eyes. This would give a wide view of the turtles' surrounds. However, light is significantly attenuated in highly coloured or turbid wetlands and within disturbed and enriched wetland environments; light is also greatly attenuated by the presence of algal blooms (Davis *et al.,* 1993). Scattering (rather than absorption), is the most important factor in restricting visibility as scattering causes light to come from all directions similar to that experienced in fog (Lythgoe, 1988). Many wetlands on the Swan Coastal Plain are tannin-stained, in particular, those wetlands occurring on the Bassendean sand land formations. The colouration is mainly due to the decomposition of plant material from the fringing vegetation such as the Paperbarks e.g. *Melaleuca rhaphiophylla,* into humic and fulvic acids (Davis *et al.,* 1993). The low light levels in these wetlands, combined with the limited range of visibility due to fallen logs and aquatic vegetation, may mean that sound could be an important sense for *C. oblonga* existing within this type of environment. For those species living at depth or in turbid waters, it is suggested that auditory and vibratory stimulus would be an important mode of communication (Scott, 1968). For example in crocodilians; specialised sensory organs, known as 'Dome Pressure Receptors', are located around the crocodilian face detecting surface ripples when they are partially submerged (Soares, 2002). However, sound has the advantage in that it can be transmitted in every direction or can be narrowed to a single direction to transmit information rapidly using a range of frequencies and temporal variations (Hawkins and Myrberg, 1983). Mrosovsky (1972) proposed that auditory (as well as olfactory) sense might be a useful means of communication in

Leatherback turtles (*Dermochelys coriacea*) existing in turbid coastal conditions in the Guianas. Other aquatic animals living in similar types of conditions, for example: the Atlantic bottlenose dolphin (*Tursiops truncatus*), which live in turbid, coastal marine environments; uses an echo-location system for detection of prey and navigation - even though it is likely that they are able to see objects clearly (in Wood and Evans, 1979); Catfish (e.g. the Upside-down Catfish (*Synodontis contractus*)) and many of their relatives; mostly inhabiting freshwater environments often in conditions of limited visibility - have an acute sense of hearing due to their Weberian apparatus (which is a system of specialised vertebrae transmitting sound waves to the inner ear via the swim bladder) and are also very vocal animals, communicating with each other via sound (Burnie, 2001). However, not all animals occupying aquatic environments of limited visibility use sound or vibratory stimuli. The Platypus (*Ornithoryncus anatinus*), which occupies a similar ecological niche as *C. oblonga* in some eastern Australian wetlands*,* is a nocturnal hunter. When the Platypus dives, it closes its eyes ears and nose (Pettigrew, 1999), and uses electric fields both a.c. and d.c., to locate and avoid objects or detect prey items in the water (Scheich *et al.,* 1986). Numerous mechanoreceptors and electroreceptors are located in the bill skin of the Platypus and together, coordinates information on prey items in the water body (Pettigrew, 1999).

Freshwater turtles can be found throughout the more coastal regions of Australia and further inland in parts of eastern Australia with well-established drainage systems (see Cann, 1998). In temperate Western Australia, turtles occupy top-end predatory positions in the food chains of these wetlands, similar to that occupied by both fresh and saltwater crocodiles in tropical freshwater systems. It is already known that crocodilians, including juveniles, produce vocalisations, although these are in-air (Britton, 2001; Britton in Richardson *et al.,* 2002). While it was understood that sound-producing apparatus is not present in all reptile species (Bogert, 1960); this research began on the assumption that a freshwater turtle would have the necessary morphological characteristics for sound production - given that terrestrial chelonians are able to vocalise in-air with the types of sounds being produced indicating that a sound-producing mechanism is present (Kelemen, 1963). However, as these turtles spend the majority of their time submerged, the major research question addressed by this project was to determine if *C. oblonga* vocalised underwater and whether their vocal activity could be related to behavioural or ecological aspects of their lives.

1.2 Bioacoustics

Sound is produced when an object moves or vibrates in a gas, liquid or solid medium (a vacuum cannot conduct sound). Sound consists of particles of the medium oscillating in a to and fro movement. This oscillation is parallel to the direction of sound transmission and results in variations in pressure. Both particle oscillation and pressure variations are determined by the impedance i.e. the density and elasticity, of the medium (Urick, 1983; Hawkins and Myrberg, 1983).

In an aquatic environment sound travels approximately four and a half times faster than it does in air (however, it is about 3% slower in freshwater compared to seawater (Rogers and Cox, 1988)). This increase in sound speed means that the wavelengths are also four and half times longer in the aquatic environment (Urick, 1983; Hawkins and Myrberg, 1983; Tyack, 2001). The absorption of sound is less in this media compared to air and similarly, absorption increases with frequency so that low frequencies are used for long-range communication. However, as attenuation of higher frequencies is less in water, high frequencies can be utilised for communications over longer distances compared to similar frequencies in-air (Forrest, 1994). In air, there are large differences in impedances between: the source of the sound e.g. a Bat; the medium e.g. air; and the target e.g. moth (prey item) or rock (avoidance strategy) (Sales and Pye, 1974). Aquatic organisms, such as fish, have a density similar to that of the surrounding media and are relatively 'transparent' to sound (Popper *et al.,* 1988). Turtles would not be as transparent to sound as fish due to structures such as their skeleton, carapace and plastron, which have a greater density than water (Lenhardt, 1982).

Animals may utilise sound vibrations for communications intraspecifically (socially), interspecifically (between species) and also autocommunication (echo-location) for navigation and prey detection (Sales & Pye, 1974). Sounds that utilise variations in pitch, loudness and patterns are known as complex sounds and are able to communicate much more information than simple tones (Bright, 1984). 'Vocal' animals such as birds or humans blow air from the lungs across membranes which makes them vibrate. Fish can produce a variety of sounds including 'scraping' sounds and grinding their teeth; and they are able to vibrate an internal structure known as the swim bladder (Bright, 1984). Insects use 'stridulation' which is the vibration of various parts of their exoskeleton (Sales & Pye, 1974). They are known as 'instrumentalists' and examples include spiders and crustaceans, which rub or beat appendages such as legs or wings (Bright, 1984). Sound emitting organs are often arranged bilaterally in invertebrates, but usually they are unpaired in vertebrates. Sometimes sound emission is performed by one sex only - for example used by males in mating. If both sexes use sound, there may even be differences in the sounds produced between the sexes (Busnel, 1968).

Animals can produce sounds that are beyond human hearing either at infrasonic frequencies below 20 Hz, or at ultrasonic frequencies above 20,000 Hz (Bright, 1984). Sales and Pye (1974) suggest that use of ultrasound is likely to be more widespread than is realised. For example; nearly all the small mammals hear in the ultrasonic range of frequencies (Bright, 1984). Infrasonic frequencies are utilised by large mammals including marine mammals such as the Cetaceans e.g. the Blue whales (*Balaenoptera musculus*), where this form of sound is able to propagate over long distances and for these mammals - is thought to be important for communication and mating (Hart, 1996).

1.2.1 Chelonian Vocalisations

Research on the vocalisations produced by chelonians was briefly reviewed in Gans and Maderson (1973). However, there appears to be a paucity of research on these animals, which means that the role and importance of sound in the ecological and behavioural aspects of their life history remains largely speculative (Campbell and Evans, 1972); with research tending to focus more on their hearing mechanisms. Most of the research on vocalisations was conducted during the 1960's and 70's on a small number of tortoise species e.g. the Red-footed tortoise (*Geochelone carbonaria*) (Campbell and Evans, 1967; 1972), *Geochelone travancorica* (Auffenberg, 1964; Campbell and Evans, 1972) and the Galapagos tortoise (*Geochelone elephantopus*) (Bogert, 1960; Jackson and Awbrey, 1978). Recent research has been conducted on two subspecies of the Asian forest tortoise *Manouria emys emys* and *Manouria emys phayrei* (McKeown *et al.,* 1990) and on Marginated tortoises (*Testudo marginata*) (Sacchi, *et al.,* 2003) as well as Hermann tortoises (*Testudo hermanni*) (Galeotti *et al.,* 2004). The only marine turtle that has been investigated was the Leatherback turtle (*Dermochelys coriacea)* (Mrosovsky, 1972).

While most of the research has tended to focus on sounds emitted during breeding activities, there are other references on vocalisation. Carr (1952) mentions calls produced by marine turtles when being hurt or killed - which he considered were sounds incidentally produced by the exhalation of breath. He also described 'short rasping calls' or a noise similar to that of a 'mewing kitten' produced by the Gopher tortoise (*Gopherus polyphemus*). Hissing has been described in agonistic encounters by the Wood turtle (*Clemmys insculpta*) (Kaufmann, 1992). Goode (1967) has also observed hissing noises from *C. oblonga* and other Australian chelids such as *Chelodina expansa* and *Elseya latisternum,* which he considered to be involuntary exhalations associated with aggressive behaviour. On handling, mouth-opening or gaping, combined with inflation of the buccal-pharyngeal region is often observed in *C. oblonga.* This is generally considered to be a sign of stress / or threatening behaviour by this animal. Buccal expansion has been described for many reptiles and is often viewed as a defensive display (Pough *et al.,* 1998). The Chameleon for example; will inflate laryngeal air sacs when irritated, portraying a more formidable appearance (Kelemen, 1963) and 'gaping' has also been described in crocodiles as visually threatening behaviour (Britton, 2001). Young *et al.,* (1999, 2001) also found buccal expansion in Puff Adders (*Bitis arietans*) to be part of their defensive behaviour and it was also associated with sound production, in particular hissing.

The single audible vocalisation I heard produced by *C. oblonga* was similar to a bellow or roar. This animal was wandering around the bottom of a canoe trying to find a way out. In a similar situation, Campbell and Evans (1967) recorded *Gopherus agassizii* emitting a 'low piteous cry' as it attempted to climb out of the sink that contained it. Their observations indicated that these sounds were not emitted passively. While the biological significance of the calls made by *G. agassizii* are unknown, given its situation, Campbell and Evans (1967) concluded that the calls were likely to be distress calls. Similarly, the bellow emitted by *C. oblonga* was also considered to be a distress call.

Roaring or bellowing has been observed in the Galapagos tortoise (*Testudo elephantopus*) (Bogert, 1960; Crawford and Awbrey, 1978), which is associated with breeding activities. Sounds produced by *G. carbonaria* during mating are described as 'a series of clucks', similar to the calls produced by chickens (Campbell and Evans, 1967). The vocalisations of *Geochelone travancorica* appear to be unique, as it is the only species reported to call in 'chorus' where a number of individuals call together at regular intervals. Only the young of an aquatic Asian species, *Platysternon megacephalum* have been observed to 'squeal', particularly when disturbed. Loss of this ability to vocalise appears to be related to a certain level of maturation and corresponds with a change in appearance i.e. loss of bright colours, when the carapace length measures around three inches and when jaw development was such that they could bite (Campbell and Evans, 1972).

The Malagasy radiated tortoises (*Geochelone radiata*) vocalise in synchrony with xiphiplastral blows (mounted male thrusting on the lower rear edge of the female's carapace by the xiphiplastron, with vocalisations thought to immobilise the female) (Auffenberg, 1978). Auffenberg (1978, P.282) describes the vocalisation being similar to "… a nail being pulled from a board in two short jerks." While he noted some individual variations between the male vocalisations, he did not consider that auditory cues were important in courtship and breeding. Mrosovsky (1972) noted that Leatherback turtles produced a variety of calls when nesting, but also considered that sound production in turtles was probably of minor functional importance. However, recent investigations by Sacchi *et al.,* (2003) and Galeotti *et al.,* (2004) on *Testudo* *marginata* and *Testudo hermanni* respectively, would suggest that the importance of vocalisations were otherwise in these species.

1.3 Ambient Noise in Wetlands

Ambient noise in the marine environment comprises contributions from the biological components as well as contributions from the prevailing weather conditions and anthropogenic sources (Urick, 1983). Research on ambient noise has mostly focused on the marine environment with very little known on ambient noise in freshwater environments.

1.3.1 The Biological Component

Biological noise is superimposed on the natural background noise; such as that generated from the action of wind, rain or waves (Busnel, 1968). There would be a number of sources of biological sound contribution to ambient noise in a freshwater environment; particularly the sounds produced by invertebrates. Only a few species have been studied in any detail – for example, the Waterboatman (Corixidae) and their underwater stridulation. As these organisms are a common prey item for the Oblong turtle (Woldring, unpublished), they are discussed in some detail in the following section.

The mechanism of stridulation in corixids occurs when the femur of each foreleg (the file or pars stridens), which has specialised structures known as 'pegs', is stroked in a downward movement over a thickened area on the side of the head (known as the 'plectrum') (Theiβ *et al.,* 1983). However, there are some sounding stroke and sounding structure variations between species (see Bailey, 1983; Jansson, 1972); including the rare occurrence of the plectrum and pars stridens occurring on opposite sexes i.e. one structure on the female and the other on the male (Aiken, 1982b). While stridulation is commonly associated with mating behaviour and indeed, appears crucial for successful mating to occur, its function is not limited to mating as Jansson (1973a) also suggests male stridulation acts agonistically to space males.

Within the genus *Cenocorixa,* songs vary between species and sex, consisting of one or more verses in a complex system of communication. In particular, it is the temporal patterning of pulse-trains and the amplitude of the signals, which distinguishes the main differences occurring between signals of various species (Jansson, 1973a). Aiken (1982b) divided the calls from males into spontaneous calls; courtship calls; and mounting and copulatory signals. Females give an answer signal, which Aiken (1982b) has only heard given in response to a courtship call. Jansson (1972) also described 'cleaning sounds', which were produced by both sexes of *Cenocorixa spp.* where the hind legs move over various parts of the body, particularly after being handled.

The air bubble surrounding submerged corixids, plays a primary role in production and receipt of sound (Prager and Streng, 1982). As a sound receiver has not been detected in some organisms e.g. in the Caddis fly larva (*Hydropsyche spp.*) it has been suggested that the minute hairs which cover part of the bodies of macroinvertebrates, may pick up the vibrations of the water molecules (Frings and Frings, 1967; Bright, 1984). For example in some Crustacea, small tufts of hair are present on various parts of the body. Some of these hair tufts have a chemoreceptive function while others detect water displacement (Bright, 1984).

Other biological contributions to ambient noise include freshwater fishes. Although sound production is more common and more complex in marine fish than in freshwater fish, the few freshwater species known to be sound producers includes those from the family Cyprinidae e.g. the Goldfish (*Carassius auratus* Linnaeus) (Moulton, 1963); which is an introduced species and observed in many wetlands on the Swan Coastal Plain. Other noise contributions would include calls made by frogs and also those associated with feeding and breeding activities from the many waterbirds which utilise these wetland habitats.

1.3.2 The Natural Component: Wind and Rain

The prevailing wind and weather conditions can heavily influence ambient noise (Hawkins and Myrberg, 1983), in particular, wind speed can cause noise over a wide frequency range (Urick, 1983) and combined with the seasonality of the biological contribution, there is considerable variation over time. But variability in ambient noise may also occur as sound transmission conditions alter (Urick, 1983), which might occur as water-levels in wetlands drop over summer and rise with winter rain-fall.

Knudsen *et al.,* (1948) considered one of the main causes of noise underwater in shallow marine waters was produced by the breaking of waves and was related to wave height. However, it has since been shown that it is not the actual 'breaking' of the wave that causes the noise but when the waves break, it is the resultant creation and entrainment of bubbles, which are the sources of transient sound and is correlated with wind speed rather than wave height (Medwin, 1995; Cato and McCauley, 2002). Rain also contributes to changes in the ambient noise spectrum. Noise contribution from precipitation is dependent on the rate of fall (Urick, 1983; Förster, 1995) and variations in the drop size - even light rainfall contributes significantly to an increase in underwater sound levels (Förster, 1995). However, noise arises not from rain-drops impacting on the waters surface, but from the oscillations of the resultant transient microbubbles formed from the impact that is the primary source of sound, particularly for frequencies over 500 Hz (Medwin, 1995). Interestingly, it has also been shown experimentally; there is a linear increase in sound energy as water drops fall on water that is of increasing freshness (where temperature of the water drop and receiving water are the same) (in Medwin, 1995).

1.4 Background to Shallow Water Acoustics

Research into shallow water acoustics has traditionally been conducted in the marine environment, with freshwater acoustics receiving much less attention. The wetlands of the Swan Coastal Plain are surface expressions of an unconfined aquifer. They are shallow, permanent or seasonal waterbodies; with depths that rise or fall according to the height of the water table (Chambers and Davis, 1988). In shallow water environments, sound propagates by being alternately reflected from the air/water interface and the bottom boundary i.e. sound travels in a waveguide, and the nature of these boundaries influence transmission characteristics (Ingenito *et al.,* 1978; Urick, 1983; Rogers and Cox, 1988; Forrest *et al*, 1994). The limitations placed on signal propagation in shallow water environments, is considered greater than that experienced by animals communicating terrestrially (Forrest *et al.,* 1993).

Aquatic organisms can alter the propagation or reception characteristics of their signals by altering the depth of the water that they are signaling or listening in – that is moving into shallower or deeper water accordingly (Forrest, 1994; Forrest *et al.,* 1993). In much the same way, terrestrial animals can alter their propagation characteristics whereby the signaler can elevate itself above the ground, allowing low frequency sounds to propagate greater distances with minimal attenuation (Marten and Marler, 1977). Forrest *et al.,* (1993) explains that for an animal to send a signal, it would need to move into a depth of water that allowed the transmission of the frequency of its signal i.e. the signal was above the cut-off frequency.

Shallow water has been shown to act as a steep highpass filter (Forrest *et al.,* 1993; Boatright-Horowitz *et al.,* 1999), with the optimum frequency of propagation in a shallow water environment determined by the depth of the water, the temperature gradients and the nature of the bottom sediments (Jensen and Kuperman, 1982). In water depths of 1-2m with slow sound speed sediments, the usable frequency range for organisms is extremely limited particularly for those using signals with lower frequency components (Fine and Lenhardt, 1983; Rogers and Cox, 1988). As water levels drop in a wetland - as they do in the summer months; the usable frequency range would be restricted to the higher frequencies. If however, the sediment contained a sand component or gravel (where sound is rapidly attenuated (Aiken, 1982a)); then cut-off frequencies would be lowered (Rogers and Cox, 1988). Many wetlands on the Swan Coastal Plain display high organic and silt, gas-rich sediments.

Generally, the sediment in a wetland consists of three different layers: an oxidised zone which lies at the soil water interface; an alternately aerobic / anaerobic zone which lies beneath the oxidised zone; and below this, a permanent anaerobic zone (Reddy and D'Angelo, 1994). In these freshwater organic rich sediments, two major pathways of movement are available for organic molecules. Either they combine with metal cations and thus become a part of the sediment, or they form gas. Gases produced under both aerobic and anaerobic decomposition includes: CO_2 , NH_3 , N_2 , H_2S and CH_4 (Anderson and Hampton, 1980a; Reddy and D'Angelo, 1994) and these bubbles are stored in the sediment or they escape into the water media above (Anderson and Hampton, 1980a). The presence of gas bubbles in both water and in sediments can greatly influence the acoustical properties within an aquatic environment (see Anderson and Hampton, 1980a,b).

Attenuation and sound speed within sediment and in water are dependent on the frequency of propagation of the sound wave and the resonance of the gas bubbles present. Gassy water acts as a highly dispersive medium of a propagating sound wave and is greatest when the frequency is near the resonance frequency of the bubbles (Anderson and Hampton, 1980a; Urick, 1983). Anderson and Hampton (1980b) found that gassy sediments form highly reflective boundaries and that gas content in sediments slowed the sound speed at even small concentrations as low as 0.1% (sound speed in sediment were slowest in clay and silt compared to fine / coarse sands). However, bubble resonance also influences propagating conditions in sediments with their maximum influence seen in the mud and silt sediments, which are the predominant substrates found in many wetlands along the Swan Coastal Plain.

To highlight the important influence that gas in sediments have on acoustical properties in a waterbody; Jackson and Williams (1996) found that scattering from the bubble layer (which was about one metre below the sediment/water interface) dominated scattering occurring due to other features such as roughness of the sediment surface. Interestingly though, echoes from the substrate could be used for organisms to 'hide' in, which Astrup (1999) has suggested maybe one reason why some marine fish have been observed swimming to the bottom sediments - to escape echo-locating predators. Under relatively undisturbed conditions in a wetland, the type of backscattering described in Jackson and Williams (1996) is likely to be accounted for by the organisms present in wetlands, however it is not known what impacts might occur for organisms within degraded wetlands where anaerobic decomposition dominates resulting in unnaturally high gas content in the substrate or water.

1.5 Objectives

In order to assess sound production by *C. oblonga* within an aquatic setting, the objectives of this study were:

1. To describe the ambient noise in freshwater systems and to ascertain the extent of spatial and temporal variations in the sound field.
2. To determine if *C. oblonga* emits or produces sound and to develop methods to detect these sounds.
3. To analyse turtle vocalisations and establish a categorisation of calls, determining if there were differences between males / females / and juveniles.
4. To trial play-back of vocalisations in order to determine if there were any behavioural or auditory responses by turtles to these sounds.

Chapter 2.0 GENERAL METHODS

2.1 Introduction

In total, over 230 days were spent in the field and more than 500 hours of tape recordings were made for this research. The research consisted of three major components: 1.) Monitoring ambient noise in a range of wetlands, 2.) Recording the sounds made by *C. oblonga* within a natural setting and in an experimental setting and 3.) The response of *C. oblonga* to acoustic stimuli. For each of the above components, details of the specific methodology are located within the relevant chapters. General methods are discussed below.

2.2 Field Sites

Study sites were selected as wetlands known to support populations of turtles. Wetlands were within the Perth Metropolitan area and included sites on the Swan Coastal Plain as well as two inland wetlands on the eastern borders of the metropolitan area (See Map. 2.1).

2.2.1 Blue Gum Lake:

Blue Gum Lake Reserve is located at latitude 32° 2' 31" South, and longitude 115° 50' 43" East (Map. 2.2).

Blue Gum has a permanent hydrologic regime, but is a relatively shallow water body, which diminishes in size due to evaporation during the summer months. The southwestern end often dries completely during summer. Blue Gum is characterised by large dead trees in the central body of water and has a small terrestrial buffer of native vegetation on the eastern aspect. Nuisance algal blooms have been a problem at this wetland in the past (Street, 1992) and at times, *Azolla filiculoides* forms extensive mats on the surface of the lake.

2.2.2 Lake Leschenaultia:

Lake Leschenaultia lies within the Shire of Mundaring 38km inland from the coast, and east of the Perth Metropolitan area. It lies at latitude 31º 51' 10" S and longitude 116º 15' 07" E (Map. 2.3).

Lake Leschenaultia was constructed in 1897 by the West Australian Government Railway. This lake was built to provide water to the steam engines that came through John Forrest National Park on their way out to Northam and York, and other locations further east. Leschenaultia has a permanent hydrologic regime. Water in this lake is contributed to by five different catchments. Surrounding the lake, terrestrial vegetation consists of forested areas of Marri (*Eucalyptus calophylla*), Jarrah (*Eucalyptus marginata*) and Wandoo (*Eucalyptus wandoo*) (Notice board at Lake Leschenaultia). Within the lake, large stands of the emergent native rush - the Jointed Twigrush (*Baumea articulata*) grows around much of the lake. The northern shore is cleared of emergent vegetation for public bathing as well as other locations intermittently spaced around the lake to provide public access to the water. Public usage of the lake is passive recreation - limited to swimming, picnicking and canoeing. One outboard, motorised boat is used for rescue on the lake.

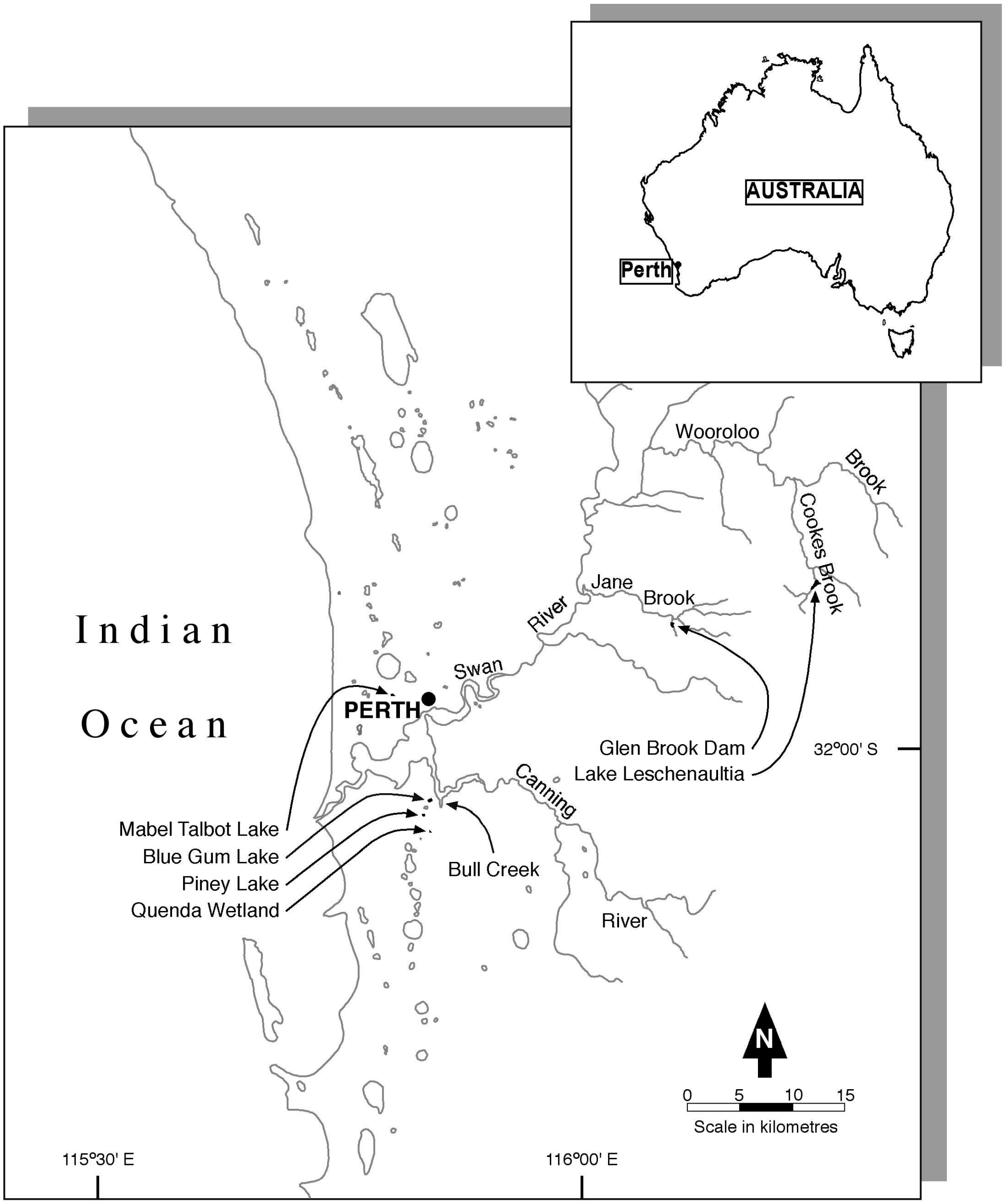
2.2.3 Glen Brook Dam:

Glen Brook Dam lies within the John Forrest National Park on the Darling Scarp 26.5km inland from the coast. It lies at latitude 31º 53' 16" S and longitude 116º 05' 29" E (Map. 2.3).

A telephone interview was conducted with Mr George Duxbury (12th March, 2003), formerly the ranger in John Forrest National Park, who was able to give a history of Glen Brook Dam. The dam was constructed during the mid-sixties, under the authority of the Public West Department (PWD) as it was envisaged that it was needed as a 'backup' reservoir for the Mundaring Weir during very dry periods. However, it was mainly used to provide flow-through to the swimming pool within the National Park and for irrigation purposes. Today it is reserved for wildlife usage, continued irrigation of park lands and also for fire-fighting purposes. Some recreational use (swimming) occurs, but this is outside its designated use. Similar to Leschenaultia, terrestrial vegetation surrounding this dam are Marri (*Eucalyptus calophylla*), Jarrah (*Eucalyptus marginata*) and Wandoo (*Eucalyptus wandoo*), with a small stand of the Jointed Twig-rush (*Baumea articulata*) which occupies approximately one-quarter of the western margin of this dam.

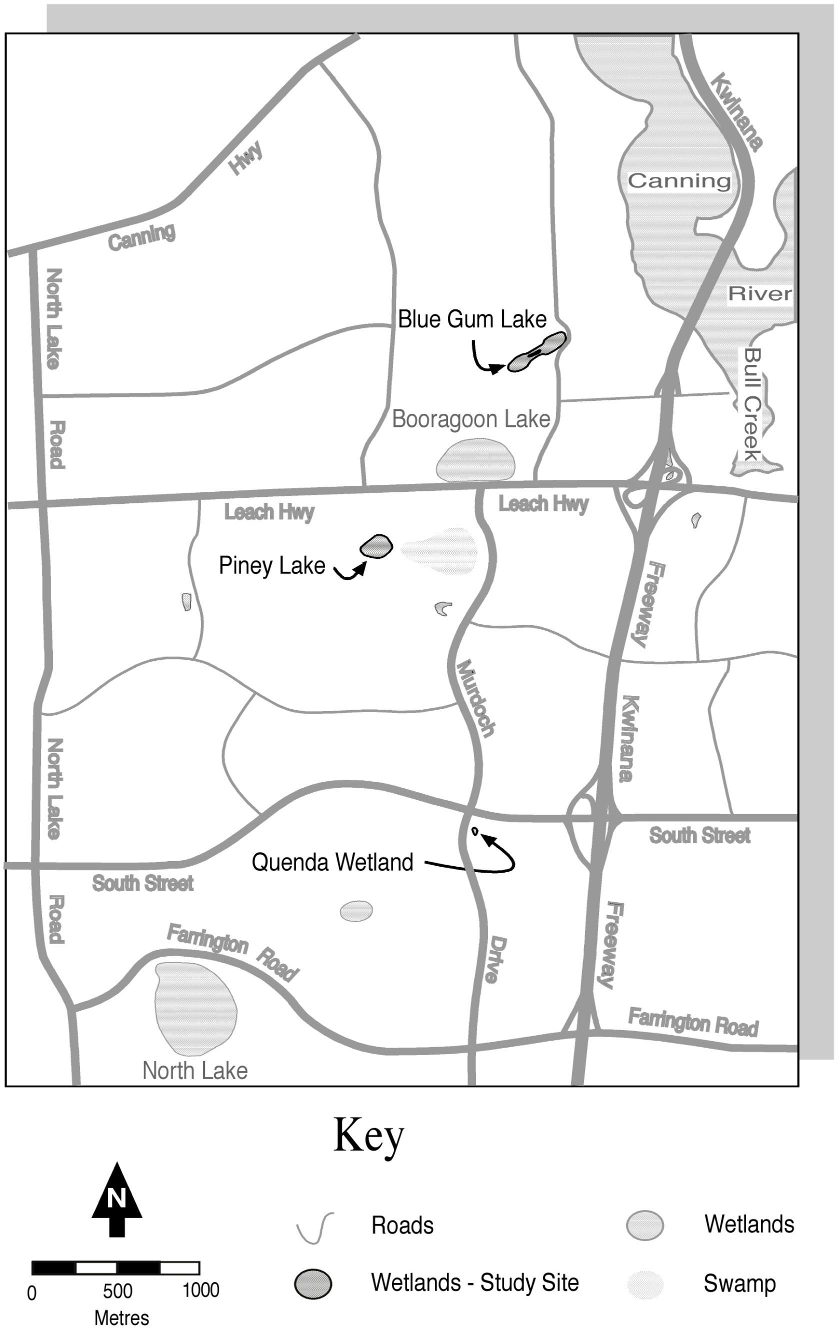
2.2.4 Quenda Wetland

Quenda Wetland is a small waterbody, which lies to the east of Murdoch University at the intersection of South Street and Murdoch Drive. It is located within native vegetation. Small stands of emergent reeds and rushes have been planted in recent years. Logs lie within this wetland providing hiding and basking sites for turtles. The substrate is sandy with an organic mix and reaches a winter maximum of around 1.8m depth.



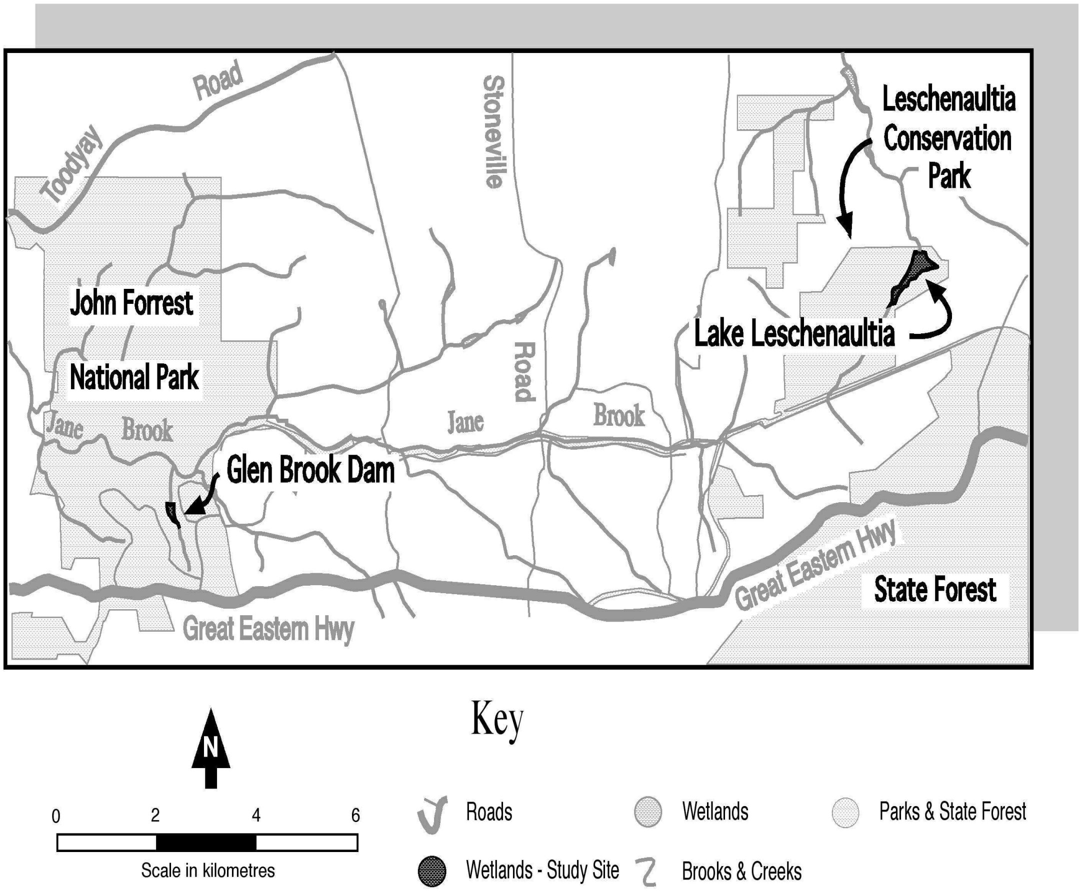
Note: For an expanded view of the 'Study Sites', please refer to the maps that follow.

Map.2.1. Map of the Swan Coastal Plain showing distribution of wetlands and study sites. (Map produced by Steven Goynich, Murdoch University)



Map.2.2. Map showing expanded view of the urban study sites Blue Gum Lake, Piney Lake and Quenda Wetland, which also shows their proximity to urban roads and the Leach Highway.

(Map produced by Steven Goynich, Murdoch University)



Map.2.3. Map showing expanded view of the inland study sites: Glen Brook Dam and Lake Leschenaultia.

(Map produced by Steven Goynich, Murdoch University)

2.3 Equipment and Procedures

2.3.1 The Animals

Most of the turtles used in this research were hand-captured with only a small number being trapped. Traps used were baited, modified funnel traps (Kuchling, 2003). Most of the female turtles were ultra-sounded by Dr Gerald Kuchling from the University of Western Australia to ascertain follicle development and receptivity for breeding. The ultra-sound equipment used to determine gravid females was a *Toshiba Sonolayer-L Sal 32B tomographic scanner*. The probe was a linear array probe (*Toshiba IVB 505S)* with a centre frequency of 5MHz, the focal distance of the acoustic lens was 30mm and a field depth of 113mm and width 56mm (Kuchling, 1989).

2.3.2 Recording Sounds within Wetlands

Although the acoustic characteristics of underwater sounds within a wetland or produced by turtles was unknown, recording equipment was chosen to detect within the audible range. This seemed reasonable considering that vocalisations of terrestrial chelonians studied to date lie within the audible range (e.g. Campbell & Evans, 1967, 1972; Auffenberg, 1964, 1978; Jackson & Awbrey, 1972) and also the in-air auditory ability for freshwater turtles appears to lie within this range (e.g. Wever, 1978; Fettiplace *et al.,* 1972). Although the hydrophones covered a frequency range extending into the ultrasonic frequencies, the costs involved in obtaining custom-built recording equipment that covered the audible and ultrasonic frequencies were outside budgetary constraints.

In an attempt to control for the effects of human disturbance when placing the hydrophone in the water, recordings began approximately two minutes after exit from the water. Noise and conversations were also kept to a minimum and when possible, sitting occurred rather than standing so as to minimise any effects that a human observer may have on turtles.

Two types of hydrophones were utilised in this study. These were: the Cetacean Research Technology (CRT) C53 hydrophone model and the Hightech HTI-96-MIN. An identical HTI-96-MIN was loaned by the Centre for Marine Science and Technology (CMST), Curtin University in 2004. The C53 was used in 2003 but was damaged and replaced with the HTI-96-MIN hydrophone.

The C53 hydrophone had a frequency response of 14Hz to 60KHz (±4.5 dB) and was omnidirectional below 10 kHz. Two high-pass filter options were available on the hydrophone: one at 16 Hz and the other at 350 Hz. The C53 had a nominal sensitivity of -165 dB, re 1V/µPa (this included the gain from the hydrophone preamplifier). The HTI-96-MIN hydrophone had a sensitivity of -164 dB re: 1V/µPa with a frequency response of 20Hz to 30KHz (-3dB). The sensitivity was flat within this band and was reference calibrated to USRD standards.

Analog recordings were made on a TASCAM DA-P1 DAT recorder, using a sampling rate of 48 kHz in the short play mode. The specifications of the tape deck were: a linear frequency response of 20 Hz – 20 kHz ± 0.5 dB (in short play mode); a 16 bit resolution; a tape speed of 8.15 mm/s with signal to noise ratio of >90 dB playback and wow and flutter unmeasurable at <0.001% (Technical Documentation sheet). The input level was set at ‘8’ and fixed by taping the dial to prevent movement. The analogue input was set on PAD 20 dB to ensure peaks from the source didn’t exceed the OVER setting in the peak level meter. The PAD 20 dB setting reduces the signal by 20 dB. As the recording equipment did not detail local time, but rather gave a time signature (ABS) from the start of each tape; local times were synchronised against the ABS time on the recorder.

Sounds recorded were digitised using a Sound Blaster Audigy DE 24-bit/96kHz stereo sound card with a noise level of 100 dB SNR in an Intel Pentium 4 PC. The sound card recording levels remained fixed throughout all analysis. The sound analysis software was SpectraPLUS version 2.32.04. This is a 32 bit Windows program, with a maximum sampling rate of 48 kHz and a sampling precision of 8 or 16 bit. It has a maximum of 2 channels with modes in real-time, recording and post-processing. Adobe Audition version 1.0 was used for filtering functions.

2.3.3 Calibration of Equipment

DAT recorder:

Recording equipment was checked using a calibrated signal generator to measure the response of the recorder to a range of frequencies using input levels from 0 to 10 on both left and right channels to ensure gain was the same in both channels. The signal generator was a TRIO 15 MHz oscilloscope CS-1560-AⅡ. The 0 dB mark on the recorder was also checked to ensure that no clipping occurred at this point. A 1 kHz signal at an input level of 660 mV peak to peak (or 482 mV RMS value) was used to check this didn’t occur. Results indicated gain was the same for both channels at all input settings. No clipping occurred at the 0 dB mark. To check the frequency response of the tape deck, a white noise signal was generated at -90dB re 1 V^2/Hz and was fed directly into the recorder (using the right channel) revealing a flat frequency response (input-2) (Figure. 2.1).

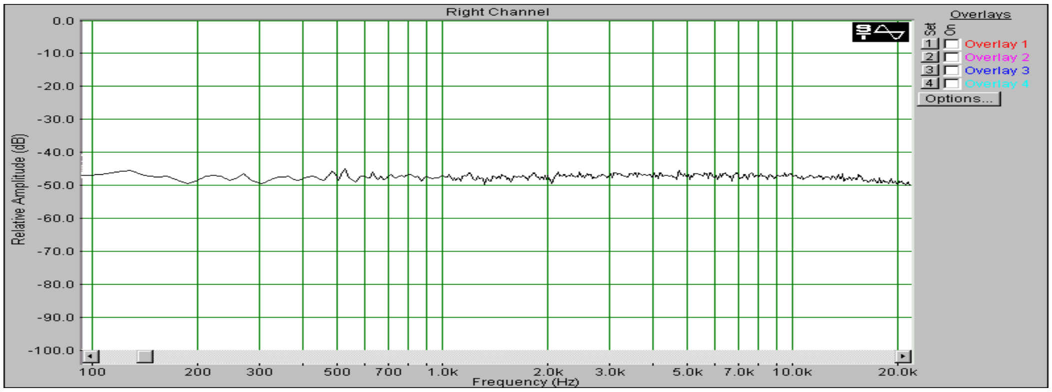


Figure.2.1. Spectrum of the calibrated white noise into the right channel of the TASCAM DAT-P1 recorder. A flat response is evident (FFT 4096 points, sample average of 60)

To determine the gain at the different input settings on the recorder, a 1 kHz signal (at 500 mV) was fed directly into the right channel.

To calculate gain at the different input levels on the DA-P1 DAT recorder using line of best fit: $y = mx + c$

(where y = relative amplitude in dB, m = slope, c = y-intercept) (Figure. 2.4).

Gives calculated values of $m = 3.837$, $c = -39.05$, $r^2 = 0.9701$

∴ the difference between the gain at an input of 8 (used throughout the research) and gain at input of 2 (used in calibration) gives:

→ -8.354 dB - -31.376 dB = 23.022 dB

i.e using an input-8 gives an overall gain to the signal of 23 dB relative to the input-2 setting.

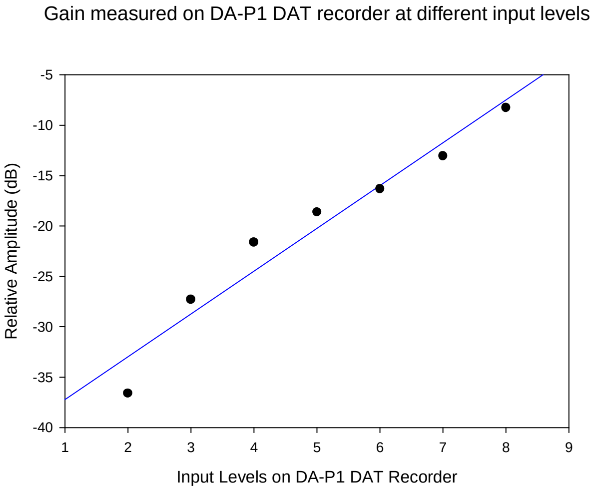


Figure.2.2. Linear plot of gain on the Tascam DA-P1 DAT recorder using the input settings 0-8.

Hydrophones:

Each hydrophone was calibrated once throughout this research. The hydrophone, recording gear and sound card were calibrated as a single unit by comparing the spectral output of a calibrated hydrophone against the C53 and the HTI-96-MIN hydrophones. Deterioration of equipment was not expected within the time frame of this research and so more frequent calibrations were considered unnecessary.

Calibrated hydrophones were supplied by the Centre for Marine Science and Technology (CMST) from Curtin University. Calibrated hydrophones used were: 1.) a MASSA TR–1025 C hydrophone (serial number 495) and a sensitivity of

-195 dB, re 1V/µPa; and 2.) a RESON TC 4033 hydrophone (serial number 4703110) and a sensitivity of -202 dB, re 1V/ µPa. For the MASSA hydrophone, a 20 dB preamplifier was used and a 40 dB preamplifier was used for the RESON. Hydrophone calibrations were undertaken on the Armaments wharf at the West Australian Naval Base on Garden Island, under the authority of Dr Darryl McMahon from the Defence Science and Technology Organisation (DSTO). Calibration took place under the supervision of Dr Alec Duncan from CMST, Curtin University and also Dr David Matthews from DSTO.

The method of calibration consisted of a calibrated hydrophone initially taped next to the C53 hydrophone (using the 16 Hz high-pass hydrophone filter) and later, the HTI-96-MIN hydrophone. A UW 30 speaker (signal source) was secured 1m from the hydrophones and this gear was then lowered 3m into the sea (max. water depth 14m). In calibration of the C53, a calibrated sound source was played through the speaker using frequency sweep's and white noise. In calibration of the HTI-96-MIN, a CD was used with recorded test signals of constant amplitude sinusoids using frequency sweeps from 50 Hz to 10 kHz in 30 seconds and tones at 50 Hz, 100 Hz, 750 Hz and 1000 Hz. The input level on the recorder was set at 2 for calibration, with the analog input set on PAD 20 dB to ensure that no clipping occurred. Calibrated hydrophones were plugged into the right channel of the tape deck with the left channel being used for the non-calibrated hydrophones. Using the onscreen frequency cursors to take an average across the entire spectrum, the output of the C53 hydrophone was measured to be 10 dB higher than that of the MASSA hydrophone with a 20 dB preamplifier (see Figure. 2.3). The sensitivity of the MASSA hydrophone is –195 dB re 1 V/ µPa but with the 20 dB pre-amp gain the sensitivity at the output of the preamplifier would be -175 dB, re 1V / µPa. Since the sensitivity of the C53 is 10 dB higher than this, its sensitivity is therefore -175 dB, re 1V / µPa + 10 dB = -165 dB, re 1V / µPa.

Similarly, the output for the HTI-96-MIN hydrophone was measured to be 2 dB lower than that of the RESON hydrophone with a 40 dB preamplifier (see Figure. 2.4). The sensitivity of the RESON hydrophone is -202 dB re 1V / µPa but with the 40 dB preamplifier gain, the sensitivity at the output of the preamplifier would be

-162 dB, re 1V / µPa. Since the sensitivity of the HTI-96-MIN is 2 dB lower than this, its sensitivity is therefore –162 dB, re 1V / µPa - 2 dB = 164 dB, re 1V / µPa.

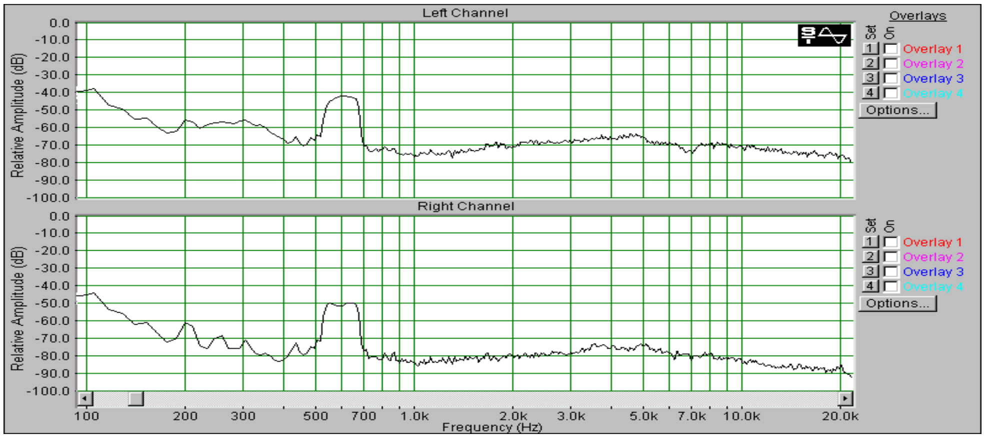


Figure.2.3. Spectra obtained in hydrophone calibration comparing the output of the CRT C53 hydrophone (left channel) with that of the calibrated hydrophone the MASSA TR-1025 (right channel) using frequency sweeps (the peak at 600 Hz indicates the relative outputs from each hydrophone from the frequency sweeps by the calibrated sound source) (FFT 4096 points with an averaging of 60 samples).

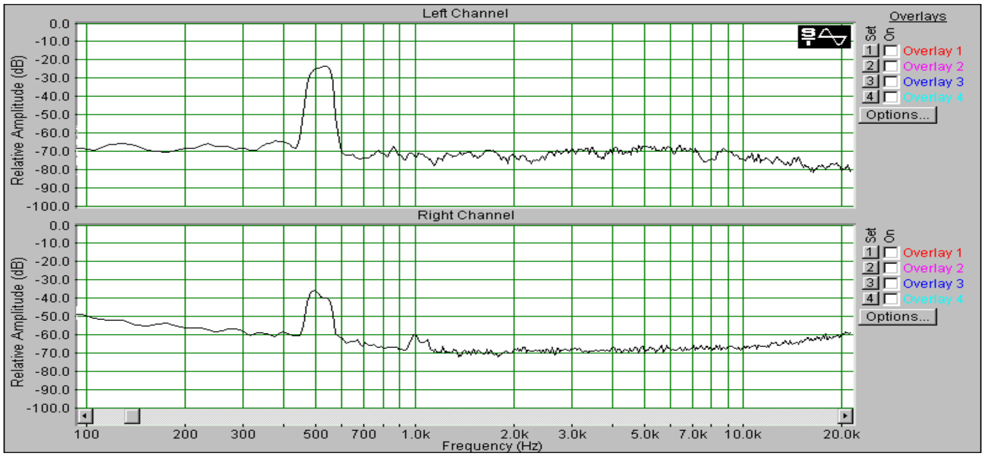


Figure.2.4. Spectra obtained in hydrophone calibration comparing the output of the HTI-96-MIN (left channel) with that of the calibrated hydrophone, the RESON TC 4033 (right channel) using frequency sweeps (the peak at 500 Hz indicates the relative outputs from each hydrophone from the frequency sweeps by the calibrated sound source) (FFT 4096 points with an averaging of 60 samples).

2.3.4 Analysis of Acoustic Sounds

- To calculate pressure spectrum levels in dB re 1 μPa^2 / Hz, this was given by the equation:

Equation.1. i.e. Pressure spectrum levels in dB re 1 μPa^2 / Hz = output voltage spectrum level (dB re 1 V^2 / Hz) + correction factor (dB re V/μPa)

(Where: Output voltage spectrum level = voltage level in analysis frequency band f_{rs} - 10 $\log_{10}$ (f_{rs}) and correction factor = -hydrophone sensitivity – gain)

- Spectral values output by SpectraPLUS were converted to units of dB V^2 / Hz by subtracting from the relative amplitudes 10 $\log_{10}$ (f_{rs}) where f_{rs} = the frequency resolution (from McCauley, 2001, P.46).

Equation.2. i.e. For FFT of 4096 points with a frequency resolution of 11.6 Hz

$$\rightarrow 10 \log_{10} (11.6) = 10.64$$

∴ 10.64 was subtracted from the relative amplitudes to give dB V^2 / Hz when using an FFT of 4096 points.

- To calculate the gain of the analysis system at each session, a tape with white noise recorded at -90 dB re 1 V^2/Hz (input 8) was played into SpectraPLUS. The level of the white noise was analysed using the onscreen frequency / amplitude cursors and gain was calculated by subtracting the known input level (-90 dB re 1 V^2/Hz) from the measured amplitude (corrected to V^2 / Hz by subtracting 10.64 for FFT 4096 points).

The correction factor then utilised for spectral analysis was →

the sensitivity of the hydrophone minus the gain of the recording set-up.

Equation.2. Correction factor = - Hydrophone Sensitivity - gain

This was then added to the corrected relative amplitude values (in V^2 / Hz) of the sound field to give pressure spectrum levels in dB re 1 μPa^2 / Hz .

- To analyse the noise levels in a wetland, one minute time averages were used (averaging of 700). Number of samples were calculated as follows:

Equation.3.

$$\frac{4096 \quad \text{(FFT)}}{48{,}000 \quad \text{(sampling frequency)}}$$

which gives the number of samples per second 0.08533

$\therefore$ 703 samples were required per minute

Statistical analyses were performed in EXCEL. Graphs were produced in SigmaPlot 2002 version 8.0.

2.3.5 Classification of Acoustic Units

Terminology used for signals used by aquatic invertebrate organisms was taken from Broughton (1963) and Janssen (1973a, p.3.).

Pulse:

The shortest identifiable unit of a signal seen in a spectrogram.

Pulse-train:

A series of pulses, which are usually separated by an interval of silence before another series of pulses.

Pulse-train Group:

A group of more or less similar pulse-trains.

Dominant Frequency:

Is that frequency with the greatest amplitude.

Temporal Pattern:

The way in which the units are arranged in a signal.

Signal:

A signal is a complete set of pulse-trains or group of pulse-trains

For the turtle vocalisations, classification of sounds was based on terminology used in both insect (e.g. Broughton, 1963; Janssen, 1973a), bird song (e.g. Shiovitz, 1975; Thompson *et al.,* 1994) and some of the aquatic mammalian groups such as cetaceans (e.g. Clark, 1982).

2.3.6 Chorusing

Cato (1978, p.737) defined a continuous chorus as: "when the noise from many individuals is continuously above the background for an extended period (usually an hour or more) using an equipment averaging time of 1 second".

McCauley (2001) added a further category to include what he termed a 'dis continuous' chorus where discernable, non-overlapping calls occurred often enough to produce a significant increase above the ambient sound when using an averaging time of 1 minute in the signal analysis. In this study, a third category was added to include the contribution from a single organism that produced a persistent call (where each bout lasted 35 seconds or longer, punctuated by short silent intervals of just over 1 second) which continued throughout the entire recording and was sufficient to produce dominant peaks in the ambient noise spectra well above the background noise levels. These calls were termed 'persistent non-chorus' contributions.

2.3.7 Cut-off Frequencies

To calculate the absolute cut-off frequency for propagation of sound in the three wetlands, water depths obtained from bathymetry measurements were used in the calculations and gave a maximum summer depth at Blue Gum Lake of 1.0m and in winter a maximum of 2.0m. At Lake Leschenaultia and Glen Brook Dam (which remain relatively deep throughout winter and summer) - a summer water depth of 1.0m was used as turtles were noted to use the margins of the wetlands in the evenings. In winter an 8m depth was used in the calculations (although this could be at least 2m deeper in winter at the maximum allowable height for these wetlands).

Initially, the sound speed for the water was calculated using the formula given by Medwin (1975):

<u>Equation.4.</u>

$$c = 1449.2 + 4.6T - 5.5 \times 10^{-2}T^{2} + 2.9 \times 10^{-4}T^{3} + (1.34 - 10^{-2}T)(S - 35) + 1.6 \times 10^{-2}D$$

Where D is the depth (m), S is the salinity of the water (parts per thousand) and T is temperature of the water (^{o}C).

Medwin's formula was chosen because the limits specified for use of the formula allowed for the fluctuations experienced within these wetlands ($0 \leq T \leq 35^{0}$, $0 \leq S \leq 45$ ppt, $0 \leq D \leq 1{,}000$ m) (N.B. For some inland West Australian wetlands; where salinities can exceed those of sea water (Lien Sim, pers. com) this formula may not hold). Salinity was taken at 4 ppt in summer and 1.5 ppt in winter.

Water temperatures used in the calculations were 28 °C in summer at Blue Gum Lake and 24 °C at Lake Leschenaultia and Glen Brook Dam. The winter temperature of 15 °C was used for all three wetlands. These temperatures reflected those recorded throughout the study period, but were below the maximum and above the minimum.

CALCULATIONS FOR SOUND SPEED IN WATER AT BLUE GUM LAKE

In summer	In winter
Using approximations:	Using approximations:
$T_S = 28$ °C	$T_W = 15$ °C
$D_S = 1$m	$D_W = 2$m
$S_S = 4$ppt	$S_W = 1.5$ppt

Sound speed in the water was calculated using Medwin's (1975) formula to give:

$C_S = 1508.4$ m/s in summer $C_W = 1467$ m/s in winter

CALCULATIONS FOR SOUND SPEED IN WATER AT GLEN BROOK DAM AND LAKE LESCHENAULTIA

In summer	In winter
Using approximations:	Using approximations:
$T_S = 24$ °C	$T_W = 15$ °C
$D_S = 1$m	$D_W = 8$m
$S_S = 4$ppt	$S_W = 1.5$ppt

Sound speed in the water was calculated using Medwin's (1975) formula to give:

$C_S = 1497.84$ m/s in summer $C_W = 1467.06$ m/s in winter

The sediment sound speed was then calculated from ratios in Jensen *et al.,* (P.38, 2000) for sound speed in silt (predominant constituent in sediment at Blue Gum Lake) and in gravel (predominant constituent in sediment at Lake Leschenaultia and Glen Brook Dam), using the calculated sound speed in water from Medwins (1975) formula above. Using the ratio's given in Jensen *et al.,*(2000) for a silt bottom (1.05), sound speed in the sediment at Blue Gum Lake was calculated as follows:

Equation.5.

$$\frac{C_{sediment}}{C_{water}} = 1.05 \quad \therefore \; C_{sediment} = 1583.82 \text{ m/s for summer}$$

$$\frac{C_{sediment}}{C_{water}} = 1.05 \quad \therefore \; C_{sediment} = 1540.4 \text{ m/s for winter}$$

Using the ratio's given in Jensen *et al.,*(2000) for a gravel bottom (1.2), sound speed in the sediment at Glen Brook Dam and Lake Leschenaultia was calculated as follows:

$$\frac{C_{sediment}}{C_{water}} = 1.2 \quad \therefore \quad C_{sediment} = 1797.41 \text{ m/s for summer}$$

$$\frac{C_{sediment}}{C_{water}} = 1.2 \quad \therefore \quad C_{sediment} = 1760.47 \text{ m/s for winter}$$

The absolute cut-off frequency for propagation of sound was then calculated using the formula given by Urick (1983) and Rogers and Cox (1988):

Equation.6.

$$F_c = \frac{c_w / 4h}{(1 - c^2_w / c^2_s)^{½}}$$

Where c_s is the sound speed of the sediment (ms^{-1}), c_w is the speed of sound in water (ms^{-1}), h is the depth of the water (m) and f_c has the units Hz.

The theoretical estimate for cut-off frequencies was considered reasonable in the absence of empirical data.

2.3.8 Video Equipment

In order to associate sound with behaviour, an underwater camera system was used to view in real time. An infra-red (IR) camera was used, although the light emitting diodes (LED's) emit a red light source and may possibly attract turtles to it, the infra-red camera was considered a better option for use in tannin-stained waters of the wetlands as well as night-time viewing, as opposed to the use of a conventional camera.

This equipment was supplied by Underwater Camera Systems and consisted of a K-OL-2000 WP Lipstick camera with IR illumination using six LED's. Camera lens angle was 75º and the image device was a 1/3" Sony CCD with 400 TVL resolution. This later needed to be modified, as the six LED's were insufficient to obtain sensible images, and also the shape of the camera was difficult to manoeuvre. An array of 32 LED's were placed around the original 6 LED's on the camera face. The water-proof housing was also changed from a long cylindrical shape to a broad, short cylindrical shape, which was constructed to be neutrally buoyant.

The camera was mounted on an underwater tripod arrangement as well as hand-held. For extra illumination, four M120 infra-red spotlights consisting of a panel of 28 LED's were sometimes used. For the night-time behavioural observations, conventional underwater lighting was used consisting of a 'Waterwerks Underwater 50 watt Aqua Light'.

2.3.9 Wind Speed

Average wind speed was taken at each site whilst recordings were taking place to ensure influence of wind was accounted for in the data. Initially, this was measured by a Monitor Sensor AND-02 Anemometer, which was loaned by the Centre for Water Research at the University of Western Australia. This anemometer had three conical heads, giving it a near linear relationship between rotational speed and wind speed. It had a threshold of 0.3m/s and counts every four revolutions – measuring 1/100th of a kilometer for each digital cycle (UWA / Instruments, 13.2.03). Unfortunately, this stopped working and was replaced with a simple, hand-held CASELLA anemometer with wind speed scales in m/s, Beaufort, km/hr and knots.

2.3.10 Invertebrate Statistical Analysis

Invertebrate samples collected with sound recordings were identified to class or order for the microinvertebrates and the macroinvertebrates were identified to family or genus. Statistical analysis was performed by PRIMER 5 version 5.2.2.

2.4 Pilot Studies

Initially a number of pilot studies were conducted within: an artificial pond; at Blue Gum Lake; Glen Brook Dam; and Lake Leschenaultia. Four approaches were used to determine turtle vocalisations:

2.4.1 Isolating Turtle Vocalisations

<u>Study.1.</u>

Method

Initially nine adult Blue Gum Lake turtles (8 females; 1 male) were placed into a large round tank with dimensions of 1.8 m dia. x 0.65 m depth. The hydrophone was suspended from the center of the tank and recordings occurred over several hours. Turtle behaviour was also filmed during this period.

Results

Only scraping and scratching sounds were heard with no vocalisations. Turtles 'huddled' together as a close group or on top of each other throughout the trial with some biting and chasing observed.

<u>Study.2.</u>

Method

The second approach used was to see if turtles emitted a distress call. So turtles were trapped in the usual way – using baited, modified funnel traps. The hydrophone was suspended in front of the trap but no vocalisations were recorded. The trap was then raised up and down quickly in the water three or four times to distress them.

Results

No discernable vocalisations were heard.

Study.3.

Method

As animals may go quiet under laboratory settings and also, due to the known difficulties for making acoustic recordings in laboratory aquariums (Parvulescu, 1966; Hawkins and Myrberg, 1983; Yager, 1992); the experimental design and equipment used for the third experiment was based on a similar arrangement to that used in Yager (1992). In order to approximate 'free-field' sound conditions and obtain representative acoustic recordings of *C. oblonga,* a sound-transparent floating cage was constructed to constrain a turtle within a wetland setting. The frame of the cage was constructed of 50mm poly-piping (depth 1.0m x width 0.75m) which was then covered with non-reflective material (polyester fabric (shade-cloth)). The poly-piping was chosen for its sound transparency and also buoyancy properties. The hollow poly pipes were filled with water to the water-line of the wetland. This ensured the removal of air in the pipes that were submerged, making the structure as acoustically sound-transparent as possible and also weighted the cage to ensure only partial submersion in the waterbody, which enabled constraint of the turtle as well as enabling the turtle(s) to surface for air as well. The cage was tethered by rope to four plastic stakes to prevent it tipping over or drifting.

The turtle was placed within this cage with the hydrophone suspended in the middle and was monitored for several hours.

Results

No discernable vocalisations were heard.

Study.4.

Method

As the unnatural settings may have been the reason why turtles weren't vocalising, a more natural setting was arranged in Blue Gum Lake. Bait (chicken livers) was tied in a cotton cloth and then tied to a stake placed in the wetland. This was used to attract the turtles to the site. Camera equipment was suspended from the tripod arrangement and filmed throughout with the hydrophone suspended from a separate pole.

Results

Again no vocalisations were heard - the only sounds were those of feeding.

2.4.2 Ambient Noise

See chapter 3 for details of this investigation.

Chapter 3.0 AMBIENT NOISE IN WETLANDS

3.1 Introduction

The objective of this investigation was to describe the ambient noise in freshwater systems and was undertaken on the assumption, that if turtles vocalised, then their calls would make up part of the ambient noise in a wetland and would establish the context in which turtles were vocalising. However, as there is a paucity of research on the underwater sound field in freshwater systems to guide this investigation; this study was largely conducted: 1.) As a pilot study to ascertain how best to approach sound recordings in a wetland setting; 2.) To determine if there were temporal and spatial differences in the sound field; and 3.) To determine if differences existed between wetlands. These investigations allowed trial and error in use of the equipment such as hydrophone placement and weather conditions, and to establish a 'best practice' recording regime for the remainder of this research.

3.2 Methods and Materials

3.2.1 The Wetlands

The ambient sound recordings consisted of a series of recordings made in three clear-water systems which were known to support turtle populations: Blue Gum Lake; Glen Brook Dam; and Lake Leschenaultia. These wetlands differed in terms of their location, degree of nutrient enrichment, water depth and complexity within the habitat. Clear-water systems in particular were chosen, because Blue Gum is a wetland that was estimated to support a large population of turtles (2040, 95% CI 1562 - 2937) (Giles, 2001), and was therefore a wetland predicted to be utilised for part of the acoustic research on the turtles. The clear-water also enabled some observation of turtles without the need for camera equipment. However, Blue Gum Lake is an urban wetland and some enrichment has occurred. Therefore, the other wetlands were chosen as relatively undisturbed sites to provide comparison of ambient sounds found in undisturbed, clear-water wetlands.

Blue Gum Lake is dominated by submerged macrophytes with a small section of emergent macrophytes. It is shallow (1-2m) and demonstrates seasonal drying in one half of the wetland. The substrate is predominantly mud / silt with a mix of sand. Lake Leschenaultia is dominated by submerged and emergent macrophytes and is a permanent waterbody with shallow and also relatively deep (max. depth ~9m) sections. The substrate is largely gravel and sand with an organic mix. Glen Brook dam is a steep-sided and also relatively deep (max. depth ~8m) permanent waterbody. Water levels have declined considerably in this dam but have the potential to reach close to 15m when the dam is full. There are no submerged macrophytes in this dam but is dominated by benthic algae with only a small section of emergent vegetation. Glen Brook Dam has large granite boulders within this wetland, which provide a complexity of habitat in contrast to that seen at the other two wetlands. The substrate is largely gravel and sand with some clay content.

3.2.2 The Recording Regime

Blue Gum Lake, Glen Brook Dam and Lake Leschenaultia were monitored over a four week period and occurred in the last month of summer and included the first week of autumn (Feb-Mar 2003). Recordings were made every week at the three wetlands within the time periods dawn (5am-7am), midday (11am-1pm), dusk (5pm-7pm) and midnight (10.30pm-12.30am) (Table. 3.1) (The 2-hour time blocks were necessary to accommodate the time taken to walk around the perimeter of the wetland for each recording period e.g. it took 1 hour to walk around Glen Brook Dam and Blue Gum Lake, but 2 hours to walk around Lake Leschenaultia). Recordings occurred at locations north, south, west and east around the wetland and lasted for approximately four to five minutes at each location and only the presence or absence of a call was noted.

Table.3.1. Summary of recording regime that was undertaken from February – March 2003 for the ambient sound recordings. Recordings occurred at each location (North, South, West and East) for approximately five minutes.

	Mon	**Tues**	**Wed**	**Thurs**	**Fri**	**Sat**	**Sun**
Blue Gum	Dawn NSWE	Dusk NSWE					
	Midday NSWE	Midnight NSWE					
Glen Brook			Dawn NSWE		Dusk NSWE		
			Midday NSWE		Midnight NSWE		
Leschenaultia				Dawn NSWE		Dusk NSWE	
				Midday NSWE		Midnight NSWE	

This recording regime was undertaken on the assumption that the recordings would reveal any diurnal changes in the sounds and provide a representative sample of sounds heard within these wetlands. In total, 25.4 hours of wetland noise recordings were made with 192 separate recordings. Recordings were made at a distance from the shoreline that was accessible in wading gear with the hydrophone suspended between two star-pickets above the bottom sediments. This method of recording at a distance from the shoreline was employed, as researchers would normally access wetlands in wading gear to obtain invertebrate samples to this distance and hydrophone placement represented the ‘sweep’ area that invertebrates would be taken from.

Recordings of ambient noise included windy days to ensure the natural component was accounted for in the recordings to represent natural variations encountered in ambient noise for the organisms present in wetlands. No precipitation occurred during the summer and so could not be accounted for during these recordings. Vehicle traffic including aircraft was noted and time recorded to ensure that anthropogenic noise contributions were also accounted for in the spectral outputs.

At each recording - water temperature, moon phase, and presence of waterbirds were noted. Wind speed was taken three times during the recording session. This was averaged and rated; Beaufort Wind Scale 0= no breeze (<1.8 km/hr or Beaufort Wind Scale 0), Beaufort Wind Scale 1= light breeze (1.8 km/hr to 6.12 km/hr or Beaufort Wind Scale 1), Beaufort Wind Scale 2= medium breeze (6.12 km/hr to 11.88 km/hr or Beaufort Wind Scale 2), Beaufort Wind Scale 3= strong breeze (11.88 km/hr to 19.44 km/hr). A sediment core sample was taken at each wetland using an Auger bit (internal diameter of 10cm x 12cm length), which was twisted down into the substrate. Bathymetry was also determined at each wetland by taking water depths at approximately 20m intervals from a canoe rowed across each wetland, using a grid overlay on an aerial map of the wetland.

3.2.3 Invertebrates

Invertebrate samples were taken over a period of a fortnight, at each recording session at each location. Sweeps for the invertebrates were made in accordance with the wetland macroinvertebrate rapid bioassessment protocol (Davis *et al*., 1999). A fine mesh sweep net (25 μm) was utilised and this was moved around the hydrophone in a zig-zag manner from surface to the bottom sediments – but not inclusive of the bottom sediments (bottom sediments were not usually included in the sweep but in shallow areas it was difficult not to obtain some bottom sediments in the sample). It wasn't the intention to determine which organisms were calling as sound can propagate over long distances underwater and the distance of the calling organism from the hydrophone was not known. Invertebrate samples provided data on the abundance and diversity as well as a presence / absence of invertebrate families at the wetlands for comparison and provided a rating of disturbance. Importantly, invertebrate samples were taken to find out if there were corresponding spatial and temporal differences in support of any variations found in the sound recordings. In total, 96 samples were collected to view what organisms were present in the water column near the hydrophone. The invertebrates were preserved in ethanol. The presence / absence of crustaceans such as Gilgies (*Cherax quinquecarinatus*) and Marron (*Cherax tenuimanus*) seen in a torch beam from shore to hydrophone were also noted (but not included in the data set) and also the presence of Billabong Mussels (Class Bivalvia).

Invertebrates were identified using Gooderham and Tsyrlin (2002); Davis and Christidis (1997).

3.2.4 Acoustic Signals

Acoustic signals are presented as narrow-band spectra produced from the Fast Fourier Transforms. They were digitised at a sampling rate of 48 kHz, FFT size 1024 points, giving a time resolution of 5.33 msec and a frequency resolution of 46.875 Hz with an averaging of 4 (with a 75% overlap). A Hanning smoothing window was used and due to concerns of aliasing, signals were filtered using the Butterworth low-pass filter using a corner frequency of 15 kHz. For each distinct signal presented, four main parameters were measured from the spectrograms: 1.) Frequency range in kHz (from the lowest to the highest measurable frequency), 2.) Average duration of the signal in seconds, 3.) The dominant frequency in kHz (frequency of that harmonic with the greatest amplitude) and 4.) For those signals with measurable pulses – the average inter-pulse interval (IPI) in seconds. The IPI was measured from the end of one pulse to the beginning of the next pulse (using the spectrograms). Examples of each call were selected to show them at their maximum level with well-defined spectral contours.

For the ambient noise section (biologics and wind), spectra were obtained from ⅓-octave band measurements made using SpectraPLUS software which synthesised ⅓-octave bands from FFT 4096 points and an averaging of 700 samples per minute (with no overlap), which gave a frequency resolution of 11.7 Hz and a time resolution of 85.3 msec. The recorded signal spectrum was converted to pressure using calibration factors (see Ch. 2 General Methods: section 2.3.4) and these results were bandwidth corrected by subtracting the appropriate ⅓-octave bandwidth correction (see Beranek, 1988), to give received sound levels in dB re 1μPa^2 / Hz.

Terminology used by Broughton (1963) and Jansson (1973a) has been adopted in the analysis of acoustic signals (Ch. 2 General Methods: section 2.3.5) for those organisms that had identifiable calls and able to be separated from the ambient noise recordings. Some calls may have been over-looked because they were not recognised as a call due to: 1.) their infrequent occurrence; or 2.) they could not be distinguished from the self-noise of the measuring and recording set-up, particularly on windy days. Some signals may have been masked in the ambient noise; such as bubbles produced by escaping gas. As most of the species that produced sounds have not been identified, the sounds were described according to their aural character or failing a suitable aural description; were described according to some spectrographic feature of the call.

3.2.5 Ambient Noise and Wind

Ambient noise was recorded at the three wetlands at each north, south, west east site for four different wind speeds – Beaufort Wind Scale 0,1,2 & 3 in the absence of intermittent noise such as gas bubbles and biologics. Wind speed measurements made during the summer study ranged from 0km/hr to 12.4 km/hr (Beaufort Wind Scale 0-3), so the wind speeds assessed were considered reasonable for this study. Wind recordings took place in the morning during the winter months as this time period was quietest of all periods of the day and also this season was relatively quiet biologically (compared to summer and autumn). While spring was predicted to be the most important period for turtles, winter ambient noise levels were used as the baseline to represent the spring ambient sound conditions. This seemed reasonable as little evaporation was expected to occur from winter water levels to spring and from some previous recordings undertaken in spring 2002, the biologics were active in this period and thus would have made an even greater contribution to ambient noise than the few 'Cork on Glass' callers recorded in winter.

Spectrum levels for recordings made at sites (N S W E) were tested using ANOVA tables for one-way analysis of variance to see if there was any difference in ambient noise between sites (Appendix 1).

3.3 Results

3.3.1 The Biologics Present

The frequencies that freshwater organisms utilised were between 3 kHz to around 14 kHz, with the exception of the 'bird-like song' which extended from 500 Hz up to around 10 kHz. Except for those sounds that have been categorised under the 'chorusing' section, there were four distinctive calls recognised throughout the study period. A summary of the acoustic properties of calls is presented in Table 3.2 and each call is described below with the spectrographic outputs and diel periods of calling activity revealing the temporal and spatial nature of these sounds.

Table.3.2. Summary table of the acoustic properties of the biological calls heard at Lake Leschenaultia, Glen Brook Dam and Blue Gum Lake, summer 2003.

Call (Aural character)	Wetland(s)	Spectral output	Mean duration (s)	Mean Number of pulses	Frequency range (kHz)	Dominant frequency (kHz)
'Tick, Tick…' (n=103)	Leschenaultia Glen Brook	pulse repetition	1.34 ±0.28	11.3 ±2.4	3 - 6	equally weighted
'Cork on Glass' (n=20)	Blue Gum Glen Brook	usually single pulse	.134 ±0.21	2 ±1	4.5 - 10	7 - 8
'Ratchet' (n=12)	Blue Gum	pulse repetition	0.721 ±0.391	*U/C	1.6 – 7	2.8 – 3.3
'Bird-like song'	Leschenaultia Glen Brook	complex	Variable	None	.5 - 10	1.8 - 2
Chorusing (C) and persistent non-chorus calls (PNC)						
'Tinsel caller' (PNC)	Blue Gum	pulse repetition	>1 min	numerous	7-9	7.5
'12/6 kHz rattle' (PNC)	Blue Gum	pulse repetition viewed as continuous spectral output	>1 min	numerous	5.8-13.8	10.6-13.8
'5.5 kHz rattle' (PNC)	Blue Gum	Pulse repetition viewed as continuous spectral output	>1 min	numerous	5-6.8	5 - 5.5
'Cork on Glass' (C)	Blue Gum	pulse Repetition viewed as continuous spectral output	>1 min to hours	numerous	4.5-10	4.5-10

*U/C Number of pulses unable to be counted

‘Tick, tick’ Call

The sound of the pulse-train in Figure 3.1 was heard as ‘tick, tick, tick, tick…’. The frequency range of pulse-trains mostly ranged from around 3 - 4.5 kHz with some extending up to 6 kHz. From a randomly selected pulse-train of 10 pulses, the average inter-pulse interval was 0.122 seconds (SD=0.013). This pulse-train was occasionally heard when recording intermittently during the winter at Lake Leschenaultia, and it was noted to be significantly slower (water temperature 14.5ºC). From a randomly selected winter pulse-train of 10 pulses, there was an average inter-pulse interval of 0.319 seconds (SD=0.022) (T= -22.61, P<0.001, DF=16). The highest number of pulses in a pulse-train was 18, heard once at Glen Brook Dam during the summer recordings and the lowest number of pulses was five, recorded at Lake Leschenaultia.

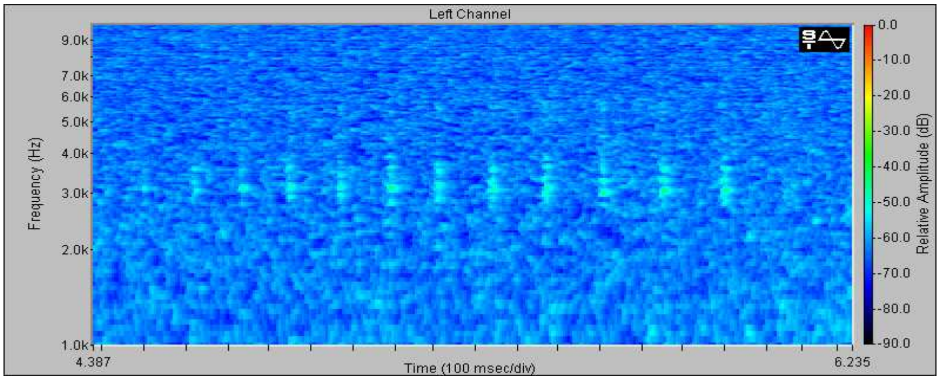


Figure.3.1. Spectrogram of a representative pulse-train of ‘tick-tick’ calls frequently heard at Lake Leschenaultia and Glen Brook Dam but were not heard at Blue Gum Lake. Recording taken at dusk, March 2003. Water temperature 23ºC.

The ‘tick, tick...’ organism was active throughout all time periods at Lake Leschenaultia but vocal activity was more widespread at midnight in particular, and to a lesser extent at dawn (Table. 3.3). Glen Brook Dam also reveals more activity in the midnight recordings (Table. 3.4) but unlike this caller at Lake Leschenaultia – was rarely heard throughout the other time periods. The main sites of occurrence for this call were south, west and east for all time periods at Lake Leschenaultia and at Glen Brook Dam – the south and west sites were predominantly utilised.

Table.3.3. Spatial and temporal calling patterns of the ‘tick, tick…’ call at Lake Leschenaultia, summer 2003 (‘+’ the call is present and ‘ – ’ the call is absent).

Lake Leschenaultia

Week	Dawn				Midday				Dusk				Midnight			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	-	-	+	+	-	+	+	+	-	-	+	-	-	+	+	+
3	-	+	+	-	-	-	+	-	-	+	+	+	-	+	+	+
2	-	+	+	+	-	-	-	-	-	-	-	-	-	-	+	-
1	-	-	-	-	-	+	-	-	-	-	+	-	+	+	+	+

Table.3.4. Spatial and temporal calling patterns of the ‘tick, tick…’ call at Glen Brook Dam, summer 2003 (‘+’ the call is present and ‘ – ’ the call is absent).

Glen Brook Dam

Week	Dawn				Midday				Dusk				Midnight			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	-	-	-	-	-	-	-	-	-	-	-	-	+	-	+	-
3	-	-	+	-	-	-	-	-	-	-	+	-	+	+	+	-
2	-	-	-	-	-	-	+	-	-	-	+	-	-	+	-	+
1	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	+

‘Cork on Glass’ Calls

The call depicted in Figure 3.2 sounded like ‘a piece of cork rubbed quickly on wet glass’, and consisted of either a single pulse, or two pulses made rapidly together; which are shown on the right in Figure 3.2. Occasionally there were three or more rapid pulses in the signal. Spectral components in the signal ranged from 4.5 kHz and at times, reached above 10 kHz, however greatest output occurred between 7-8 kHz. These calls were heard as isolated calls at dawn and midday (depicted in Figure. 3.2). However, by dusk, in particular, and to a lesser extent at midnight; individual calls were indistinguishable and organisms were in chorus (see Figure. 3.7). Interestingly, not all sites at Blue Gum Lake revealed this chorusing at dusk and midnight (see below under chorusing).

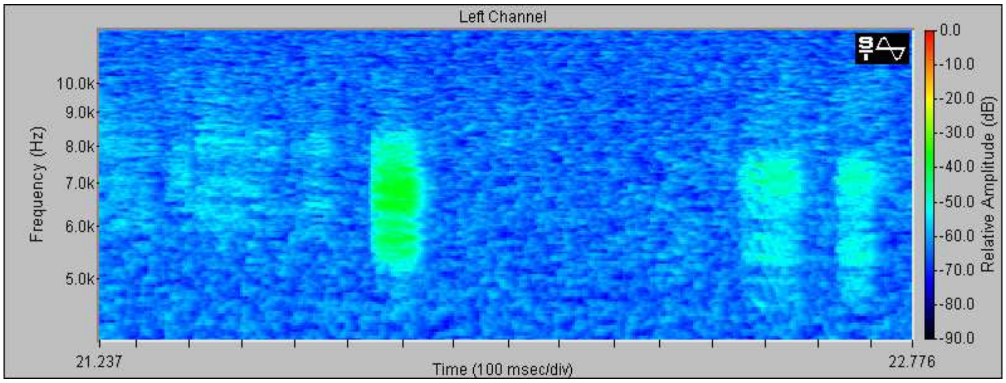


Figure.3.2. Spectrogram of the 'cork on glass' call regularly heard at Blue Gum Lake and Glen Brook Dam. This recording was made at Blue Gum Lake, dawn February, 2003. Water temperature 23°C.

'Cork on Glass' calls were present in all recordings at Blue Gum Lake (Table. 3.5) indicating they were widely distributed throughout the wetland and dominated the sonic activity within this wetland. At Glen Brook Dam, 'Cork on Glass' calls were only heard faintly in the recordings and only dis continuous chorusing was evident. The midnight recordings revealed that sonic activity was widespread throughout the wetland. At dawn, the distribution of calls did not appear to reveal any site preference. However by midday and dusk, 'Cork on Glass' calls were present in all the southern recordings and to a lesser extent the eastern sites were also favoured (Table. 3.6).

Table.3.5. Spatial and temporal calling patterns of the 'Cork on Glass' calls at Blue Gum Lake, summer 2003 ('+' the call is present and ' – ' the call is absent).

Blue Gum Lake

Week	**Dawn**				**Midday**				**Dusk**				**Midnight**			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
3	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
1	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

Table.3.6. Spatial and temporal calling patterns of the 'Cork on Glass' calls at Glen Brook Dam, summer 2003 ('+' the call is present and ' – ' the call is absent).

Glen Brook Dam

Week				
4	- + - +	- + - +	- + - +	+ + - +
3	- - - -	- + - -	- + - +	+ + + +
2	- - - -	- + - +	- + - -	+ + + +
1	+ - + +	+ + + -	- + - +	+ + + -
	N S W E	N S W E	N S W E	N S W E
	Dawn	**Midday**	**Dusk**	**Midnight**

The 'Ratchet' call

The sound of the call in Figure 3.3 was likened to the sound of a 'ratchet'. The call spans a broad spectrum from around 1.6 kHz extending up to around 7 kHz with a dominant frequency of around 3 kHz with a lesser peak at around 4.2 kHz.

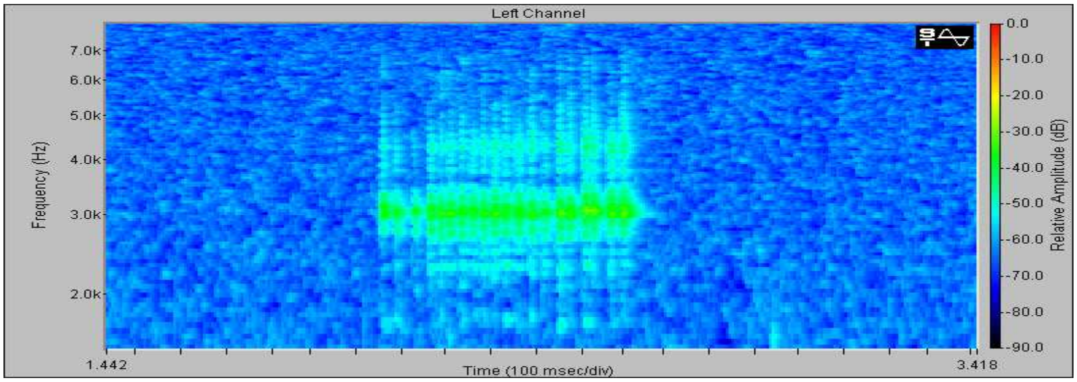


Figure.3.3. Spectrogram of the 'Ratchet' call only heard at Blue Gum Lake, in particular at midnight. The recording was made on the western aspect of the waterbody at midnight, February, 2003. Water temperature 29°C.

The 'ratchet' organism appears to be more active at midnight, occurring in the highest number of recordings in this period. In particular, the 'ratchet' call was heard at all the northern and southern sites (Table. 3.7), but was only heard in two dusk and a single dawn and midday recording.

Table.3.7. Spatial and temporal calling patterns of the 'Ratchet' call at Blue Gum Lake, summer 2003 ('+' the call is present and ' – ' the call is absent).

	Blue Gum Lake															
	Dawn				**Midday**				**Dusk**				**Midnight**			
Week	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	-	-	-	-	+	-	-	-	-	-	-	-	+	+	-	-
3	-	-	-	-	-	-	-	-	-	-	-	-	+	+	-	+
2	-	-	-	-	-	-	-	-	-	-	-	-	+	+	-	+
1	-	-	-	+	-	-	-	-	+	-	+	-	+	+	+	-

'Birdlike Song'

The call represented in Figure 3.4 was particularly interesting, because of its complex structure - unlike the percussive sounds which have dominated the recordings made in these freshwater environments. This call had a 'birdlike' quality to it and at the time of recordings, there was no corresponding aerial bird song.

Components of the call were likened to 'mouse-like squeaks', or heard as a single 'squeaky kiss' (Figure. 3.5), with other components sounding similar to the 'fluttering of bird wings' (Figure. 3.6). This call was not heard at Blue Gum Lake. Spectral components in this call ranged from around 500 Hz to nearly 10 kHz with the dominant frequency around 1.8 kHz to 2 kHz.

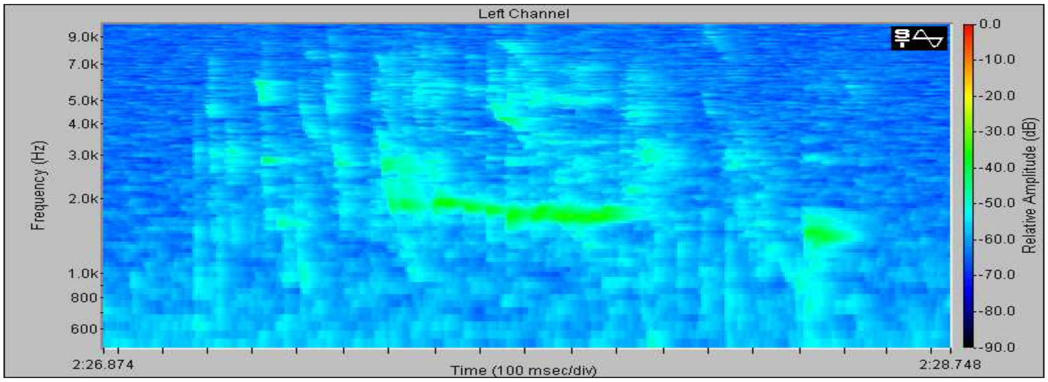


Figure.3.4. This spectrogram reveals the most complex of the calls recorded at the three wetlands. This was the longest section of the 'bird-like song' call recorded at midnight east, Lake Leschenaultia. The responsible organism is unknown. Water temperature 28°C.

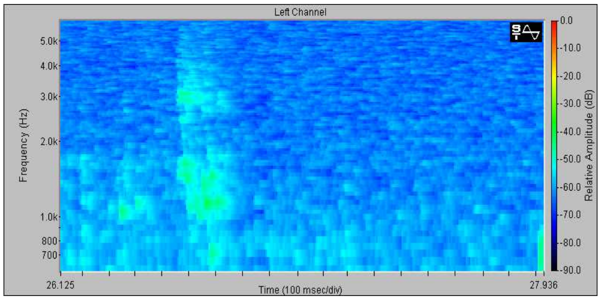


Figure.3.5. A single 'squeaky kiss' heard occasionally.

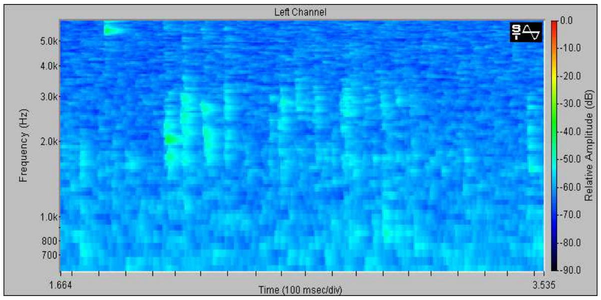


Figure.3.6. The 'Birdlike fluttering ' component of this signal sometimes heard on its own.

At Lake Leschenaultia, this organism was only an intermittent caller throughout the time periods (Table. 3.8). However, the longest and most spectacular call was made at this wetland in a midnight recording at the eastern site. This organism was a more frequent caller at Glen Brook Dam, with the northern site being consistently favoured at midnight (Table. 3.9).

Table.3.8. Spatial and temporal calling patterns of the 'Bird-like song' at Lake Leschenaultia, summer 2003 ('+' the call is present and ' – ' the call is absent).

Lake Leschenaultia

Week	**Dawn**				**Midday**				**Dusk**				**Midnight**			
4	+	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-
3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-
2	-	-	-	+	-	-	-	-	-	+	-	-	+	-	-	+
1	-	-	-	+	+	-	-	-	-	-	-	-	-	-	-	+
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E

Table.3.9. Spatial and temporal calling patterns of the 'Bird-like song' call at Glen Brook Dam, summer 2003 ('+' the call is present and ' – ' the call is absent).

Glen Brook Dam

Week	Dawn				Midday				Dusk				Midnight			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	+
3	-	-	-	-	-	-	+	-	+	-	-	+	+	-	-	-
2	-	-	+	-	-	-	-	-	-	-	+	-	+	-	-	-
1	-	-	-	-	+	-	-	+	-	-	+	-	+	-	-	+

3.3.2 Chorusing

Chorusing (continuous or dis continuous) and persistent non-chorus calls (For definition see Ch.2 General Methods; section 2.3.6) occurred either separately or at the same time. Examples of chorusing were selected to reveal chorusing at its maximum level and these are shown in the spectrograms as well as the spectrums (Figures 3.13 - 3.15). The spectra reveal that most of the energy lies from around 5 kHz to14 kHz.

The 'Cork on Glass' Chorus

Spectral components of the 'Cork on Glass' calls in chorus dominated the frequency band from around 4 kHz up to 10 kHz (Figure. 3.7). Continuous chorusing was only heard at Blue Gum Lake and occurred at most north, south and east recordings at dusk (Table. 3.10), but was mostly heard as a dis continuous chorus at the western site at dusk. By midnight, most of the calls were in a dis continuous chorus (Figure. 3.8) / or isolated calls, occurring mostly in the northern site with isolated calls in the west. Dis continuous chorusing could be heard at Glen Brook Dam, in particular at the southern sites at midnight (Table. 3.11), but received levels were at a low signal to noise ratio. Midnight appeared to be the period of greatest sonic activity for the 'Cork on Glass' callers at Glen Brook Dam, unlike that heard at Blue Gum Lake, which occurred at dusk.

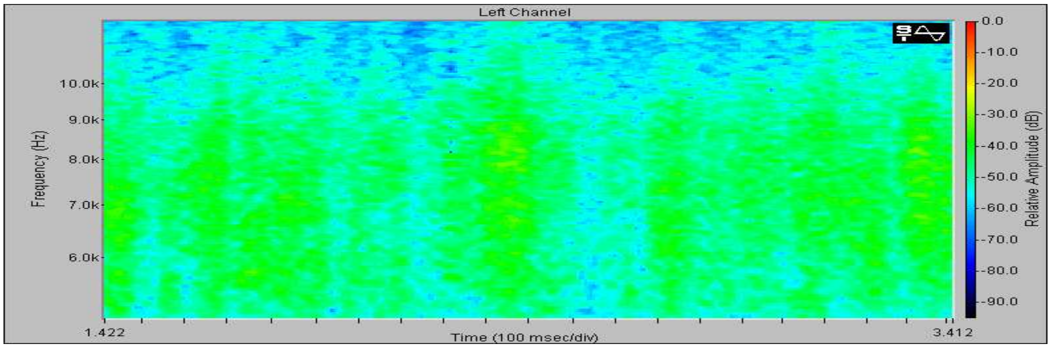


Figure.3.7. A continuous 'Cork on Glass' chorus heard at dusk on the northern aspect of Blue Gum Lake in February, 2003. Individual calls overlap and are indistinguishable. Water temperature 25ºC.

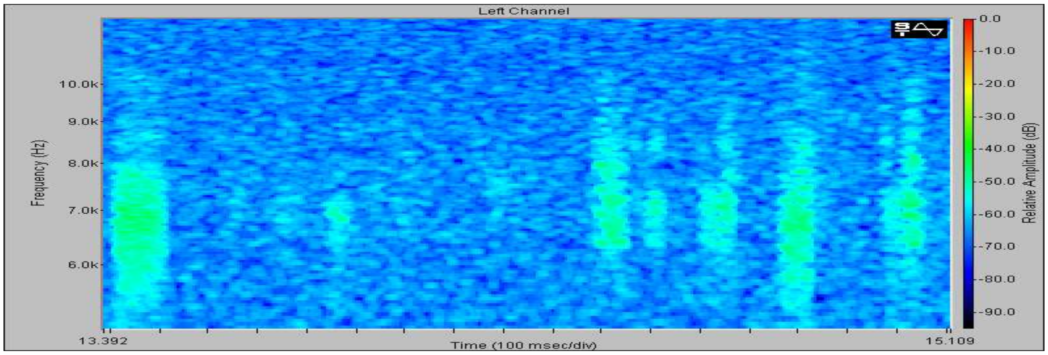


Figure.3.8. A dis continuous chorus of 'Cork on Glass' calls heard at midday at the northern aspect of Blue Gum Lake in February, 2003. Individual calls are distinct and do not overlap. Water temperature 25ºC.

Table.3.10. Spatial and temporal calling patterns of the 'Cork on Glass' chorus at Blue Gum Lake, summer 2003; based on a presence / absence of chorusing as either continuous chorusing (C), or as dis continuous chorusing (d). Isolated calls are shown as ' I '.

Blue Gum Lake

Week	**Dawn**				**Midday**				**Dusk**				**Midnight**			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	d	I	I	d	d	I	I	I	C	C	I	C	C	C	I	C
3	I	I	I	I	d	I	d	I	C	C	d	d	d	C	I	C
2	I	I	I	I	I	I	I	I	d	C	d	C	d	d	I	d
1	I	I	I	I	I	I	I	I	C	C	d	C	d	C	d	C

Table.3.11. Spatial and temporal calling patterns of the 'Cork on Glass' dis continuous chorusing at Glen Brook Dam, summer 2003.

Glen Brook Dam

Week	**Dawn**				**Midday**				**Dusk**				**Midnight**			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	-	I	-	I	-	I	-	d	-	I	-	I	I	I	-	d
3	-	-	-	-	-	d	-	-	-	I	-	I	I	d	I	I
2	-	-	-	-	-	I	-	I	-	d	-	-	I	d	I	d
1	I	-	I	I	I	d	I	-	-	I	-	I	I	d	I	-

The '12 / 6 kHz Rattle'

The call in Figure 3.9 was heard as a 'high-pitched-rattle'. Spectral components reveal two distinct bandwidths being utilised, with a dominant frequency of around 12 kHz with a lesser peak at 6 kHz - thought to be either a sub-harmonic or more likely, the opposite stroke as the organism stridulated. The '12 / 6 kHz rattle' occurred at a single midday recording but was mostly heard at midnight; in particular it was heard at all sites in the third week of recordings (Table. 3.12). It was thought to be produced by a single organism, and has been classified as a 'persistent non-chorus' call as it called continuously for more than 35 seconds and was sometimes heard for periods of up to 2 minutes with a short break of 1 second, before continuing. The amplitude of this call was sufficient to raise time-averaged ambient noise levels producing persistent spectral peaks at midnight (Figure. 3.15)

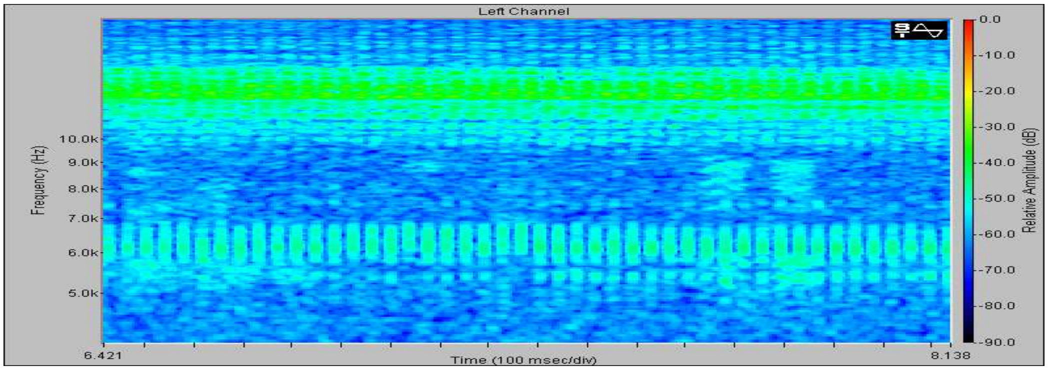


Figure.3.9. The '12 /6 kHz rattle' at Blue Gum Lake recorded here in a midnight west recording. The 6 kHz pulse repetition always occurred with the 12 kHz rattle and was thought to be the 'opposite stroke' as the organism stridulated. 'Cork on Glass' callers were occasionally heard calling between these two band-widths. Water temperature 27.5°C.

Table.3.12. Spatial and temporal calling patterns of the '12 /6 kHz rattle' at Blue Gum Lake, summer 2003 ('+' the call is present and ' – ' the call is absent).

Blue Gum Lake

Week	Dawn				Midday				Dusk				Midnight			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
3	-	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+
2	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-
1	-	-	-	-	-	-	-	+	-	-	-	-	-	-	+	-

The ‘5.5 kHz Rattle’

The ‘rattle’ of Figure 3.10 has also been classified as a ‘persistent non-chorus’ sound; thought to be produced by a single organism calling for extended periods so that at times was seen as a near continuous line on the spectrograms. The ‘5.5 kHz rattle’ was a frequent caller at midday and appeared in the second week in 3 dusk recordings, but this caller was never heard at midnight. Spectral components ranged from around 5 kHz to around 6.8 kHz and at midday - appeared to favour the southern and eastern sites (Table. 3.13). It was not heard at the western site and was present in only one north recording.

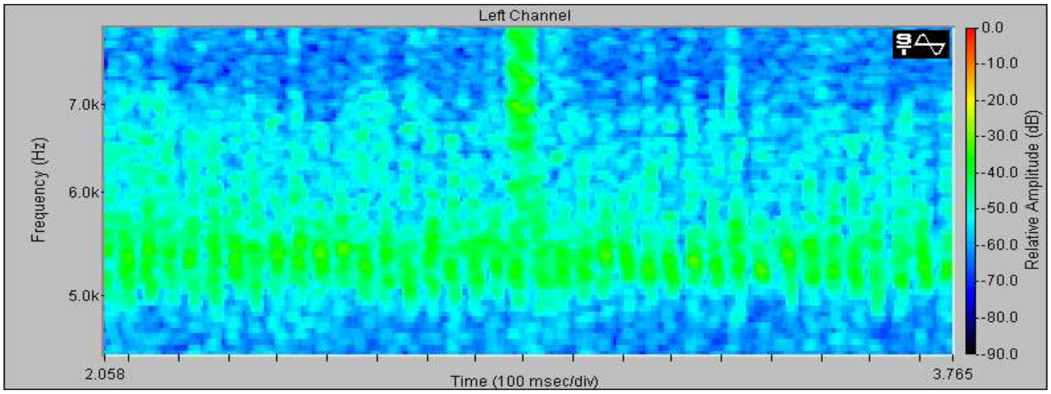


Figure.3.10. Spectrogram of the ‘5.5 kHz Rattle’ only heard at Blue Gum Lake and was a frequent caller at midday. Call shown in the center was from a ‘Cork on Glass’ caller. Recording taken at midday. Water temperature 29°C.

Table.3.13. Spatial and temporal calling patterns of the ‘5.5 kHz rattle’ at Blue Gum Lake, summer 2003 (‘+’ the call is present and ‘ – ’ the call is absent).

Blue Gum Lake

Week																
4	-	-	-	-	-	+	-	+	-	-	-	-	-	-	-	-
3	-	-	-	-	-	+	-	+	-	-	-	+	-	-	-	-
2	-	-	-	-	+	+	-	+	+	+	-	+	-	-	-	-
1	-	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
	Dawn				**Midday**				**Dusk**				**Midnight**			

The 'Tinsel caller'

The call of Figure 3.11 sounded very similar to the sound produced when shaking a piece of Christmas tinsel. Mostly the spectral components ranged from around 7 kHz to 9 kHz. This organism was a frequent caller at midday and favoured the eastern site in particular (Table. 3.14). This call was recorded in only two midnight recordings and could be heard between intermittent pulses of the 'Cork on Glass' chorus.

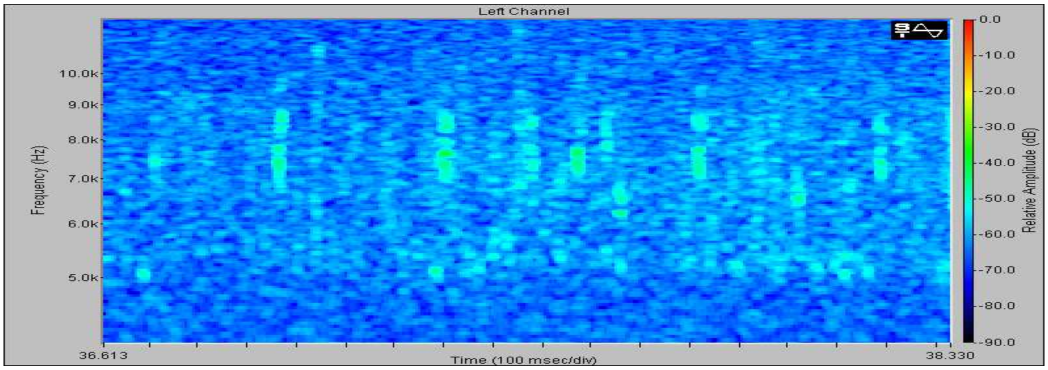


Figure.3.11. Spectrogram of the 'Tinsel caller' at Blue Gum Lake. Recording taken at midday. Water temperature 28°C.

Table.3.14. Spatial and temporal calling patterns of the 'Tinsel caller' at Blue Gum Lake, summer 2003 ('+' the call is present and '–' the call is absent).

Blue Gum Lake

Week	Dawn				Midday				Dusk				Midnight			
	N	S	W	E	N	S	W	E	N	S	W	E	N	S	W	E
4	-	-	-	-	-	+	-	+	-	-	-	-	-	-	-	-
3	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-
2	+	-	-	-	+	-	-	+	-	+	-	-	-	-	-	-
1	-	-	-	-	-	+	-	+	-	-	-	-	-	+	-	+

3.3.3 Spectrum Levels

The Biologics

Examples of both continuous and dis continuous chorusing were evident at Blue Gum Lake, as well as 'persistent non-chorus' contributions. Glen Brook Dam had evidence of only dis continuous chorusing but at a low signal to noise ratio and is therefore not shown here. The outputs depicted in Figures 3.13, 3.14, and 3.15 reveal noise spectrum levels when each call type (chorus and/or persistent non-chorus calls) were evident in the time periods: midday, dusk and midnight. Each call is revealed at its maximum and is superimposed on the ambient background noise (Beaufort Wind Scale 0-2). Midday recordings revealed a distinct peak between 5 kHz – 6 kHz (in green)(Figure. 3.13), produced by the '5.5 kHz rattle' and a lesser peak (in red) from the 'Tinsel caller', which is around 12 dB below the '5.5 kHz rattle'. At dusk, continuous 'Cork on Glass' chorusing dominated this time period producing a dominant peak at 6 kHz - 8 kHz, which was slightly higher – around 8 dB above the maximum spectra at midday and around 3 dB above the midnight maxima (Figure. 3.14). Midnight recordings revealed two dominant spectral peaks: one at a frequency of around 6.3 kHz and the other peak around 12.5 kHz (Figure. 3.15) - produced by the '12 /6 kHz rattle' (blue line). This organism occasionally called with the dis continuous 'Cork on Glass' chorus, revealed as a lesser peak between the 12 kHz and 6 kHz peaks (lime green line). The 'Cork on Glass' calls had largely subsided into dis continuous chorusing by midnight, with noise levels dropping from around 72 dB at dusk to around 62 dB by midnight (broken pink line).

Background Ambient Noise

A wetland surface state corresponding to wind speed criteria was established (Table 3.15) for Beaufort Wind Scale 0,1,2,3. Speeds above the Beaufort Wind Scale of 3 produced extraneous noise at some sites and therefore this wind speed could not be assessed using the onshore method of recording. It was difficult to record at Blue Gum Lake in the complete absence of biologics and a small peak between 6-8 kHz is evident in the averaged ambient noise spectra (Figures. 3.12-3.15).

Table.3.15. Relation between wind speed and wetland surface state (adapted from Wenz, 1962, p.1937)

Surface Criteria	**Breeze**	**Beaufort Wind Scale**	**Wind Speed (km/hr)**
Mirror-like	none	0	<1.8
Ripples	light	1	1.8 - 6.12
Basket-weave pattern	medium	2	6.12 - 11.88
Wavelets	strong	3	11.88- 19.44

It was found at Beaufort Wind Scales of 0, 1 and 2 there was no difference overall to ambient noise in either Blue Gum Lake or Glen Brook Dam (Appendix.1.). Due to the buffering effects of stands of reeds and rushes near shore at Lake Leschenaultia which resulted in a surface state that was mirror-like or rippled in-shore while the center of the lake experienced greater wind speeds; Beaufort Wind Scale of 2 or above could not be assessed at Lake Leschenaultia from shore. Therefore, only wind data for Blue Gum Lake and Glen Brook Dam are presented (Figure. 3.12). Wind speed data for Blue Gum Lake was then averaged to give a single curve for ambient noise (Figures 3.13-3.15)

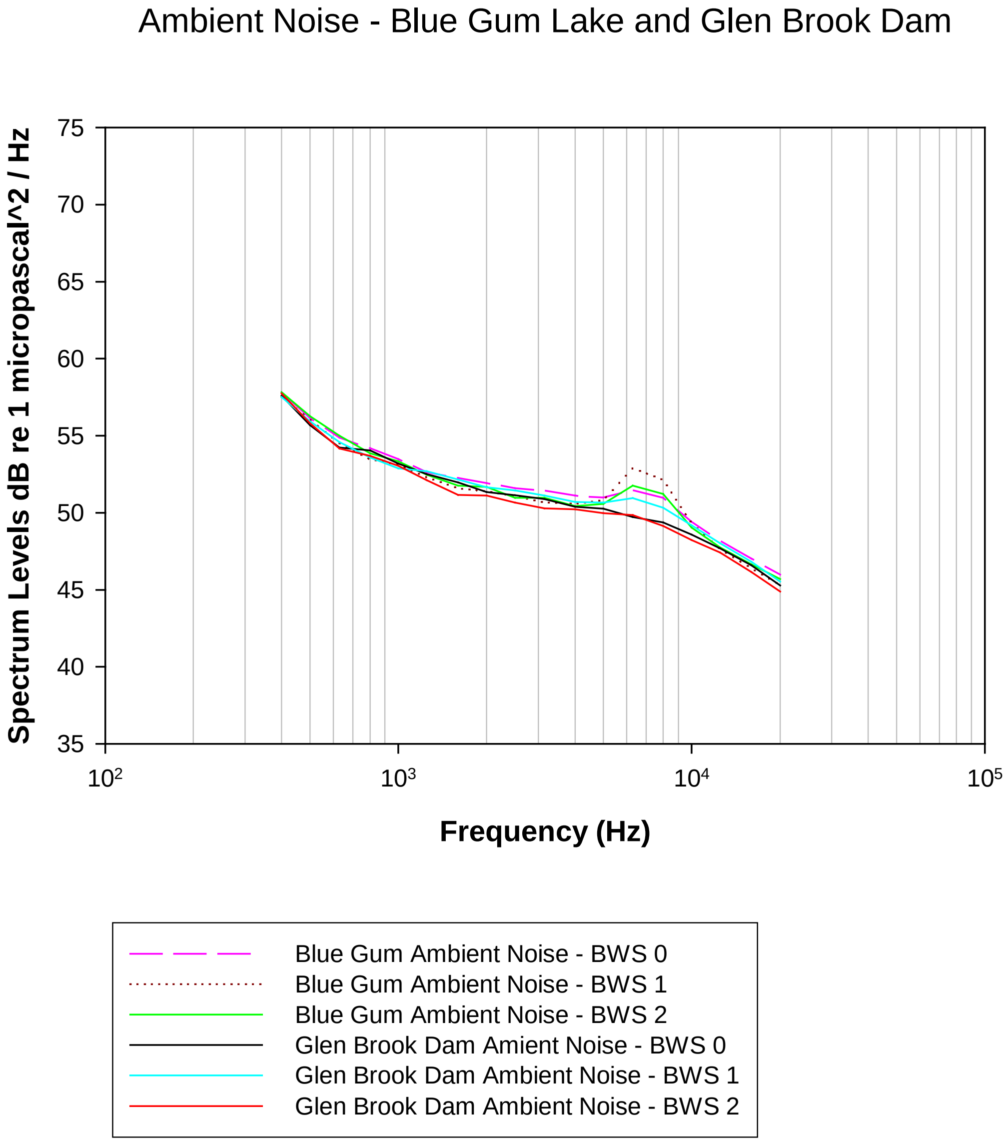


Figure.3.12. Spectra of winter ambient noise levels at wind speeds of Beaufort scale 0, 1 and 2 revealing very little difference between each wind speed. The small peak at 6-8 kHz is from the ‘Cork on Glass’ callers, which call throughout all time periods and all seasons. Blue Gum Lake and Glen Brook Dam, winter 2003.

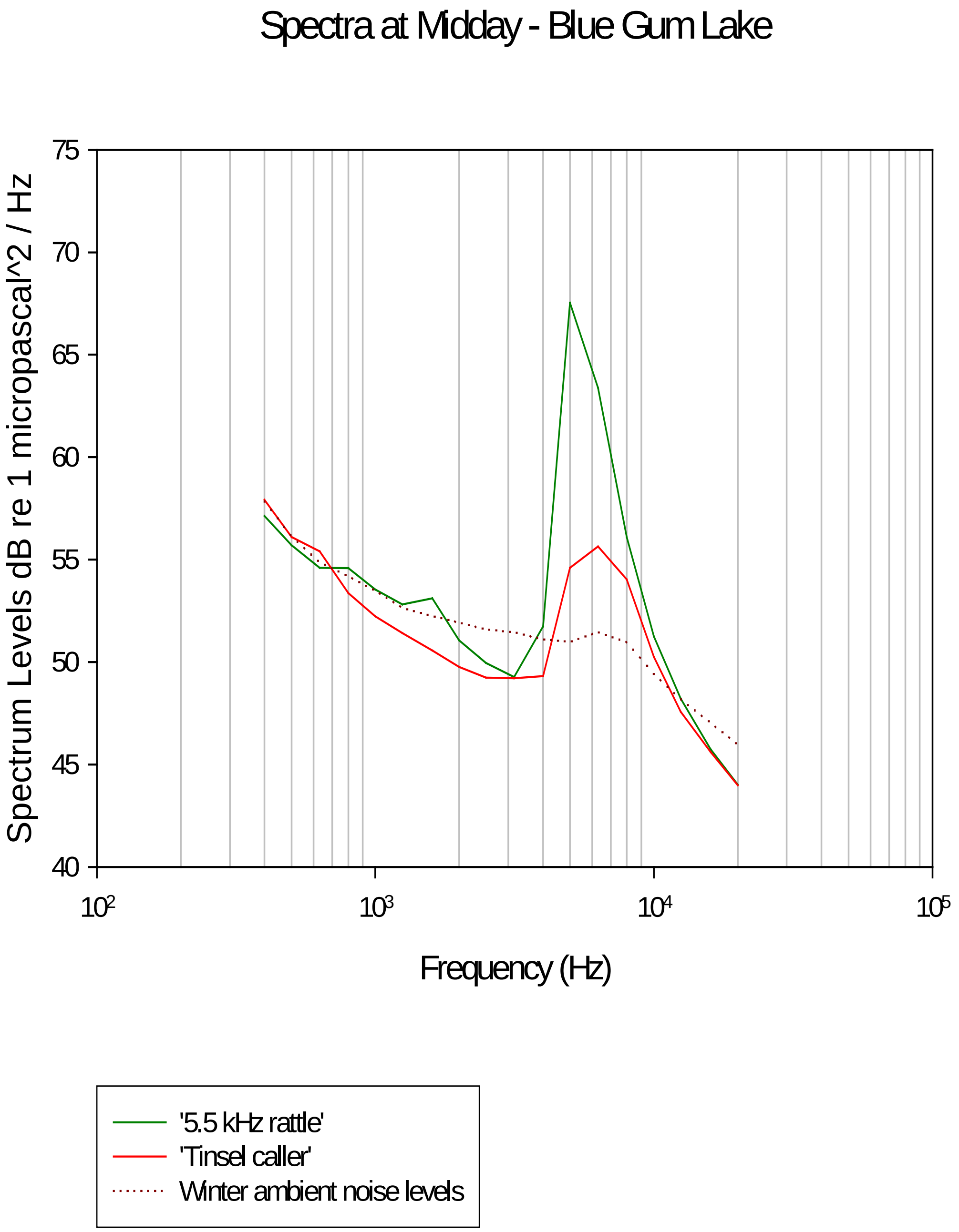


Figure.3.13. Spectra of dominant biologic activity at midday from ⅓-octave band measurements. The dominant peak at 5kHz – 6kHz was produced by the '5.5 kHz rattle' with the lower peak being produced by the 'Tinsel' caller. Blue Gum Lake summer 2003.

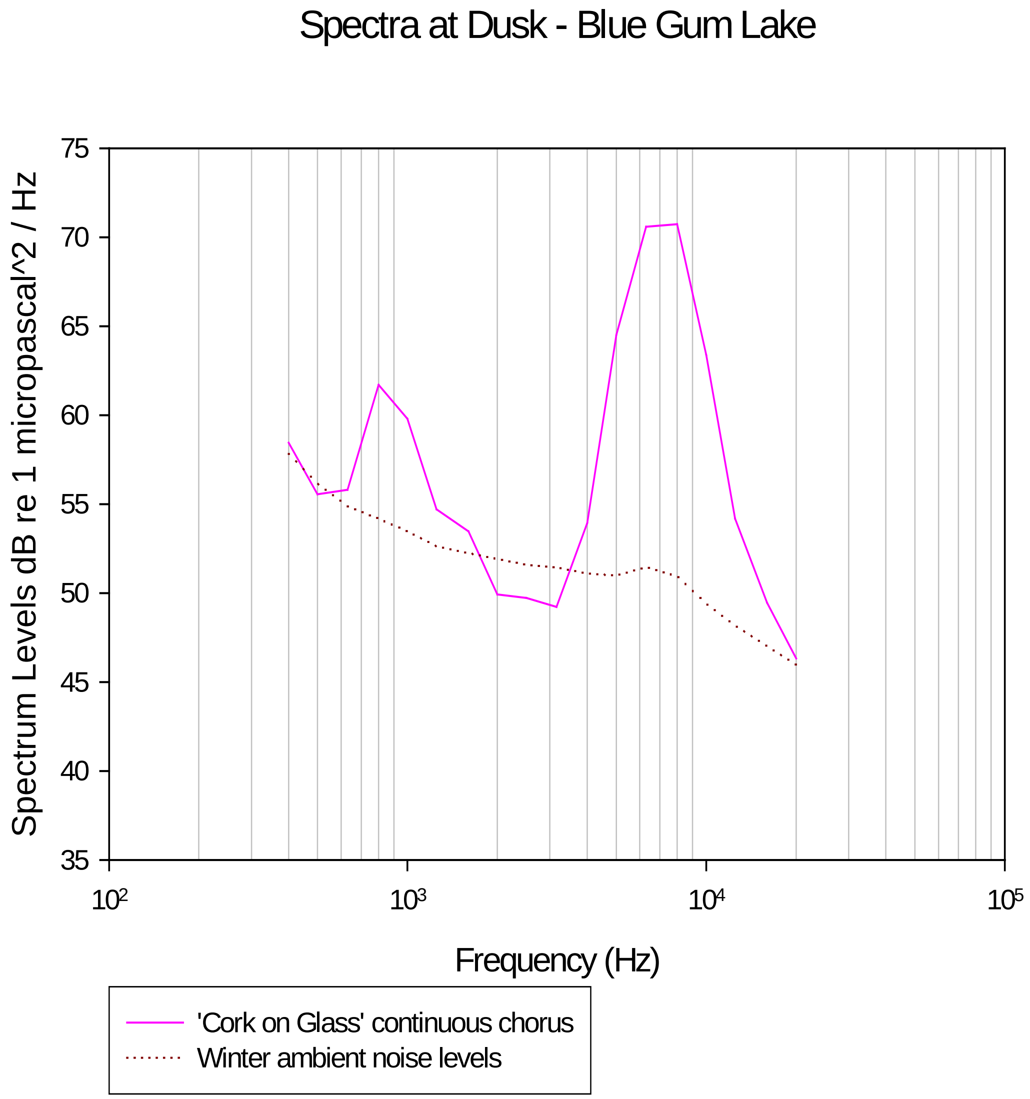


Figure.3.14. Spectra of dominant biologic activity at dusk from ⅓-octave band measurements. The dominant peak at 5kHz – 10kHz are from the 'Cork on Glass' callers in continuous chorus. The peak at 800 Hz was produced by gas bubbles escaping from the sediments. Blue Gum Lake summer 2003.

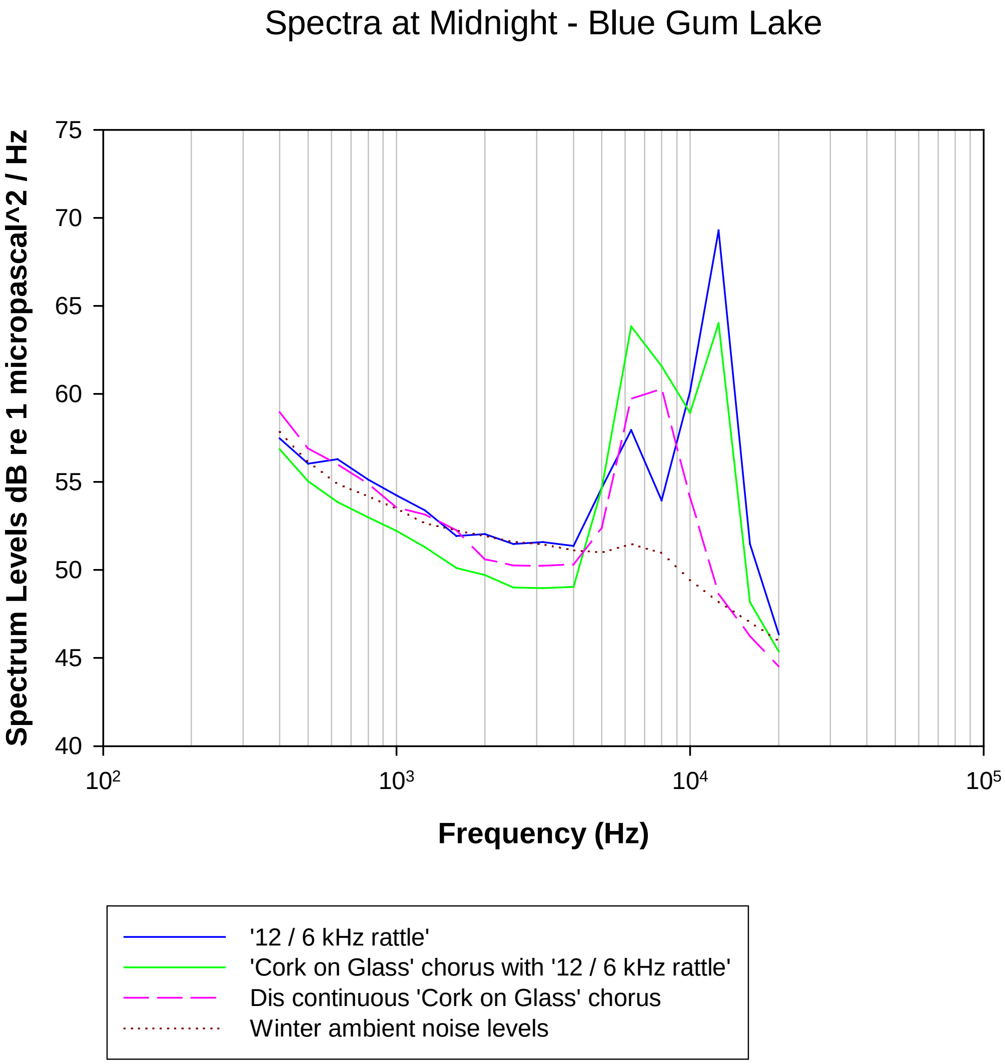


Figure.3.15. Spectra of the dominant biologic activity at midnight from ⅓-octave band measurements. The persistent non-chorus contributions are shown as separate spectra as well as the dis continuous chorusing, which was more prevalent at midnight. Blue Gum Lake summer 2003.

3.3.4 Anthropogenic Noise Contributions in Wetlands

In total, there were four sources of anthropogenic noise - the helicopter and the speed-boat (recorded at Lake Leschenaultia), and the water aerator and heavy haulage vehicles (recorded in urban wetlands at a later date).

Helicopter

Noise produced by a helicopter directly overhead at Lake Leschenaultia revealed a dominant frequency band of around 500 Hz to 2 kHz (Figure. 3.16). The CRT 350 Hz hydrophone filter was being used at the time and consequently, if there were any low frequency contributions from the helicopter, they were not revealed in this output.

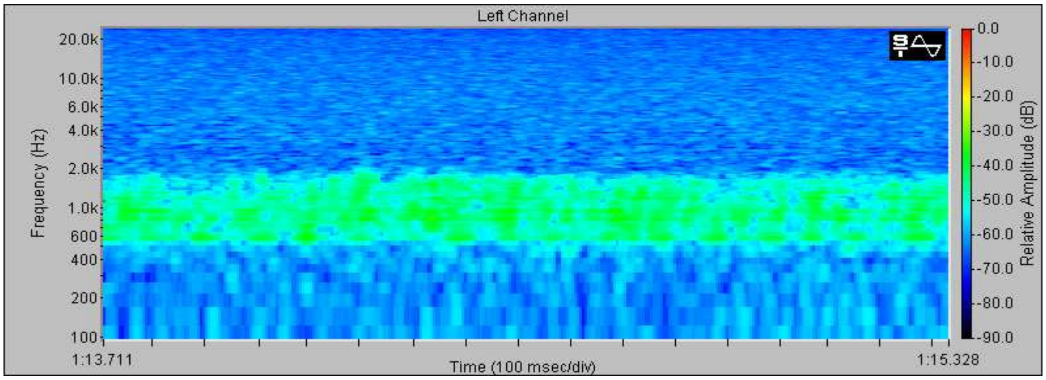


Figure.3.16. The spectral output revealing noise produced by a helicopter directly overhead at Lake Leschenaultia during the summer, 2003. The 350 Hz Hydrophone filter was being used at the time of this recording.

Speed Boats

Use of a dinghy with an outboard motor (25 Hp) is occasionally used at Lake Leschenaultia for rescue activities. The spectral output of Figure 3.17 reveals a broad frequency bandwidth from around 4 kHz up to around 20 kHz with a dominant frequency at around 11 kHz. The spectrogram reveals the boat at its closest approach and was approximately 10m from shore (speed unknown).

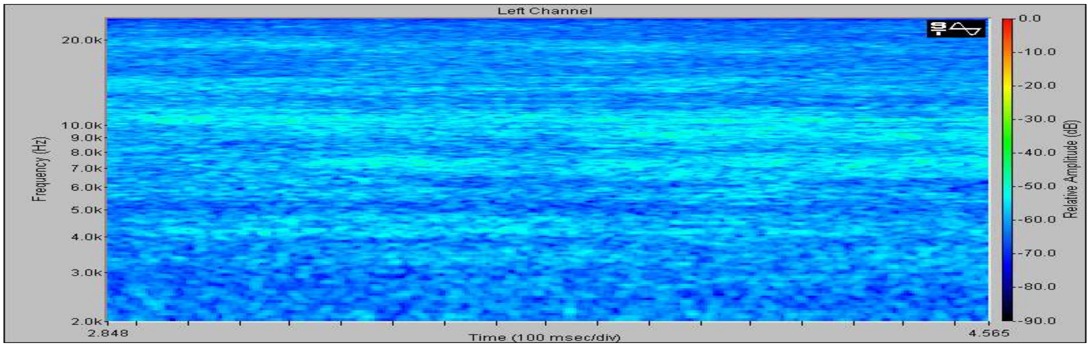


Figure.3.17. Spectrogram of the noise produced by a passing outboard motor boat at its closest approach at Lake Leschenaultia. Summer, 2003. Water temperature 28°C.

Water Fountains (aerators)

The underwater sound field at Mabel Talbot Lake, Subiaco when a single aerator was operating (approximately 20m from the hydrophone); revealed a band of noise from 2 kHz to nearly 20 kHz (Figure 3.18). If any invertebrates were calling within this lake, they would be completely masked by this noise.

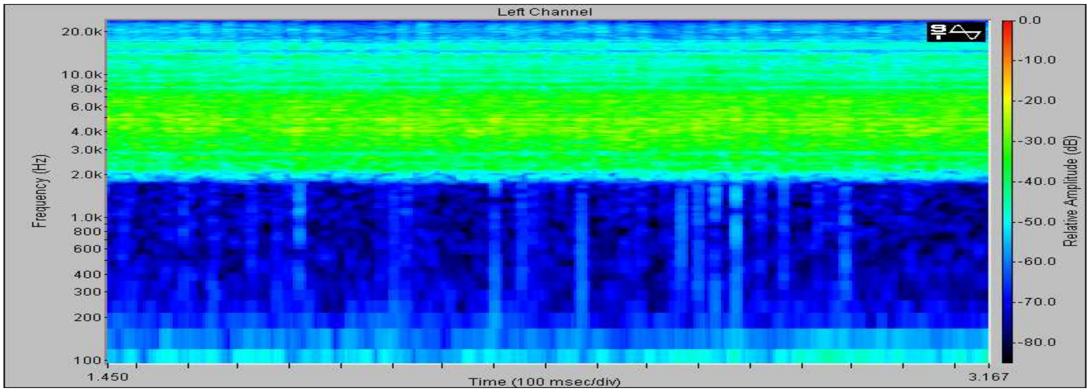


Figure.3.18. Spectrogram of the ambient sound field in Mabel Talbot Lake, Subiaco whilst a water aerator was operating. Recorded in winter, 2005. Water temperature 15°C.

Heavy Vehicles

Heavy vehicle noise was distinctive in the recordings, particularly at low frequencies from around 100 Hz to just over 200 Hz (Figure 3.19). This recording was made at Piney Lake - located approximately 200m from the Leach Highway, which is a major arterial highway used extensively by heavy haulage vehicles. Low frequency noise was also evident from light vehicles such as cars (<150 Hz) which were heard at Blue Gum Lake, however this wetland lies in close proximity to the road (<20m).

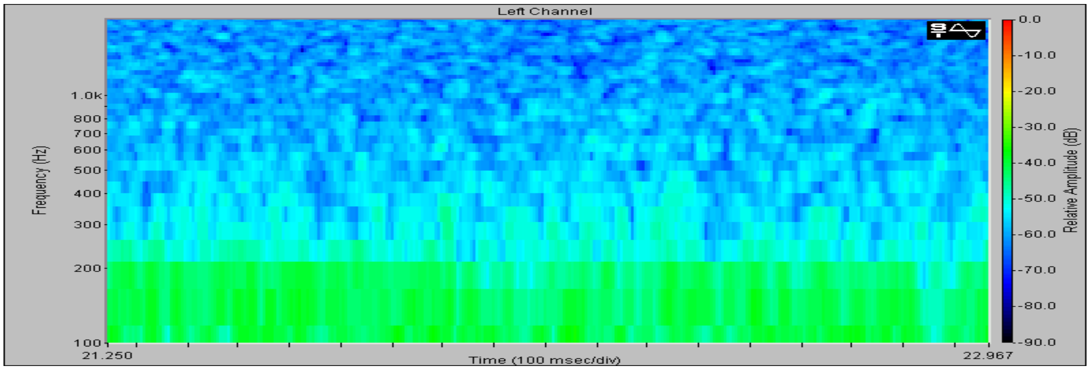


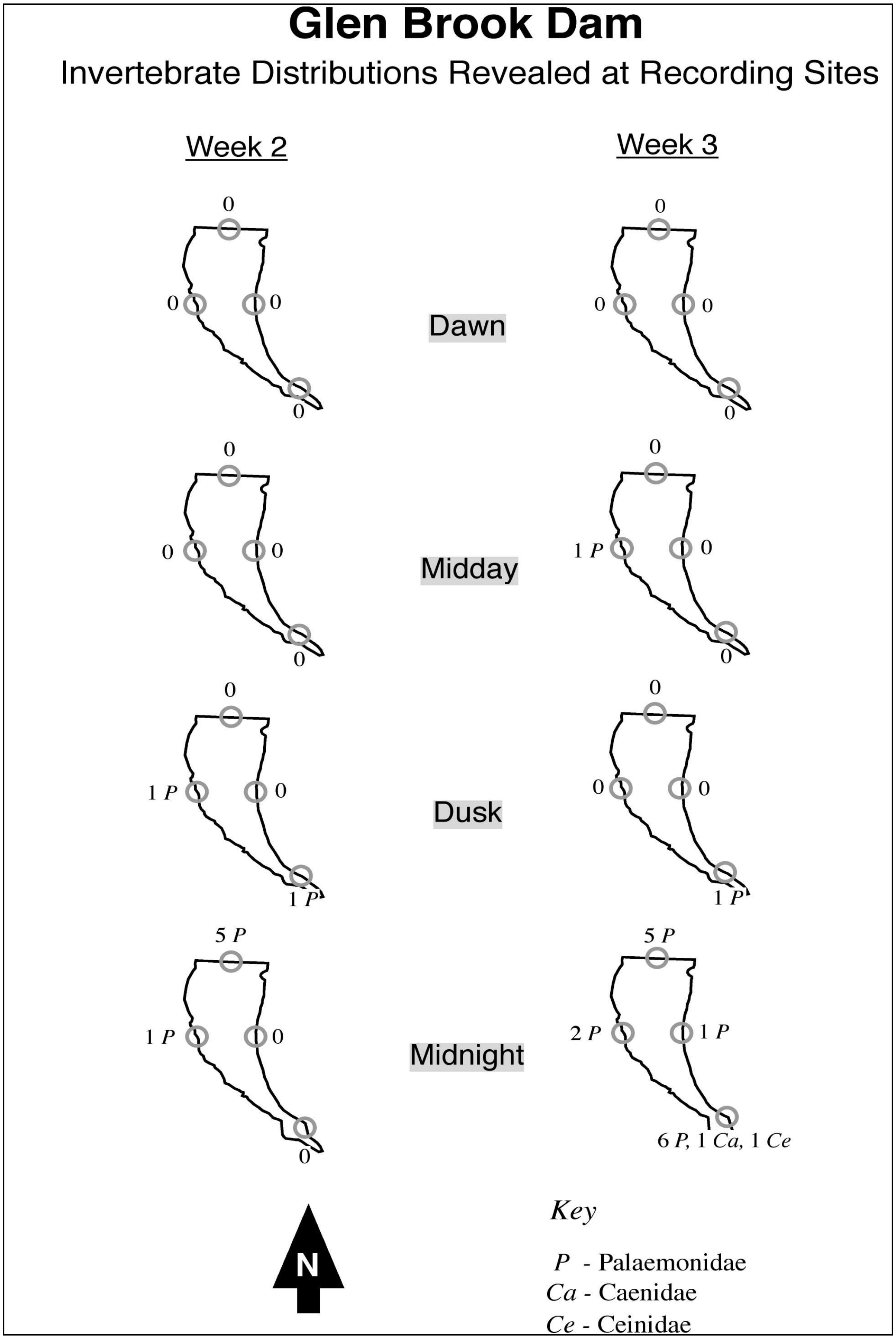
Figure.3.19. Spectrogram of low frequency noise produced by a heavy haulage vehicle passing around 200m from Piney Lake along the Leach Highway. Recorded on 4th July, 2003 at 9.45pm. Water temperature 13ºC.

3.3.5 Invertebrates

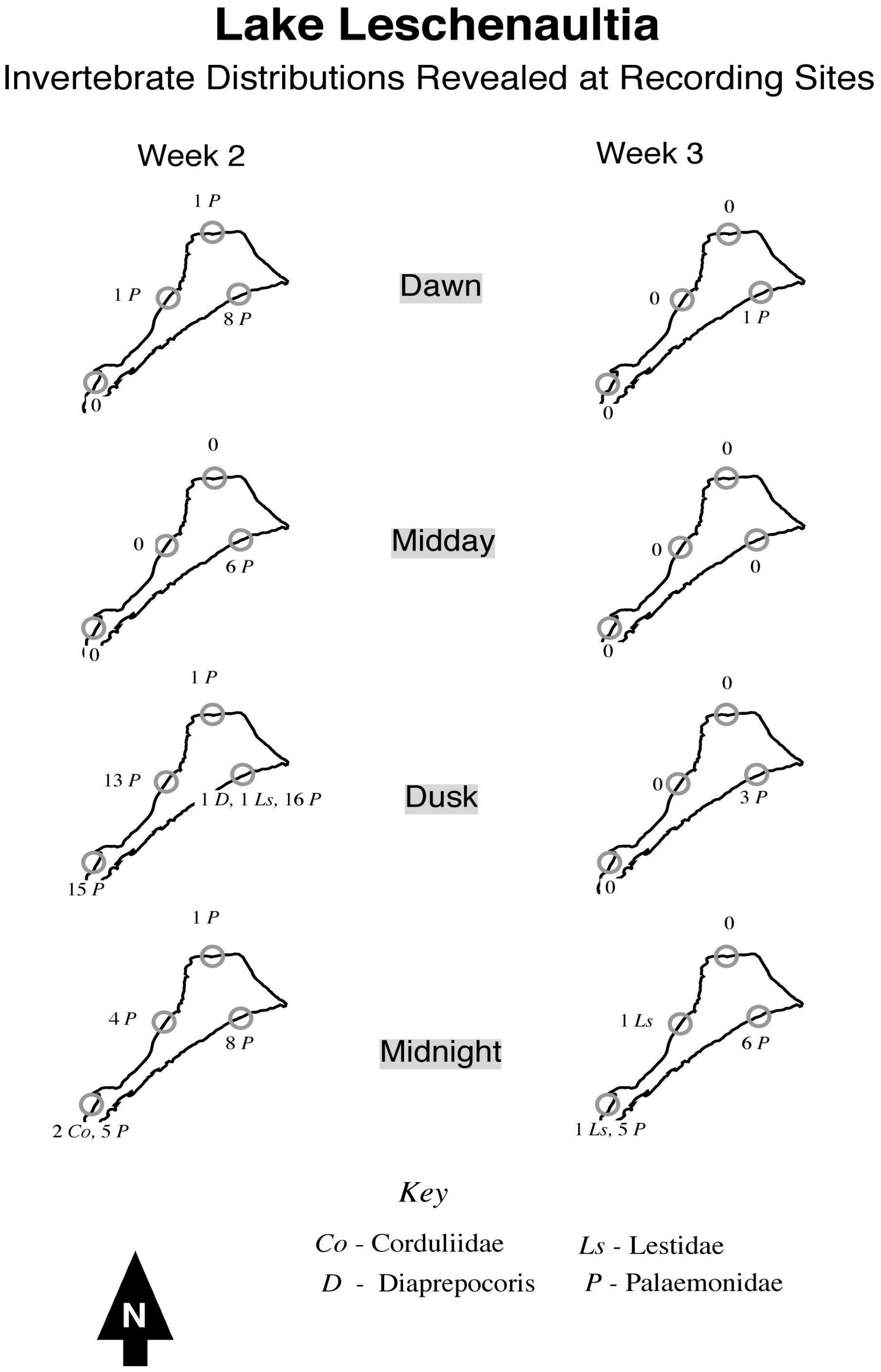
Invertebrates at the three wetlands were separated into: microinvertebrates (identified to Class or Order) and macroinvertebrates (identified to Genus) (Table. 3.16). Midnight at Lake Leschenaultia and Glen Brook Dam was revealed as the time period with the greatest number of invertebrate families present (10 and 8 respectively), of which three were macroinvertebrates. At Lake Leschenaultia, the macroinvertebrate families present were: Shrimps (Palaemonidae), Dragonflies (Corduliidae) and Damselflies (Lestidae) and at Glen Brook Dam: the Shrimps (Palemonidae), Mayflies (Caenidae) and Amphipods (Ceinidae) families. The Shrimps dominated both dusk and midnight at these wetlands with very few organisms present at dawn and midday unlike the number and diversity of macroinvertebrates present at Blue Gum Lake in these two time periods (Maps. 3.1, 3.2 & 3.3). For Blue Gum though, it was dusk that had the greater number of invertebrate families represented (22), of which six were macroinvertebrates. At midnight, 19 families were present, of which seven were macroinvertebrates. The Amphipods, Mayflies and to a lesser extent Water Boatmen (Corixidae), dominated dusk and midnight samples (Map. 3.3).

Table.3.16. List of invertebrates found at Lake Leschenaultia, Glen Brook Dam and Blue Gum Lake.

	Lake Leschenaultia	*Glen Brook Dam*	*Blue Gum Lake*
Microinvertebrates:			
Copepoda	√	√	√
Ostracoda	√	x	√
Collembola	√	√	√
Hydracarina	√	x	√
Hirudinea	√	x	√
Gastropoda	√	√	√
Cladocera	√	√	√
Diptera	√	√	√
Oligochaeta	x	x	√
Macroinvertebrates:			
Lestidae (genus:*Austrolestes*)	√	x	x
Palaemonidae (genus:*Palaemonetes*)	√	√	x
Caenidae (genus: *Tasmanocoenis*)	x	√	√
Corduliidae (genus: *Hemicordulia*)	√	x	x
Corixidae (genus: *Diaprepocoris*)	√	x	x
Corixidae (genus: *Agraptocorixa*)	x	x	√
Corixidae (genus: *Micronecta*)	x	x	√
Ceinidae (genus:*Austrochiltonia*)	x	√	√
Leptoceridae (genus: *Oecetis*)	x	x	√
Notonectidae (genus: *Anisops*)	x	x	√
Ecnomidae (genus: *Ecnomus*)	x	x	√
Vertebrate:			
Gambusia	√	√	x

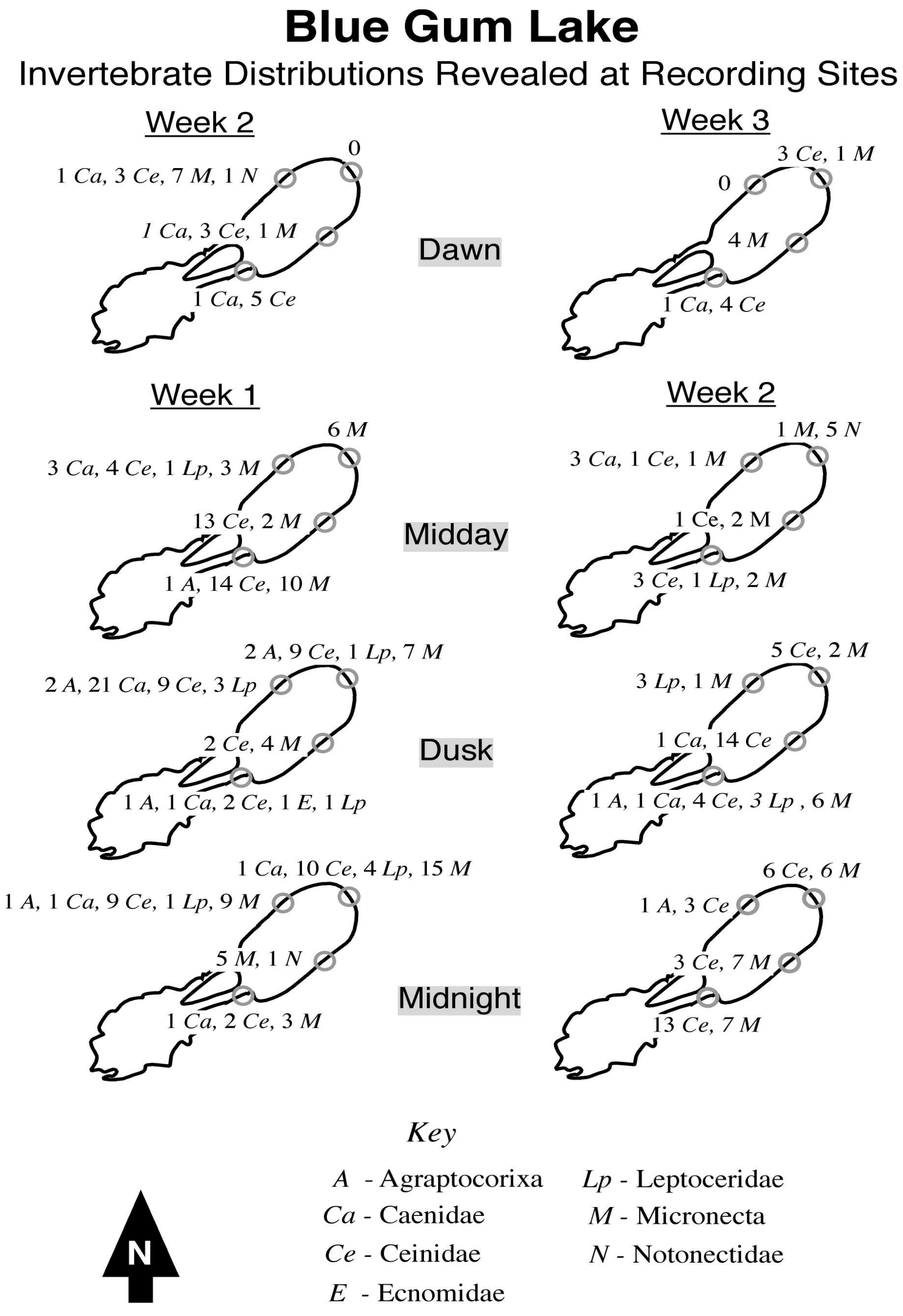


Map.3.1. Actual number of macroinvertebrates found from sweeps (equivalent to sampling 1 m^3 of water) taken at recording sites in weeks 2 & 3. More organisms were found in both the midnight samples. Note the comparative prevalence of the Shrimp (Palaemonidae) and the lack of organisms found at dawn and midday within Glen Brook Dam.
(Graphics by Steven Goynich, Murdoch University)



Map.3.2. Actual number of macroinvertebrates found from sweeps (equivalent to sampling 1 m^3 of water) taken at recording sites in weeks 2 & 3. More organisms were found in both the dusk and midnight samples compared to the dawn and midday samples. Note the prevalence of Shrimp (Palaemonidae) at Lake Leschenaultia also.

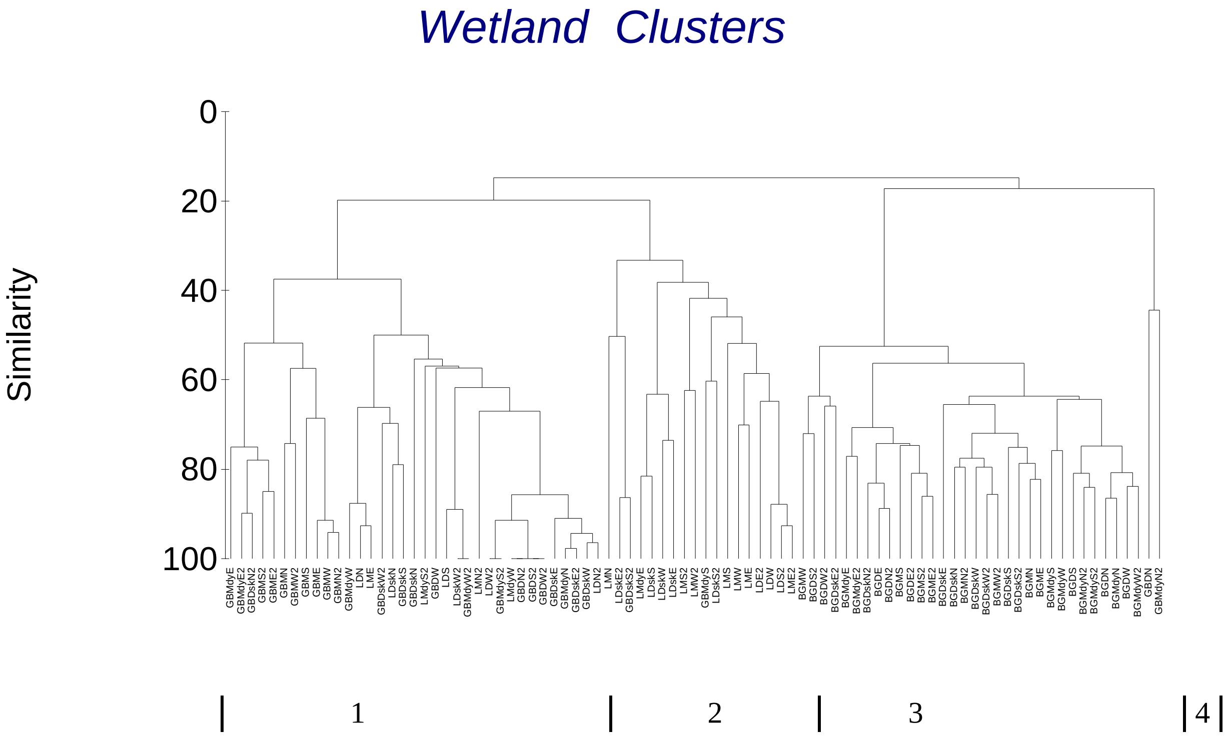
(Graphics by Steven Goynich, Murdoch University)



Map.3.3. Actual number of macroinvertebrates found from sweeps (equivalent to sampling 1 m^3 of water) taken at recording sites in weeks 2 & 3 at dawn and weeks 1 & 2 at midday, dusk and midnight. More organisms were found from midday through to midnight compared to dawn. Overall, abundance and diversity was very different to that found at Lake Leschenaultia and Glen Brook Dam and also the prevalence of organisms at midday, which was not seen at the less disturbed sites. (Graphics by Steven Goynich, Murdoch University)

Initially the entire data set was analysed using the Bray-Curtis similarity clustering and was transformed by 4th root transformation (Figure. 3.20). Those sites where no organisms were found have been deleted from the data set. At about 15% similarity, two broad groups were present. Blue Gum Lake separated out from Lake Leschenaultia and Glen Brook Dam; which appeared as a mixed grouping – except for Glen Brook dawn north and midday north (week 2), which separated as a group on its own at about 20% similarity due to the abundance of Water Fleas (Daphniidae). Blue Gum Lake separated out as a distinct group on its own due to the abundance of Water Fleas, Waterboatmen (*Micronecta*) and Copepoda. The main species responsible for the division of the first cluster were the Copepoda and Water Fleas and for the second cluster, the Shrimp were responsible for this division and to a lesser extent, Mosquito's and Midge Larvae (Dipterans).

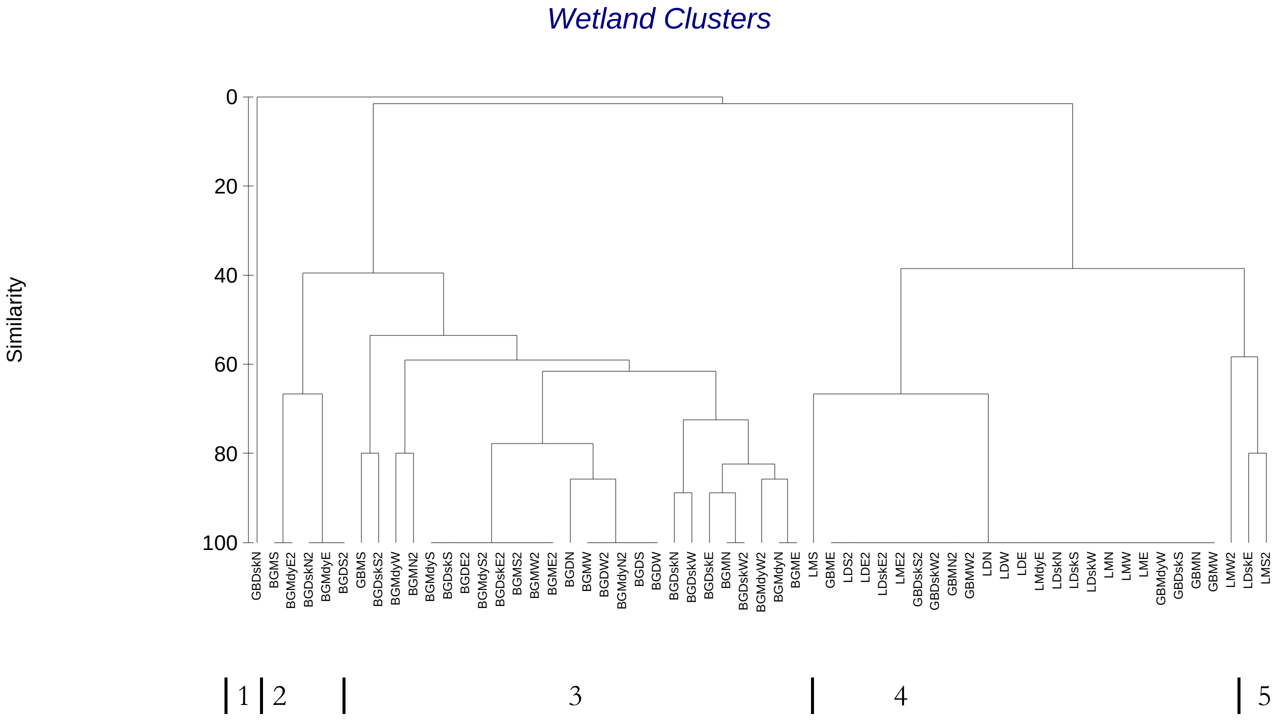
The average dissimilarity values for groups 1 & 3, 2 & 3, 4 & 3 were very high (90.3, 96.3, & 93.3) which highlights the distinct difference between Blue Gum Lake and the other two wetlands. This was largely driven by the high abundance of the Water Fleas and the obvious absence of organisms between sites such as the Shrimp only being found at Lake Leschenaultia and Glen Brook Dam but not at Blue Gum Lake, or the Waterboatmen (*Micronecta*) only found at Blue Gum and not at the other two sites.



Key to groupings: Wetland initials (egBG= Blue Gum, L=Leschenautlia, GB=Glen Brook), time of day (egD=Dawn, Dsk= Dusk, M=midnight, Mdy=Midday), site (eg N= north, S=South, W=West, E=East), the number 2 designates the 2nd week of sampling.

Figure. 3.20. Dendrogram displaying the wetland clustering based on macro and micro-invertebrate abundances. Data was transformed by the Bray-Curtis similarity using the 4th root.

It was believed unlikely that microinvertebrates were contributing to the audible sound field in a wetland, so microinvertebrates were separated from the macroinvertebrates. Sites where no macroinvertebrates were found were deleted from the analyses. Based on a presence / absence transformation at the 2% similarity – again cluster analysis (Figure. 3.21) largely separated the three wetlands into two distinct groups: 1.) Blue Gum Lake and 2.) A mixed grouping of Glen Brook Dam and Lake Leschenaultia – however a single site at Glen Brook Dam (MS midnight south) did appear with the Blue Gum Lake group and dusk north at Glen Brook Dam appeared as a distinct group on its own due to only a single macroinvertebrate organism found at this site. At around 40% similarity, cluster analysis revealed five distinct groups (shown at the bottom of Figure. 3.21). The main organisms responsible for each separation were: for group 1 - the presence of only a single organism; for group 2 - the Waterboatmen (*Micronecta*) & Back Swimmers (Notonectidae); for group 3 – Waterboatmen (*Micronecta* & *Agraptocorixa*) and Amphipods; for group 4 – Fairy Shrimp; and for group 5 - the Dragonflies and Shrimps. The average dissimilarity values for groups 2 & 3 (mostly Blue Gum Lake sites) against all other groups were extremely high ranging from 98.3 to 100 % which again highlights the distinct difference between Blue Gum Lake and the other two wetlands which was confirmed in the sound recordings.



Key to groupings: Wetland initials (egBG= Blue Gum, L=Leschenautlia, GB=Glen Brook), time of day (egD=Dawn, Dsk= Dusk, M=midnight, Mddy=Midday), site (eg N= north, S=South, W=West, E=East), the number 2 designates the 2nd week of sampling.

Figure. 3.21. Dendrogram displaying the wetland clustering based on macro-invertebrates only at the wetlands. Data was transformed by the Bray-Curtis similarity using the presence/absence transformation.

3.4 Discussion

There is an extensive body of literature describing the acoustic field of the marine environment, but by contrast there is very little published data on the acoustic field in a freshwater environment. To provide the freshwater data within a contextual frame, comparisons have been drawn in this thesis with the marine environment and interestingly, similarities were found between the two environments.

In a marine environment, ambient noise varies both temporally and spatially (Cummings *et al.,* 1964; Urick, 1983) as well as seasonally (McCauley, 2001). Similarly, spatial and temporal trends were also evident in this study. Despite the brevity of the sampling time and the distance between each recording site which may have introduced some bias, there did appear to be variability between north, south, east and west recordings at all three wetlands, as well as the time of day that the recordings were made; including some weekly variation. From the invertebrate samples taken, variability in presence/absence and distributions of aquatic organisms was also confirmed.

3.4.1 Natural Contribution to Noise

Wind

Unlike the ocean, these wetlands have some buffering to the influences of wind as they occur in natural depressions and are often surrounded by terrestrial vegetation and, in some cases urban dwellings. Therefore, wind in these wetlands do not reach the wind speeds that the ocean would be subjected to and consequently only the low wind speeds at Beaufort Wind Scale of 0,1,2 & 3 were the most relevant speeds. The contribution to ambient noise in a wetland as a result of wind action on the waters surface was negligible at low wind speeds of Beaufort Wind Scale 0,1 & 2 and it appears that water depths from 1-8m did not influence the levels attained at these wind speeds either. Dietz *et al.,* (1960) also found no correlation between wind speed and sound pressure levels in shallow waters (7 fathoms or 12.8m) when wind speeds were lower than 9.76 km/hr, but variations have been found in the noise spectra at these wind speeds in a slightly deeper freshwater system at 15m (see Hawkins and Myrberg, 1983). The averaged ambient noise spectra (from the average of Beaufort Wind Scales 0,1 & 2) revealed that higher noise levels were present in a wetland compared to spectra produced for some northern hemisphere 'shallow' marine environments (see Wenz, 1962). While the definition of a 'shallow marine environment' (< 100 fathoms or <183m) is obviously different to an inland wetland and would be considered deep; this was used as a basis for some comparison. Using Wenz (1962) Beaufort Wind Scale of 2 – Blue Gum Lake and Glen Brook Dam ambient noise levels were very similar to some shallow marine locations. Similar spectrum levels were also found with those of ambient noise in tropical seas surrounding Australia (see Cato, 1976). At much lower frequencies still (between 11-45 Hz), investigations by Lomask and Saenger (1960) found at zero sea-state ambient noise was quieter in a deep lake (750 ft or 228m) compared to the marine environment. Low frequency noise in the marine environment has been attributed to shipping traffic (Wenz, 1962; Cato, 1976). The frequency response of equipment used here was not able to reveal noise levels at the low frequencies discussed in Lomask and Saenger (1960) and therefore no comparisons could be made at similar frequencies.

An increase in noise was readily apparent when wind speeds reached a Beaufort Wind Scale of 3 or greater. This was due to mechanical noise as a result of the recording set-up. In the marine environment, the hydrophone and cables are subjected to forces from currents and sea-surface movement, which produces self-noise in the measurement system (Wenz, 1971). While these lentic wetlands have no turbulence from tidal currents to contend with, noise from flow and water surface movement was evident as wind speed increased. When using the hydrophone suspended between the two star-pickets, the cloth used for suspending the hydrophone vibrated at higher wind speeds causing 'humming' and wavelets slapped on the star pickets. While the hydrophone would detect sounds best when suspended vertically in the water column, the issue of self-noise meant that this method of recording was not used in later recordings. Instead, the hydrophone was allowed to lie coupled with the bottom sediments. Any attenuation that might occur from lying with the bottom sediments was considered acceptable in this study. However, Other issues with noise as a result of wind action also came from: movement of the 'surf' on shore moving the hydrophone cable, with this 'surf' often carrying debris into the shore which would rub against the hydrophone cable; waves slapping on nearby structures such as the height datum poles; the wooden jetty at Lake Leschenaultia; protruding dead wood at Blue Gum; and also granite boulders in Glen Brook Dam. Also, due to the shallow nature of Blue Gum Lake - wind speed at Beaufort Wind Scale of 3 or greater was probably sufficient to move the water column and resulted in movement and aggravation of the bottom-mounted hydrophone. Therefore, future recordings needed to be undertaken on calm days or alternatively, in the case of movement of the hydrophone on the bottom sediments by a moving water column; the hydrophone would need to be fixed rigidly to the bottom.

Bubbles

Release of gas from disturbed bottom sediments was an intermittent source of noise. When wading out into the waterbody to place the hydrophone on the star picket mountings, many gas bubbles were released from the sediment - more than would normally be released at any one time. Bubbles contributed significant 'clutter' to the ambient noise recordings over a broad frequency range and in some cases, masked the biological sounds. As a consequence, the hydrophone was deployed from the shore for the remainder of this research which meant there was less noise contribution into future recordings from bubbles and also this procedure minimised disturbance in the wetland allowing relatively undisturbed acoustic behaviour to then be recorded (see Watkins and Daher, 1992).

Biological

It appears there is a relationship between urbanisation and the diversity of invertebrate species present in wetlands, in particular, the detrital food chain becomes dominant in urban wetlands (Chambers and Davis, 1988). Greater diversity and abundances were revealed in the invertebrate samples from Blue Gum Lake (the urban wetland) compared to samples from the least disturbed sites - Lake Leschenaultia and Glen Brook Dam, with wetland clustering supporting the differences between these wetlands. In support of the differences in diversity and abundances, there were variations heard in the recordings. A paucity of calls (two types) were recorded at Lake Leschenaultia and Glen Brook Dam which were different to calls recorded at Blue Gum Lake which had a greater diversity of calls (five types) as well as the presence of chorus activity which was not heard at the two least disturbed sites. From the maps of macroinvertebrate presence, there appears to be temporal and spatial variations in their distributions including some variation between the two weeks, although consistent trends of dusk and midnight were the periods of greatest diversity and abundance of macroinvertebrates. The diversity and abundance at dusk and midnight was synonymous with the increased sonic activity in these two time periods.

In these clear freshwater environments, noise levels were greatest at dusk in particular and to a lesser extent at midnight due to choruses. Similar trends in chorus activity have been recorded in a marine environment (e.g. Fish, 1964; Clapp, 1964; McCauley *et al.,* 1996), although unlike the freshwater recordings, dawn chorus are also evident at some marine locations (Cato, 1978). Continuous contributions to ambient noise levels with the highest amplitude are often produced by invertebrates and also fish (Fish, 1964). However, the bandwidth of greatest output in these shallow freshwater environments were at higher frequencies than that found in the marine environment. In the freshwater environment, the greatest output was found to be from 6 kHz to around 14 kHz, while in recordings made in the Timor Sea, East Indian and the West Pacific Ocean, Cato (1978) found the bandwidth of greatest output was from around 400 Hz to 4 kHz, with Clapp (1964) reporting most energy between 100 Hz to 1000 Hz in coastal waters off San Diego. Chorus spectrum levels between dusk and midnight in the Timor Sea were around 10 dB higher than the greatest output revealed in the freshwater dusk 'Cork on Glass' chorus and around 12 dB above the greatest output at midnight from the '12/6 kHz rattle', but comparable spectrum levels were seen in the West Pacific and East Indian Ocean spectra.

Generally, there is a paucity of research on freshwater invertebrate organisms and their calling behaviour. For those that have been studied (e.g. Jansson, 1973a & b; King, 1999a & b), particularly from the family Corixidae; Jansson (1973b) found diel periodicity of stridulation with periods of maximum activity occurring at different times of the day for different species. He believed light was the controlling factor in determining periodicity. In this study, the 'Cork on Glass' callers were found to call in all time periods, but dominated the dusk and midnight recordings in summer by their chorusing activity - particularly at the disturbed wetland – Blue Gum Lake. Chorusing activity, has been described in only one other Australian freshwater invertebrate, *Micronecta concordia,* which was distinctive in itself as it was synchronised (King, 1999b). Interestingly, King (1999b) noted that he could move his microphone (microphone encased in a rubber membrane) to locate different spots where sound production was higher, indicating that males possibly aggregate. This movement and aggregation may explain why chorusing was not recorded at all sites or at the same site each week.

Cato (1978, p.737) specifically defined a chorus as: *1.The rise and fall of noise as a function of time in a frequency band containing the spectral peak. 2.The presence of a characteristic peak in the spectrum. 3.The characteristic sound.* The dusk 'Cork on Glass' chorus was an obvious chorus event producing well-defined spectral peaks and dominated the recordings at Blue Gum Lake. However, defining the calls for the '12/6 kHz rattle', '5.5 kHz rattle' and the 'Tinsel caller' was not so obvious, as these calls appeared to be produced by a single organism. It was due to the energy in the call and their persistence in a recording that these callers were included in the chorusing section.

McCauley (2001) considered sounds produced by marine fish to be associated with breeding activities or possibly used in communication and he noted that their calling activity was suggestive of its importance in their daily behaviour. The only freshwater fish encountered in this research were the Mosquito fish (*Gambusia holbrooki*), and occur in high densities; but no calls could be associated with them. Unlike the marine environment; inland freshwater environments don't have the intermittent contributions such as those produced by the marine mammals (see Cato & McCauley, 2002), but it was found from the latter part of this research (see chapters 4 & 5); that intermittent sounds did come from freshwater turtles and that some of the over-looked intermittent sounds in the ambient sound recordings were actually produced by turtles.

The 'Bird-like' call of Figure 3.4 was a distinctive call due to its complexity and considered to have been produced by a vertebrate animal. One suggestion is that it could have been produced by a waterbird. As many of the waterbirds spend a large proportion of their time submerged e.g. Australasian Grebe (*Tachybaptus novaehollandiae*) and the Musk Duck (*Biziura lobata*), it would be reasonable to consider that they may make sounds underwater. Musk Ducks have been observed producing sounds, which include thumps, splashes and booms (Mr Alan Hill, the Park Ranger from Lake Leschenaultia, personal conversation). However, during the day, waterbirds could be heard producing intermittent sounds from dunking, diving and landing on the water but no real relationship between a call and a bird underwater could be made. Other vertebrates utilising this wetland include a number of frog species, the Mosquito fish (*Gambusia holbrooki*) and also the Western minnow (*Galaxias occidentalis*) and the turtle, *C. oblonga* – although, the 'Bird-like call' was not heard in the repertoire of the freshwater turtle.

Marron and Gilgies were observed at Glen Brook Dam and known to exist in Lake Leschenaultia (Hill, pers. comm., 2003), however, is was considered unlikely for Marron to still be present at Blue Gum Lake as Marron do not occur in degraded wetlands (Smith *et al,* 1997). While no bioacoustical analyses have been undertaken on the Marron or Gilgies; it is highly likely that they may be biological contributors to ambient noise in wetlands. This is due to the morphology of Crustacea generally, where many have the stridulatory apparatus consisting of the pars stridens-plectrum type (Dumortier, 1963), or even the 'stick and slip' mechanism, which has been described for the Spiny Lobster (*Panulirus argus*) (Patek, 2001). There were a number of sounds heard in the recordings that weren't identifiable calls but may have been produced by these larger crustaceans.

3.4.2 Anthropogenic Contribution to Noise

Wetlands on the Swan Coastal Plain and those further inland, are isolated from industrial and shipping noise contributions that shallow and deep water marine environments are subjected to (e.g. Epifanio *et al,* 1999; Finneran *et al*, 2000; Thompson and Richardson, 1995; Potter and Delory, 1998) and to a lesser extent, experienced in southern oceans (Cato, 1976). In addition, inland waters are isolated from sounds such as the damaging high intensity sounds experienced in the marine seismic petroleum industry (McCauley *et al.,* 2003) and from other sources such as marine geophysical surveys (Greene and Moore, 1995). However, wetlands were not without noise contributions from anthropogenic sources. Some opportunistic anthropogenic sounds encountered in this research included: helicopters; road vehicles; motor-boats; and water aerators. Most of these sounds were intermittent, although water aerators were generally used continuously in block periods of time (i.e all day or all night).

Both Lake Leschenaultia and Glen Brook Dam lie beneath aircraft flight paths, but sounds produced by aircraft did not appear in any of the recordings, neither did a light aircraft making repeated sweeps above Quenda Wetland. This lack of appearance in the recordings may have been due to the altitude at which the craft were flying and the angle of incidence of sound at the waters surface (see Greene and Moore, 1995). More likely though, the low frequency response of the hydrophone (CRT C53) may have resulted in masking of low frequency noise contributions produced by these aircraft as later recordings using the HTI-96-MIN hydrophone did reveal noise in wetland recordings from overhead aircraft.

Noise produced by the helicopter used at Lake Leschenaultia for helicopter search and recovery practice was very distinctive and dominated the recordings. Overhead helicopter flights have elicited short-term responses from Bowhead whales (*Balaena mysticetus*) and also Beluga whales (*Delphinapterus leucas*), but it was unknown whether the response was from the sound or the sight of the aircraft (Patenaude *et al.,* 2002). The underwater recording taken at Lake Leschenaultia revealed the noise produced by the helicopter lay in a frequency band considered to be within the audible range of *C. oblonga.* In shallow water bodies, particularly where there are reflective bottom boundaries – lateral propagation is enhanced as multiple reflections effectively lengthens the time that sound is received underwater (see Greene and Moore, 1995) and the sudden appearance of a noise produced by a helicopter into an environment that would normally be relatively quiet at these frequencies, is something that needs to be investigated if this type of activity is to continue. Interestingly, in Desert tortoises (*Gopherus agassizii*), it was found that they lacked a startle response when subjected to a simulated overhead jet flight. Instead, the tortoises displayed a physiological response to this noise by freezing for up to 113 minutes (Bowles and Eckert, 1997). This type of response / behaviour has not been investigated in *C. oblonga,* but if a similar physiological response was elicited in *C. oblonga*, for an aquatic animal this would no doubt prove fatal. At a later date, a helicopter flying above the artificial ponds elicited no vocal response from the turtles, unfortunately behavioural responses could not be observed at the time.

The only water-craft encountered in this research was the occasional use of a small motor-boat at Lake Leschenaultia to assist those people having difficulties with their canoes. The noise contribution appeared to be brief as the motor-boat passed by and utilised a high frequency bandwidth. While the noise contributions from the helicopter and the speed-boat were intermittent, the urban contributions were more constant and lasted for longer periods. The low frequency 'rumbles' produced by heavy haulage vehicles were heard up to 200 m from the highway at Piney Lake. This might be expected to result in more or less permanent low frequency noise into wetlands that lie in close proximity to highways, particularly those utilised by heavy haulage vehicles.

The use of water aerators was another source of more or less consistent sound as they were generally utilised for long periods of time. The sound produced by aerators resulted in noise at frequency bandwidths utilised by invertebrates. This would result in persistent masking of their calls and if sound is important in invertebrate ecology, then masking at the frequencies of use could play a part in driving community composition or possibly, even collapse of entire invertebrate communities.

3.4.3 Influence of Temperature

The influence of temperature wasn't an aspect directly tested for in this research, but its potential importance is recognised. As invertebrates appeared to dominate the ambient sound recordings, the influence of temperature on these organisms will be important.

Temperature may be an important factor in driving the 'sound cycles' within a wetland - determining sound production in some invertebrate communities within an optimal range. Temperature has been shown to determine the onset of stridulation in both *M.*

Concordia (King, 1999b) and in cenocorixids (Jansson, 1974). In addition, Jansson (1974) found in cenocorixa that stridulation ceased when the water temperature reached 29ºC. The influence of temperature may be one reason why biological sounds were not heard at the western site at Blue Gum Lake towards the end of the recording regime. The maximum temperature reached at this site was 33°C. From the occasional recordings undertaken during the winter months, it appeared that very few organisms were calling and for the 'tick, tick' wave-train (the only recognisable call from the summer recordings), there was a longer inter-pulse-interval than the interval for the same wave-train called during the summer. Similarly, in calls produced by organisms from the family Corixidae; e.g. *Cenocorixa* (Jansson, 1974) and *Micronecta* (King, 1999a,b), variations in temperature resulted in changes in the pulse rate, pulse-train rate and signal duration, with no observable changes in the frequencies or temporal patterns of the sounds produced by these organisms.

The lack of calls generally in the winter months may be due to the inability of some organisms to stridulate below a certain temperature. King (1999b) found stridulation ceased when temperatures fell below 7.4°C for species within the genus *Micronecta*. Interestingly, when listening for turtle calls during the winter months and also in spring two years later at Blue Gum Lake there was a new suite of singers present (except for the 'Cork on Glass' callers), which may be reflecting some seasonal use of 'sound space' in a wetland and that these wetlands are biologically dynamic systems.

Chapter 4.0 THE ACOUSTIC REPERTOIRE OF *Chelodina oblonga*: Individual sounds

4.1 Introduction

Chelonians are not highly vocal animals and for many years were thought to be 'the silent group' of animals (Campbell and Evans, 1972). As such, there is a paucity of acoustical research on chelonians, with research mainly restricted to those in-air vocalisations produced by terrestrial chelonians predominantly during the breeding season (e.g. Bogert, 1960; Auffenberg, 1964; Jackson and Awbrey, 1978; McKeown *et al.,* 1990; Sacchi *et al.,* 2003 and Galeotti *et al.,* 2004). However there are reports of other sounds produced by chelonians outside breeding activities; such as those produced by freshwater turtles in defence or agonistic encounters (Goode, 1967; Kaufmann, 1992) and even some reports of in-air sounds produced by marine turtles when nesting (Mrosovsky, 1972) and when being hurt or killed (Carr, 1952). There are also unpublished observations of tortoise vocalisations made by amateur 'turtle and tortoise clubs' or by those who keep these animals as pets (e.g. 'Tortoise Calls' http://www.tortoise.org/tortcall.html.)

It was not known whether turtles produced underwater vocalisations. Kumpf (1964) reported hearing 'roars' in their acoustic-video recordings (located in 65 ft of water near Bimini, Florida) when two Green turtles (*Chelonia mydas*) were present, but as to whether the turtles produced the 'roars' remained unconfirmed. There are some recent unpublished observations by J. Little (pers. correspondence, 2004), who believed he heard 'low and high-pitched' sounds produced by juvenile and sub-adult

Green turtles. However, no published studies deal specifically with the underwater vocalisations of freshwater turtles so there is no precedent for this research.

4.4.1 Objectives

1. To identify and describe the elements in the vocal repertoire of *C. oblonga*.
2. To identify and describe differences in vocalisations between males, females and juveniles.

4.2 Methods and Materials

4.2.1 Experimental Procedures

Two recording regimes were used which consisted of both random and structured recordings: 1.) Recordings were undertaken in a natural setting at various wetlands and 2.) In an artificial environment.

Natural Wetlands

As there were no recognisable sounds emitted by turtles in the pilot studies, recordings resumed at a number of wetlands in winter. This season was chosen for several reasons: 1.) Winter / spring is known to be the breeding season for *C. oblonga* (Burbidge, 1967); 2.) Most of the previous research regarding tortoise vocalisations had occurred in the breeding season (e.g. Bogert, 1960; Campbell and Evans, 1967; Auffenberg, 1978; Crawford and Awbrey, 1978); and 3.) Rain sufficient to fill wetlands did not occur until the end of June / early July in 2003.

Recordings were undertaken at several urban wetlands. These included: Piney Lake - which was known to have a comparatively healthy population of turtles where successful breeding had occurred in previous years (Giles, 2001); Blue Gum Lake which contained a large population of turtles (estimate >2000 turtles) (Giles, 2001);

and Quenda Wetland, which was known to contain a small population of adult turtles (Giles, 2001) and where contributors to background noise such as that produced by birdlife, wind and heavy vehicle traffic, were minimal. Recordings occurred during the months of July, August, through to mid September, 2003 and occurred during the day as well as evenings, up to midnight, in blocks of two to five and half hours.

Artificial Environment

The second component of this research involved listening to turtles contained in artificial ponds. This had the advantage of ascribing calls definitively to turtles in the absence of other biologics - particularly calls by invertebrates (although these later colonised the tanks in small numbers - they were by then, easily recognised). Extraneous urban noises, bird-calls and falling twigs and seeds into the ponds were also occasionally heard in recordings, but as all recordings were made with a human listener, these sounds were easily accounted for. Initially, recordings of turtles were not undertaken in an artificial environment as animals are known to go silent in laboratory settings (McCauley, pers. comm., 2002) and there are difficulties and inaccuracies associated with recording in aquaria (Parvulescu, 1966; Hawkins and Myrberg, 1983; Yager, 1992). However, an opportunity arose to work with the entire population of turtles (42) from a small urban wetland; Masons Gardens in Nedlands, Perth; while the wetland was undergoing restoration. Turtles were sexed, measured, weighed and micro-chipped. There were 19 males of which 8 were sub-adults and 23 females of which 9 were sub-adults. Males (including sub-adults) ranged in size from 10.89cm - 19.47cm Carapace Length (CL), mean=13.60cm, SD=2.03 and females (including sub-adults) ranged from 11.10cm - 21.92cm CL, mean=16.24cm, SD=3.57. Sub-adult turtles were not separated from the group (a carapace length of 10cm was used as the cut-off length for juveniles). From the ultra-sound examinations conducted by Dr

Gerald Kuchling, nearly all the females (except one) had follicular development. From their physical condition, they were considered to be a healthy population and had a near 1:1:1 ratio of females, males and sub-adults. In the following year from May - November of 2004 and in January 2005, sounds were recorded from two other turtle groups; from Pinweryning Dam and Blue Gum Lake respectively. For details of these populations, see Chapter 6. Also, four turtles that had been seized by customs officers from an attempt to be smuggled out of the country, were recorded whilst recuperating at the authors residence for two days prior to release.

The artificial ponds were round plastic tubs with dimensions of 0.65m depth x 1.80m diameter and were assembled to recreate small wetlands with logs and emergent/ floating aquatic plants which provided suitable habitat with hiding places for turtles (Figure. 4.1). The bottom was covered with a mix of soil and sand. The Masons Gardens turtles were initially kept as an entire population in a single artificial pond and after five weeks of recording as a group, females were then separated from the males and placed into two separate ponds of similar dimensions.

Figure.4.1. Artificial ponds used to contain turtles in which to make recordings of their underwater vocalisations. Each pond was set-up to re-create a small wetland and umbrella's were used to keep the afternoon sun off the ponds during summer.

Hatchlings and Juveniles

Seven hatchling and five juvenile turtles were utilised to ascertain whether very young turtles were also vocalising. Both groups were sourced from two populations of turtles: the hatchlings came from clutches from Lake Leschenaultia (3) and Bandyup Women's Prison population (4); the juveniles (CL: 5.90 cm to 9.93 cm) came from Bandyup Women's Prison and Pinwernying Dam populations. Juveniles were placed into a below-ground outdoor pond (1m length x 0.5m width x 0.4m depth). Hatchlings were placed into a small glass aquarium (35.5cm length x 20cm width x 22.0cm depth). Both the pond and the aquarium were set up to recreate small wetlands with aquatic vegetation and woody material for cover and were lined with a sand substrate. Hydrophone placement was limited in both ponds and lay in approximately 7cm of water in the glass aquarium and round 30cm in the juvenile pond. Hatchlings and juveniles were recorded over the months May through to December 2004.

4.2.2 Recording Regime

Recordings were made in the artificial ponds during the months of September and October 2003, February to December, 2004 and January 2005. Recordings occurred from as early as 4.30am (dawn recordings) to as late as 1.30am (evening recordings). The hydrophone was suspended in the center of each pond at a depth of 0.5m.

4.2.3 Acoustic Signals

Turtle vocalisations are presented as narrow-band spectra produced from Fast Fourier Transforms (FFT). They were digitised at a sampling rate of 48 kHz and FFT size 1024 points with an averaging of 4 - giving a time resolution of 5.33 ms (using a 75% overlap) and a frequency resolution of 46.875 Hz. A Hanning smoothing window was used.

'Mechanical' noise such as sounds associated with movement or feeding activities such as the lunge-snaps (or known as 'strike-and-gape' (Georges *et al.,* 1999)) at food items; have been excluded from the acoustic repertoire as well as the sounds produced when turtles burped/ hiccupped, scratched themselves, or were heard breathing at the waters surface – including 'whistle-breathing' (which has been noted by Legler quoted in Gans and Maderson, 1973).

4.3 Results

Only a small number of calls (3x 'short chirps', 2x 'medium chirps', many single 'staccato' pulses) were recorded in wetlands that could be attributed to turtles. There were also a number of other calls recorded at Quenda Wetland and Piney Lake, but could not be attributed to turtles (even though they occurred at a similar frequency to turtle calls e.g. 'zippers' and 'shakes'). As wetlands are relatively large areas and biologically active with birdlife and other organisms; it was impossible to entirely exclude their input (birdcalls were sometimes heard underwater). As a consequence, most of the data presented here are from recordings made in controlled conditions of the artificial ponds.

Most of the turtle calls could be separated into distinctive categories although there was some variation in the spectral nature within these groupings. In total, seventeen categories of vocalisations were recorded and are summarised in Table 4.1a. (juvenile calls are summarised in Table 4.1b). Vocalisations consisted of: a) clacks; b) clicks; c) squawks; d) hoots; e) short chirps; f) high short chirps; g) medium chirps; h) long chirps; i) high calls; j) cries or wails; k) cat whines; l) grunts; m) growls; n) blow bursts; o) staccatos; p) a wild howl; and q) drum rolling. Some categories occurred together in a bout (for definition see Table. 4.2). For example: grunts were heard at the beginning and end of the ‘wild howl’ call. Due to the polymorphic nature of the ‘chirp’ vocalisations, it was difficult to separate these sounds into distinctive categories. While they were intuitively similar because of the way in which they sounded, use of the name ‘chirp’ is essentially generic as phonetically, ‘chirp’ vocalisations consisted of a range of sounds such as ‘eeaow’, ‘MmM’, ‘M’, ‘oi’, ‘ar’. A format was used similar to that of Coscia *et al* (1991) for the vocalisations of Timber wolf (*Canis lupus*) pups. ‘Chirp’ calls were identified and separated according to at least two acoustic parameters. For example, the ‘short chirp’, the ‘high short chirp’, the ‘medium chirp’ and the ‘long chirp with the long frequency up / down-sweep elements’ were a similar type of sound to listen to, but they differed in their duration and their spectral structure – in particular, the long up / down-sweep elements were very distinctive and long chirps often contained three harmonics. ‘Short chirps’ were brief calls containing two harmonics or none, but the ‘high short chirps’ contained three harmonics and higher frequency elements compared to the ‘short chirps’. A summary of the range of turtle vocalisations recorded in this research and their acoustic parameters are presented in Table 4.1a & b. All sounds presented here were recorded in the artificial ponds. As calls recorded in aquaria can result in some signal distortion (Parvulescu, 1966; Yager, 1992), in particular the length of a vocalisation due to reverberation (Herzel *et al.,* 1998), it is understood that not all acoustic descriptions will reflect free-field recordings.

The sustained turtle vocalisations are described in Chapter 5.

Table.4.1a. Summary of adult turtle vocalisations and their acoustic properties recorded from three populations: Masons Gardens (MG), Pinweryning Dam (PD) and Blue Gum Lake (BG), including the four seized turtles* (ST) (NB These calls were all recorded in the artificial ponds).

Vocalisation: Aural Character, Category & Population	**Spectral output**	**Mean Duration (s)**	**Frequency Range (kHz)**	**Periodic/ Aperiodic**	**Dominant frequency (kHz)**	**Sex**
Clacks (pulse) (numerous: using random n=17) *MG, PD*	Continuous	0.050 ±0.011	1.4 - 2.1	periodic	1.5 – 2.0	M/F
Broadband Clicks Echo-location pulse? (numerous: using random n=18) *MG, PD*	Continuous	0.046 ±0.009	0.100 - > 20	aperiodic	8.0 – 16.0 0.85 - 1.70	M/F
Short Chirp (single note) (n=10) *MG, PD, BG*	Harmonics (1-2)	0.074 ±0.021	0.600 - 2	periodic (complex)	0.90 & 1.80	M/F
High Short Chirp (syllable) (n=7) *MG, PD, BG*	Harmonics (3-5)	0.114 ±0.024	0.400 - 2.8	▪periodic (complex) ▪Frequency modulation	0.88	M
Hoots (syllable) (n=12) *MG, BG*	Richly Harmonic (10)	0.152 ±0.075	0.117 - 2.3	periodic (complex)	0.16 -0.20	M/F
Squawks (syllable) (n=5) *MG*	Harmonics (=2)	0.207 ±0.038	0.600 - 1.8	periodic (complex)	1.5 – 1.8	M
Medium Chirp (syllable) (n=27) *MG, PD, BG*	Sparsely harmonic (≤3) with short up-sweep & down-sweep elements	0.290 ±0.124	0.780 -1.6	▪periodic (complex) ▪Frequency modulation	0.60 - 0.80	M/F
SH Long Chirp (syllable) (n=31) *MG, PD, BG*	Sparsely harmonic (=3),with long up-sweep &/or down sweep elements	0.360 ±0.144	0.650 - 1.6	▪periodic (complex) ▪Frequency modulation	0.70 - 0.80	M/F

* Seized Turtles came from a group of turtles rescued from smuggling activities at Perth International Airport.

Table.4.1a. Summary of adult turtle vocalisations and their acoustic properties (cont.).

Vocalisation: Aural Character, Category & Population	**Spectral output**	**Mean Duration (s)**	**Frequency Range (kHz)**	**Periodic/ Aperiodic**	**Dominant frequency (kHz)**	**Sex**
RH Long Chirp (syllable) (n=34) *PD*	Richly harmonic (=5-6), finishes in rapid up-sweeps Biphonation evident	0.342 ±0.121	0.255 - 3.8	▪periodic (complex) ▪Frequency modulation	0.46 - 0.65	M
High Calls (syllable) (n=6) *PD, BG*	Sparsely Harmonic (=3) rapid upsweep slow long downsweep	0.464 ±0.186	0.21 - 3.5	▪periodic (complex) ▪Frequency modulation	0.95-1.8	F
Cat Whines (syllable & burst) (n=4) *ST*	Tonal & Noisy elements	2.100 ±1.260	1.8 - 2.8	aperiodic (longest part of call)	2.4 - 2.6	F
Grunts (pulses) (numerous using n=12) *PD*	Noisy	0.075 ±.0.27	0.100 - 2.5	aperiodic	≤0.36	M/F
Growls (bursts) (numerous) *PD*	Noisy, unstructured	varied ≤2.0	0.100 - 1.1	aperiodic	≤0.20	M/F
Blow bursts (bursts) (numerous) *PD*	Noisy, spectrally coherent	varied ≤3.0	0.100 - 10	aperiodic	≤0.30	M/F
Wild Howl (syllable & (pulses) (n=1) *PD*	contains richly harmonic (=7), pulsed & noisy elements	10.263	0.100 - 3	periodic/ aperiodic	'howl' 0.21-0.39 0.57-0.70 'growling rattle' ≤0.17	F

Table.4.1a. Summary of adult turtle vocalisations and their acoustic properties (cont.).

Vocalisation: Aural Character, Category & Population	**Spectral output**	**Mean Duration (s)**	**Frequency Range (kHz)**	**Periodic/ Aperiodic**	**Dominant frequency (kHz)**	**Sex**
Wails (syllable) (n=5/2) *PD, BG*	Sparsely harmonic (≤4) finishing in rapid down-sweep	long wails 1.324 ±0.39 short wails 0.565 ±0.023	0.19-1.47	▪periodic (complex) ▪Frequency modulation	0.42	M/F
Drum Rolls (Pulses) (n=5) *PD, BG*	Coherent repetitive pulses	2.717 ±1.288 (Males)	0.100 up to 0.75	aperiodic	≤0.21	M/ F
Staccato (Pulses) *PD, BG, MG*	Rapid pulse series	varied	0.100 - 1.0 0.100 - 10.0	aperiodic	<0.12 <0.21	M/F

Table.4.1b. Summary of juvenile turtle vocalisations and their acoustic properties (NB These calls were recorded in artificial environments).

Vocalisation: Aural Character, Category & Population	**Spectral output**	**Mean Duration (s)**	**Frequency Range (kHz)**	**Periodic/ Aperiodic**	**Dominant frequency (kHz)**	**Sex**
'Duck honks' (syllable) (n= 11)	noisy, coherent structure	0.108 ±0.015	0.10 – 3.15	periodic	0.175	sub-adult male
Short Chirp (single note) (n=1)	short Up-sweep No harmonics	0.049	1.0 – 1.2	periodic	1.1	unknown
Staccato (Pulses)	single pulses or short series	varied	0.100 - 1.0	aperiodic	<0.12 <0.21	unknown

4.3.1 Terminology

Turtles produced a variety of sounds and classification of acoustic units were divided into five categories: pulse, note, syllable, bout and burst (Table. 4.2).

Table.4.2. Classification of acoustic units

Pulse: Defines the shortest and simplest of sounds produced by turtles. Pulses had a duration of around 0.05 seconds.

Note: Defines the shortest of the complex sounds, with a duration of around 0.075 seconds usually with first and second harmonics present with little to no frequency modulation.

Syllable: Defines a longer duration (around 0.30 seconds or longer) and more complex call. Harmonically structured with different rates of frequency modulation throughout the call and often finishing with well-defined up / down sweep elements.

Bout: Defines a sequence of three or more pulses, notes, syllables or bursts; called at intervals of around 1-25 seconds and could occur over a period of several minutes.

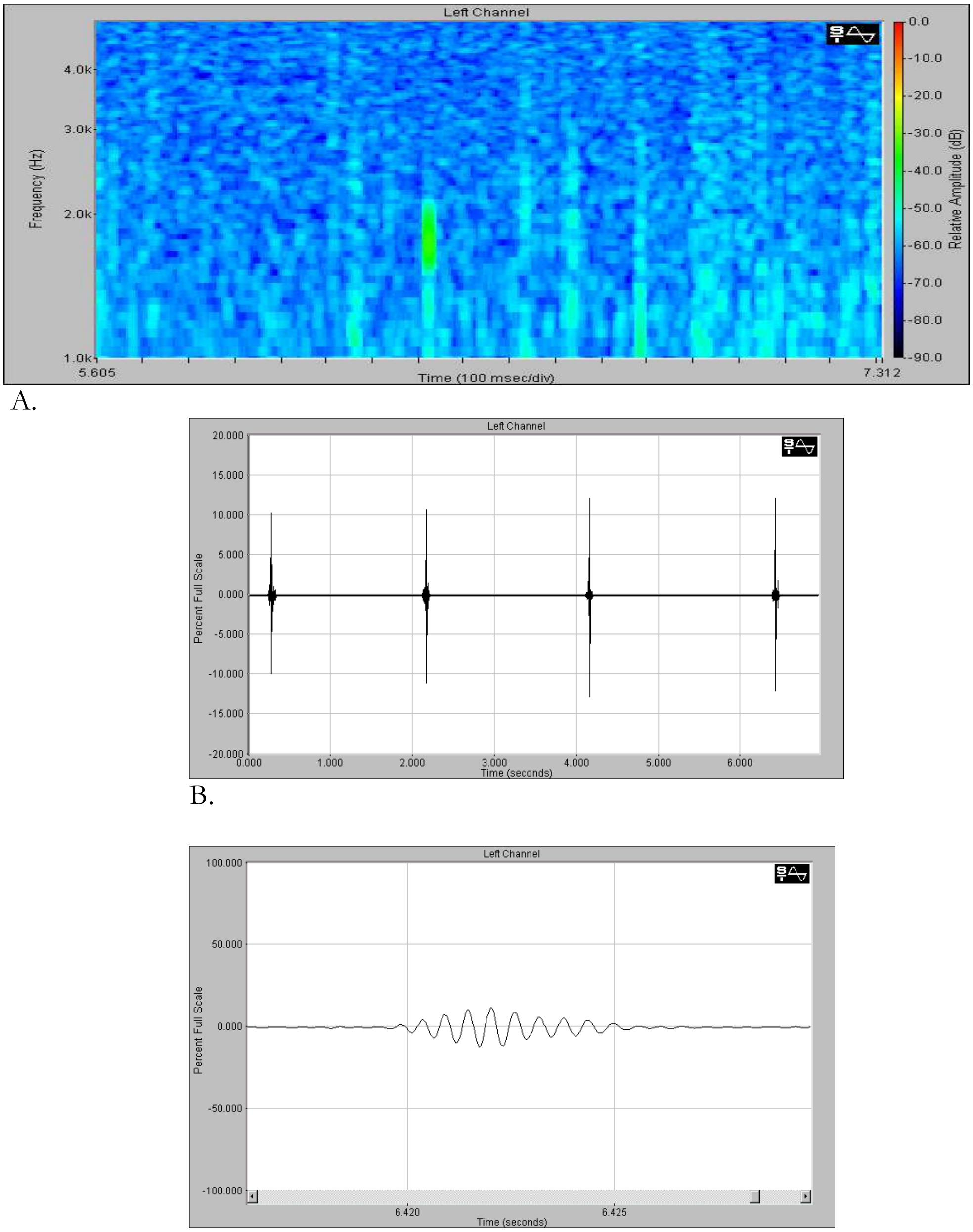
Burst: Defines those calls that were noisy and had a harsh strident sound quality. These chaotic segments (Herzel *et al.,* 1998) had a spectral structure either coherent or incoherent, but there were usually no harmonics or frequency modulation (aperiodic). The call length was of variable duration, but usually longer than syllables.

4.3.2 Descriptions

Vocalisations were generally named according to their aural character or failing a suitable description – some feature in the spectral structure of the call. For each call presented, three main parameters were measured from the spectrograms: 1.) Frequency range in kHz (from the lowest to the highest measurable frequency); 2.) Average duration of the signal in seconds; and 3.) The dominant frequency in kHz (frequency of that harmonic with the greatest amplitude). Usually, the dominant frequency was also the fundamental frequency. Also noted for each call, was the presence of frequency modulation and harmonics. Examples of each call were selected for their high signal-to-noise ratio with well-defined spectral contours.

'Clacks'

'Clacks' consisted of a number of pulses of brief duration (mean=0.050 sec, SD=0.011, n=17) (Figure. 4.2A & B). The frequency range of 'clacks' was around 1.4 kHz up to 2.1 kHz consisting of well-defined sinusoidal elements (Figure. 4.2A & C). 'Clacks' occurred as either relatively even-spaced calls (Figure. 4.2B) or occurred irregularly. Sometimes 'squawks' or 'short chirps' were heard interspersed between 'clacks' or at the end of a 'clack series'. From a randomly selected segment consisting of five 'clack' pulses and one 'short chirp', there was an average inter-pulse-interval (IPI) (measured from the end of one pulse to the beginning of the next pulse) of 4.504 seconds (SD=0.751) with a comparatively long separation from pulse five to the 'short chirp' of 12.558 seconds.



A.

B.

C.

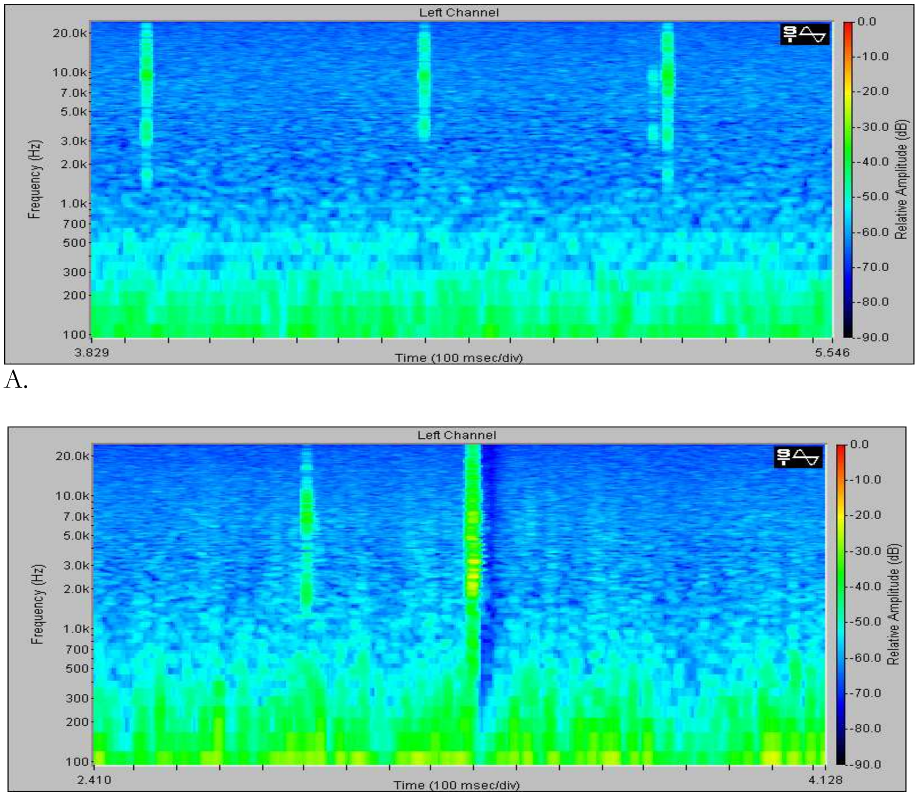
Figure.4.2. A. Spectrogram of one 'clack' pulse, which was the last pulse in the waveform spectra in B. **B**. Waveform of a 'clack' series consisting of four pulses.

C. Expanded view of the fourth 'clack' shows the fine structure of 'clacks' revealing well-defined sinusoidal elements. 'Clacks' were a frequent call by the Masons Gardens Turtle population. Occasionally, 'clack trains' were interspersed or ended with either a 'squawk' or 'short chirp'. This type of sequence was recorded in the artificial pond but was also heard during the summer in an isolated recording at Glen Brook Dam, 2003, where the 'clack train' ended with a 'duck-like squawk'. Recording was digitally filtered using a passband of 1150-2500 Hz.

‘Broadband Clicks’

Broadband ‘clicks’ were sudden onset sounds of short duration (mean=0.046 sec SD=0.009, random n=18), heard either singly or in multiples of three or more ‘clicks’ (Figure. 4.3A) and at times were heard as distinctive ‘double-clicks’, where two successive ‘clicks’ occurred with a brief silent interval between each ‘click’ (Figure. 4.3B). Other ‘clicks’ had dominant frequencies in the mid-frequency range with the pulse extending from below 100 Hz up to around 8 kHz (Figure. 4.4). All ‘click’ types were recorded in the artificial ponds and in the field. The most obvious ‘clicks’ produced in the field were those made by a turtle investigating the hydrophone at a distance of < 30cm (Figure. 4.4).

The ‘double click’ had an intra-click interval (time between the two audible ‘clicks’ on the spectrogram measured from the end of one pulse to the beginning of the next pulse) of 0.362 seconds (SD=0.148, n=9) (Figures. 4.3B & 4.5). Figure 4.5A depicts some of the variability seen in the ‘double-click’ presentation. There was variation in the intra-click interval and duration of the ‘clicks’ with the second ‘click’ having a longer duration than the first (Figure. 4.5B & C). Energy in the ‘clicks’, in particular the second ‘click’ (Figure. 4.5B & C), often overloaded the recording gear with frequency range outside recording capabilities. The expanded waveform of the second ‘click’ in the second ‘double-click’ presentation (Figure. 4.5C) reveals what appears to be echoes. Frequency range of ‘clicks’ varied. Some ‘clicks’ were high frequency pulses (Figure. 4.3A) from around 3 kHz to above 20 kHz extending into ultrasonic frequencies.



A.

B.

Figure.4.3. Broadband 'clicks' recorded in the artificial ponds produced by both male and female turtles. **A.** Spectrogram revealing the higher frequency 'clicks'. Recorded on the 23rd September, 2003 at 10.30pm. Water temperature 15 °C. **B.** Spectrogram revealing the 'double-click'. The second 'click' contains more energy than the first 'click' and has a frequency spectrum beyond the capabilities of the equipment. Recorded at dawn, February 4th, 2004. Water temperature 24°C.

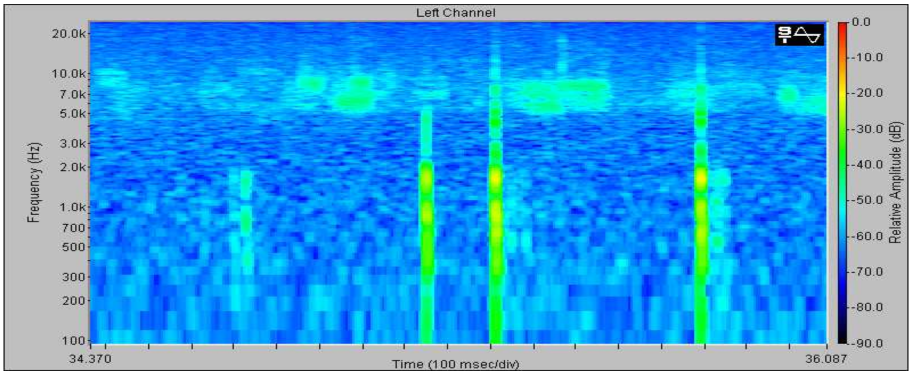


Figure.4.4. Spectrogram revealing three rapid 'click's from a series of 'clicks' recorded at dawn - Blue Gum Lake in the summer, 2003. A turtle had swum nearby to investigate the hydrophone and was within 0.3m of the hydrophone when this recording was made. 'Cork on Glass' callers are utilising the frequency band between 5-11 kHz. Water temperature 23°C.

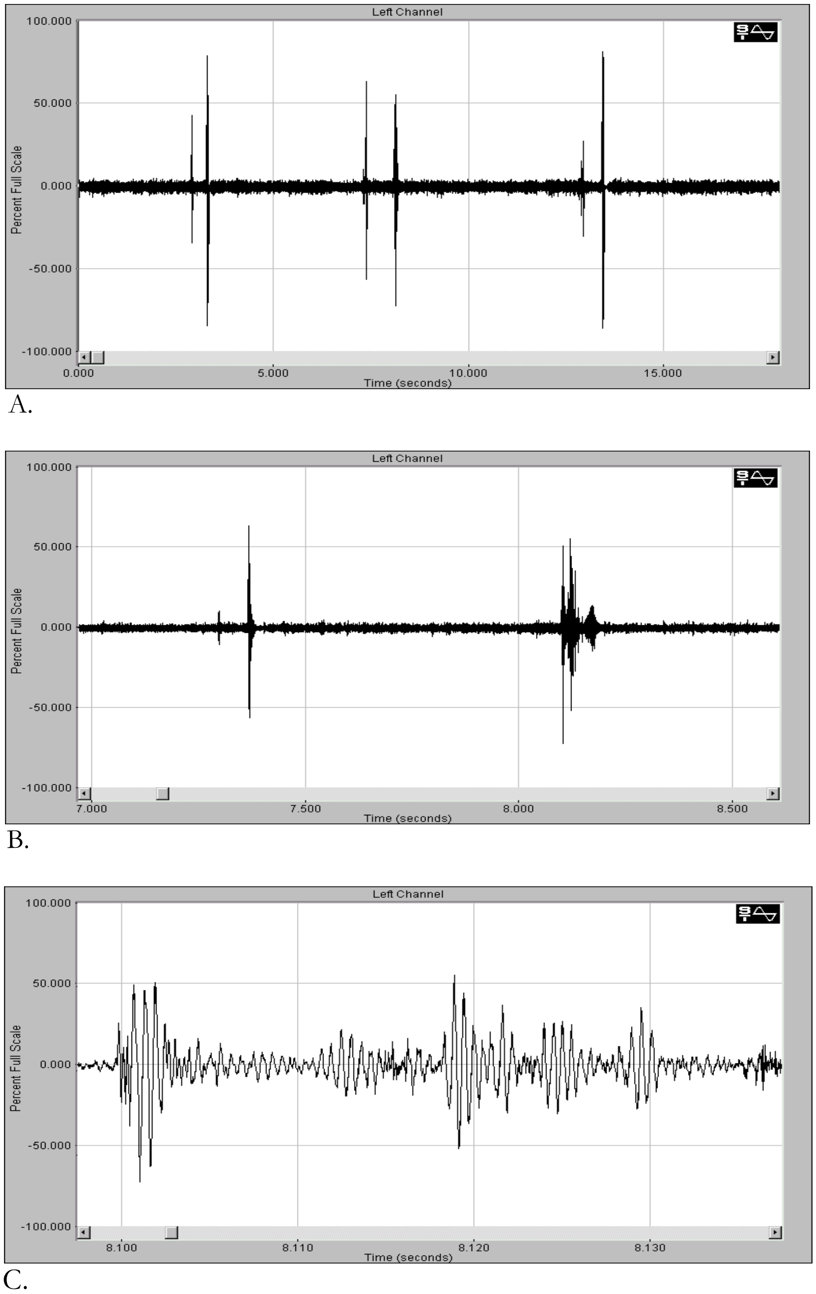
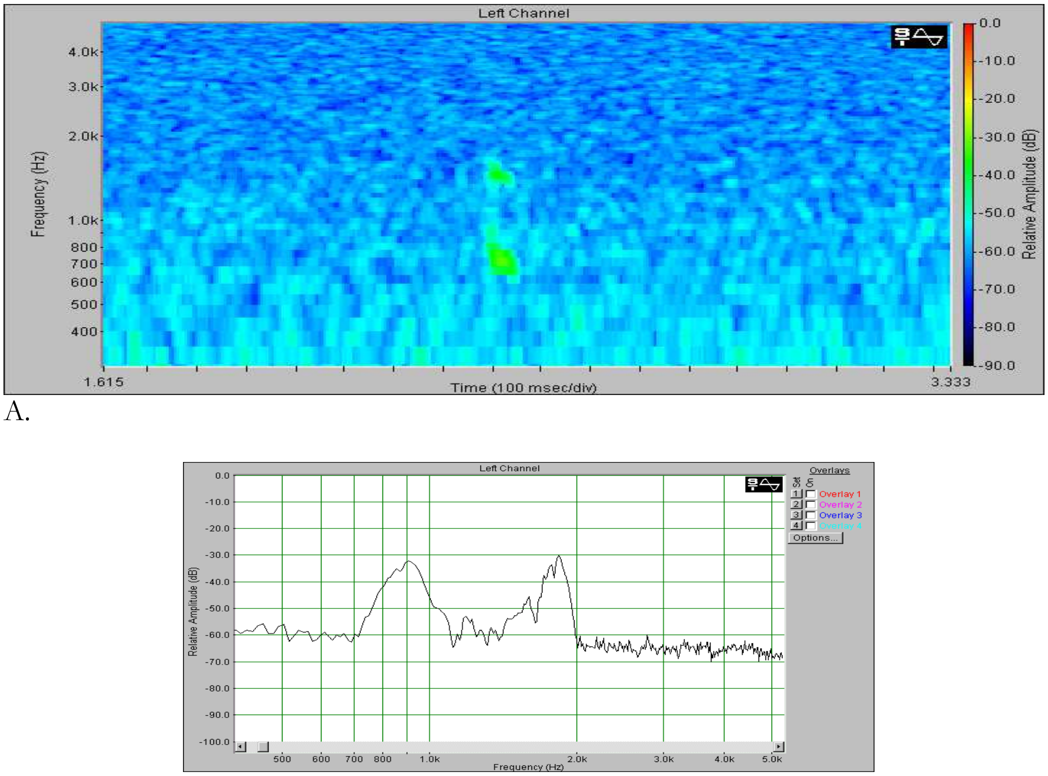


Figure.4.5 A. Waveform characteristics of the first three sets of 'double-clicks' in a dawn recording consisting of eight 'click' sequences which lasted around 50 seconds revealing some of the variations that occurred in 'double-click' presentation. The 'double-click' in spectrogram 4.3B is the first 'double-click' shown in A. **B.** Expanded view of the second 'double-click' pattern from A. **C**. Expanded view of the second 'click' in B. Recordings were digitally filtered using a highpass filter with a cutoff frequency of 300 Hz. Water temperature 24°C.

‘Short Chirps’

Most of the ‘short chirp’ calls consisted of only first and second harmonics at similar amplitudes (Figure. 4.6A& B) with generally little or no frequency modulation. They were of short duration (mean=0.074 sec SD=0.021, n=10) and sounded similar to those produced by kittens. These calls could be heard as ‘oi’, ‘orr’, ‘arr’, ‘m’, ‘ha’ or ‘ow’. ‘Short chirps’ were considered to be a single note with calls utilising a frequency range from around 500 Hz to just under 2 kHz.



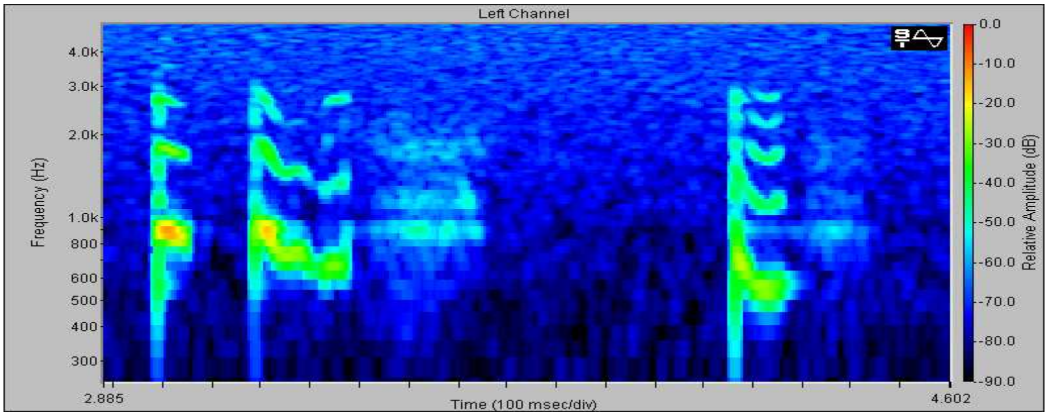
A.

B.

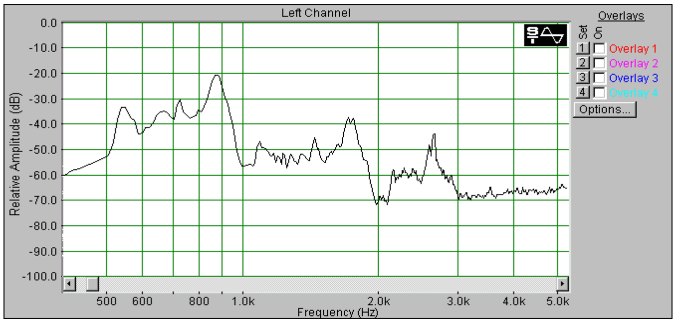
Figure.4.6. A. Spectrogram of a typical ‘short chirp’ revealing two harmonics with dominant frequencies in both harmonics.. **B.** Amplitude spectrum of the ‘short chirp’ revealing the greatest energy in the first harmonic is around 900 Hz and around 1.8 kHz in the second harmonic (Amplitude spectra: FFT 4096 points with 75% overlap). Water temperature 15.5°C.

‘High Short Chirps’

This category has been separated on the basis of the distinct frequency modulation (FM) and the presence of three or more related harmonic components (Figure. 4.7) usually at higher frequencies than the ‘short chirp’. ‘High short chirps’ were recorded during February 2003 and only male turtles produced these calls. They were of slightly longer duration than the previous 'short chirp' category (mean=0.114, sec SD=0.024, n=7). The number of harmonics ranged from three to five with the frequency range extending from around 400 Hz - 2867 Hz. In the chirp bout shown on Figure 4.7A, greatest spectral energy for the first 'high short chirp' was around 830 Hz - 910 Hz but in the second 'high short chirp' (third syllable), there were two main spectral peaks ranging from 640 Hz – 730 Hz and 530 Hz – 575 Hz (Figure. 4.7B). The second syllable was assigned to the vocal category of 'long chirp with long down-sweep'. This particular sequence of vocalisations had a melodic quality to it*.



A.



B.

Figure.4.7. A. Spectrogram of a 'chirp bout' emitted by a male turtle in the artificial pond. The first and third syllables in this bout have been separated into a separate category due to the frequency modulation and the presence of three or more harmonics. **B.** Amplitude spectrum of the last syllable shown in A with four distinct peaks. (Amplitude spectra:FFT size 4096 points, 75% overlap). Recorded on the 12th February, 2004. Water temperature 28°C.

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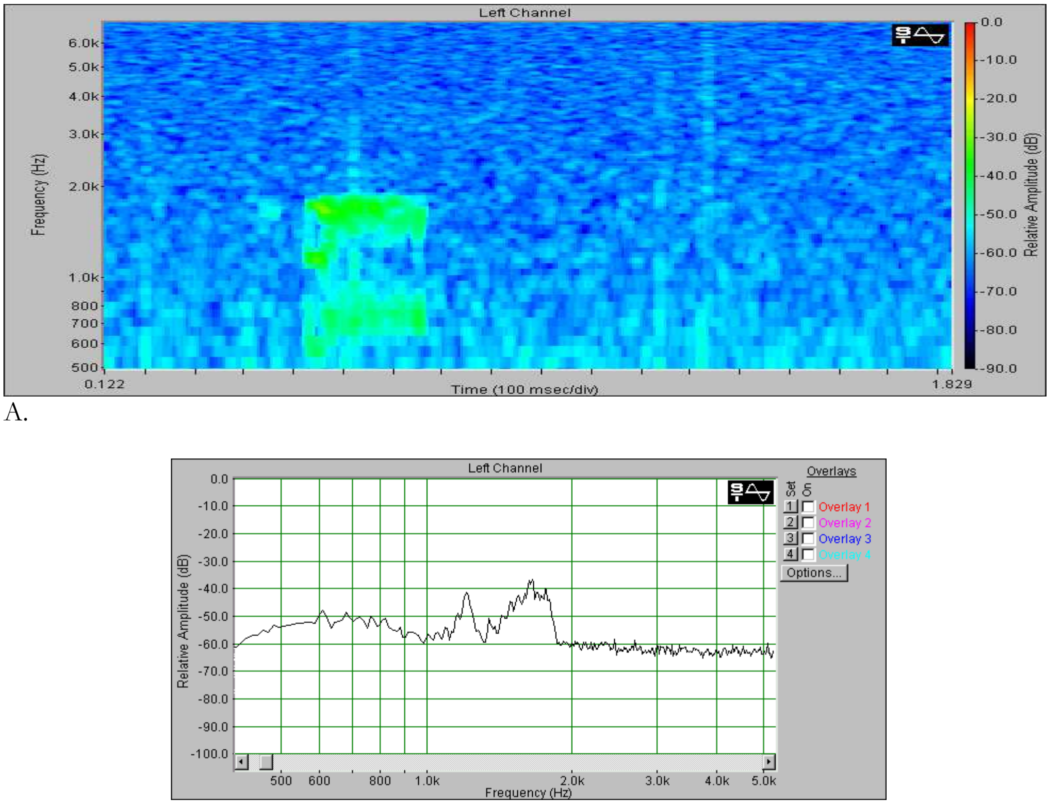
* ..And was truly lovely to listen to…

'Squawks'

'Squawks' sounded very similar to a duck quack and had a mean duration of 0.207 seconds (SD=0.038, n=5), similar to the duration of 'medium chirps'. 'Squawks' ranged in frequency from around 600 Hz up to 1.8 kHz with the dominant frequency around 1.4 kHz to 1.8 kHz (Figure. 4.8A & B). Very little frequency modulation was evident in this call (except at the beginning of the call) and had a 'flat' sounding quality to it.



A.

B.

Figure.4.8. A. Spectrogram of a typical 'squawk', which was sometimes heard interspersed between 'clacks' or at the end of a 'clack train'. **B.** Amplitude spectrum of the 'squawk' revealed a wider frequency use in the higher frequencies. (Amplitude spectra:FFT size 4096 points, 75% overlap). Recorded October 30th, 2003 at 6.30pm. Water temperature 15.5°C. Digitally filtered using a highpass band filter at 500 Hz.

‘Medium Chirps’ and ‘Long Chirps’

The ‘medium and long chirps’ were longer duration calls compared to the ‘short or high short chirps’ (‘Medium chirps’: mean=0.290 sec, SD=0.124, n=27; ‘SH long chirps’: mean=0.360 sec SD=0.144, n=31 and ‘RH long chirps’: mean=0.342 sec SD=0.121, n=34) and were frequency modulated containing short or long, up/down sweep elements - with the sweep often being rapid. ‘Medium and long Chirps’ sounded similar to ‘MmM’, ‘Mew’ or ‘eeiow’. Structurally, the ‘medium and SH long chirps’ were sparsely harmonic (SH), with two to three harmonically related elements. In the example of Figure 4.9A, three harmonically related components are evident; first, second and fourth harmonics – the third was missing. The ‘RH long chirps’ were richly harmonic (RH) (Figure. 4.10A) and had five or more harmonically related elements.

Dominant frequencies were below 1 kHz for these longer chirps. For the ‘medium chirps’, dominant frequencies were around 600 – 800 Hz and around 700-800 Hz for the ‘SH long chirps’ (Figure. 4.9B). For the ‘RH long chirps’, maximum energy was lower - at 460-650 Hz, but unlike the ‘short and SH long chirps’, spectral peaks also occurred at higher frequencies from 900-1500 Hz (Figure. 4.10A & B). The ‘RH long chirps’ were only produced by male turtles from Pinweryning Dam (CL 20.55-22.62cm). Examples of the differences encountered in the ‘medium chirp’ calls are clearly shown in Figure 4.11a-e which reveals a bout of ‘chirp’ calls (over seven minutes) which appeared to be a turtle(s) responding to the barking of a dog (poodle) on a neighbouring property. The poodle barks could be heard under-water and occurred at a similar frequency to the turtle calls. No calls were heard more than an hour prior to the poodle barking and did not continue after the poodle stopped barking. Generally, ‘chirp’ calls occurred either as isolated calls or in a sequence of three or four calls. On only two other occasions were longer bouts of ‘chirp’ calls heard (32 calls in 16 mins / 34 calls in 7 mins - heard in the Pinweryning Dam turtle population). In the ‘chirp bout’ below, the turtle called immediately after or almost on top of the poodle call on three occasions - depicted in spectrogram Figure 4.11 (2), (24), (28) & (29) (average time between poodle call and turtle response call 0.00347 sec, SD=0.00055). The ‘medium chirp’ usually followed the bark on average 0.0753 sec (SD=0.116). Spectrogram Figure 4.11 (5) and (9) reveals the nature and frequency of the poodle call (1 kHz to 1.8 kHz at times 1.43 and 2.33 minutes respectively) and the rapid call response of the turtle.

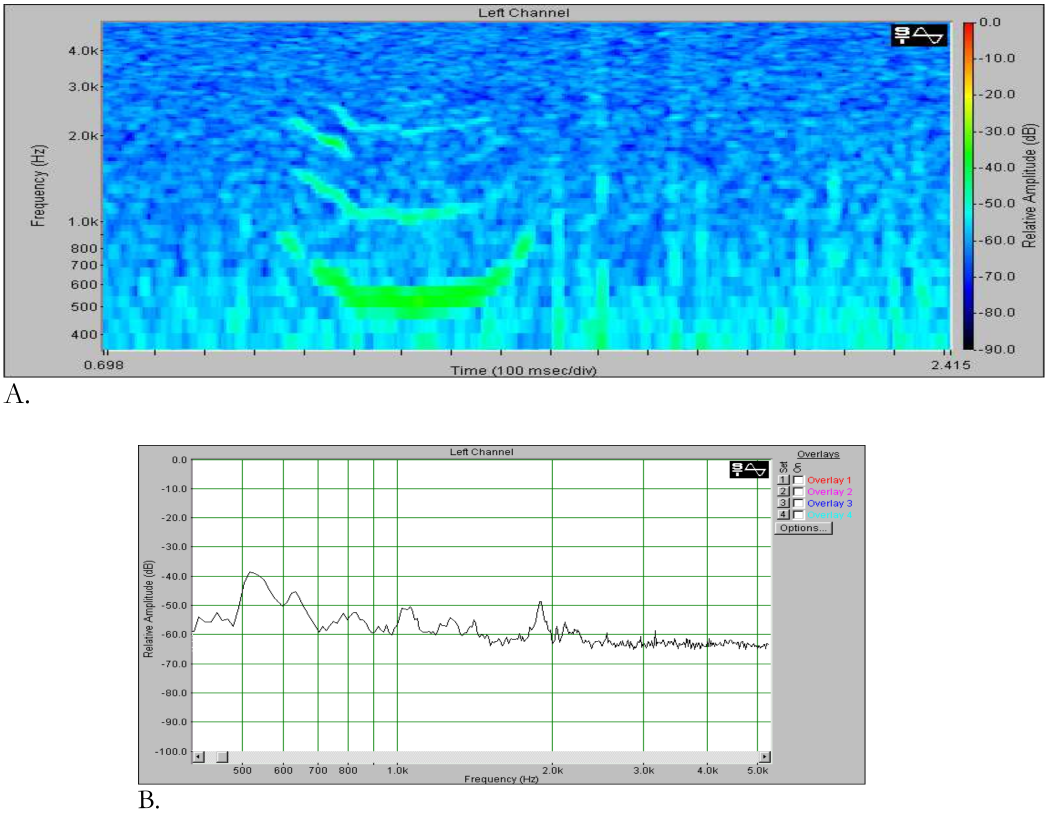


Figure.4.9. A. Spectrogram of a ‘SH (sparsely harmonic) Long Chirp’ (heard as ‘eeiow’). Note the rapid frequency down and up sweeps. **B.** Amplitude spectrum revealing a comparatively small dominant peak at around 520 Hz with harmonics at lower amplitudes. (Amplitude spectra: FFT size 4096 points, 75% overlap). Recorded at Midnight, October 2003. Water temperature 14.5°C.

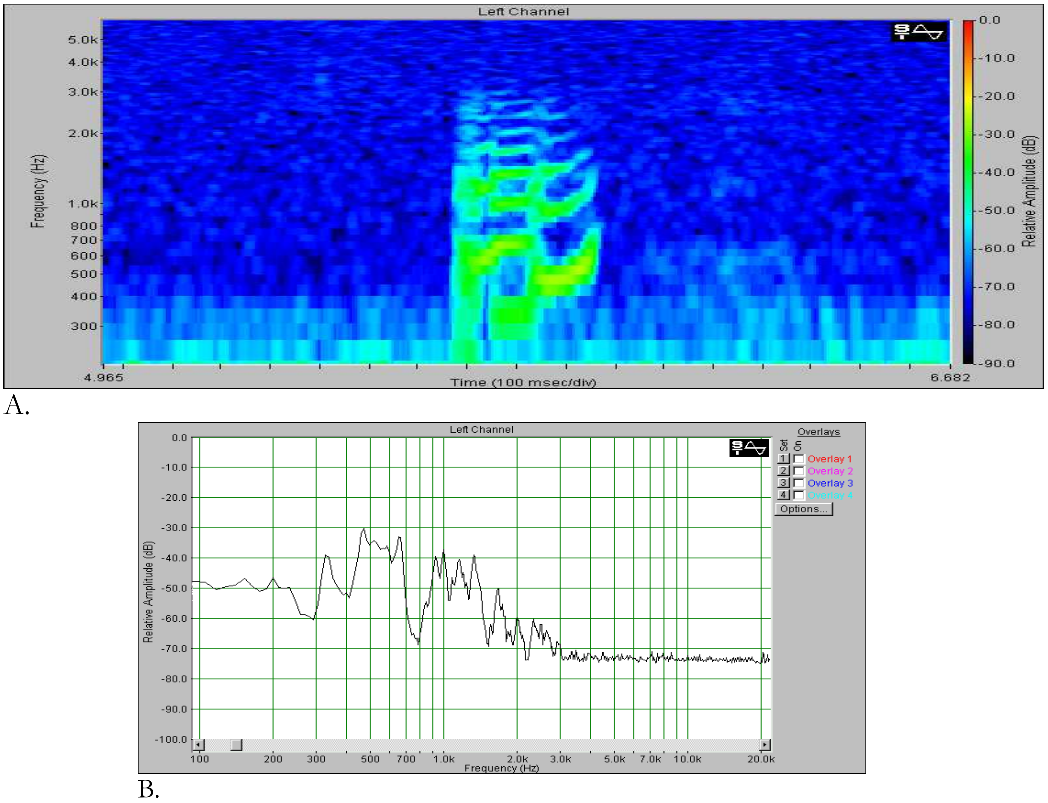


Figure.4.10. **A.** Spectrogram of a 'RH (richly harmonic) Long Chirp' call clearly revealing the rich harmonic structure and the rapid frequency up sweeps. **B.** Amplitude spectrum revealing a dominant peak at around 460 Hz with spectral peaks evident between 900-1500 Hz. (Amplitude spectra: FFT size 4096 points, 75% overlap). Recorded at 7pm, August 2004. Water temperature 13°C.

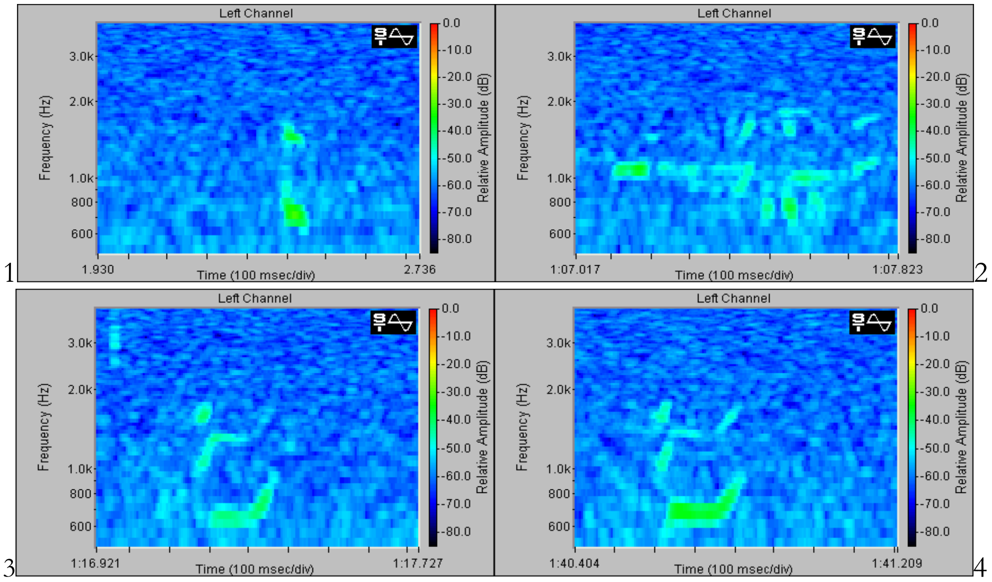


Figure.4.11a. Spectra revealing a 'chirp bout' believed to be in response to a poodle barking from 0-1.41 minutes in a 7 minute chirp bout. Spectrogram # 2 reveals the turtle vocalisation almost on top of the poodle bark. Recorded September 2003. Water temperature 15°C.

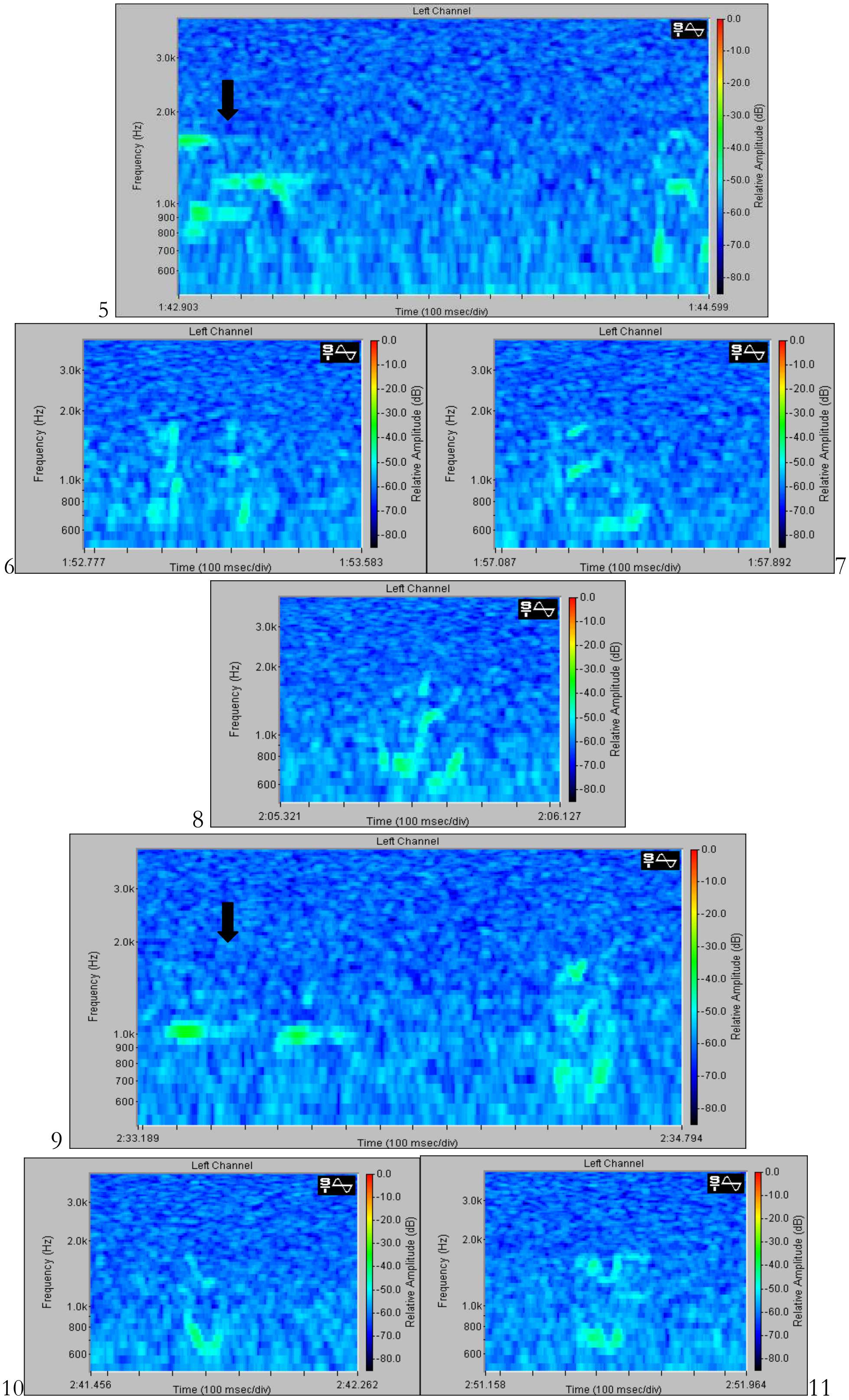


Figure.4.11b. Spectra revealing a 'chirp bout' believed to be in response to a poodle barking from 1.42-4.03 minutes in a 7 minute chirp bout. Spectrogram #5 reveals the poodle bark on the left (at time 1.43 minutes) followed by the turtle vocalisation. Spectrogram # 9 reveal the poodle barks on the left followed by a turtle call. Recorded September 2003. Water temperature 15°C.

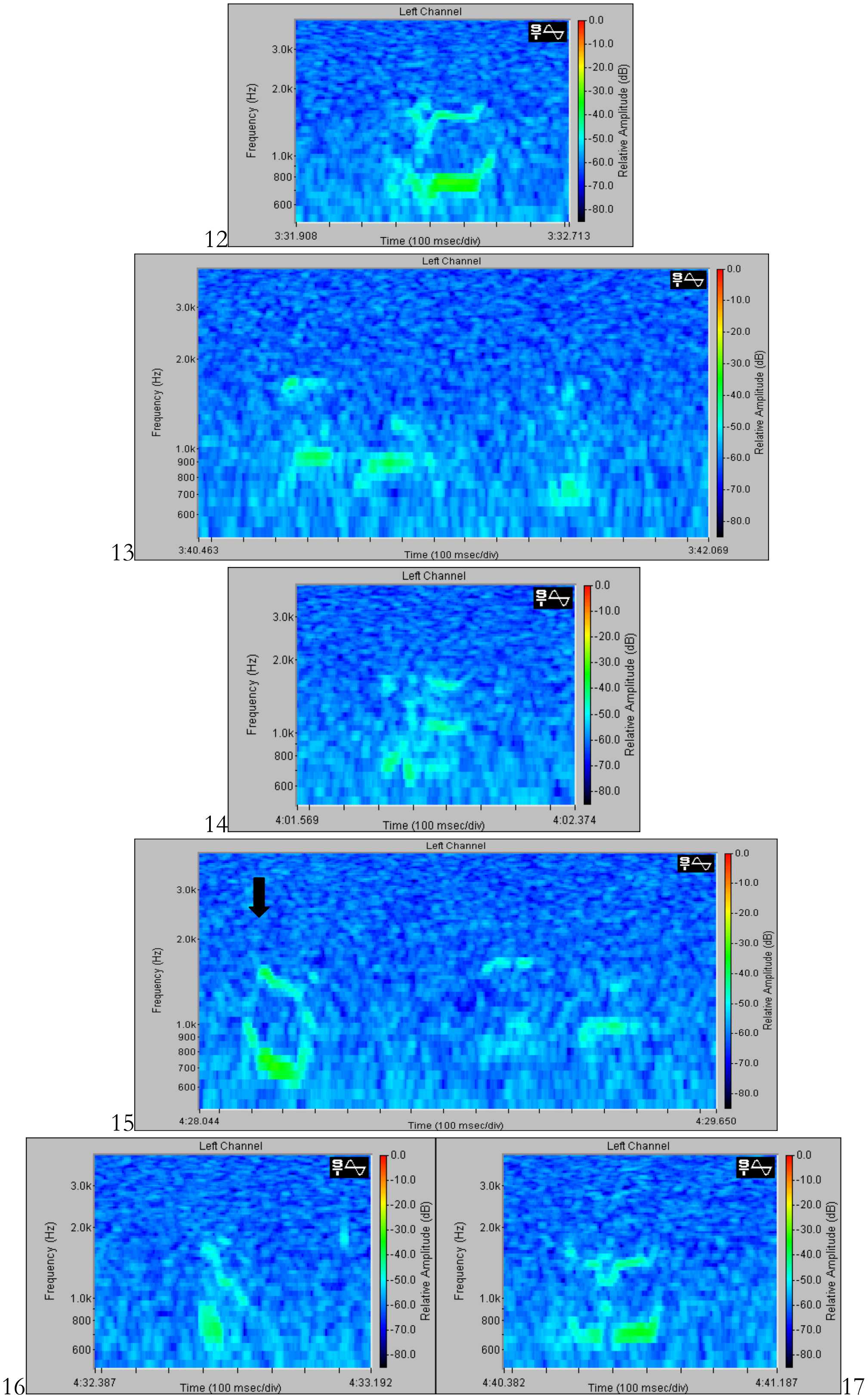


Figure.4.11c. Spectra revealing a 'chirp bout' believed to be in response to a poodle barking from 3.31-4.41 minutes in a 7 minute chirp bout. Spectrogram #13 reveals the poodle barks on the left followed by a turtle call; #14 reveals the turtle and poodle bark on top of each other and #15 has the turtle call on the left followed by the poodle bark. Recorded September 2003. Water temperature 15ºC.

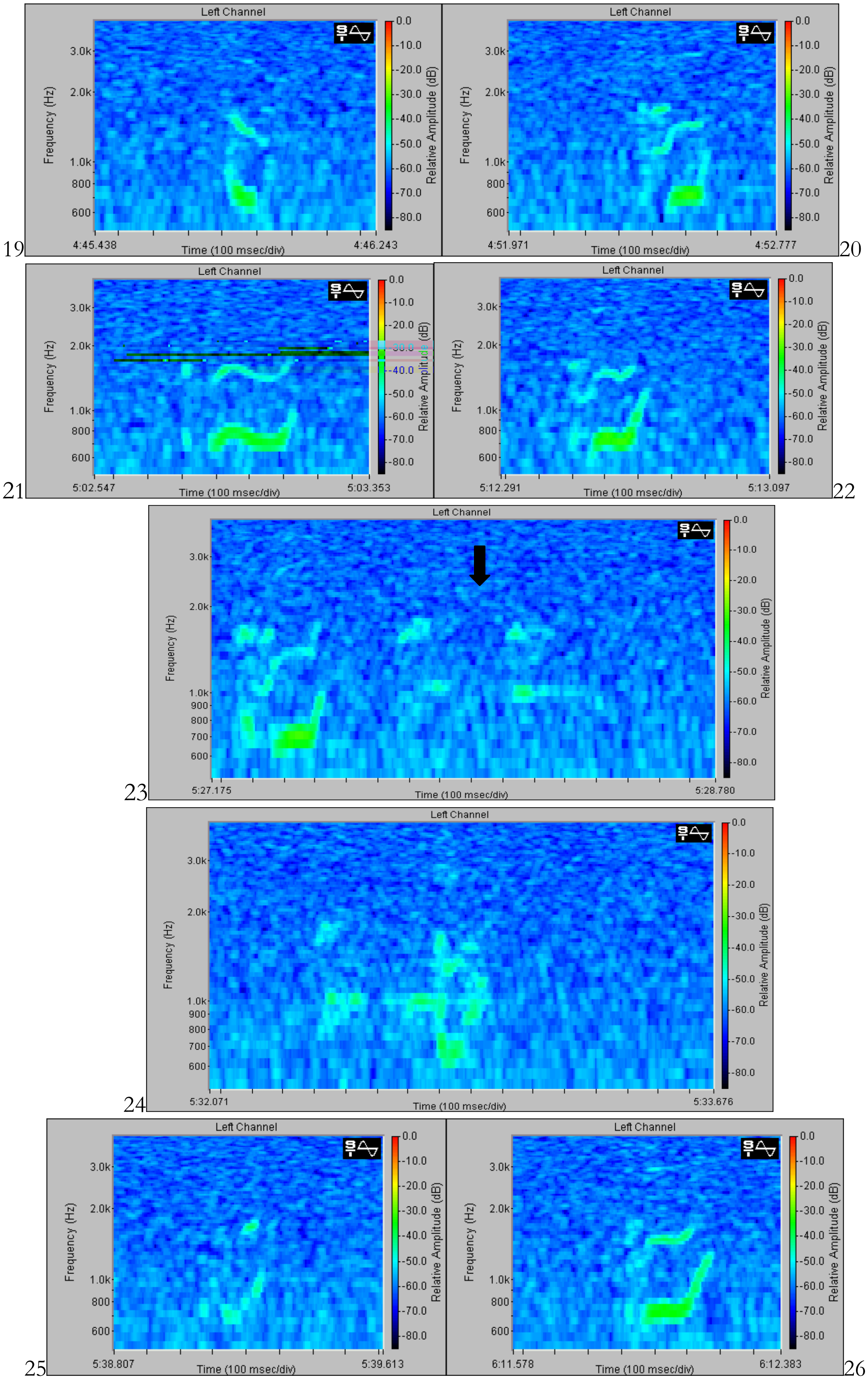


Figure.4.11d. Spectra revealing a 'chirp bout' believed to be in response to a poodle barking from 4.45-6.12 minutes in a 7 minute 'chirp bout'. Spectrogram #23 reveals the poodle bark following the turtle call on the left, but it was not considered that the poodle could hear the turtle. Spectrogram # 24 reveals the turtle calling almost on top of the poodle bark. Recorded September 2003. Water temperature 15ºC.

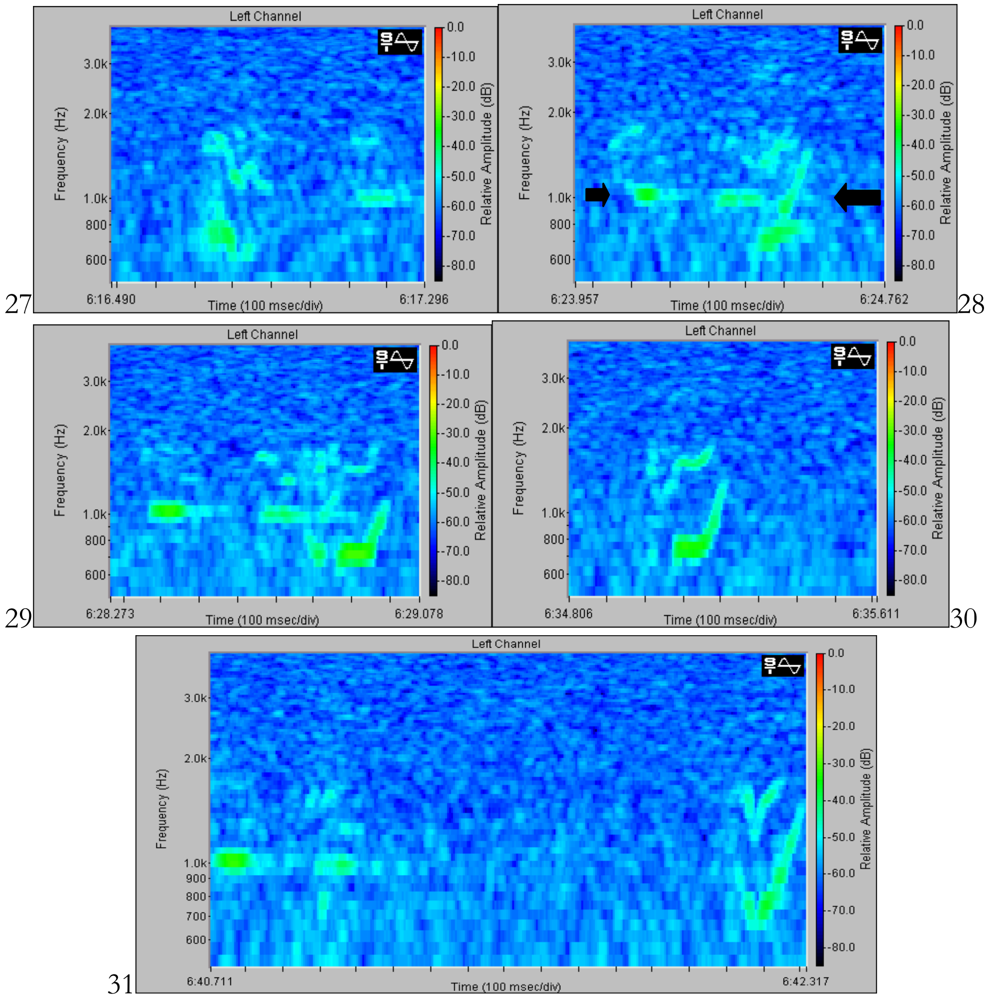
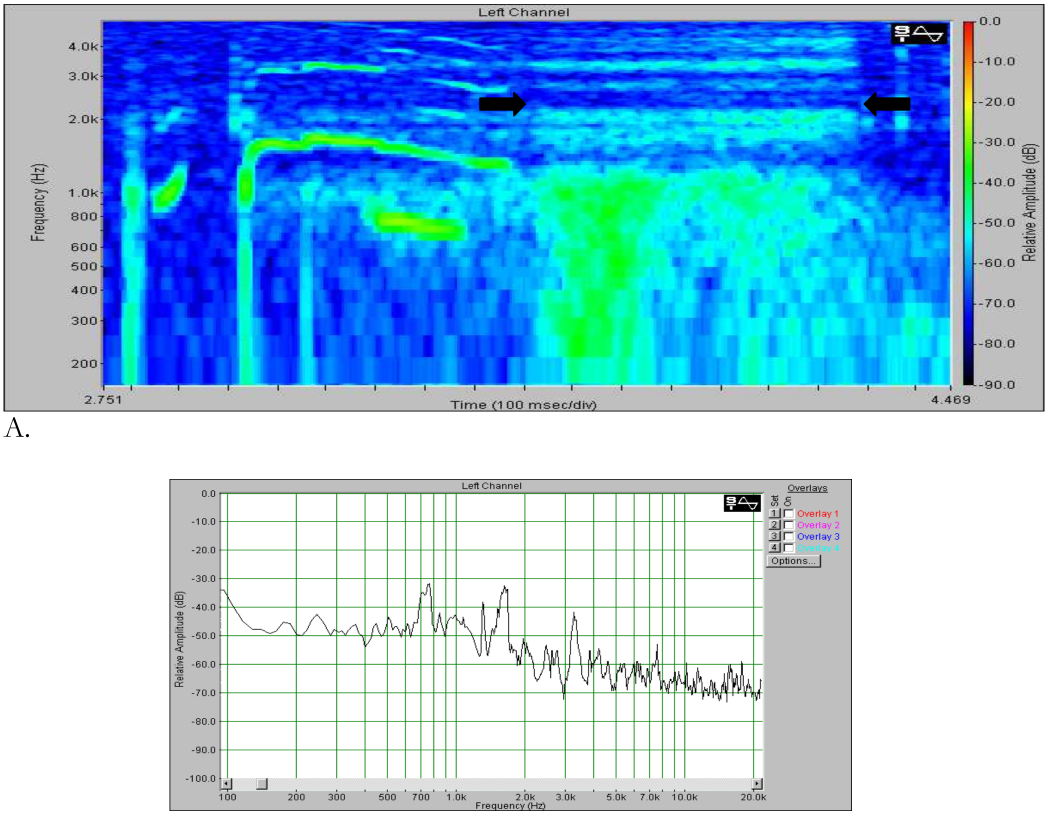


Figure.4.11e. Spectra revealing a 'chirp bout' believed to be in response to a poodle barking from 5.16-6.42 minutes in a 7 minute 'chirp bout'. Spectrogram #'s 28 & 29 reveals the turtle calling almost on top of the poodle call (In #28 turtle call is on the right). Recorded September 2003. Water temperature 15ºC.

'High Calls'

'High calls' were mostly produced by Pinweryning Dam female turtles (CL: above 24cm) (Figure. 4.12). 'High calls' by this group typically began with a short pre-syllable (0.160 sec for example shown), with a rapid up-sweep; followed by a longer syllable (0.569 sec in Figure. 4.12A) with a slow down-sweep, finishing with a 'breathy' component (shown between arrows). Harmonically related elements were also present in calls made by this group of turtles with frequency range extending from around 440 Hz up to 3.5 kHz (Figure. 4.12B). The high frequency range was due to the presence of harmonics but dominant frequencies were usually below 1 kHz at around 950 Hz. The single ‘High call’ recorded in the Blue Gum female group (carapace lengths below 20cm) had dominant frequencies from 950 Hz up to 1.8 kHz and occurred in a series of peaks (Figure. 4.13B). Harmonics were not evident in this call and neither was the pre-syllable or the ‘breathy’ component a part of this call. The Blue Gum ‘High call’ had a lower frequency range from 209 Hz to 1.87 kHz (Figure. 4.13B). Mean call duration of ‘High calls’ was 0.464 sec (SD= 0.186, n=6). The ‘breathy’ component in the Pinweryning Dam ‘High calls’ (seen from time 3.5 sec to 4.3 sec in Figure. 4.12A) were not measured in the call duration.



A.

B.

Figure.4.12. A. Spectrogram of a ‘High call’ call from the Pinweryning Dam females. The call begins with a rapid up-sweep pre-syllable and finishes with a long slow down-sweep. Harmonic elements are evident (‘Breathy’ component shown between arrows). **B.** Amplitude spectrum revealing several peaks, but dominant frequency is below 1 kHz (Amplitude spectra: FFT size 4096 points, 75% overlap). Recorded at 3.45pm, July 2004. Water temperature 15°C.

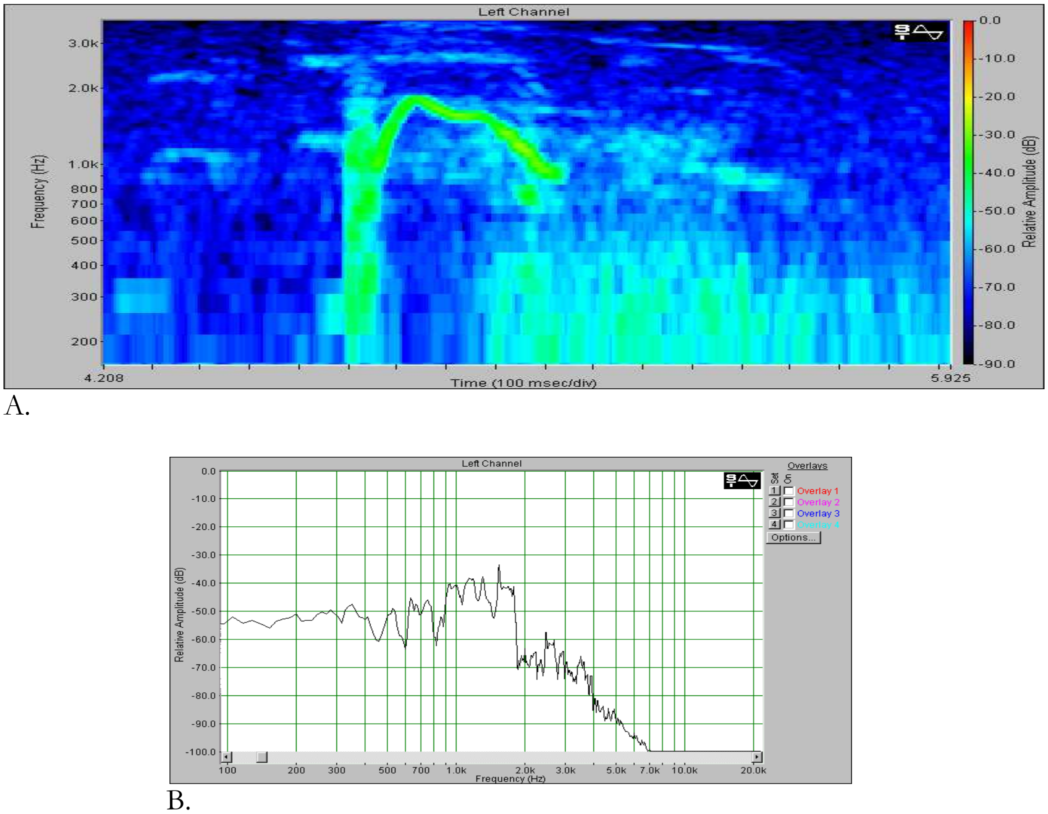


Figure.4.13. A. Spectrogram of a 'High call' call from the Blue Gum Lake females revealing similarity in structure to the 'High calls' of the Pinweryning Dam females with the exception of no harmonics or pre-syllable being present. **B.** Amplitude spectrum revealing a band of dominant frequencies (Amplitude spectra: FFT size 4096 points, 75% overlap). Recorded at dawn, January 2005. Water temperature 23°C.

'Wails'

'Wails' are syllables with a sparse structure composed of at least four harmonically-related elements. Frequency modulation in the long 'wails' occurred as a gentle undulation, finishing with a rapid downsweep. Short 'wails' (Figure. 4.14 #3) had a flat sound quality, with little to no frequency modulation. The bout of 'wails' revealed in Figure 4.14 lasted over a minute (78.86 sec) and occurred as a pattern of - a long 'wail' followed by a short 'wail' (this pattern was more obvious when listening). The long 'wails' became progressively longer and finally finished on a short 'FM wail'. Occasional single 'wails' were heard in other turtle groups; with a bout of 'wails' only recorded on a single occasion. The 'wail' bout was preceded by a number of 'blow bursts', with 'grunts' being heard throughout. Single 'grunts' were called between the 'wails' and one is evident in the last spectrogram – # 8 at time 1.07.233. Frequency use ranged from 190 Hz to 1468 Hz, with a mean duration for long 'wails' of 1.324 seconds (SD=0.39, n=5) and a mean duration for the short 'wails' of 0.565 (SD=0.023, n=2). Dominant frequency use was just above 400 Hz with lesser peaks above this but below 1 kHz.

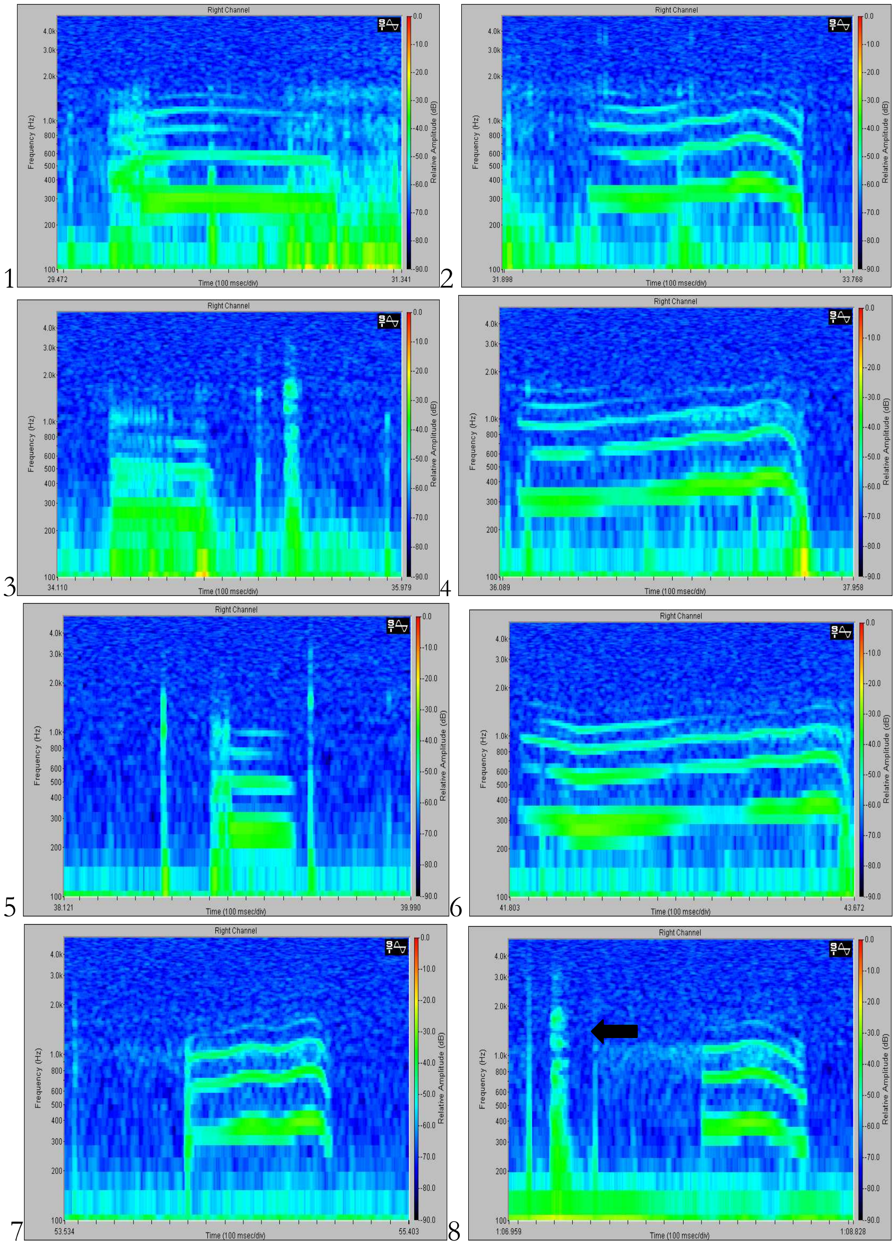
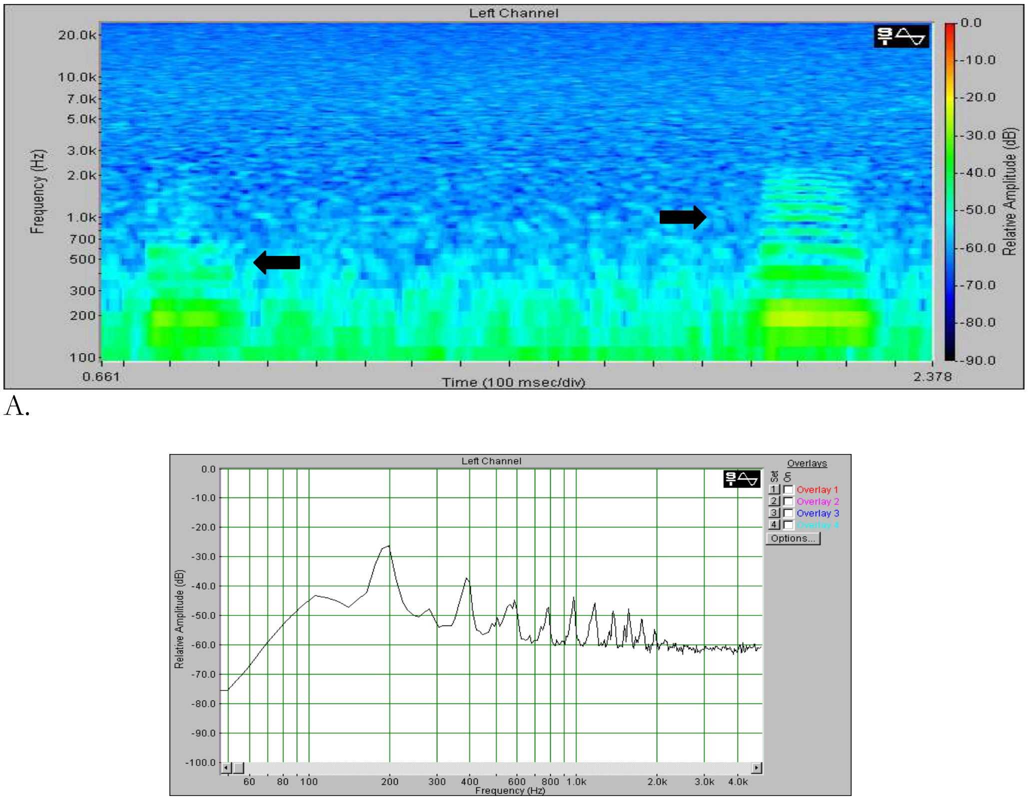


Figure.4.14. Spectrograms 1-8 depicting the bout of 'wails' produced by the large females (CL 24.21cm –28.24cm). Note the single 'grunt' pulse at time 1.07.23 minute in spectrogram 8. This was thought to have been produced by another female nearby. Recorded November 12th, 2004 at 7.40pm. Water temperature 20°C.

‘Hoots’

‘Hoots’ are syllables with a dense structure of harmonically-related components (around 10 harmonics)(Figure. 4.15A). They have a flat sound quality to them with only slight frequency modulation and were produced by both male and female turtles. ‘Hoots’ appeared either singly or several and utilised a broad frequency band ranging from around 120 Hz to just over 2 kHz, with a mean duration of 0.152 seconds (SD= 0.075, n=12). The dominant frequency was from around 160 Hz to just over 200 Hz with nine lesser peaks (Figure. 4.15B).



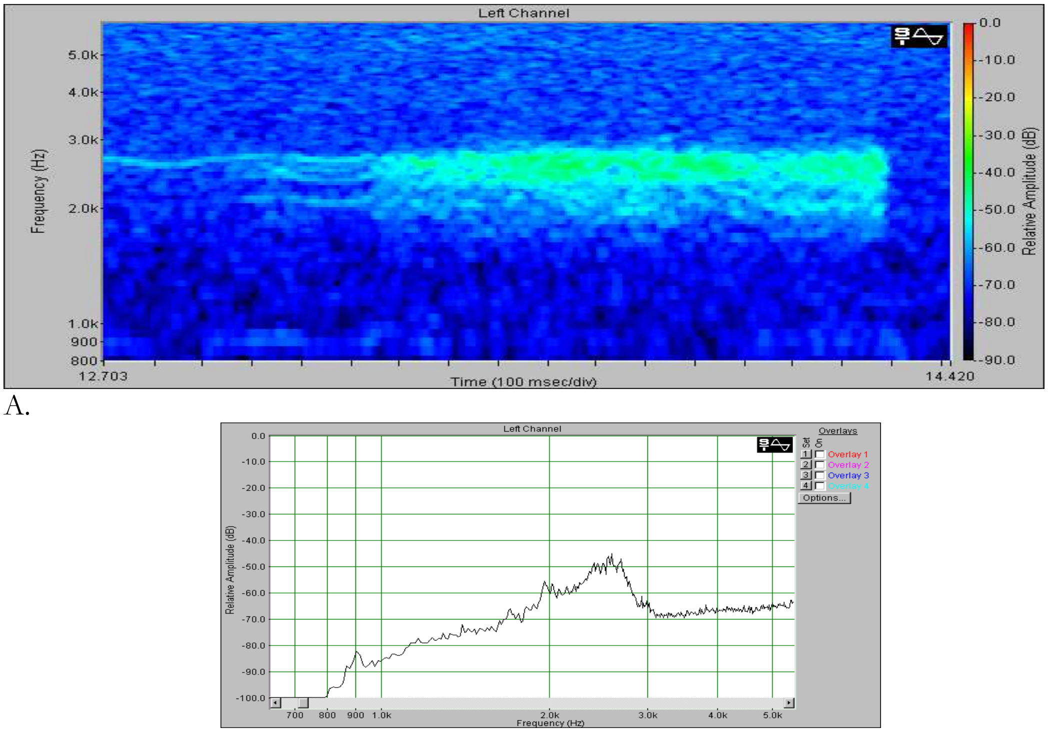
A.

B.

Figure.4.15. A. Spectrogram of two ‘hoots’ produced by male turtles. **B.** Amplitude spectrum of the last ‘hoot’ in A. The first harmonic at 200 Hz has the greatest amplitude decreasing in the second harmonic and with the higher frequency harmonics tapering off. (Amplitude spectra: FFT size 4096 points, overlap 75%). Calls were recorded on the 16th October, 2003 at 11.50pm. Water temperature 15°C.

'Cat Whines'

'Cat whines' were a similar sound to those produced by cats in aggressive encounters and were only recorded by the 'seized' turtles (CL>24cm). These calls were one of the longer calls in *C. oblonga's* repertoire (mean=2.10 sec, SD=1.26, n=4). Spectrograms revealed a transition from periodic phonation to aperiodic chaos with an abrupt finish (Figure. 4.16A). The call began with a constant frequency; with harmonics evident in the middle of the call (periodic), but most energy in this call was contained in a noisy bandwidth in the last two-thirds of the call (aperiodic), producing a harsh strident sound. The noisy part of this call ranged from around 1.8 kHz to 2.8 kHz with the dominant frequency occurring from around 2.4 kHz to 2.6 kHz (Figure. 4.16A & B).



A.

B.

Figure.4.16. A. Spectrogram of a 'cat whine' produced by the 'seized turtles' revealing the single frequency use at the beginning of the call, changing to a harmonic structure with an abrupt transition to chaotic phonation in the last two-thirds of the call. **B.** Amplitude spectrum of the 'cat whine' revealing similar relative amplitude throughout the higher frequency components in this call. (Amplitude spectra: FFT size 4096 points, overlap 75% and digitally filtered at 1200 Hz). Recorded March 24th, 2004. Water temperature 22ºC.

‘Grunts, Growls and Blow bursts’

‘Grunts’, ‘growls’ and ‘blow bursts’ occurred either singly or in combination with each other and were numerous among the Pinweryning Dam turtles. ‘Grunts’ were the shortest duration of these sounds (mean=0.075 sec, SD=0.027, using n=12), with ‘blow bursts’ the longest, usually lasting more than one second – occasionally up to three seconds. ‘Grunts’ were pulsed sounds ranging from around 100 Hz to usually around 2.5 kHz or even higher as shown in Figure 4.17 with dominant frequencies below 360 Hz. On one occasion, the seized turtles were recorded producing a series of ‘grunts’ (8) followed by a ‘moan’. On this occasion, the ‘moan’ appeared to be a ‘grunt’ of longer duration (1.174 sec n=1) and was frequency modulated. ‘Growls’ and ‘blow bursts’ (Figures. 4.18 & 4.19 respectively) were continuous chaotic (noisy), broadband calls with no frequency modulation. While ‘growls’ revealed no harmonics, the ‘blow bursts’ were spectrally coherent with some harmonics evident and were often interspersed with ‘staccato’ pulses. Most of the energy in the ‘growl’ extended from 100 Hz to around 1100 Hz with a concentration of energy below 200 Hz (Figure. 4.18A & B). ‘Blow bursts’ had a distinctive sound, similar to someone blowing vigorously into a glass jar. Most of the energy range in this call was higher than ‘growls’ extending from 100 Hz up to around 10 kHz with a concentration of energy below 300 Hz (Figure. 4.19A & B). For both chaotic calls, the low frequency concentration of energy was so intense it overloaded the recording equipment at the settings used for all the complex, periodic calls. ‘Blow bursts’ had a sudden onset and finish while ‘growls’ had a slow onset and finish with a build up to intense energy in the middle of the call.

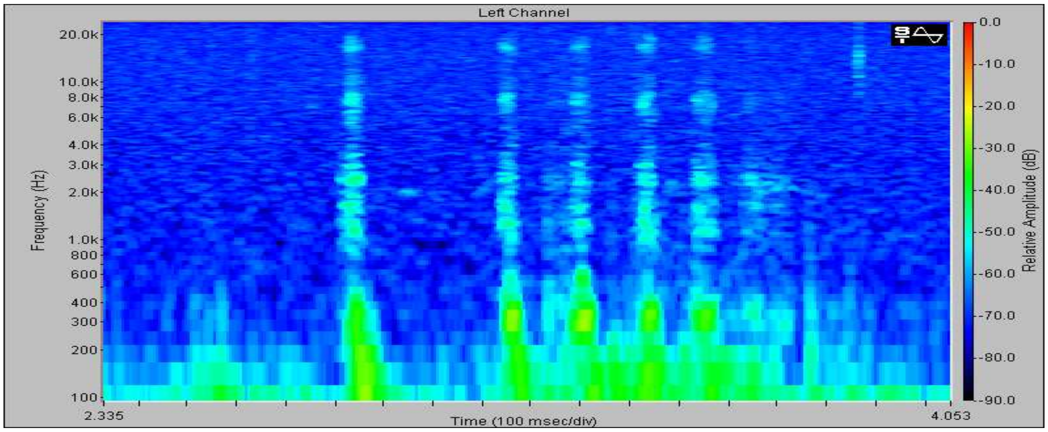
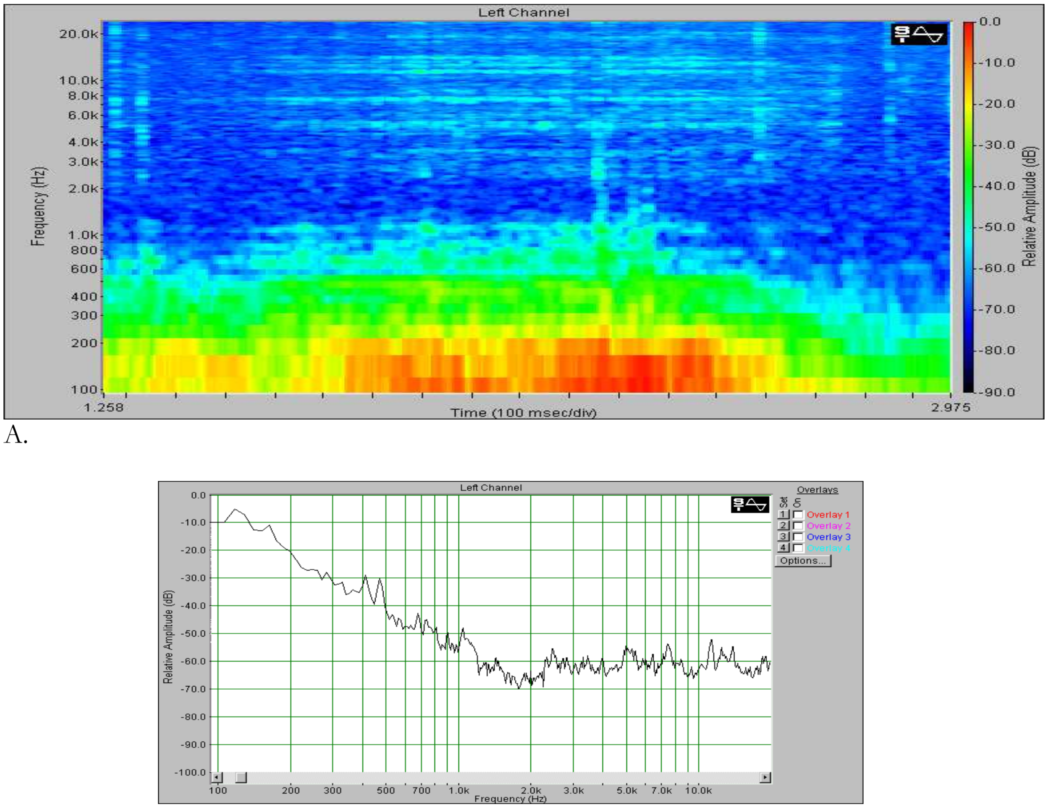


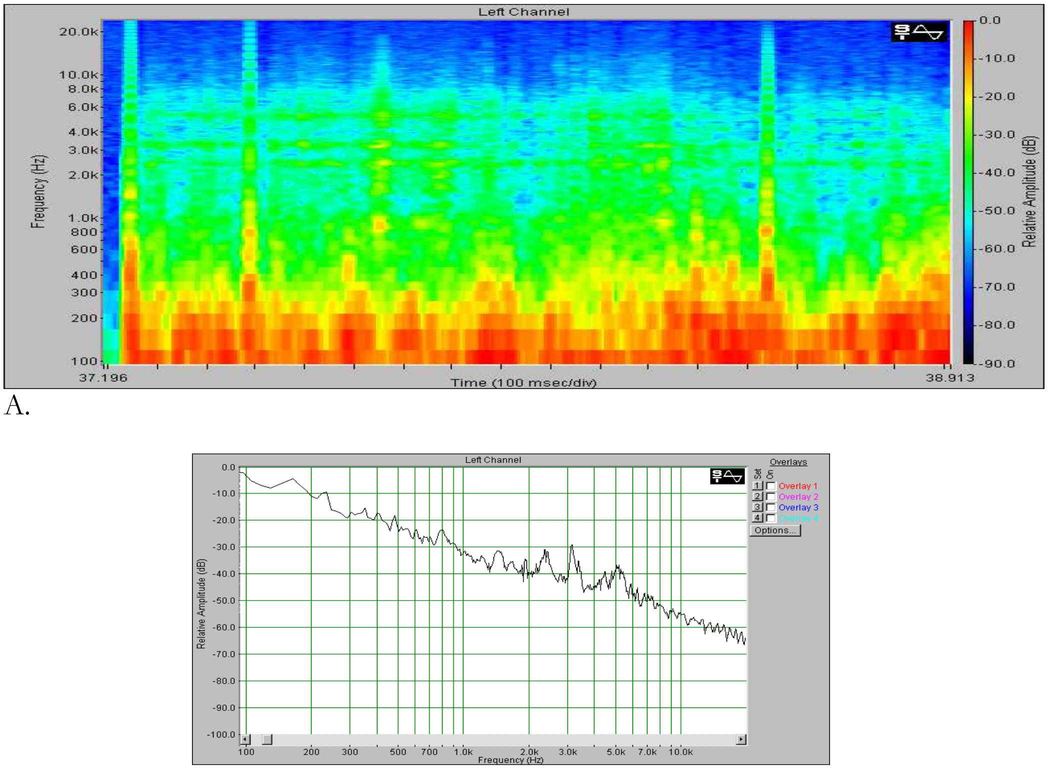
Figure.4.17. A series of 'grunts' produced by the Pinweryning Dam 'females with follicles' (CL 19.95-23.05cm). 'Grunts' occurred singly or could be heard forming a staccato-like effect. Recorded September 29th, 2004. Water temperature 13°C.



A.

B.

Figure.4.18. A. Spectrogram of a 'growl' recorded in the artificial ponds by the Pinweryning Dam large female group (CL 24.21-28.24cm). 'Growls' are noisy unstructured sounds. **B.** Amplitude spectrum of the 'growl' depicted in A, revealing the intense energy at frequencies below 250 Hz. (Amplitude spectra: FFT size 4096 points, overlap 75%). Recorded October 28th, 2004. Water temperature 23.5°C.



B.

Figure.4.19. **A.** Spectrogram of a 'blow burst' by the Pinweryning Dam large female group (CL 24.21-28.24cm) which was recorded in the artificial ponds. 'Blow bursts' are noisy but spectrally coherent sounds **B.** Amplitude spectrum of the 'blow burst' depicted in A, revealing the intense energy at frequencies below 300 Hz. (Amplitude spectra: FFT size 4096 points, overlap 75%). Recorded October 28th, 2004. Water temperature 23.5°C.

On one occasion, large females were recorded producing three 'growl squawks' (Figure. 4.20). Calls began with rapid downsweeps (periodic), with an abrupt transition to aperiodic chaos in the main body of the call. This noisy but coherently structured burst finished abruptly and ranged from 100 Hz up to around 1194 Hz. Each call was progressively longer by about 1 second (1.047 sec, 2.023 sec, 3.162 sec) and was separated by 21.638 secs (SD=1.835, n=2).

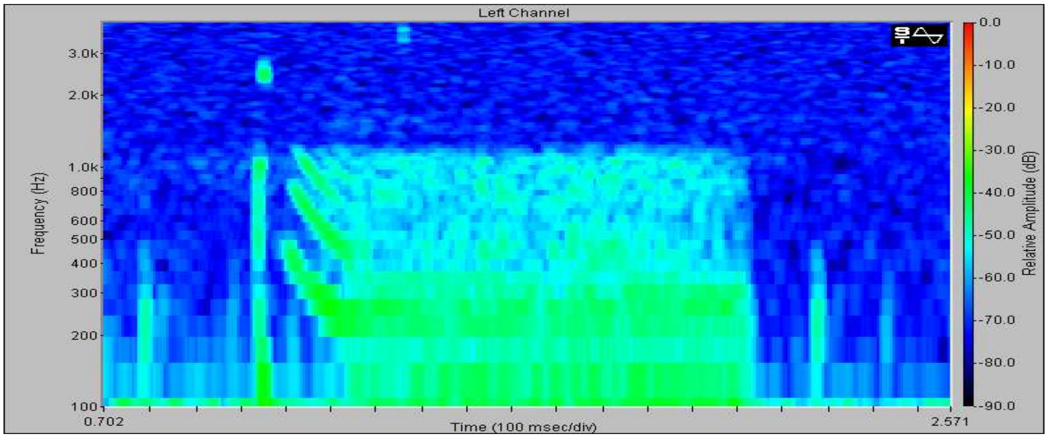


Figure.4.20. Spectrogram of a 'growl squawk' by the Pinweryning Dam large female group (CL 24.21-28.24cm) which was recorded in the artificial ponds. 'Growly squawks' began with rapid downsweep elements finishing in a noisy but spectrally coherent burst. Recorded November 12th, 2004. Water temperature 20ºC.

'Wild Howl'

The 'wild howl' call was the longest of all calls (lasting 10.263 sec) (excluding bouts) recorded for *C. oblonga* and was only recorded on one occasion by Pinweryning Dam females (CL: 19.95-23.05cm). This call dominated the frequency spectrum from 100 Hz to 3 kHz and consisted of: 1.) A number of high energy pulses ('grunts') (Figures. 4.21#1 &#2) occurring at the beginning of this call; 2.) A 'howl' at time 7.4 - 8.0 seconds (Figure. 4.21#3); and 3.) A 'growling rattle' (sounding similar to a lion) at time 8.8 – 11.2 seconds (Figure. 4.21#4). The 'howl' had a dominant frequency at about 280 Hz with a lesser peak at about 610 Hz. The 'howl' was richly harmonic revealing at least 7 harmonically related elements and frequency modulation. The 'growling rattle' had dominant frequencies below 134 Hz with lesser spectral peaks at 638 Hz and 1835 Hz.

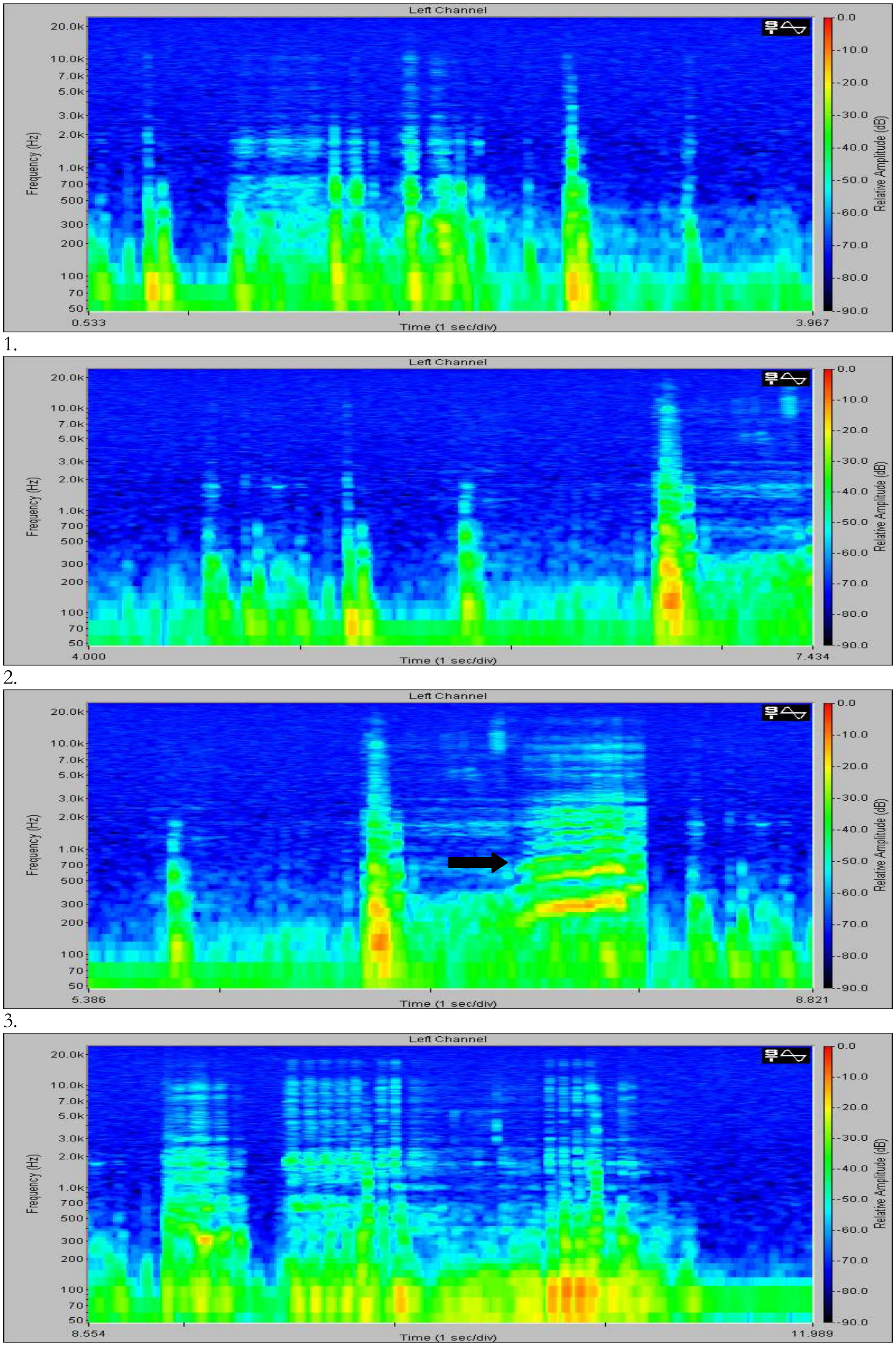
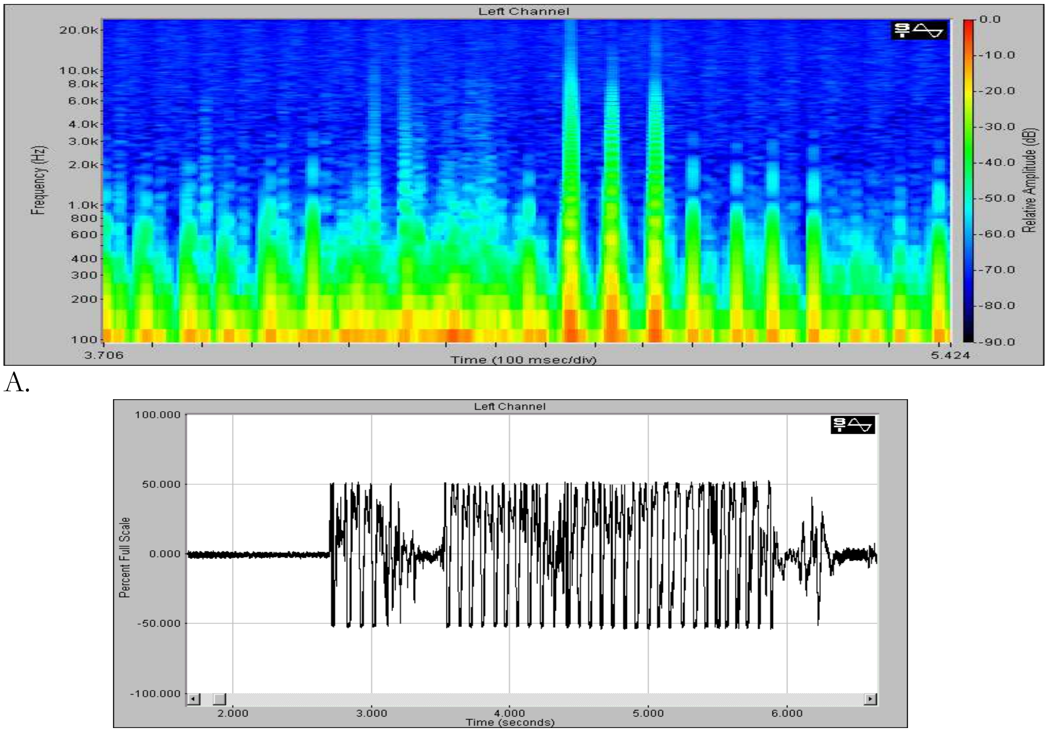


Figure.4.21. A sequence of spectrograms of the 'Wild howl' by a female from the Pinweryning Dam population. #1 & #2 reveal the grunts produced in the early part of the call finishing with the 'Howl' in spectrogram #3 at time 7.4 – 8.0 seconds followed by the 'growling rattle' in spectrogram #4 at time 8.8 – 11.2 seconds. Dawn recording, September 29th, 2004. (FFT 2048 points, averaging of 4 & 75% overlap). Water temperature 13°C.

'Staccatos'

'Staccatos' consisted of a series of rapid, 'thump-like' pulses producing a sound considered more 'mechanical' than biological. Thump pulses occurred singly or typically in short series (see the 'Wild howl' Figure. 4.21 spectrogram #1 at time 1.7 – 2.5 seconds). The longest and most distinctive bout of pulses produced a 'staccato' lasting 6.225 seconds – part of which is revealed in Figure 4.22A. Generally, 'staccatos' had low dominant frequencies below 117 Hz. Most pulses in the 'staccato' extended up to or below 1 kHz, but in the three pulses extending up to around 10 kHz (at time 4.6-4.8 sec), dominant frequencies in these pulses were up to 210 Hz. Similar to the 'growls' and 'blow bursts', the output at the dominant frequencies resulted in clipping of the waveform (Figure. 4.22B) at the settings used for all other vocalisations.



A.

B.

Figure.4.22. A. Spectrogram of the most distinctive 'staccato' which was recorded in the juvenile pond that an adult male had inadvertently made his way into. Greatest output is evident in the lower frequencies up to around 210 Hz. **B.** Waveform characteristics of 'staccato' at time 2.7 – 6.3 seconds. Waveform is clipped due to the output in the lower frequencies. Dawn recording, September 29th, 2004. Water temperature 16°C.

‘Drum rolls’

‘Drum rolls’ consisted of coherent and repetitive ‘rolling-pulse-like’ sounds occurring as a bout of five separate rolls in both Pinweryning Dam and Blue Gum males (Figure. 4.23 & 4.24 respectively). The ‘drum roll bout’ in Pinweryning Dam males lasted around 1 minute but was longer in Blue Gum males, lasting around 5 minutes. Pinweryning Dam ‘drum rolls’ lasted on average 2.717 seconds (SD=1.288, n=5) whereas Blue Gum Lake ‘drum rolls’ were slightly longer with an average duration of 2.717 (SD=1.288, n=5). In both bouts, separation between first and second ‘drum rolls’ was brief (Pinweryning Dam = 0.762 secs; Blue Gum Lake=2.8 secs). For the remaining ‘drum rolls’, average time between ‘drum rolls’ in the Pinweryning Dam males was 15.846 seconds (SD=9.846, n=3) but in Blue Gum males, this was longer (as it was spread over 5 minutes) with a ‘drum roll’ on average every 55.155 seconds (SD=37.840, n=3). A doublet pattern was evident in the ‘drum rolls’ of the Pinweryning Dam males (Figure. 4.23) but individual pulses could not be counted, whereas Blue Gum male ‘drum rolls’ had an average of 24 pulses per ‘drum roll’ (SD=6.033, n=5). Both ‘drum roll’ patterns had dominant frequencies below 210 Hz, but frequency range was greater in the Blue Gum male ‘drum rolls’ extending from 100 – 750 Hz, with Pinweryning Dam male ‘drum rolls’ ranging from 100 – 346 Hz.

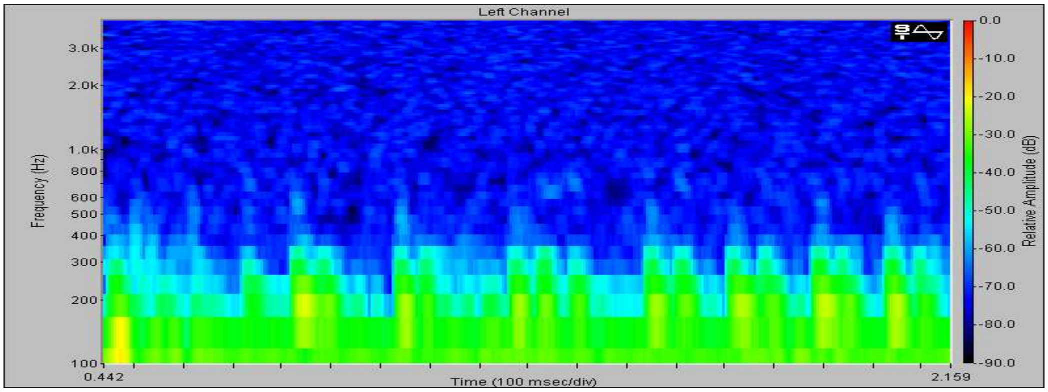
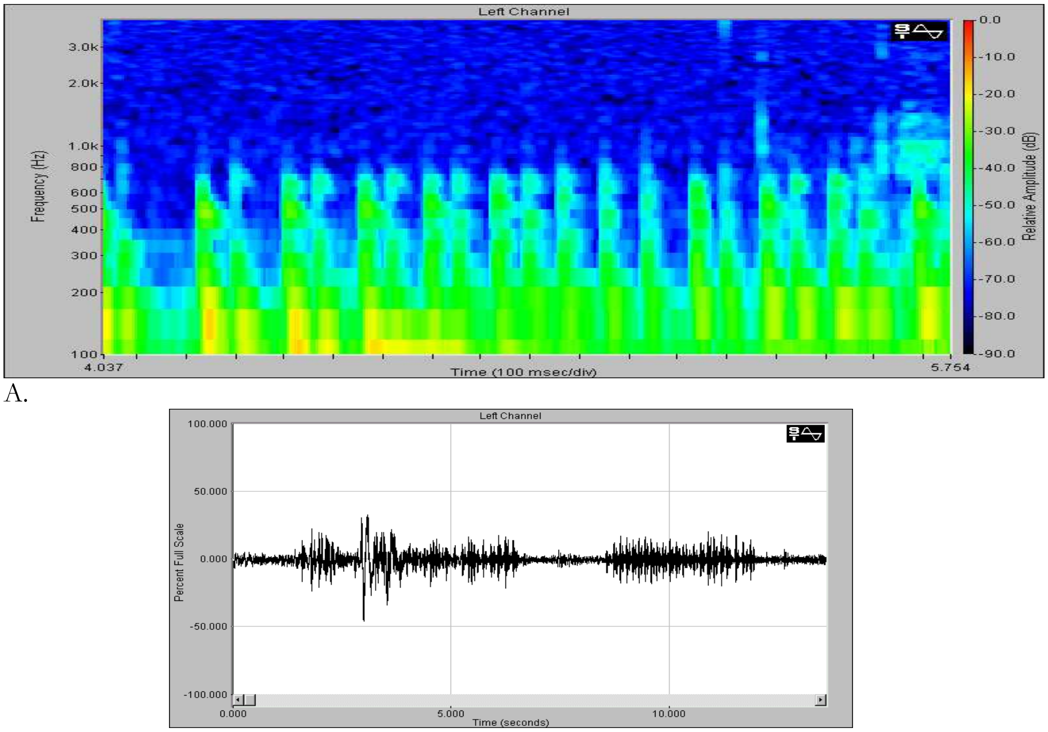


Figure.4.23. Spectrogram showing a short section of the ‘drum rolls’ recorded in the Pinweryning Dam large male group (CL:20.5-22.6cm). Recorded July 22nd at 6.20pm. Water temperature 15°C.

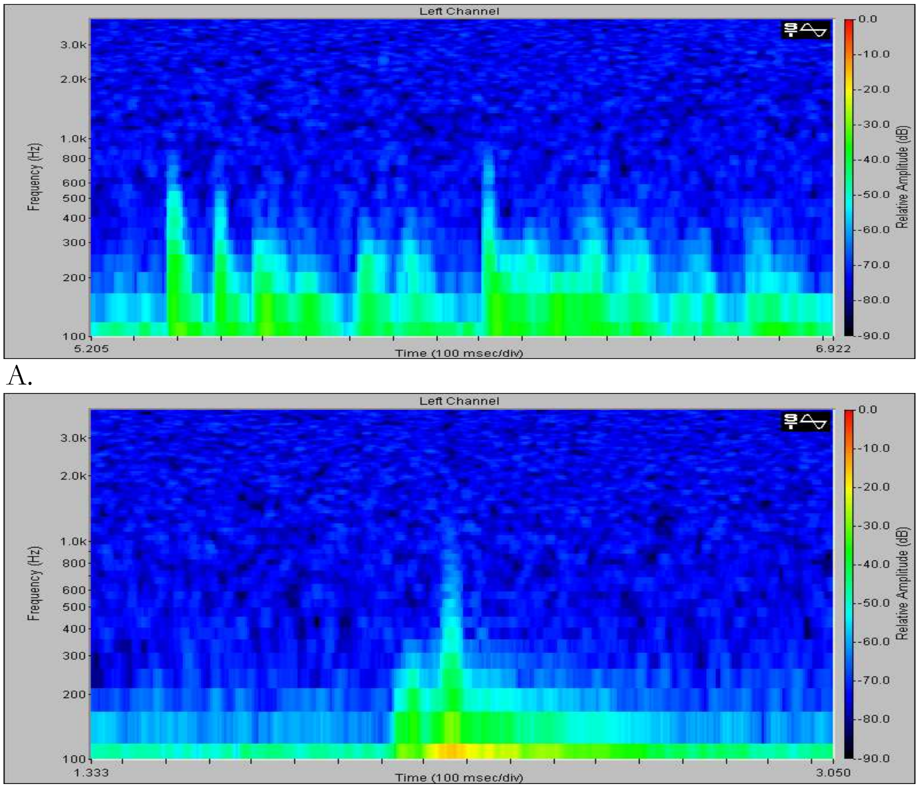


A.

B.

Figure.4.24. **A**. Spectrogram of a short section of the first ‘drum roll’ from a bout lasting just over 5 minutes by Blue Gum Lake males. **B**. Waveform character of the first and second ‘drum roll’. Recorded January 12th, 2005 at 4.30pm. Water temperature 28°C.

On occasion, Pinweryning Dam females produced short ‘drum rolls’ during July and August. Female ‘drum rolls’ were less distinct than the long male ‘drum rolls’ and typically lasted < 2 seconds. The ‘drum roll’ of Figure 4.25A was distinctive for its rhythm and echoing drum beat sound, while the ‘drum roll’ of Figure 4.25B consisted of a bout of five brief ‘drum rolls’ (which sounded like ‘ker-thump-thump’). Frequency range extended from around 100 Hz to 515 Hz with dominant frequencies below 164 Hz.



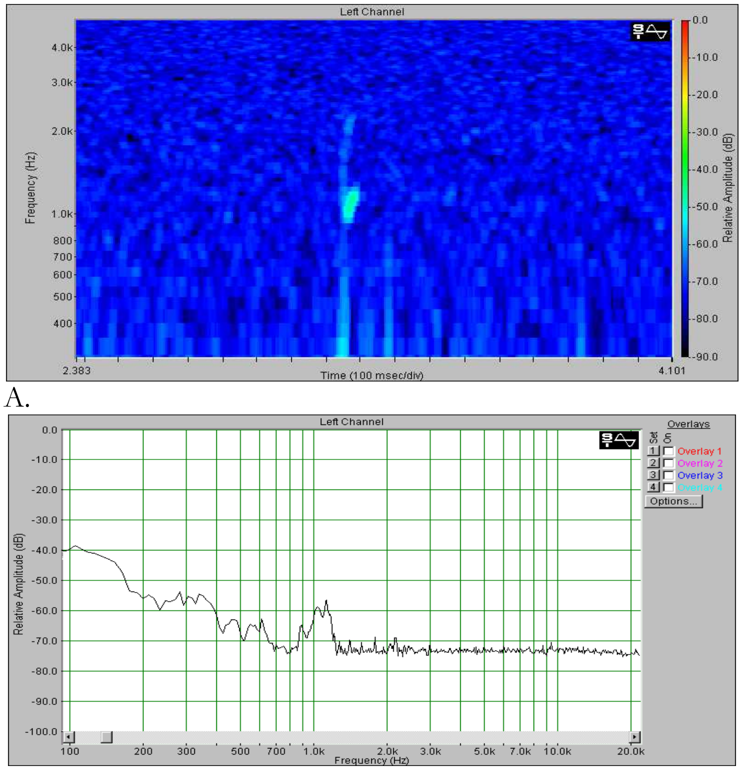
A.

B.

Figure.4.25. A. Spectrogram showing the short 'drum roll' produced by a large female from Pinweryning Dam (CL 24.21cm – 28.24cm). Recorded on August 28th, 2004 at 1.50pm. Water temperature 11.5°C. **B.** Spectrogram of a 'Drum roll' from a bout of five rolls produced by a smaller Pinweryning Dam female (CL 19.95cm – 23.05cm). Recorded on August 29th, 2004 at 5.30pm. Water temperature 9°C.

Juvenile Calls

There were no discernable vocalisations emitted by hatchling turtles. Juveniles (<10cm) rarely vocalised using complex calls. Only a single short chirp of brief duration (0.049 sec) was recorded (Figure. 4.26A) and consisted of a rapid up-sweep and no harmonics. The dominant frequency was around 1113 Hz (Figure. 4.26B). 'Staccatos' were however, a prevalent call and occurred in single pulses or short series and had a similar structure to the adult 'staccato'.

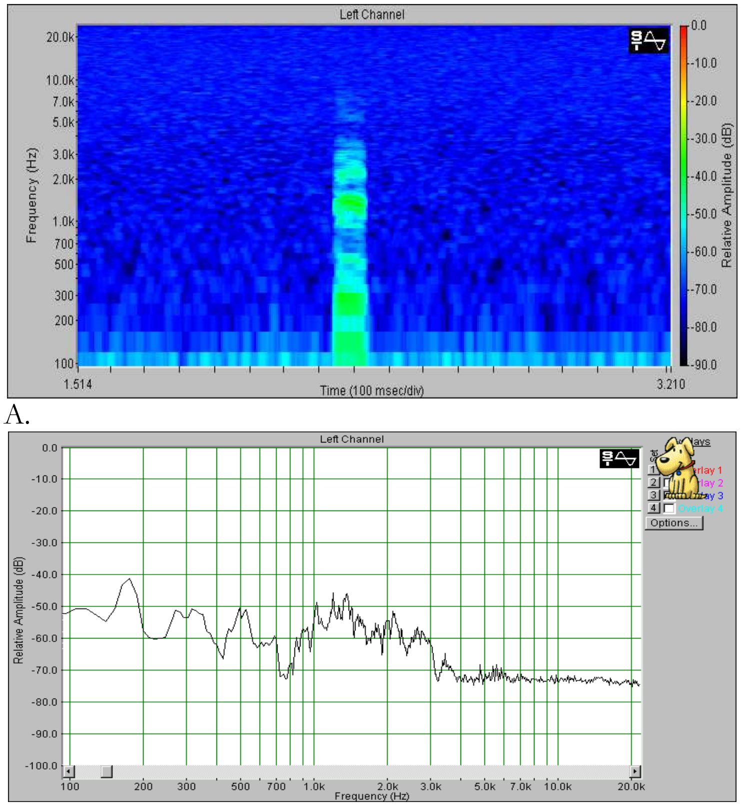


A.

B.

Figure.4.26. A. Spectrogram of a single chirp produced by juvenile turtles (CL:<10cm), consisting of a short up-sweep signal of short duration. **B.** Amplitude spectra of the juvenile 'short chirp' revealing a dominant peak at around 1113 Hz (Amplitude spectra: FFT size 4096 points, overlap 75%). Recorded on November 29th, 2004. Water temperature 22 °C.

The 'Duck honk' call was recorded over a two-hour period produced by a single sub-adult male turtle (carapace length 11.3cm) in a separate below-ground pond. This call is a broadband, discordant sound ranging from around 100 Hz up to 3.15 kHz (Figure. 4.27A) and had an average duration of 0.108 sec (SD=0.015, n=11). Several spectral peaks are evident with a dominant frequency of around 175 Hz (Figure. 4.27B).



A.

B.

Figure.4.27. A. Spectrogram of the 'Duck honk' call produced by a sub-adult male turtle with a carapace length of 11.3 cm. **B.** Amplitude spectra of the sub-adult 'duck honk' revealing several spectral peaks (Amplitude spectra: FFT size 4096 points, overlap 75%). Recorded in a below ground pond, May 14th, 2004 at 9 pm. Water temperature 11°C.

In-air Vocalisations

Turtles were noted to occasionally vocalise at the waters surface. This occurred only at night (from dusk through to midnight) and sounded similar to the 'chirp' calls underwater. No recordings were made in this research of in-air vocalisations.

4.3.3 Cut-off Frequencies

The absolute cut-off frequencies were calculated (using formulas in Ch 2 General Methods; section 2.3.7) to give an estimate of the frequency below which sound will not propagate at each wetland (Table. 4.3). At Blue Gum Lake, the low maximum summer water depths (1.0m) combined with the sediment type (silt) has resulted in a high cut-off frequency (1237 Hz), nearly twice (1.8) the cut-off frequency calculated for Glen Brook Dam and Lake Leschenaultia for a similar depth. In winter, when water depths were deeper, cut-off frequencies were lower than summer cut-off frequencies which was expected; but cut-off frequencies remain relatively high at Blue Gum Lake (601 Hz) due to a shallow maximum depth combined with the silt sediment. Cut-off frequencies were particularly low at Glen Brook Dam and Lake Leschenaultia (83 Hz) due to deeper water depths available and their gravel sediments.

Table.4.3. Estimates of the cut-off frequencies calculated in the wetlands for summer and winter using two water depths available for calling activity.

	CUT-OFF FREQUENCIES (Hz)	
	Summer	**Winter**
LAKE LESCHENAULTIA	677.4 Hz (d=1.0m)	82.9 Hz (d=8.0m)
GLEN BROOK DAM	677.4 Hz (d=1.0m) (T=24°C, d=1.0m max)	82.9 Hz (d=8.0m) (T=15°C, d=8.0m max)
BLUE GUM LAKE	1236.75 Hz (T=28°C, d=1.0m max)	601.4 Hz (T=15°C, d=2.0m max)

4.4 Discussion

Chelodina oblonga utilises an underwater repertoire of complex and percussive sounds with short, medium and potentially long-range propagation characteristics. This species is not a 'vocal specialist', but rather, they were intermittent callers. Seventeen (17) vocal categories were recorded for *C. oblonga*, but while this number of categories is not as extensive as some of the more social aquatic mammalian groups, e.g. Harp seals (*Pagophilus groenlandic*) with at least 27 underwater and two in-air calls described (Serrano, 2001); the turtle vocal repertoire was however extensive – particularly so for reptilians as a group. Such an extensive repertoire is suggestive of complex social organisation. Frequency use was broad with a number of calls extending beyond the current in-air sensitivity measured for a freshwater turtle from the family Chelidae (*Platemys platycephala*) (see Wever, 1978). Frequency use started from around 100 Hz in some of the percussive displays, extending as high as 3.5 kHz in some of the complex calls such as the 'high calls', with 'clicks' extending beyond the upper 20 kHz limit of the recording equipment. However, most turtle vocalisations had dominant frequencies below 1 kHz.

Transmission of Turtle Vocalisations

Vocalisation frequencies are most likely to be influenced by transmission characteristics of the environment (see Marten and Marler, 1977; Bradbury and Vehrencamp, 1998; Tyack, 2001) and sediment types, sediment gas, temperature and water depths would be expected to be important for propagation of turtle vocalisations (see Anderson and Hampton, 1980a,b; Forrest *et al.,* 1993). The prevalence of low dominant frequencies in turtle vocalisations would mean wavelengths at these frequencies would be longer relative to water depths in some wetlands (e.g. using calculated summer sound speed from Ch 2 General Methods 2.3.7; v=1508.4 m/s, f=700 Hz gives a wavelength of around 2.15 m). In Lake Leschenaultia and Glen Brook Dam, with their predominantly gravel and sand sediments combined with water depths up to 8-9 metres; the estimated cut-off frequencies (at 8m=82.9 Hz) would enable the transmission of all turtle vocalisations (e.g. using summer sound speed of v=1497.84 m/s and one of the lower dominant frequency in the repertoire f=200 Hz will give a wavelength of 7.5 m). However, turtles vocalising at shallower depths (0.2m - 1m) in wetlands would find the usable frequency range restricted to higher frequencies (see Forrest *et al.,* 1993; Boatright-

Horowitz *et al.,* 1999). In contrast though, an enriched wetland such as Blue Gum Lake, with gas-rich and predominately sand/silt sediments, combined with very shallow water depths (1-2m maximum levels during winter/spring and shallower in summer); would represent a more problematic environment for communication in turtle populations. In mud sediments, even despite the characteristics of the underlying substrate (see Jones *et al.,* 1964), attenuation increases, particularly at relatively high frequencies (and also increases with increasing sea state particularly for frequencies 400 Hz and above) (Marsh and Schulkin, 1962;). Also, gas in sediments forms a highly reflective boundary resulting in phase reversal and alterations to the sound speed in sediments (Anderson and Hampton, 1980b). In the absence of empirical data, the cut-off frequencies in Blue Gum Lake were estimated at 1237 Hz in summer and 601 Hz in winter. The shallow-water of Blue Gum Lake acts as a high-pass filter (Forrest, 1994) and only the higher frequency calls will propagate readily. While turtles may be able to alter the propagation of their call by moving into deeper or shallower water when they signal; cut-off frequencies are still determined by the shallowest position of the sender or the receiver (Forrest, 1994) and in summer, deeper water levels may not be available to turtles to enable propagation of the lower frequency elements in their calls.

From the signal characteristics in the complex turtle calls i.e. the ‘chirp’ calls which are considered to be their main contact call; it appears that turtles may have already accounted for high cut-off transmission characteristics in their environment. ‘Chirp’ calls were harmonically structured which would enable transmission of at least part of the call (i.e. the higher frequency components) if the lower frequencies weren’t able to propagate due to insufficient water depths. In addition, Wever (1978) found the auditory sensitivity of the turtle ear to be a function of head temperature, with a maximum response seen at a head temperature of around 30°C. This may indicate that turtles have heightened sensitivity to auditory signals during the summer months when water temperatures are particularly high (maximum recorded temperature during summer was 33°C), and conditions are not optimal for sound transmission of lower frequencies. The higher harmonics in the complex calls, which are the components in the call that would be transmitted in the shallow water; were usually at lower amplitudes than the first harmonic, but this heightened auditory sensitivity may be enabling turtles to hear the low amplitude higher harmonics.

Wetlands are acoustically complex environments and factors such as rocks, logs, trees and submerged /emergent vegetation would likely contribute to degradation of signal characteristics (see Wiley and Richards, 1977; Forrest, 1994; Edds-Walton, 1997). However, wetlands have many open water habitats and as turtles are very mobile animals, they could utilise open water areas to reduce degradation of signal characteristics. With some of the limitations imposed on the propagation of turtle vocalisations within a wetland, it might be expected that most turtle communications would occur at relatively close range. Signal characteristics of the short and medium 'chirp' calls suggest that these are likely to be relatively short to medium range calls with the 'longer chirps' with the distinct frequency modulated elements possibly intended for communications further afield (although empirical measurements need to be undertaken to determine these distances). Frequency modulation is used to encode information and is a useful means to transmit information over longer distances. Frequency modulation also produces a signal pattern that is dissimilar to the background noise making it stand out (Wiley and Richards, 1978). For the calls with predominantly low frequency components – these were potentially long-range calls (again empirical measurements need to be undertaken to determine how far 'long-range' actually is). Although, for some animals e.g. the Oyster toadfish (*Opsanus tau* L.,) (Fine and Lenhardt, 1983), their communication is thought to take place only over small distances despite the energy and frequency used in their call. While this may be the case for some of *C. oblonga's* calls, for example, the 'blow bursts' were considered for close range interaction, this was not considered to be the *modus operandi* for all *C. oblonga's* predominantly low frequency calls. In particular, as this species appears to be located throughout all parts of a wetland, it would make sense to be able to transmit a call over longer distances (e.g. Blue Gum Lake has a width of approximately 88m and a length of 450m, depending on rainfall) particularly during the mating season (see chapter 5) when water levels are deeper.

Turtles and Tortoises

Extensive vocal repertoires have not been revealed in the literature for those chelonians studied (or recorded) to date, but rather examples of a single type of call are mostly presented. McKeown *et al* (1990) were the only researchers to suggest the existence of an extensive repertoire within a single chelonian species i.e. *Manouria e. emys* with distinct differences between male and female vocalisations. For many of the chelonian calls presented in the literature, calls often extended above 1 kHz, but similar to *C. oblonga,* most of the energy in adult calls were below 1 kHz - including the in-air calls from a single Leatherback turtle (*Dermochelys coriacea)* (Mrosovsky, 1972). Examples of calls included those from: *Gopherus agassizii* (Campbell & Evans, 1967), *Geochelone radiata* (Auffenberg, 1978), *Geochelone elephantopus* (Jackson and Awbrey, 1978). A number of unpublished chelonian calls, which were accessed from the internet (URL:http://www.tortoise.org/tortcall.html 7 .08.04) were analysed and also found to have most energy in their calls below 1 kHz e.g. the African spurred tortoise (*Geochelone sulcata*) (recording by S. Levine), the Red-footed and Yellow-footed tortoises

(*Geochelone carbonaria* and *Geochelone denticulate* respectively) (recordings by B. Morris), the Egyptian tortoise (*Testudo kleinmanni*) (recording by Michael. J. Connor) and the Bell's hinged-back tortoise (*Kinixys belliana nogueyi*) (recording by Brad Morris).

C. oblonga also produced calls where there was more energy above 1 kHz such as the clicks, 'short and high chirps', 'squawks' and the 'cat whines', but there are fewer examples of this occurring in the calls of other chelonians. The few calls mentioned included from: *Geochelone travancorica* and *Platysternon megacephalum* (Campbell & Evans, 1972) and the Chaco tortoise (*Geochelone chilensis*) (recording by Laura Rico) (URL:http//wwwtortoise.org/tortcall.html 7.08.04). Many of the complex calls produced by *C. oblonga* were harmonically structured and frequency modulated which was also evident in many of the terrestrial chelonian calls e.g. *Platysternon megacephalum* (Campbell & Evans, 1972) and *Testudo marginata* (Sacchi *et al.,* 2003) as well as those accessed from the internet (URL:http://www.tortoise.org/tortcall.html 7.08.04) e.g. the African spurred tortoise (*Geochelone sulcata*) (recording by S. Levine), Bell's hinged-back tortoise (*Kinixys belliana nogueyi*) (recording by Brad Morris) and the Chaco tortoise (*Geochelone chilensis*) (recording by Laura Rico). *C. oblonga* calls were generally sparsely harmonic containing from two to three harmonics, which was similar to a number of the chelonians mentioned above e.g. Bell's hinged-back tortoise (recording by Brad Morris) and the Chaco tortoise (recording by Laura Rico). However, 'RH long chirps' and 'hoots' were richly harmonic calls with around 6 and 11 harmonic elements respectively. This was comparable to calls produced by *Testudo marginata* (Sacchi *et al.,* 2003) which had up to 13 harmonics and a call by the African spurred tortoise (*Geochelone sulcata*) (recording by S. Levine) revealed the most harmonic elements at 15. Apart from *C. oblonga,* the only chelonian in the examples above shown to have frequency sweeps in their call was the Bell's hinged-back tortoise (*Kinixys belliana* *nogueyi*) (recording by Brad Morris), which had distinctive down-sweeps, sweeping from around 1 kHz to 428 Hz in 0.67 seconds.

Hearing in Long-necked, Freshwater Turtles

Like all turtles, *C. oblonga* has no external ear, but the tympanic membrane is visible and is distinguished by a small oval at the side of the head. The membrane forms the outer wall of the air-filled, tympanic cavity and lies closed forming a continuous surface of smooth skin (Wever, 1978; Legler, 1993).

Research by Wever (1978) revealed that the turtle ear is a well-developed organ. However, only in-air sensitivities are shown for one species within the family Chelidae (*Platemys platycephala*). These in-air sensitivities reveal that detection of sound was particularly good between the frequencies of 60 - 600 Hz with sensitivity dropping away steeply above 1500 Hz at a rate of around 50 dB per octave or more. In Cryptodires, hearing sensitivities reach their maximum between 100 to 700 Hz. Wever (1978) largely attributed the auditory competence in the lower frequencies to the amplifying ability of the inner ear and the large number of hair cells present. For the upper frequency limit of hearing, Manley (1990) considers the flexibility of the middle ear to be a major influence in reptiles generally. In recent investigations into the physiology of the ear; Fettiplace *et al.,* (2001) found that sound is detected by vibrations of hair bundles on the sensory hair cells in the cochlear. When these hair bundles move as a result of the force from ion channels opening and closing, Fettiplace *et al.,* (2001) suggest that these movements may play a role in the amplifying ability of the cochlear and perceive differences in frequency.

Chelodina oblonga are mostly aquatic animals – but leave the water to nest (females) and migrate between wetlands (males and females) (Burbidge, 1967). Wever (1978) suggested that the structure and nature of the turtle ear was a compromise between its ability to receive both aerial and aquatic sounds. Measurements made by Wever (1978) using the Common box turtle (*Terrapene c. carolina*) revealed this dual receiving ability, but this turtle had greater sensitivity for aerial sounds. While this turtle has a preference for damp areas such as floodplains, it was largely land-based (Burnie, 2001), and as Wever (1978) noted, the greater aerial sensitivity would not be unexpected. For *C. oblonga* though, given that it is largely aquatic, it might be expected to have a greater sensitivity in an aquatic setting, however this still needs to be tested for.

Sound Production and Individual Variation

There is a paucity of information on the sound producing mechanisms in reptiles generally. Although reptiles are known to vocalise using a modified glottis (Gans and Maderson, 1973), most reptiles, with the exception of the gekkonid lizards e.g. the Tokay gecko (*Gekko gecko*) (Moore *et al.,* 1991) which have true vocal cords; animals within the reptilian group are not known to have true vocal cords but membranous folds housed in a cartilaginous larynx (Schumacher, 1973). Use of sound underwater maybe a more widespread phenomena in Australian long-necked turtles than is realised. Molecular analysis indicates a monophyletic lineage for Australian chelids, with *Chelodina oblonga* revealed as being closely related to the long-necked forms in the Chelodina longicollis group (i.e. *canni, novaeguineae, longicollis, steindachneri, reimanni, pritchardi*) (Seddon *et al.,* 1997; Georges *et al.,* 1999; Georges per. Comm., 2005). Morphologically, *C. oblonga* shares the characteristic long, thick neck found in the macrochelodina group - *Chelodina rugosa* and its relatives (*parkeri, burrungandjii, expansa*, etc). Given the close relationships (at a molecular level or morphological level)(Georges and Kuchling, pers. comm.), it is highly likely these animals would share similarities in the morphology of sound producing mechanisms with *C. oblonga* and therefore, highly possible that they would also vocalise.

A closed mouth appeared to be used in the underwater vocalisations of *C. oblonga*. While it was usually difficult to determine which animal produced a call (particularly when recording animals in a group), on the few occasions when a 'chirp' call was emitted and the turtle was very close to the hydrophone (with the call sounding close and loud), the call appeared to be produced internally without accompanying open mouth movement. Using a closed mouth in sound production may not be unusual in chelonians as Campbell and Evans (1967) noted the mouth was closed when vocalisations were produced by the tortoise *G. agassizzi*, but in both *G. agassizzi* and *G. carbonaria*, vocalisations were accompanied by gular pumping. Gular pumping was often observed in *C. oblonga* when they surfaced, but no acoustic emissions were recorded at these times. There is some evidence that airflow from buccal pumping does not contribute to lung inflation in freshwater turtles, but rather air only moves in and out of the buccal cavity (Druzisky and Brainerd, 2001) and so vibration of sound producing apparatus is unlikely to occur in this instance.

Many cetacean sounds (Popper, 1980) and hippo calls (Barklow, 2004) also appear to be produced internally without the need for constant resurfacing for air. Although airflow, or more specifically - pressurised airflow, is considered to be involved in sound production in cetaceans - including click production (Cranford *et al.,* 1996). This same air is then recycled (Norris *et al.,* 1971), negating the need for constant resurfacing. Some aquatic animals negate the need for air by using a percussive process e.g. the African Pipid Frog (*Xenopus borealis*) (Yager, 1992). However, due to the complexity found in the structure of turtle calls, airflow was considered to be used in the production of most turtle vocalisations (although no hypothesis is offered for 'click' production), and similar to the cetaceans (Popper, 1980) and pinnipeds (Tyack, 2001); turtle vocalisations were also considered to be produced in a 'closed system' as there was no expulsion of air, which would have been seen as bubbles and there was no evidence of repeated surfacing for air. Occasionally, airflow through the turtles respiratory system (the underwater sound of airflow) could be heard when turtles were breathing at the waters surface, which sounded similar to that heard in some calls e.g. the 'breathy' component in the Pinweryning Dam female 'high calls'. There were the occasional 'chirp-like' sounds emitted when turtles appeared to burp or hiccup, which were accompanied by an air bubble, but these are not included here as they were regarded as incidental rather than deliberate vocalisations.

Although three populations of turtles were recorded in this research, this was not considered to be enough to indicate whether the repertoire presented reflects the complete extent of *C. oblonga's* vocal ability. However, the large number of categories suggests that a considerable proportion of the vocal repertoire has been described. While inter-individual variation in vocalisations has been noted in some tortoises e.g. *Geochelone radiata* (Auffenberg, 1978) and *Testudo marginata* (Sacchi *et al.,* 2003), this was difficult to test for in *C. oblonga* as previously noted; when recording a group of animals, it was difficult to know which animal called – particularly in an underwater setting (see Tyack, 2001). Seven hours of recordings were made of a lone female attempting to test for inter-individuality, but unfortunately no vocalisations were emitted. However, the polymorphic nature of the 'chirps' was considered to reflect some of the inter-individual variation that would occur in any population of animals. Some calls were recognised as being distinctively similar and were probably called by a particular individual, which was thought to be the case for the 'RH long chirps' and 'high calls with the breathy'. 'High calls with the breathy' were only recorded in the large female group (CL 24.21-28.24cm) and the female calling may have had some respiratory obstruction which produced this typical 'breathy' at the end of her calls and thus made them easily recognised. The 'RH long chirps' were only recorded in the large male group (CL 20.55-22.62cm) and typically occurred in a long bout (more than 5 minutes). 'Chirp bouts' (medium or long chirps) were usually considered produced by a single turtle due to the repetition of structurally similar 'chirps'. Calls in at least one other species - *Testudo marginata* (Sacchi *et al.,* 2003), were noted to be highly stereotypical for an individual turtle.

Differences in vocalisations between large tortoises and smaller animals have been reported. Auffenberg (1978) found a smaller and younger adult male *Geochelone radiata* produced a less distinct call than those produced by a large adult male. In other reptiles; Tang et al., (2001) found larger Tokay gecko (*Gekko gecko*) produced signals of greater amplitude in their advertisement calls than the smaller animals and they also had a longer series of 'binotes'. In crocodilians, changes have been noted to occur in the structure of calls from juvenile to adult (Garrick and Lang, 1977; Herzog and Burghardt, 1977). Vocalisations produced by a single sub-adult male *C. oblonga* (11.3cm CL) were of shorter duration than most adult male vocalisations (apart from the pulses and clicks) and sounded harsher than adult calls. The sub-adult 'Duck honk' calls were a similar type of call to the 'squawks' and 'hoots' called by Masons Gardens turtles and as this population contained sub-adult animals, may have been the calls of immature turtles. However, unlike the vocal crocodilian hatchlings (Britton, 2001), no discernable vocalisations could be attributed to hatchling turtles, but juveniles (<10cm) were recorded producing a single 'short chirp'. The only juvenile turtle recorded to date vocalising, is *Geochelone carbonaria* (Campbell, 1967) which were said to produce a bout of 'clucks' as they moved about.

'Clicks'

The frequency range of the 'Clicks' extended beyond the upper and lower frequency limits of the recording / analysing equipment (100 Hz to 20 kHz). While there are known to be mismatches between hearing ability and sound production e.g. the Oyster toadfish (*Opsanus tau* L.,) (Fine, 1981) and apparently is not considered unusual and can be found in the acoustic repertoire for many species (Marler, 1977); it would be interesting to know whether *C. oblonga* can perceive the high frequencies in these clicks and whether there are alternative structures involved in sound reception. While a possible echolocatory ability in turtles was not investigated in this study; the short duration, broadband clicks and double clicks could be used for this function. Use of echo-location would provide another means by which *C. oblonga* could fully take advantage of its aquatic environment. However, not all clicks are used for echolocation. In marine mammals, clicks produced by California sea lions (*Zalophus californianus*) (Moore and Au, 1975) and the click-trains produced by Sperm whales (*Physeter macrocephalus*) (Watkins, 1980), are believed to play some social role. It was thought the 'clacks' would likely fulfill some social function for turtles rather than echolocation, given their waveform characteristics (i.e. no echo pulse) and low frequency range (1.4 kHz - 2.1 kHz).

Interestingly, turtle 'clicks' appear to share characteristics common with some terrestrial echolocating animals, in particular the double-click frequency pattern which is found in: bats e.g. *Rousettus* fruit bats (Pye and Langbauer, 1998); echolocating birds e.g. *Aerodramus* (also known as *Collocalia*) (Pye, 1980; Suthers and Hector, 1982) and

Steatornis (Pye, 1980; Suthers and Hector, 1985); and also reported in a caecilian (Thurow and Gould, 1977). However, the intraclick interval in the audible double click produced by turtles was considerably longer than the birds or bats (from 191-457ms) (cf. 18-25ms in swiflets and 20-30ms in *Rousettus*), but were comparable to the caecilian (203 ms). The *Rousettus* fruit bats click their tongues to produce double clicks of very brief duration and utilise a bandwidth ranging from less than 10 kHz to over 60 kHz (Pye and Langbauer, 1998). The greatest energy in the double click produced by swiftlets is between 2 kHz and 8 kHz similar to the turtles, with swiftlet clicks produced in the syrinx using airflow (Suthers and Hector, 1982). With frequencies even as low as 2-8 kHz, detection of small objects (1-3mm dia) can be achieved in air (Griffin and Thompson, 1982). Underwater, at frequencies used by turtles, gives wavelengths of around 75 cm at 2 kHz; or 7.5 cm at 20 kHz (Ch 2 General Methods; section 2.3.7: using the calculated sound speed of the water at 1508.4 m/s). Wavelengths underwater are around four and a half times longer than equivalent frequencies in air and wavelengths at these frequencies would merely sweep around the small invertebrates, which are prey items for *C. oblonga* (Woldring, unpublished) with very little reflection of sound (Bradbury and Vehrencamp, 1998).

Although turtle clicks have some of the characteristics necessary to counteract the effects of higher sound speed in water, i.e. pulses were of short duration (to ensure there is no overlap of the pulse-echo) and were broadband pulses (see Zbinden, 1985-6; Bradbury and Vehrencamp, 1998); clicks produced by *C. oblonga* were not at the high frequencies expected for use in prey detection at prey sizes of 1-5mm (see Tyack, 2001). However, at the frequencies used by *C. oblonga,* echolocation could be used to estimate certain features within the wetland environment, such as water depth and distance to the banks of the wetland or location of submerged logs / boulders. As turtles disperse between wetlands (Burbidge, 1967) (or did so prior to barriers imposed by urbanisation), it would be an advantage to the newly arrived turtle to obtain a rapid 3-D picture of its new surrounds. It would also be advantageous for use in habitat where visibility is restricted due to factors such as turbidity and tannin-staining both of which significantly attenuate light (Davis *et al.,* 1993). It would make sense to conserve energy for hunting rather than utilising it investigating terrain. While there may be some auditory cues as prey items move through the water (Hawkins and Myrberg, 1983), or cues from the underwater sound component of calls e.g. from frog calls (see Dudley and Rand, 1992), it is more likely that alternative structures either singly or in combination are involved in prey detection. The eyes are likely to be very important for visual cues but also barbels may be involved by detecting vibration stimuli (Hartline, 1967; Winokur, 1973) produced by the aquatic invertebrates within or on the waters surface where their struggling movements produce concentric wave patterns (Wilcox, 1988; Bleckmann, 1988).

The Aerial Platform for Vocalisations and Observations

While this research was restricted to the underwater vocalisations of *C. oblonga*, the in-air vocalisations were considered worthy of some comment. There were two main in-air sounds produced by turtles: 'hissing' and 'chirps'. 'Hissing' was a barely audible sound and has also been reported by Goode (1967), which he suggested was produced by a sharp exhalation of breath. 'Hissing' was a sound thought to be associated with distress / defence by the animal. This sound was only heard when turtles were first hand-captured and removed from the water and was often accompanied by swinging of the neck from side to side and inflation of the buccal-pharyngeal region.

In-air vocalisations was unexpected - despite hearing the in-air 'roar' in previous research (Giles, 2001), which was thought to be anomalous behaviour at the time. However, observations by Allen (1950) would suggest that this behaviour was otherwise as he observed frequent surface calling in the Suwannee terrapin (*Pseudemys floridana suwanniensis*). It appears though, that in-air vocalisations are not unusual in semi-aquatic vertebrates e.g. Harp seals (*Pagophilus groenlandicus*) (Serrano, 2001) and Hippos (*Hippopatmus amphibious*) (Barklow, 2004) and are even known in some fish species e.g. sciaenids (Fish, 1954). Both male and female *C. oblonga* used the surface for calling and aurally, these calls sounded similar to the complex 'chirp' calls heard underwater. 'Surface chirps' were an infrequent occurrence and were only heard during the spring and summer from dusk through to midnight time periods. Similar to the Suwannee terrapin (Allen, 1950), heads were held steeply when calling, but despite borrowing some image-intensifying equipment; it was unknown whether *C. oblonga* used an open-mouth in these calls and whether the tympanic membrane was exposed (They disappeared very quickly when a torch beam was directed on them). Suwannee terrapins use a closed mouth in their surface calls.

'Surface chirps' were only heard in the ponds, if they were called in the wetlands they would not have been heard due to extraneous urban noises, or in the case of the larger water-bodies such as Lake Leschenaultia or Glen Brook Dam, 'surface chirps' would unlikely be heard from shore. As the water's surface is an excellent reflector of sound, effectively resulting in an increase in the distance of propagation of a signal compared to free-field signaling (Forrest, 1994), it would be an efficient means for turtles to communicate with other surfacing conspecifics in conditions of limited visibility. Turtles were also recorded producing a 'short chirp' directly beneath the waters surface just prior to surfacing (N.B. only a single animal was in view when these observations were made).

Amphibious communication, where animals are able to transmit and receive sound in both air and water simultaneously using impedance matching anatomical structures (Barklow, 2004), has been noted to occur in some semi-aquatic animals, e.g. Hippos (Barklow, 2004) and the Bullfrog frog *Rana catesbeiana* (Boatright-Horowitz *et al.,*1999). For example in Hippo's, Barklow (2004) explains the aerial component of a call is transmitted via the nasopharynx and out through the nostrils, with the underwater component passing from the larynx, through the throat fat and into the water. The external ears are used in receipt of an aerial call, but the lower-jaw bone is used in the underwater detection of sound. There are some studies suggesting the involvement of similar types of anatomical structures for sound conduction in chelonians. For example, transmission of audio-frequency vibrations through the bone carapace was found to occur in turtles (Lenhardt, 1982). However, in more recent investigations, magnetic resonance imaging (MRI) in sea turtles has revealed a column of fatty tissue connecting from the outer, middle and inner ears to the head which Ketten *et al* (1999) suggested may provide low impedance channels for the conduction of underwater sound. Whether amphibious communication is occurring in turtles is unknown, but warrants further investigation.

It is likely that use of the aerial platform is also important for the turtles to make visual observations. During the summer when recording from shore, many turtles were observed surfacing and remaining there for at least 30 seconds or more, appearing to watch a person standing onshore. They would then submerge and swim closer, resurface and observe again. As people occasionally throw bread in for the ducks -

which the turtles also eat, a human standing on shore probably represented a source of food. While these animals appear to be naturally curious and investigate new objects in their environment (personal obs), the constant resurfacing and watching would also enable a turtle to orient themselves and maintain their target direction. Holding the head vertical above the water is a behaviour observed in many Baleen Whales and is known as 'Spy-Hopping'. 'Spy-Hopping' is thought to be used for viewing other whales or landmarks (Burnie, 2001). In *C. oblonga*, usually only eyes and nostrils were exposed during the day when they surfaced, but at dusk and later at night, turtles extended a greater portion of their heads above the water holding the head steeply at an angle of 45° or greater. Interestingly, turtles were observed holding their heads above water during heavy periods of rain in the artificial ponds. This occurred during the winter months (beginning of the breeding season) and so may have been to view other surfacing turtles and thus locate mates. One female remained at the surface for over an hour. However, it can also be suggested that the turtles may have been driven to raise their heads above the surface to escape the white noise produced underwater by heavy rain.

Chapter 5.0 ACOUSTIC REPERTOIRE: SUSTAINED VOCALISATIONS

5.1 Introduction

From the random recordings that were made in the field and in the artificial ponds, a sustained turtle vocalisation consisting of numerous pulses was recorded once at Blue Gum Lake (2002) and once in the artificial ponds (2003) with both being recorded in the spring. These are presented here in a separate chapter because they were distinctive from the other turtle vocalisations due to their duration and rhythmic completion.

5.1.1 Objectives

1. Define and describe the elements in the sustained turtle vocalisations.
2. Determine if there were differences in the sustained vocalisations between performing adults from different populations.

5.2 Methods and Materials

To obtain other examples of these sustained turtle vocalisations, in the following year (2004), turtles from Pinweryning Dam were recorded from late May to November, 2004 (for details of this population see Chapter. 6.0).

5.3 Results

5.3.1 Summary of the sustained turtle vocalisations

Two displays of sustained vocalisations consisting of a bout of pulses were recorded in September 2002 at Blue Gum Lake and in October 2003 in the artificial ponds and have been summarised in Table 5.1. No sustained vocalisations were recorded in the Pinweryning Dam turtle population.

Table.5.1. Summary of sustained turtle vocalisations and their acoustic properties.

Vocalisation: Aural character	**Wetland**	**Spectral output**	**Duration of display (mins)**	**Frequency Range (kHz)**	**Dominant frequency (kHz)**	**Sex**
Rhythmic Knocks (n=1 'pulse-bout')	Blue Gum Lake	Pulsed several spectral peaks	3.8	0.200 – 2.00	Varied 0.800 - 1	unknown
Bongo Drums (n=1 'pulse-bout')	Artificial pond	Pulsed several spectral peaks	9.5	0.100 – 1.80	0.260 - 0.300	M

5.3.2 Descriptions

The following terms (Table. 5.2) have been used to define and describe elements of the sustained turtle vocalisations:

Table.5.2. Classification of the units in the sustained turtle vocalisations.

Pulse: Defines the individual component the sustained turtle vocalisation. Pulses ranged from 200 Hz to around 1.8 - 2 kHz often revealing three or more spectral peaks.

Inter-Pulse Interval (IPI): The inter-pulse interval refers to the time between the end of one pulse and the beginning of the next pulse in a pulse series, or between single pulses that had an interval of less than one second.

Pulse Series: Consists of a number of pulses ranging from two to 65, and were heard as a series of pulses separated by short intervals of silence. The inter-pulse-interval between pulses in a pulse series were usually irregularly spaced.

Silent Interval: This refers to the brief periods of silence separating single pulses or a pulse series from the next sequence of pulse(s). This period of silence usually ranged from one to eight seconds, but on a few occasions extended up to 35 seconds.
While the silent interval of one second was considered to be small, it was observed as a distinctive 'gap' between pulses on the spectral outputs.

Vibrato: Consists of a rapid series of pulses heard as a rhythmic percussive display appearing after the First Phase (after the single pulse and irregular pulse series). The '*Vibrato*' phrase denotes the Second Phase.

'Pulse-bout': This term encompasses the entire sequence of the sustained vocalisation (For definition of 'bout' see ch.4. Table 4.2).

5.3.3 The 'pulse-bout'

The 'pulse-bout' has been divided into 'First Phase' and 'Second' Phase. The two phases have then been further separated into four sections and these are listed and discussed below:

First Phase

1. The introductory stage. This is a single pulse /slow tempo stage.
2. The second stage has a fast tempo with minimal silent intervals.
3. The third stage has well defined shorter pulse series with silent intervals more often.

Second Phase

4. The fourth stage or *Vibrato* is the most rhythmic part of the 'pulse-bout'.

The 'pulse-bout'

The Blue Gum Lake 'pulse-bout' (henceforth known as the 'BGpb') was performed over almost four minutes (3.8 mins) and consisted of 210 individual pulses utilising a dominant frequency of around 886 Hz with lesser peaks at 461 Hz and 1645 Hz (Figure. 5.1A,B & C). The Masons Gardens 'pulse-bout' (and henceforth known as the 'MGpb') recorded in the artificial pond was a longer display and was performed over nine and half minutes, consisting of 817 individual pulses with a lower dominant frequency of around 278 Hz with lesser peaks at 800 Hz and 1892 Hz (Figure. 5.2A,B & C).

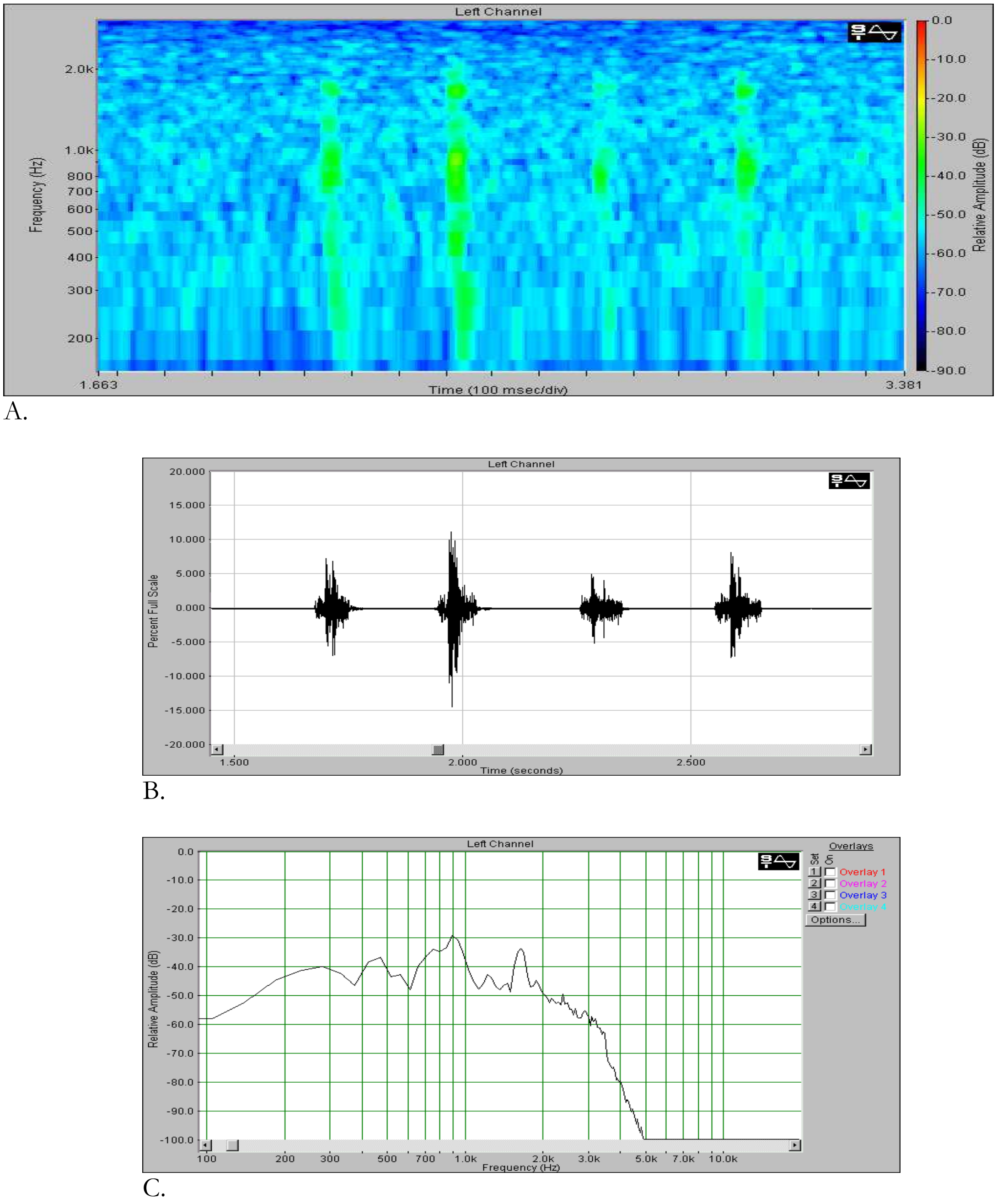


Figure.5.1. A. Spectrogram of typical pulses used in the ‘pulse-bout’ at Blue Gum Lake. **B.** Waveform characteristics of the four pulses in A. **C.** Amplitude spectra revealing dominant frequency at around 886 Hz, which is higher than the dominant frequency recorded for the male turtle from Masons Gardens. Recorded 6th September, 2002, 1 pm. Water temperature unknown. (All spectra: FFT 1024 points, 75% overlap. Digitally filtered using a Bessel pass band filter of 150 Hz to 3 kHz).

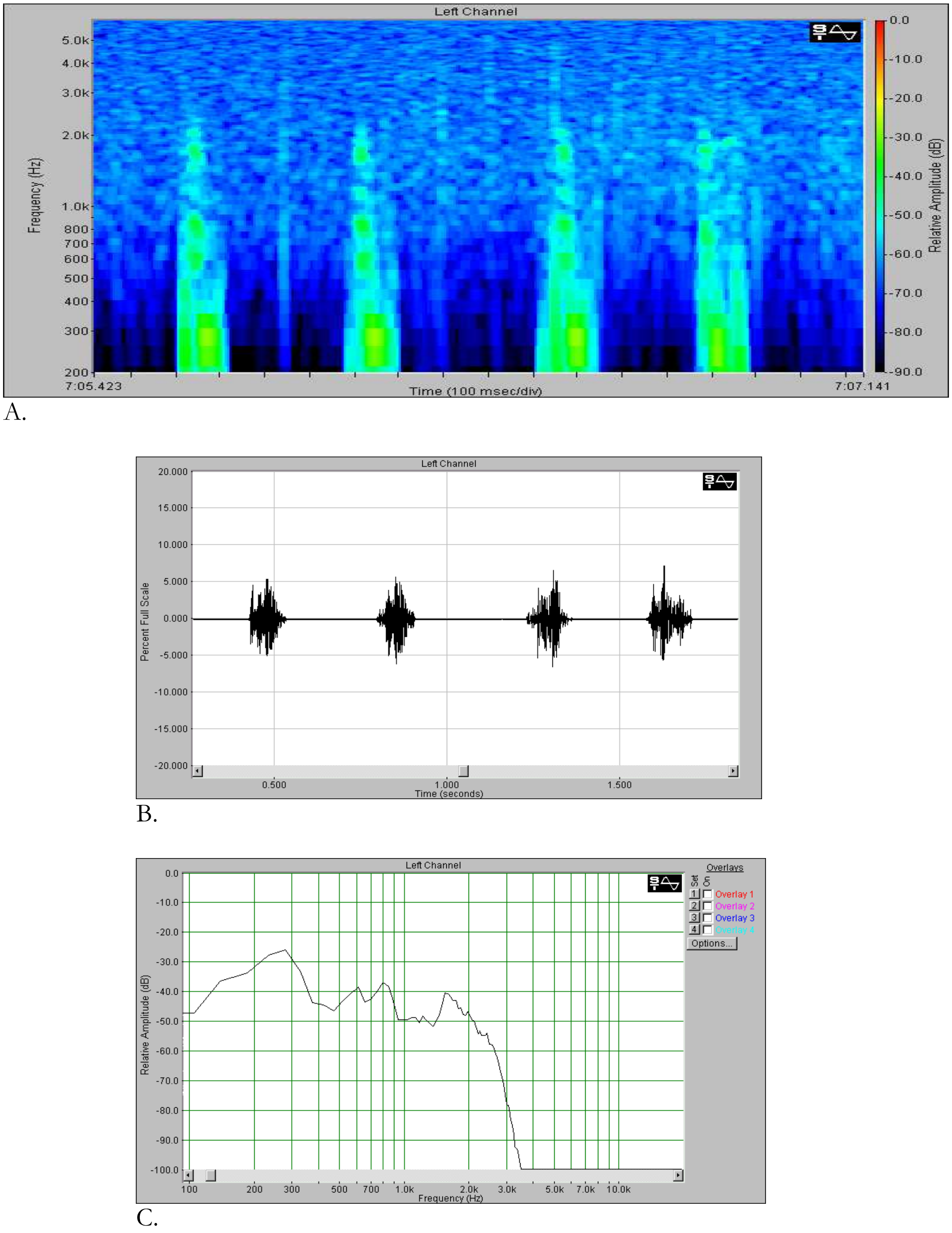


Figure.5.2. A. Spectrogram of typical pulses used in the 'pulse-bout' by the Masons Gardens male turtle. **B.** Waveform characteristics of pulses shown in A. **C.** Amplitude spectra revealing low dominant frequency at around 278 Hz. Recorded 20th October, 2003, at 5 pm. Water temperature 21°C. (All spectra: FFT 1024 points, 75% overlap. Digitally filtered using a Bessel pass band filter of 150 Hz to 3 kHz).

The First Phase

The BGpb began at the third stage of the first phase, consisting of pulse series with well-defined silent intervals. There was no introductory stage or fast tempo stage recorded in the BGpb. The introductory stage of the MGpb began with a number of single pulses (17) lasting a little over 44 seconds with each pulse being separated by silent intervals ranging from 1.3 seconds to almost 5 seconds (mean=2.7 sec, SD=1.6). In the MGpb, the second stage was differentiated by a marked change in tempo where the comparatively slow single pulses of the introductory stage were followed by a series of rapidly occurring irregular pulses (Figure. 5.3) (average IPI= 0.360 sec, SD=0.259). This stage was also the longest stage in the MGpb, continuing for nearly five minutes and could be considered more or less continuous. The brief silent intervals (n=34) averaged at 1.564 sec (SD=0.700) in duration with pulse series typically consisting of very long numbers of pulses, ranging from 7 to 65 pulses in a series. Throughout this rapid pulse period, doublets were occasionally heard which is seen in the last two pulses in Figure 5.3.

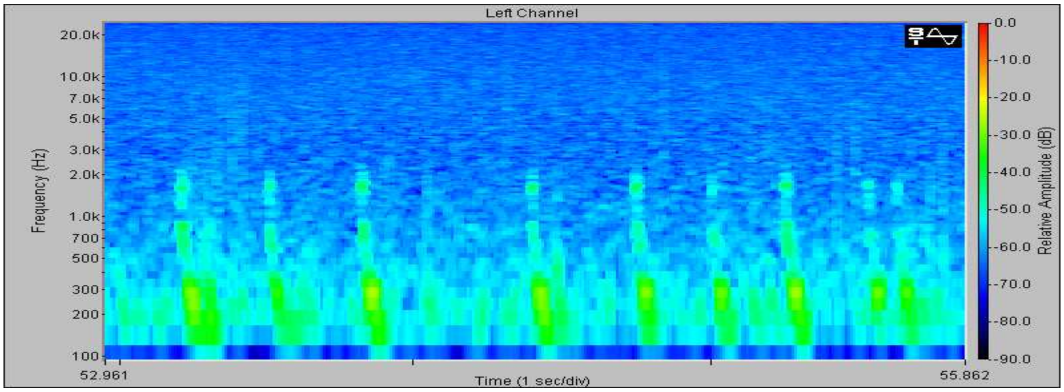


Figure.5.3. Spectrogram of an irregular pulse-series from the 'pulse-bout', typical of stage 2 in the first phase. The last two pulses are doublets, which were occasionally heard throughout phase 2 and 3 of the 'pulse-bout'. Recorded on the 26th October, 2003. Water temperature 21°C.

The third stage was differentiated by well-defined shorter pulse series combined with an increase in the number of silent intervals (24 silent intervals in nearly 1.5 minutes in the MGpb with 24 silent intervals in three minutes in the BGpb). The silent intervals in the MGpb were slightly longer in duration to those heard in the second stage (averaged at 2.009 sec; SD=1.035). Consequently, the number of pulses in a pulse series was much shorter than the second stage pulse series and ranged from 1-15 pulses but had a similar IPI average to the pulses in the pulse series in the second stage (0.394 sec, SD=0.204). By comparison, the mean IPI of this phase for the BGpb was 0.275 sec (SD=0.140), indicating a slightly quicker tempo. Doublets were also occasionally heard in this third stage for both acoustic displays.

The Second Phase

The second phase was the most rhythmic and complex of the 'pulse-bout' (Figures. 5.4 & 5.5). The tempo in this phase was more rapid than the first phase.

In the MGpb, the rhythmic displays (known as the '*Vibrato*'), occurred after almost 7.5 minutes of the first phase and lasted for two minutes, while the 'Vibrato' in the BGpb occurred after only three minutes of the first phase and lasted for less than one minute (59 secs). 'Vibrato's' in both 'pulse-bouts' continued until the end of the sustained vocalisation. In the MGpb, the identifying stage consisted of pulses occurring mainly as doublets, triplets and / or quadruplets with the mean time between pulses for this section being 0.172 sec (SD=0.145), which was very similar to the mean IPI for the BGpb of 0.201 sec (SD=0.162). However, the 'vibrato' for BGpb generally appeared as doublets and was heard as 'rocking pulses' with alternate pulses having different dominant frequencies i.e. the first, third and fifth pulses etc had dominant frequencies extending from around 548 Hz to 1030 Hz, while the second, fourth and sixth pulses etc had dominant frequencies extending from 760 Hz to 1184 Hz, which gave a 'rocking' sound to the pulse series (Figure. 5.4). The vibrato for the MGpb consisted of higher frequency elements in each pulse ‘rolling down’ from approximately 800 Hz to 620 Hz, and similarly, the lower frequency elements in each pulse rolling down from around 350 Hz to 155 Hz (Figure. 5.5). The silent intervals in the MGpb (n=26) were longer than those heard in the announcement phase with a mean interval of 3.281 seconds (SD=2.261) with the longest silent interval lasting more than eight seconds. The mean silent interval for the BGpb was similar in this section of the ‘pulse-bout’ to the MGpb silent intervals (3.115 seconds, SD=2.511) with around eight seconds also being the longest silent interval.

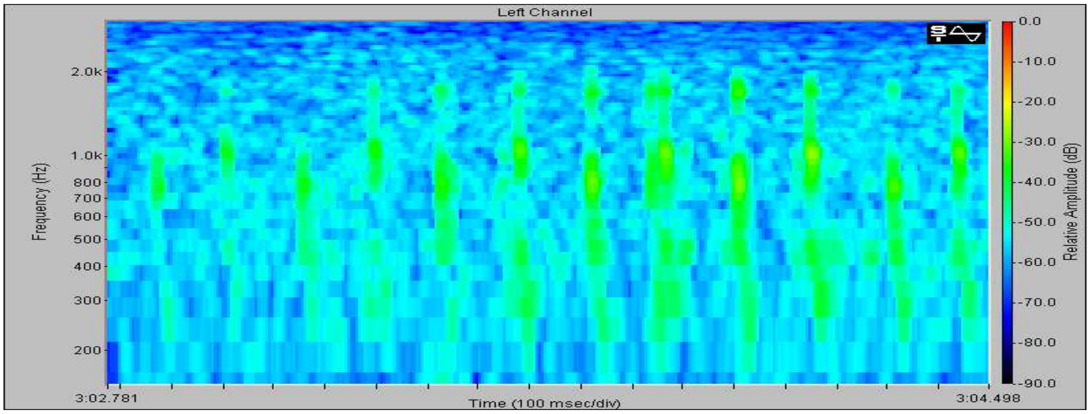


Figure.5.4. Spectrogram revealing a ‘rocking’ pulse series from the second phase of the sustained turtle vocalisation. Recorded at Blue Gum Lake on 6th September, 2002. Water temperature unknown.

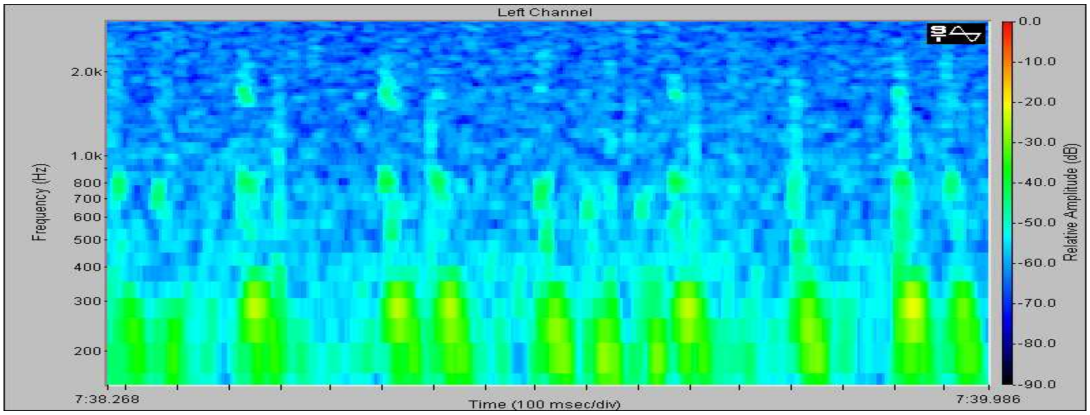


Figure.5.5 Spectrogram revealing the rhythmic pulse series or ‘*Vibrato’* from the second phase of a male turtle from Masons Gardens. Recorded 26th October, 2003. Water temperature 21°C.

5.4 Discussion

In addition to the short single calls or call bouts produced by *Chelodina oblonga,* this research has also revealed that these turtles are capable of producing sustained vocalisations lasting up to nine minutes or more. These sustained vocalisations were called 'pulse-bouts' and were divided into two phases. The 'First Phase' consisted of stereotypical single pulses and irregular pulse series. The 'Second Phase' consisted of a rhythmic display, known as the *Vibrato,* where pulses occurred in a rapid series of combinations of either doublets, roll-down triplets and quadruplets or in an alternating pattern of different frequencies. No female turtles were recorded producing a sustained vocalisation in this research, although their ability to do so cannot be discounted. Despite the opportunity to have a large sample of turtles from the same population (Pinweryning Dam) to record for 6 months during 2004 (which included the months corresponding to the mating season), neither male nor female turtles were recorded producing similar types of 'pulse-bouts' to those recorded at Blue Gum Lake or Mason's Gardens.

Reproductive advertisement displays are common in mammals, including acoustic advertisement displays which are heard in both terrestrial and marine mammals, with songs representing some of the most distinctive examples (Tyack, 1998; 2001). However, acoustic advertisement displays related to reproductive cycles in reptiles are rarely reported in the literature – only the Tokay gecko (*Gekko gecko*) (Tang *et al.,* 2001) appears to have been investigated. The acoustic advertisement displays in the Tokay gecko are seasonal and are coincident with a rise in androgen levels and gonadal masses (Tang *et al.,* 2001) and from this would also be called reproductive advertisement displays. Acoustic advertisement displays have not been reported in chelonians, with most vocalisations associated with reproductive activity only heard during mounting behaviour in the terrestrial chelonians (e.g. Bogert, 1960; Auffenberg, 1964; Campbell and Evans, 1967; Jackson and Awbrey, 1978; Sacchi *et al.,* 2003 and Galeotti *et al.,* 2004) with a single report of some pre-courtship vocalisations (McKeown *et al.,* 1990). As both of the turtle 'pulse-bouts' occurred within the breeding months (and not recorded outside this season), it is hypothesised that the turtle 'pulse-bouts' may function as an acoustic advertisement display. Interestingly the 'pulse-bouts' of *C. oblonga* appears to share a similar pattern as the acoustic advertisement display of the Tokay gecko. Tang *et al.,* (2001) described a two-phase arrangement in the advertisement display for this species which began with two to three simple multipulse sequences heard as rattles with the second phase more complex containing a series of doublets (4-11) (or referred to as 'binotes'). It is proposed that the two-phase approach to the acoustic display may perform two main functions: 1.) The first phase may act as an 'Announcement Phase' used to gain the attention of as many conspecifics (particularly females) as possible with 2.) the second phase being an 'Identifying Phase' where the male is able to 'showcase' himself as a desirable mate.

With further research into the acoustic repertoires of other reptilian animals, it may be hypothesised that this two-phase arrangement will be a typical pattern for reptilian acoustic displays - although at this stage, there is no corroborating evidence. This pattern has not been described in the crocodilian in-air advertisement displays which consists of roars/ bellows/or exhalations and are combined with visual displays such as headslaps (Garrick and Lang, 1977), however an underwater acoustic communication system hasn't been investigated to my knowledge and given their largely aquatic lifestyles – cannot be discounted.

One animal known to use pulse sequences as their reproductive advertisement displays are the male Finback whales (*Balaenoptera physalus*) (Watkins *et al.,* 1987; Croll *et al.,* 2002), which use a long series of '20 Hz' (15-30 Hz) pulsed calls occurring in signal bouts lasting as long as 32.5 hours (with rest times between bouts of 1-20 minutes). It is thought, that as this species does not aggregate in a particular area for breeding, their low frequency pulses are for attracting females over vast oceanic distances (and perhaps as a reward for the females efforts to come, the males appeared to call females to krill aggregations)(Croll *et al.,* 2002). It is unknown whether breeding aggregations occur in freshwater turtles. However, if females are widely dispersed throughout the wetland, then it could be that males need to call for females over relatively long distances, particularly given turtle size : wetland ratio (e.g. A turtle of 19cm Carapace length x 11cm Carapace width : A wetland of 450m length x 88m width). The first phase of the turtle 'pulse-bout' consisted of simple repetitions of stereotypical pulses which enhance long-range communication as simple repetitions have the effect of making the call stand out from the background noise (Wiley and Richards, 1978). Also, the low frequency components in the pulses would facilitate relatively long-range communications, particularly during winter / spring when the water levels are deeper (see Forrest *et al.,* 1993; Tyack, 2001).

Both 'pulse-bouts' exhibited lower and higher frequency elements. These elements would enable a range of propagation distances i.e. each pulse effectively has both relatively long and short-range propagation characteristics (these distances need to be confirmed in further field studies). This could be a strategy to call females at a range of distances. In addition, it would be important for the receiving female to then locate the male if she was interested. 'Pulse-bouts' used repetition, a broad range of frequencies and a broken structure, which are all characteristics to enable a receiving animal to binaurally locate the sender more easily (Marler, 1967).

As the 'pulse-bout' recorded in the artificial ponds was known to have been produced by a male separated from the females and given the acoustical characteristics of the sustained vocalisation, it is proposed that this may function as a 'calling song'. A 'calling song' is sung by an unmated male in order to attract a conspecific female, which is behaviour observed in some insects (Elsner, 1983). Using this definition, the advertisement displays used by male Finback whales (Watkins *et al.,* 1987; Croll *et al.,* 2002) would be classified as 'calling songs'. Other songs are known as 'courtship songs' which are sung as a female approaches a male and sung just prior to mating e.g. in some cricket species (Elsner, 1983), which wasn't the case in this instance (as males were separated from the females into a new tank). McKeown *et al.,* (1990) have reported various pre-courtship vocalisations in *Manouria e. emys* and *Manouria e. phayrei* and these differed between male and female tortoises. Other songs are used for male territorial spacing e.g. Agile Gibbons (*Hylobates agilis*) (Mitani, 1988), which, if this was the case for turtles, then given their close confines in the artificial ponds, it would be expected that 'pulse-bouts' would have occurred more often.

In the Hermann's tortoise (*Testudo hermanni*), Galeotti *et al* (2004) noted that successful breeding is likely to be a factor of how well a male can attract a female if they are widely dispersed rather than aggressive encounters with other males e.g. as seen in the Desert tortoise *Gopherus agassizii* (Niblick *et al.,* 1994). While there were some aggressive encounters observed throughout the study, these were intermittent and brief, consisting of biting and short chases (lasting less than a few seconds), there was no fighting observed. In the second phase of the turtle 'pulse-bout' which contained the '*vibrato*', this was considered to be the stage of the 'pulse-bout' where the male 'showcases' himself acoustically. The '*vibrato*' consisted of a complex, rapid display of pulses produced as doublets, roll-down triplets and quadruplets, or as in the Blue Gum display; 'rocking pulses'. While there have only been two examples of 'pulse-bouts' recorded to date for these animals; both '*vibrato's*' were different in terms of their structure. There is some suggestion that vocal behaviour may be costly to males and that their vocalisations would vary; containing information for females on the quality or desirable attributes of the male (Sacchi *et al.,* 2003). In Marginated Tortoises (*Testudo marginata*), Sacchi *et al* (2003) found call features differed significantly between males and certain call characteristics (call rate and duration) were well correlated with condition of a male and successful mating - indicating that aspects of vocalisations may be revealing information to the female for choice of a mate. In anurans, acoustic displays are also considered to advertise the fitness of an individual male – with females preferring calls produced by the largest and heaviest males, with calls usually consisting of relatively low frequencies (Bagla, 1999; Gerhardt and Huber, 2002) (This is usually the case, but a recent study of Tree-hole frogs (*Metaphrynella sundana*) (Lardner and Lakin, 2004) revealed no correlation with male size and call frequency). Female Hermann's tortoises (*Testudo hermanni*) were shown to prefer fast-rate and high-pitched calls (Galeotti *et al.,* 2004). The turtle '*vibrato*' consisted of fast-rate and complex sequences of doublets and triplets, and also 'rocking pulses'. Whether *C. oblonga* females exhibit a preference for a particular call type is not known. As there was only one 'pulse-bout' to use in playback (that was definitely produced by a male turtle), no comparisons for call features could be made.

Chapter. 6.0 TEMPORAL CALL PATTERNS AND BEHAVIOURAL OBSERVATIONS

6.1 Introduction

It was not known if turtles have some preference for the time of day in which to call and whether the sex of the animal and / or size and reproductive readiness has some influence on calling behaviour. Many animals exhibit preferences for vocalising at particular times of the day e.g. Male Gibbons in the mornings (Mitani, 1987); Water boatmen (genus Cenocorixa) at dusk and midnight (Jansson, 1973) and juvenile alligators (*Alligator mississipiensis*) at night (Herzog and Burghardt, 1977).

In addition to the temporal vocal activity patterns, there were many observations made on turtle behaviour throughout the study. Although no experiments were undertaken to elucidate the function of turtle vocalisations, a broad categorisation has been proposed in relation to this behaviour.

6.1.1 Objectives

1. Determine if temporal calling patterns exist in *C. oblonga* and to compare patterns between males and females and between size groupings.
2. Make observations on behaviour both in natural and artificial conditions within ecological and functional groupings (see Collias, 1960).

6.2 Methods and Materials

6.2.1 The Turtles

To determine if temporal patterns in calling exist in *C. oblonga*; turtles were collected from two populations: Pinwernying Dam and Blue Gum Lake. The Pinweryning Dam turtles (46) were obtained from a population removed from Pinwernying Dam, a Water Corporation drinking water dam, which was being cleared of vertebrate fauna prior to dredging. The dam is located just north of Katanning, approximately 360 km southeast of Perth. The 26 females and 20 males used in this research were separated into the following groups (Table. 6.1).

Table.6.1 Sizes and sexes of turtles from Pinweryning Dam used in the spring temporal calling study.

	Carapace Length(cm) (range)	**Mean(cm)**	**Standard Deviation**
Large males (LM) n=10	20.55 - 22.62	21.20	0.64
Small males (SM) n=8 (2 died in this group)	16.54 - 18.38	17.66	0.65
Large females (LFWF) with follicles 12mm-19mm n=6	24.21 - 28.24	25.85	1.40
Females (FWF) with follicles 12mm-19mm n=10	19.95 - 23.05	21.90	1.15
Females (FWOF) without follicles n=10	19.89 - 24.05	22.02	1.41

These animals were assessed as being healthy and in excellent condition based on their overall appearance. Turtles were ultra-sounded by Dr Gerald Kuchling to ascertain follicle development and receptivity for breeding. The second group of adult turtles used in the temporal calling study was a small group (20) from Blue Gum Lake and these were collected in the summer (January) of 2005. The turtles were separated into adult males and females (Table. 6.2) and were considered in good condition based on their overall appearance.

Table.6.2. Size of male and female Blue Gum Lake turtles used in the study of summer temporal calling patterns.

	Carapace Length(cm)	**Mean(cm)**	**Standard Deviation**
Females	17.48 - 20.89	19.12	1.04
Males	15.00 - 20.64	17.41	1.47

6.2.2 Recording Regime for Vocal Periodicity

The five groups of turtles from Pinweryning Dam (Table. 6.1) were held in five separate ponds (as used in Chapter. 4.) to determine if there were differences in temporal calling patterns between different size / sex classes and reproductive readiness. One hour recordings were made in each pond for the time periods dawn, midday, dusk, midnight once a week for three weeks in spring from 20th September, 2004 to the 10th October, 2004 (Table. 6.3). In total 72 hours of recordings were made. The number of calls in each size/sex category was then given as the number of calls / turtle to account for differences in the number of turtles in the artificial ponds (i.e. there were six LFWF and eight SM, with ten turtles in the remaining tanks).

Table.6.3. Recording regime used for temporal call patterns in the Pinweryning Dam turtles.

Time period for period	Date of recordings			Turtles recorded	Hours per pond per week	Total Hours 3-week
Dawn	23.9.04	30.9.04	10.10.04	LFWF, FWF, FWOF, LM,SM,J	1	18
Midday	21.9.04	1.10.04	5.10.04	“	1	18
Dusk	25.9.04	3.10.04	8.10.04	“	1	18
Midnight	20.9.04	28.9.04	6.10.04	“	1	18

Blue Gum Lake turtles were separated into males and females and were recorded in summer (11th – 31st January, 2005) at a time when the influence of breeding activities was not expected to be important. Recordings for Blue Gum Lake turtles were made at similar times as the Pinweryning Dam turtles however recording time was doubled for the summer study. In total, the equivalent of 96 hours of structured recordings were undertaken between the two populations. As there were six ponds to record within a time period in the spring study, two identical hydrophones (HTI-96-MIN) needed to be used in order to ensure that recordings took place within a reasonable time frame and each time period could be accommodated (in this case 3 hours - so actual time spent recording the Pinweryning Dam population was able to be halved and consisted of 36 field hours). Water temperatures were also recorded in each pond prior to recording.

6.2.3 Behavioural Observations

To enable viewing of turtle behaviour within the artificial ponds, four transparent perspex viewing windows (60cm width x 50cm height) were inserted into the sides of one of the artificial ponds and a single 50 watt underwater light was used which enabled night-time viewing. Field observations were made opportunistically during playback trials and when recording throughout the research. Observations were made sitting on a box approximately 1m from the water's edge and wearing Polaroid sunglasses (standing at the water's edge appeared to scare the turtles away, and sitting on a box gave a better depth of field than sitting on the ground).

6.3 Results

6.3.1 Vocal Calling Patterns

In spring, the complex turtle vocalisations consisted of the short duration 'chirps': 1.) 'Short chirps' and 2.) 'Medium chirps' - with most calls occurring at midday (Figure. 6.1). Although dusk was the warmest time period (Table. 6.5); and nearly all groups, except the large males (LM) utilised the dusk period for calling; fewer calls were made in this period compared to midday. Only females with follicles (FWF) and the small males (SM) utilised the dawn period and only males vocalised at midnight. Overall, it was the small (SM) males, with 1.88 calls per turtle (total of 15 calls in 3 hours of recordings) and the females with follicles (FWF) with 1.3 calls per turtle (total of 13 calls in 3 hours of recordings) (Table. 6.4) that were the most vocally active in this study and appeared to follow similar calling trends (except for midnight when only males were vocally active). Females without follicles (FWOF) called the least with only one call being recorded (Figure. 6.1; Table. 6.4). Large females with follicles (LFWF) called approximately the same amount as the Large males (LM) (c.f. LFWF: 0.66 calls / turtle; LM: 0.7 call / turtle).

Table.6.4. Number of 'chirp' calls made by turtles in the artificial pond in a three week period in spring 2004.

	Total number of calls	**Number of turtles**	**Number of calls per turtle**
Large Females with follicles	4	6	0.66
Females with follicles	13	10	1.3
Females without follicles	1	10	0.1
Large Males	7	10	0.7
Small Males	15	8	1.88

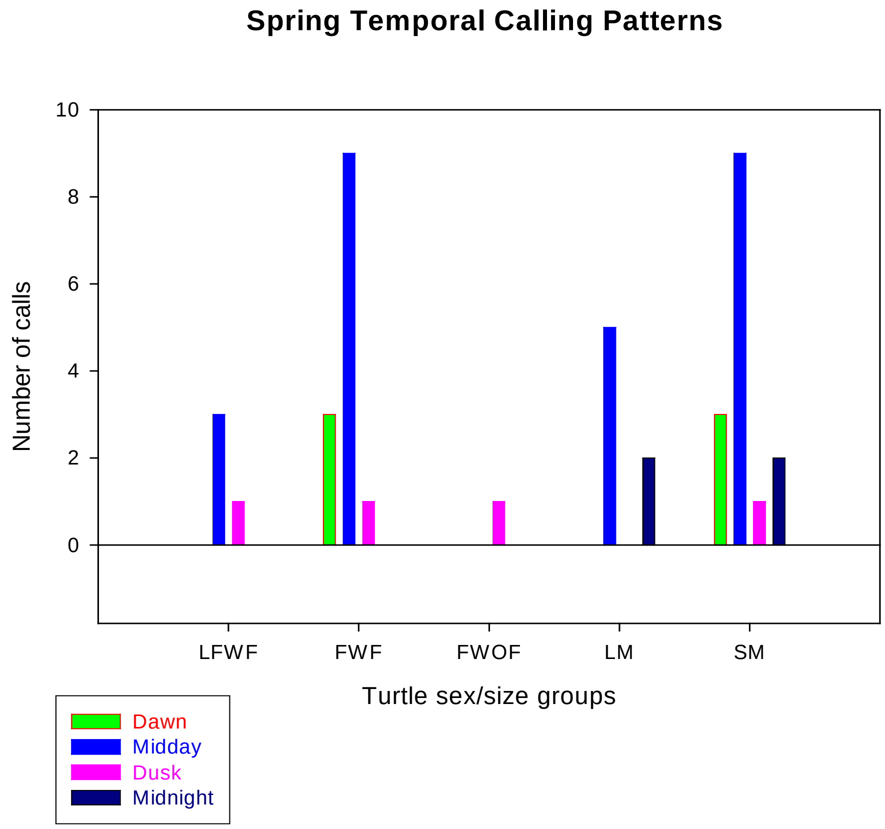


Figure.6.1. Number of calls in each sex/size class of turtles for each period of the day (LFWF=large females with follicles; FWF=females with follicles; FWOF=females without follicles; LM=large males; SM=small males).

Table.6.5. Average pond temperatures experienced by each turtle group in the spring Temporal calling study.

AVERAGE POND TEMPERATURES (°C) Sept/Oct 2004	DAWN	MIDDAY	DUSK	MIDNIGHT
Turtle Group				
Large females with follicles	13.0±2.0	15.8±2.4	17.3±1.3	13.7±0.8
Females with follicles/ & Females without follicles	12.7±2.1	15.5±1.8	16.5±0.5	14.3±0.6
Large Males	13.2±1.9	17±1.7	17.3±1.0	15.0±1.0
Small Males	12.7±2.1	15.5±2.3	15.7±0.6	13.5±0.5

Although twice as much time was spent in recording the summer calling patterns, the smaller group of Blue Gum turtles still revealed that more calls were being made overall compared to the spring study. Females called more often at dawn and midday than males (28 dawn calls & 39 midday calls by females c.f. 20 dawn calls & 26 midday calls by males)(Figure. 6.2) while males called more often at dusk and midnight than females (33 dusk calls & 25 midnight calls by males c.f. 26 dusk calls & 5 midnight calls by females). While it appeared that females may have a preference for calling in the earlier part of the day and males tending to prefer the latter part of the day for calling activity; there was however, no evidence found between the sexes and the mean number of calls in each time period (two sample t-test (two-tailed): Dawn: t-stat=0.339, df=4, p-value=0.752; Midday: t-stat=0, df=3, p-value=1.0; Dusk: t-stat=-0.717, df=3, p-value=0.525; Midnight: t-stat=-1.031, df=3, p-value=0.378). The most utilised chirp vocalisation by both Blue Gum female and male turtles was the 'short chirp', followed by the 'medium chirp' with males using these vocalisations slightly more often than females (73 'short chirps' / 20 'medium chirps' for females; 78 'short chirps' / 22 'medium chirps' for males) (Table. 6.6). Calls with high frequency components (i.e. calls containing more than two harmonics and extending above 2 kHz) were used less often in both groups.

Table.6.6. Total number of calls in each 'chirp' category using a doubled recording regime.

	FEMALES	**MALES**
January 2005		
Vocal category		
Short chirp	**73**	**78**
Medium chirp	**20**	**22**
High short chirp	**3**	**4**
High call	**2**	**0**

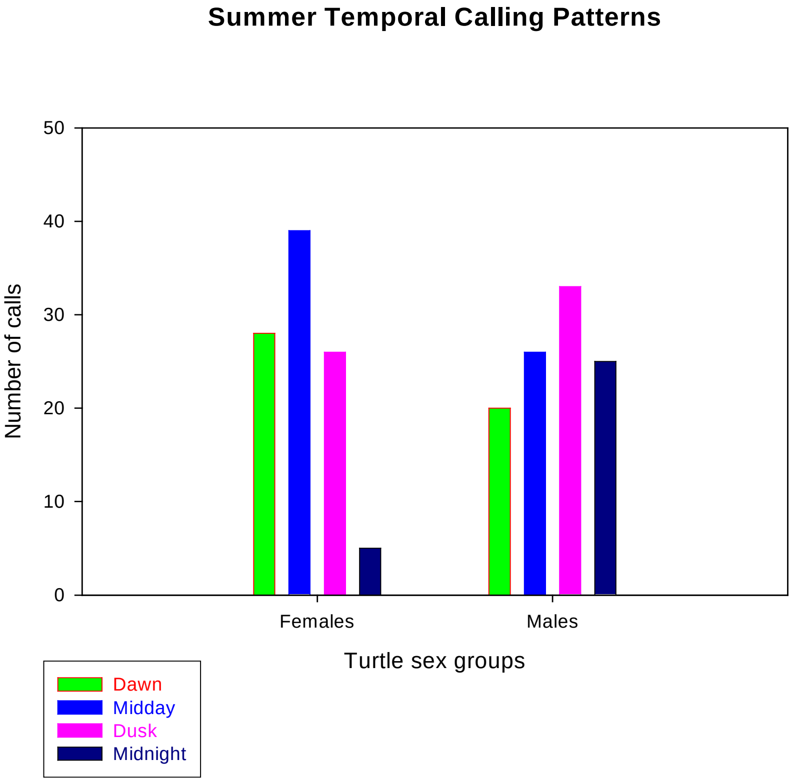


Figure.6.2. Number of calls made in summer by male and female turtles from Blue Gum Lake in the artificial ponds. Time utilised for this recording regime was double that used for the spring calling patterns.

6.4 Discussion

Animal vocalisations are usually categorised ecologically and functionally into those concerning: 1.) Food; 2.) Predators; 3.) Sexual behaviour and fighting; 4.) Parent-young interactions; and 5.) Group movement and aggregations (Collias, 1960, P.387; Busnel, 1963); with another category 6.) For those vocalisations which occur within a social context (Hopp and Morton, 1998) or what Kaufmann (1992) described as 'neutral encounters'. Within the above functional categories, Collias (1960) found that well-defined signal characteristics emerged for mammal and bird vocalisations. Campbell and Evans (1967) made some preliminary comparisons with these signal characteristics and some turtle calls, and found that similarities did exist with call structures related to the context in which they were made. Therefore, these categories have been used to place turtle calls within a contextual framework and are discussed in relationship to the signal characteristics Collias (1960) found for mammal and bird calls.

Food and Parent-Young Interactions

Unlike the biparental nurture of young which occurs in crocodilians (Garrick and Lang, 1977; Britton, 2001), or the post-ovipositional parental care observed in many reptilians (Shine, 1988); these turtles are not known to nurture their young, nor were they observed to nest-attend (There are reports by Indigenous Amazonians, that the freshwater turtle from the genus *Podocnemis sp.* waits in the water near nest sites for turtle hatchlings, and once the hatchlings are in the water, the adults guide them back to feeding grounds a few hundred kilometers away (Kuchling, pers. corr. 2005)). Therefore, the category of 'parent-young' interactions may not apply here. The lack of, or paucity of vocalisations heard in hatchlings and juveniles respectively; is likely to be a predator avoidance response by young in a species where there is no parental protection (Note that while no vocalisations were made by hatchlings, their use of ultrasound cannot be excluded). The vocal behaviour heard in juvenile crocodilians, is considered important for their survival by establishing communication with parents and siblings for protection (Britton, 2001). Turtles were not heard to vocalise within the 'food' category. While many animals emit a 'food finding' call (see Bradbury and Vehrencamp, 1998), this was not evident in *C. oblonga.* From the pilot work (Ch. 3; Study.4.); turtles were silent as they approached and consumed chicken liver bait tied to a stake in the wetland and also when kangaroo meat was placed loosely on the sediment in the wetland. This silent behaviour was also confirmed in the artificial ponds when feeding the turtles (on feeding platforms in the pond) and their nocturnal feeding on live Mosquito fish placed in their artificial ponds. The only sound recorded in feeding behaviour – nocturnally or during the day, was when turtles would strike rapidly at food items, producing a high intensity sound associated with 'snapping and lunging' movements. As *C. oblonga* appear to be non-selective feeders (Woldring, unpublished, 2001) and from observations made when using baited traps, chemoreception appears to be a well-developed sense in *C. oblonga* and other turtles (Manton, 1979) and so a 'group food call' is unlikely.

Group Movements and Aggregations

Group movements of *C. oblonga* occur when females leave the water to nest on the terrestrial buffer in spring, but no in-air vocalisations have been reported with this movement. Turtles will aggregate when there is a large food source available (Harless, 1979), which was confirmed by the numbers of turtles that were caught at any one time in a single baited trap (pers. obs). On one occasion, a large orderly feeding aggregation was observed at Blue Gum Lake in the early morning, summer 2002 (Figure. 6.3). More than 100 turtles were lined-up side-by-side around the entire NE shoreline and appeared to be feeding on a bloom of Water fleas (Daphniidae: *Daphnia*) that the wind had blown into thick masses around the shoreline. By the time the equipment was set up, some turtles had begun to disperse but while many still remained, no vocalisations were recorded. One dispersing turtle did come and investigate the equipment and a series of clicks were recorded (Ch. 4; Figure. 4.4).

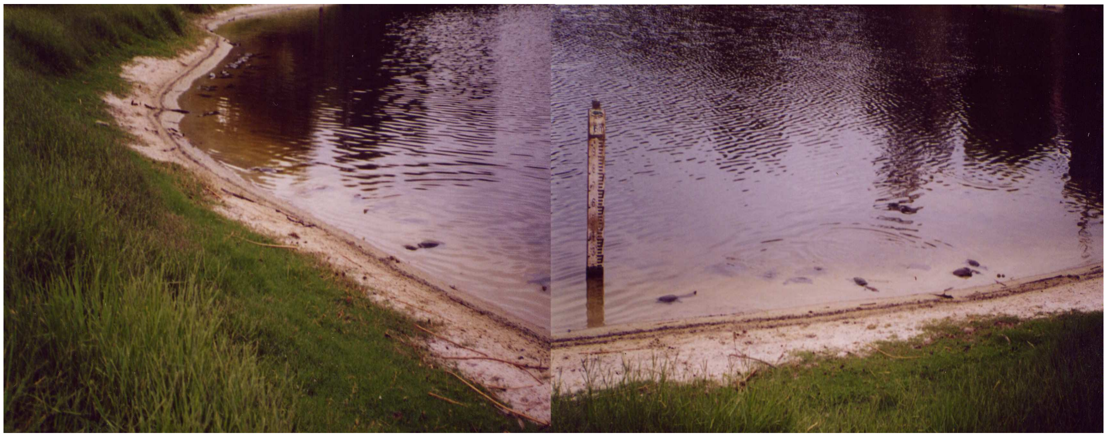


Figure.6.3 A feeding aggregation at Blue Gum Lake around the NE shoreline. Turtles had been lined up side-by-side but by the time the photo's could be taken they had begun to disperse. Dawn, summer 2002.

Smaller aggregations of turtles were observed throughout the study, in particular in the ponds where two or more turtles would sit on the bottom sediments together or on logs. At night, turtles often rested near the surface suspended within the vegetation with conspecifics nearby. No obvious vocalisations were recorded.

Social Calls

On several occasions when recording at Blue Gum Lake with two or more turtles in view; 'short chirps' could be heard as turtles were passing nearby to each other or when several turtles were investigating the speaker. When observing turtles in the wetland, they appeared to continue on with what appeared to be a 'social' or 'neutral' interaction. While several 'chirps' could be heard, it was unknown whether it was one animal calling or nearby conspecifics replying. While there were no obvious behavioural responses to the 'chirps' in playback, other than the alert posture and then swimming towards the speaker (see ch. 7); from these types of field observations and given the prevalence of 'chirps', these are hypothesised to be their main contact call for *C. oblonga* – in particular, the 'short chirp' which was the most prevalent of the 'chirps'. There were some similarities in the juvenile crocodilian contact call and the turtle contact call in that they were of low received intensity and had fewer harmonics compared to their other call types (Britton, 2001). However, crocodilian calls finished in a short down-sweep, while turtle contact calls finished in short up-sweeps. In birds and mammals, the presence of calls with changing pitch particularly the up-sweeps, evident in the turtle 'chirps', are noted as 'pleasure calls' by Collias (1960).

Predators and Threatening Situations

A predator avoidance response was observed when turtles were out of the water basking on a log and they make a visual sighting of a human or any animal which appears suddenly, particularly at close range. No warning or alarm calls are emitted, but turtles will quickly roll off the log or branch back into the water, producing a chain-effect with all nearby conspecifics performing similar behaviour. Interestingly, a series of staccato calls were recorded in the juvenile pond that an adult male had inadvertently made his way into. The most distinctive section of this call lasted 4.67 seconds with spectral characteristics very similar to the staccato call of the Squirrelfish (*Holocentrus rufus*) (Fine *et al.,* 1977) (Ch. 4; Figure 4.22). This type of call is issued as a warning when an intruder such as a predatory fish or larger fish invades the Squirrelfish territory, or the sudden appearance of any fish. Unfortunately, it was unknown whether the 'staccato' was made by the juveniles or the adult turtle, as single pulses or short series of pulses that make up a 'staccato' were produced by both adults and juveniles.

Growls in many species are associated with aggressive or agonistic encounters (see Fine *et al.,* 1977) and low-pitched, harsh sounds such as were seen in the 'growls' and 'blow bursts', are termed 'threat' calls by Collias (1960) and considered for use in repelling. The 'blow-burst' is likely to be analogous with the hissing 'threat call' well documented in reptiles e.g. crocodilians (Britton, 2001) and snakes (Kinney *et al.,* 1998). The 'growling rattle' in the turtle's 'wild howl' call described here, appears to share similar characteristics described in the snake genus *Pituophis* where their hisses produce a sort of 'staccato effect' by vibration of a special membrane at the opening of their glottis (Bogert, 1960). On one occasion, a growl was heard in the recordings when two human observers suddenly appeared overlooking the artificial ponds. In the free-field playbacks (Ch. 7), the 'growl' and 'blow burst' often resulted in the turtle turning around and quickly swimming away and it is hypothesised that this is a 'flee response' (this response was a particularly obvious response when previously, the turtle had stopped swimming in response to the 'chirp' calls). The 'growls' and 'blow-bursts' by *C. oblonga* were also high amplitude sounds (the highest in the repertoire with overloading of the equipment). Fine *et al* (1977) noted that louder than usual sounds produced by some fish were considered associated with higher emotional states. For example, in the Sea catfish (*Galeichthys felis*), high amplitude and long duration calls were elicited under situations of duress (Tavolga, 1960); with long duration of such a call noted by Collias (1960) to be associated with a greater intensity of alarm. High amplitude distress calls have been recorded in juvenile crocodilians when harassed (by pinching the toe webbing, shaking the animal or squeezing the tail base) (Herzog and Burghardt, 1977).

The 'Cat whines' produced by the turtles seized by WA Customs Officers, were made on their first night in the artificial pond. Collias (1960) noted in some bird species (e.g.

Dumetella carolinensis and *Cirus cyaneus*) that their short note alarm calls changed abruptly to prolonged, high 'cat-like' meows or screams when an enemy came too close to their nests. A high-pitched harsh continuous call is considered by Collias (1960) to be associated with a predator and meant to instill fear in the offending animal. As these turtles had come from a stressful situation (wrapped up tightly in socks and held dry in unnaturally high temperatures for several days); producing calls considered associated with high stress was not surprising. Chaotic and noisy structures in calls, such as the 'growls', 'blow bursts' and 'cat whines', are noted by Herzel *et al* (1998) to be produced under conditions when stresses are high.

Down-sweeps in calls appear to be associated with distress (Collias, 1960). Examples of calls with this type of structure included: the 'growl squawk' in Figure 4.20, which began with an obvious down-sweep structure finishing in a harsh growl; and the 'wails' consisted of slow frequency modulation but also finishing in down-sweeps. Similar types of call structural characteristics have been described in Campbell and Evans (1967) for *Gopherus agassizii,* which had been placed into a sink and was trying to find its way out. Previous to the 'wail' bout being produced, 'blow bursts' and 'growls' were heard, with 'grunts' heard throughout the bout of 'wails'. As the Pinweryning Dam turtles and the 'seized' turtles were particularly large animals and known to have been captured from large water bodies; they would be used to roaming over greater distances than were available in the small confines of the artificial ponds. With conspecifics in close proximity than would be the case in a natural environment, this would no doubt have been stressful and elicited abnormal behaviours (see Warwick, 1987; 1990) and chaotic and distressful sounds being produced. 'Cat whines' and 'blow bursts' were never recorded in the urban turtles, and 'growls' were rarely heard, which may be due to their familiarity with higher population densities and their ability to cope with conspecifics in close proximity - although other sounds may have been used as a spacing mechanism. Marcellini (1977) proposed that the commonly produced ‘multiple chirp’ sequences produced by the Gekkonid lizard *Hemidactylus frenatus* - which occurs in a range of social interactions, may act as a spacing mechanism between males – establishing and maintaining territories. However, this hypothesis was not tested for using *C. oblonga’s* ‘chirp’ calls; and as previously noted, no turtle was repelled by any ‘chirp’ in the free-field playbacks (which included both male and female ‘chirps’) and turtles were often in close proximity (and remained in close proximity) when ‘chirps’ were heard in field observations.

Aggressive behaviour did not appear to be a common occurrence in *C. oblonga* but biting and chasing was occasionally observed and is well-documented behaviour in the Wood turtle (Kaufman, 1992). In *C. oblonga,* turtles were often observed swimming close to one another, or a swimming turtle settling on top of a stationary turtle, scraping feet over the others head to settle or ‘push-off’ with the stood-upon-turtle either remaining, seemingly unaffected by the behaviour, or swimming slowly away. On the occasions that biting and chasing behaviour did occur, no vocal responses were elicited from the victim nor were there any aggressive counter-responses. Only an avoidance response to biting was ever witnessed, with the victim swimming away quickly. In the pilot work (Ch. 2; Study.1) biting and chasing was observed when turtles were placed into a stressful situation, which in this case was putting them in a tank with no hiding material. At a later date, a large female (CL: 25.8cm) (from another population) was placed into the artificial pond with the smaller adult female and male turtles from Blue Gum Lake to find out what responses would occur to a stranger and a much larger turtle placed in their midst. Initially, Blue Gum turtles appeared unperturbed by the presence of this large stranger – they neither fled from her nor investigated her and did not emit any calls. However, subtle behaviours may have gone unnoticed in this experiment and needs to be more fully investigated. After one hour (after the large female had investigated her new surrounds), the large female exhibited aggressive behaviour towards the Blue Gum turtles. The large female appeared to sniff the cloaca of passing turtles and proceeded to bite the leg of all male turtles passing or swimming nearby. Bites were sometimes brief, but a few were 'hang-on' bites where the male had to jerk his leg free. Hang-on bites have been observed in aggressive encounters in the Wood turtle (Kaufmann, 1992). There were no vocal responses by the bitten or chased turtles in this case. By the following day, all the males had climbed out of the tank and most of the smaller females, but the large female remained for several days before climbing out and returning to her own pond.

Sexual Activity

While it is hypothesised that the 'pulse-bout' (ch. 4) plays a role in breeding activities this remains unconfirmed. Similarly, observations made in the winter at Blue Gum Lake of both a courting and a copulating turtle pair (recorded on separate occasions), were not heard to vocalise. Behavioural displays are often seen in other species e.g. birds (see Ficken *et al.,* 2002; Cooper and Goller, 2004), and in chelonians, head-bobbing behaviour has been reported by McKeown *et al* (1990) in the *Manouria spp.* which is associated with the pre-courtship vocalisations. Interestingly, a type of 'head-bobbing' behaviour was observed at the waters surface in one of the courting turtles in the wetland (the turtle repeatedly lifted its head well above the waters surface and dropped it down giving a 'rocking' appearance to this activity). This 'head-bobbing' might be the equivalent to the 'headslaps' observed in the advertisement displays of some crocodilians e.g. *Alligator mississippiensis* (Garrick and Lang, 1977), however no acoustic emissions were recorded at this time from the turtles as mentioned previously.

Temperature and Vocal Activity

Ambient temperatures will influence many of the physiological processes of reptiles (see Pough *et al.,* 1998) and in some, will even change their anti-predator behaviour, from fleeing at high body temperatures to adopting aggressive body postures, vocalisations and biting at lower body temperatures (Crowley and Pietruszka, 1983). Temperature also appears to influence vocal activity in *C. oblonga.* Most vocalisations were recorded when water temperatures were above 10°C. No complex calls were heard below this temperature; only occasional, 'clicks' or 'grunts/ growls' and 'blow bursts' could be heard. The lack of complex calls (which are considered to be their social calls) produced by *C. oblonga* at <10°C would appear to correspond to the inactivity reported in some freshwater turtle species at this temperature e.g. *Chrysemys picta* (Ernst, 1971). The paucity of vocal activity by the females without follicles was a consistent trend exhibited throughout the winter / spring months and was not considered to be entirely due to the cool water temperatures. Despite water temperatures probably being cooler than those experienced in a natural wetland (as above-ground artificial ponds are known to experience more extreme temperatures – cooling and heating more rapidly than a natural environment with pond temperatures varying only slightly due to different degrees of sunlight exposure, however all ponds exhibited a trend of warming during the day and cooling at night with cool temperatures also experienced at dawn); both females with and without follicles had their ponds connected via a filtration system and therefore had similar temperatures. The paucity of vocalisations was thought likely to be related to the lack of follicle development and therefore reproductive non-receptiveness. The lack of activity - vocal or movement in non-sexually active females would be an advantage for males seeking mates, as it would be energetically expensive for males to seek out reproductively nonreceptive females. However, this explanation doesn't account for the paucity of vocal activity in the LFWF or the LM groups in the spring temporal calling study. As both these groups were vocally active prior to the temporal patterns study and following it, it is therefore considered that longer recording windows would need to be undertaken in future to account for the type of weekly variation that a short window of recording cannot account for.

The calling rate by turtles was particularly low in spring with an increase in vocalisations found during the summer period, with most turtle calls occurring throughout the warmer part of the day. 'Short chirps' were the most prevalent of the complex calls produced by turtles in all time periods and for both seasons. In summer, males called only slightly more frequently than females, but greater use was made of the dawn period by females and greater use of the midnight time period by males. Although no significant difference was found in temporal call patterns between males and females; a longer period of recording may have revealed strong evidence for a diurnal vocal activity difference between the sexes, particularly as both the spring and summer study indicated that males were more vocal at midnight than females. Nocturnal activity appeared to be common in *C. oblonga,* with turtles often seen to be active around the margins of wetlands at night, although the sex of these turtles could not be determined conclusively to make a correlation with sex of the animal and nocturnal activity. Nocturnal activity in freshwater turtles is not particularly common but has been described in some species e.g. *Chelydra serpentina* and *Macroclemys temmincki* (Ernst and Barbour, 1972), with nocturnal calling only reported in one other chelonian (and always during rain) - the terrestrial tortoise *Geochelone travancorica* (Campbell and Evans, 1972). However, the chorusing described for *G. travancorica* was not apparent in *C. oblonga,* only the occasional 'chirp' bout was recorded. While these bouts were thought to be produced by a single animal because of the similarity of structure in each call, it could not be known with any certainty whether the chirps were nearby conspecifics replying.

Chapter. 7.0 PLAYBACK STUDIES

7.1 Introduction

Acoustic communications in chelonians has not been well studied and is poorly understood. Use of sound playback was considered an important means of placing turtle vocalisations into a behavioural or communicative context, where the sound stimulus was introduced on the assumption that it would produce some change in the existing behaviour or lead to a vocal response at the time of playback. Some indigenous peoples in Madagascar use a wooden post to produce a 'thump' on the base of their canoe to call turtles to the area so as to harvest for food (Kuchling, pers. comm, 2002). Similarly, an amateur diver has described using a 'gulp' noise whilst underwater to call Hawksbill Turtles (*Eretmochelys imbricata*) - again for harvest (Palmer, pers. comm, 2002). These observations suggest that marine turtles both receive and respond to sound.

Preliminary investigations were undertaken to determine what types of responses would be elicited in *C. oblonga* when calls from their repertoire were played. Although there are numerous accounts of behaviour in freshwater turtles (see Harless, 1979; Kaufmann, 1992), behaviour under natural conditions in *C. oblonga* is largely unknown and freshwater turtles are highly mobile animals and difficult to observe directly in their aquatic environments. As a consequence, research on communication and behaviour in these animals presented as a challenge even under artificial conditions.

7.1.1 Objectives

1. Ascertain if a response, either vocal or behavioural, occurs when a sound stimulus from the acoustic repertoire is played under natural or artificial conditions.
2. Identify and describe the responses, both vocal and behavioural to playback stimuli.

7.2 Methods and Materials

7.2.1 The Turtles

Turtles use in the playback studies under artificial conditions were the Blue Gum Lake turtles that had been used in the temporal calling pattern study discussed in Chapter 6.

7.2.2 Playback Calls

The calls used for playback consisted of a number of calls that had been recorded in the artificial ponds and were selected from recordings taken from the three turtle populations: Masons Gardens, Pinweryning Dam and Blue Gum Lake. These calls were selected from within all available recording times. This selection process was undertaken in order to minimise bias in the calls presented; such as the influence of time of day (McGregor *et al.,* 1992). Calls were selected randomly to minimise any influence from a vocalisation produced by a sub-ordinate or low social-status animal (Pepperberg, 1992) (It is not known if such a hierarchy exists in *C. oblonga,* but a dominance structure has been reported in male Wood turtles (Kaufmann, 1992)). Sounds were selected for playback based on their clarity and quality; with selected calls having a well-defined spectral structure and high signal to noise ratio. While it was understood there may have been some distortion in the signals being used for playback (due to reflections and reverberation in recordings taken from artificial conditions); in the absence of calls recorded under free-field conditions, these calls were considered reasonable to trial. Also, as no previous comparisons have been made between synthetically produced signals and natural signals for attractiveness, or what acoustical properties within the signals were of importance to these turtles (see Gerhardt, 1992); only natural, unmodified signals were used.

Vocalisations were filtered to remove as much of the background noise as possible using the Bessel bandpass filter (i.e. appropriate for the frequency range in each call) in Adobe Audition v.1.0. To record to CD, the signals were converted into stereo, 44.1 kHz format using Adobe Audition v. 1.0. Trial runs were conducted in the constructed channels (see section: 7.2.7 for description) using frequency sweeps and the repertoire of calls selected for playback. Calls were filtered by Dr Alec Duncan from CMST, Curtin University, to account for the variations in output from the UW 30 speaker at the appropriate frequencies for both free-field conditions and the artificial channels (Appendix 2). While the playback calls were assessed for fidelity, subtleties in calls may have been lost through this filtering process (see Gerhardt, 1992) and also there was likely to be distortion from the limited quality of this speaker.

For the free-field playbacks ('free-field' was taken to be in a wetland setting; sections: 7.2.4 & 7.2.5), the 'AB' design was chosen where 'A' corresponded to a silent interval and 'B' a sound stimulus (see Hopp and Morton, 1998). The silent interval was taken to be up until the point at which a turtle swam into view - within 1m of the hydrophone (it was important that the turtle was still swimming when the sound stimulus was played). Turtle calls for free-field playback consisted of an artificially selected sequence of calls from the turtle acoustic repertoire (in order): 'long chirps', 'hoots' and 'wails' (in bouts) and 3 minutes of an artificially created section of the 'pulse-bout'. The 'pulse-bout' consisted of pulses from the first and second phases. These complex calls were then followed by the percussive and noisy calls from the turtle acoustic repertoire: a 'growl', a 'blow burst' and finished with the 'drum roll' bout (NB this order of complex calls to noisy structured calls were selected based on some of the observations from the pilot work section: 7.2.3).

The time between each call in a 'bout' was based on that measured in pond recordings. For example, the 'drum roll' used in playback used the entire 'drum roll' produced by the Blue Gum Lake turtles; which lasted 5 minutes and was unmodified. For playback 'chirps', a series of five similar vocalisation types were used (i.e. not the same vocalisation repeated five times), using an 'inter-chirp-interval' of around three seconds. Choosing the appropriate number of stimuli (McGregor *et al.*, 1992) was arbitrary as there had been no previous studies undertaken on freshwater turtles to guide this decision. From the recordings, most 'chirps' occurred either singly or in groups of three or four - so the five stimuli chosen for playback seemed reasonable even though on three occasions a long calling sequence did occur (32 calls in 16 minutes; 34 calls in 7 minutes; 30 calls in 7 minutes).

The list of sounds in sequence selected for playback

In order: 5 x 'long chirps' (3 seconds between each 'chirp')

5 x 'hoots' (3 seconds between each 'hoot')

All of the 'wail bout'

3 minutes of the 'pulse-bout' (pulses artificially selected from the first and second phases)

1 x 'growl'

1 x 'blow burst'

All of the 'drum roll'

For the playback of turtle 'pulse-bouts' in the artificial channels; the 'ABA' design was chosen - where 'A' corresponded to a one minute silent period prior to broadcast of 3 minutes of the 'pulse-bout' ('B'), which was then followed by another one minute of silence ('A'). The turtle was observed in both silent intervals (before and after the sound stimulus) and in the vocal sequence with responses in behaviour (phonotaxic) or vocal (antiphonal) being recorded. Pseudoreplication (McGregor *et al.*, 1992; Hopp and Morton, 1998) was not considered to be an issue in either sections of playback as this part of the research was to find out if a response would be elicited to the signal and to note what type of response(s) occurred.

In order to minimise habituation (Langbauer *et al.,* 1990; Pepperberg, 1992), the call sequence or the artificial 'pulse-bout' was played only once to each turtle. In the free-field playbacks, the call sequence was played on the assumption that turtles passed by the speaker only once and passed the speaker randomly with broadcast ceasing when the turtle swam out of sight (i.e. the full sequence of calls was usually not played).

7.2.3 Pilot Study: Playback in Free-field Conditions

Playback of turtle sounds was initially conducted in five wetlands under free-field conditions as a pilot study. The wetlands were: Glen Brook Dam, Lake Leschenaultia, Leschenaultia Pool, Neil MacDougall Lake and Mabel Talbot Lake. For the pilot study, calls from the repertoire were played in a random order and were broadcast through the underwater speaker at varying water depths. The first free-field trial conducted at Glen Brook Dam occurred at the deepest end (max. depth ~7m) (northern aspect), and was clear of boulders and aquatic vegetation with little to attenuate the broadcast sound, so was considered to provide ideal conditions for propagation of turtle calls. The speaker could only be placed in 1.0m of water as this was the limit of visibility for an on-shore observer. Speaker placement in the remaining wetlands was limited by the depth of water available, and the distance at which the speaker was visible from the shore. This regime was used to determine the responses that might be elicited from turtles under natural conditions.

Pilot Study: Results and Discussion

While a field situation for playbacks was considered an ideal setting in which to observe natural behaviour, it was understood that there was no control over factors such as the influence of conspecifics (e.g. territoriality, agonistic encounters, competition etc) and other influences such as wind, waves and habitat conditions which might result in the degradation of signal characteristics (see McGregor *et al.,* 1992; Gerhardt, 1992). At Glen Brook Dam, an almost immediate response was elicited to the 'drum rolls' which were the first calls to be played. After three 'drum rolls' (which lasted 2.42 minutes), a large turtle surfaced and raised its head well above the water looking in my direction and the recording equipment onshore, where the turtle remained in this orientation for around one minute, then disappeared without resurfacing again. No other responses could be observed or heard for the remainder of the playback trial (lasting 25 minutes). The appearance of the turtle may have been unrelated to the broadcasts as spy-hopping or any other observable responses were not evident at the trials conducted at Lake Leschenaultia, Leschenaultia Pool and Neil MacDougall Lake.

Initially, the broadcast of calls in Mabel Talbot Lake in Subiaco did not appear to elicit any behavioural responses (e.g. spy-hopping). However, it wasn't until a turtle swam nearby the speaker that a response could be observed. Of the eight turtles that were seen swimming by, five turtles exhibited responses to the broad-cast calls. The five turtles which exhibited a response were within 1m of the speaker while the remaining three turtles were approximately 3m from the speaker and swam past without stopping or moving towards the speaker. When the 'drum roll' was played, one of the nearby turtles turned around and very quickly swam away from the speaker. Therefore, for the remainder of the field trial, only 'chirp' calls and the artificial 'pulse-bout' were played. The behaviour of the next three 'swim-by' turtles consisted of turtles ceasing swimming and remaining completely still. Necks were either flexed or outstretched and heads either turned away or directed towards the speaker. This behaviour was considered important to test for in formal playback trials. All turtles swam out of sight before completion of the artificial 'pulse-bout'. One female began to swim away, but when the 'vibrato' section of the 'pulse-bout' began, she stopped and remained motionless for approximately one minute with her carapace facing the speaker (she appeared to stand vertical in the water) and her head held to one side. No vocal responses were elicited.

Pilot Study: Summary

The results of the pilot study indicated that: 1.) The order of presentation of calls was considered to be important due to the response elicited in one turtle where it swam away quickly from the speaker when the 'drum roll' was played. In addition, as it was thought that noisy structured calls were associated with highly emotive states or aggressive encounters – an avoidance response wasn't being tested for at this stage; 2.) Turtles needed to be within 1m of the speaker as: a.) only near-by turtles could be observed adequately (if a response occurred when turtles were further away from the speaker, these responses could not be accounted for); and b.) due to attenuation of signal characteristics within a natural setting and the relatively short range propagation properties of the 'chirp' calls, responses of turtles at 1m was considered reasonable

(NB when the hydrophone was thrown out at least 3m beyond the speaker 'chirp' calls were not readily detected); 3.) The behaviour seen in the three turtles of stopping swimming with their necks flexed or outstretched was considered an important behavioural response that would be used to test whether turtles were attracted to broadcast turtle calls; and 4.) Playback studies need to be undertaken on calm days as surface ripples from wind obscured the vision into the water for observing and timing how long a turtle remained in this posture.

7.2.4 Free-field Playbacks

To test whether turtles were responding to turtle calls or the presence of the speaker or a new sound in their environment; free-field playbacks continued at Mabel Talbot and Blue Gum Lakes during the months of April and May, 2005. The speaker remained *in situ* from one to three hours within each waterbody, using a repetitive playback sequence of 1.) Turtle calls, 2.) Silence and 3.) White noise - where turtle calls were played to the first 'swim-by' turtle, silence occurred with the second 'swim-by' turtle and white noise was played to the third 'swim-by' turtle. As previously noted, turtles investigate new objects in their environment, so to control for the presence of the speaker, swimming towards or around the speaker was not timed or included as part of this research - only time spent in the above described posture. As the control acoustic stimulus; 30 seconds of white noise was played to every third 'swim-by' turtle. White noise was generated digitally in SpectraPLUS and recorded to CD. This playback regime occurred on the assumption that turtles swam by randomly and that the turtle swam past the speaker only once.

7.2.5 Playback Trials under Artificial Conditions

Use of an artificial environment was considered appropriate for playback of the turtle ‘pulse-bout’ so that male/ female responses can be noted specifically, as sex of a turtle was unable to be conclusively determined in the wetland setting. A purpose-built artificial environment was constructed, which consisted of a below ground rectangular channel (2.4m length x 0.8m width x 0.6m deep). Wooden railway sleepers formed the channel boundaries and it was lined with plastic polythene sheeting (Figure. 7.1). The use of a below-ground channel was to provide a more controlled setting in which to conduct playbacks. As *C. oblonga* are cryptic animals, an artificial environment enabled viewing of turtles to record their responses to the sound stimulus. While it was understood that turtles would normally be communicating in acoustically compromised situations (e.g. due to bubbles, water depth, influence of wind etc) and not in artificial channels; this set-up was constructed to ensure turtles could hear the calls and to control for a number of variables that may be present in a natural setting e.g. wind, noise produced from wavelets slapping on the cable or water birds, and this set-up also controlled for the sound contributions from other aquatic organisms (most of which are unidentified). Two sleepers lay across the center of the channel to simulate ‘hide’ for the turtle. This was to encourage the turtle to be central in the artificial channel for playback experiments and also to provide an alternative place for turtles to hide instead of around the speakers. Two speakers were used – one at either end of the channel with one being a ‘dummy’ speaker (these were swapped around) and the hydrophone placed centrally.

Figure.7.1. The below ground channels used for playback of the turtle 'pulse-bout'.

7.2.6 Playback Responses

Each turtle was allowed a minimum of one-hour acclimatisation in the artificial channel prior to playback. As the underwater behaviour of these turtles was unknown and any subtle behaviour would likely go unnoticed, only simple responses were recorded (see Falls, 1992). These consisted of: 1.) Movement towards or away from the speaker; and 2.) A vocal response and if so, what category of call and how many calls were made; and lastly 3.) No response (i.e. no change from the 'zero state').

7.3 Results

7.3.1 Free-field Playback Trials

In this experiment, 29 wild turtles swam past the speaker. One-way analysis of variance revealed that the time spent remaining still with necks flexed or out-stretched during turtle calls, white noise or silence were not equal between the groups ($F_{2,24}=23.04$, p-value< 0.001) and that the mean time spent in this posture when turtle calls were played (mean=50.75 sec, SD=26.13, n=9) was significantly longer compared to the other two groups: white noise (mean=3.5 sec, SD=4.72, n=8) (t-stat=-5.126, df=15, p-value<0.001); and silence (mean=6.62 sec, SD=4.90, n=12) (t-stat=5.347, df=19, p-value<0.001).

7.3.2 Playback of the 'pulse-bout' in the constructed channels

There were no antiphonal (vocal) responses to the 'song' and neither was there an obvious phonotaxic (behavioural) response by either male or female turtles when the 'song' was played under artificial conditions. Half the females showed no response to the song with two moving away from the sound source. No trend was evident in responses by males (Figure. 7.2).

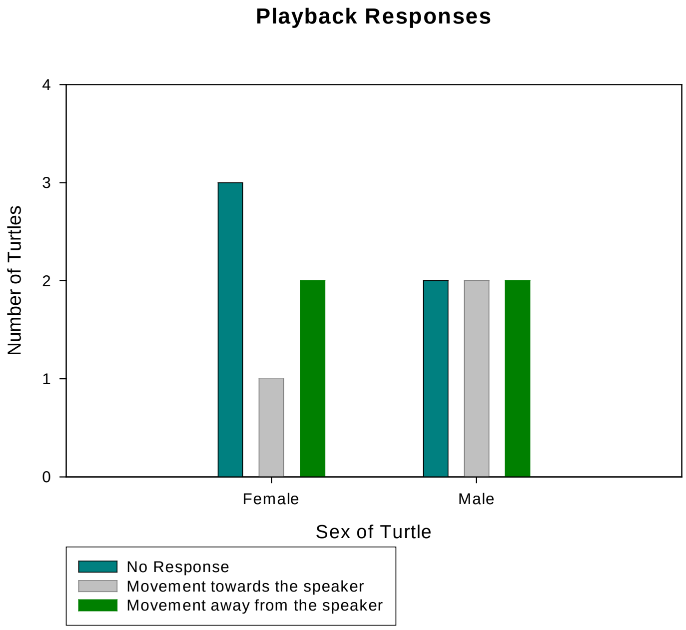


Figure.7.2. Responses of turtles to playback of the 'song' under artificial conditions in the constructed channel. Water temperature 11ºC.

7.4 Discussion

The behavioural responses observed in the preliminary free-field play back trials where the turtle stopped swimming and either had its neck flexed or outstretched and may or may not have looked a the speaker; was considered to be an 'alert posture'. A similar type of posture has been described in Galeotti, *et al* (2004) where the Hermann's tortoises stopped moving, extended their necks and appeared to look at the speaker when broadcast calls were made of their calls. The results are also suggestive of vocalisations being 'signals' as defined by Busnel (1963) - where an observable and (sometimes) measurable reaction occurs in a receiving animal. However, as playback studies could only be examined at a rudimentary level here, longer-term studies are further needed to fully elucidate the meaning of the turtle vocalisations and this posture.

It is understood that not every signal will have a specific communicative meaning separate from other calls (Hailman and Ficken, 1996) and some may even have multiple meanings. For example, the juvenile crocodilian contact call is made in non-threatening situations such as when they move around or when they are stationary; or when approaching other juveniles and also in periods of excitement (Britton, 2001).
Edds-Walton (1997, p.49) defined a 'contact call' as being "a call made by a single animal (of either sex or size class), which is physically separated from conspecifics and when called, leads to the approach or acoustical response between the caller and a conspecific". In turtles this was difficult to determine. From the free-field playbacks, turtles usually moved towards the speaker, but it could not be known entirely whether it was the calls or the speaker, or both, which interested them. These turtles appear to be naturally curious animals and will investigate new objects in their environment (pers. obs) and so curiosity may play a factor in the approach towards the speaker. An attempt was made to hide the speaker (in a black plastic rubbish bag) but this resulted in complete attenuation of the signal so this was abandoned.

The position of the head was not considered an indication of interest in the playback call (it was the turtle actually stopping swimming and remaining still that was the important criterion in the 'alert posture' being adopted). While turtles may hear the sound, lack of orientation of turtles towards the sound source is not necessarily an indicator that a turtle has not heard the sound. The 'surface chirps' would have shorter wavelengths in-air at the frequencies used in the 'chirp' calls and the turtles would be expected to obtain directional information for calls elicited in air, but head-turning or movement towards the caller was not readily obvious even when using image intensifying eyewear or when moonlight enabled good vision. Given the paucity of research concerning communications in reptilians as a group and in turtles specifically, it maybe that other more subtle behaviours are occurring in turtles that might be overlooked by a human observer. They may also be using other sensory organs or structures to acquire the necessary information. Aquatic turtles may not need to react visibly or acoustically to acoustic stimuli because they may have the information already by other means e.g. detection of vibrations by barbels or tubercles (Hartline, 1967; Winokur, 1973).

While the 'pulse-bouts' described in Chapter. 5 are hypothesised to be acoustic advertisement displays and associated with reproductive activity; playback of the artificially constructed sequence of the 'pulse-bout' in the channels (and in some free-field broadcasts) did not appear to motivate females or males one way or another. The lack of a response to the playback 'pulse-bout' may have been due to: 1.) Wild-caught turtles being stressed in an artificial setting – particularly as the artificial channel was not set-up to recreate a small wetland and therefore contained no hiding material; 2.) The time of year may have had some influence as the 'songs' were only recorded in September and October (when water temperatures were also warmer) and playback of the ‘pulse-bout’ was conducted in August (although this was still within the breeding months); 3.) Call characteristics may not have been ‘attractive’ to females seeking a mate; or 4.) Females may have already mated or were reproductively unreceptive, and 5.) Distortion from the speaker may have made the ‘pulse-bout’ unrecognisable or unattractive to turtles.

Chapter. 8. GENERAL DISCUSSION

8.1 Overview

This thesis provides evidence that *Chelodina oblonga* utilises an underwater acoustic communication system. *Chelodina oblonga* is not a vocal specialist, i.e. they do not vocalise continuously, and nor they appear to vocalise within all categories of ecological and functional groupings (see Collias, 1960, Busnel, 1963), but the 17 categories described in the vocal repertoire, are suggestive of complex social roles. The present study provides a preliminary categorisation of turtle vocalisations. Future research may elicit information on the significance of the variation within each call, particularly within the polymorphic 'chirp' calls, which may change these categories or even produce a much larger repertoire than described here. Further research on different populations of *C. oblonga*, including those from geographically remote populations or from those populations isolated by urbanisation may reveal additional variations in their vocalisations.

As recorded in many of the terrestrial chelonians, dominant frequencies were more often below 1 kHz with frequency use extending from around 100 Hz to 3.5 kHz in both the percussive and complex calls. 'Clicks', however extended beyond the limit of the recording capabilities of the equipment. While many of the vocalisations are probably utilised for relatively short to medium-range communications, transmission capabilities over longer distances were likely in the calls with dominant low frequency elements e.g. the 'drum rolls' and the sustained vocalisations – although these distances still need to be determined (short, medium and long-range distances are unlikely to correspond to distances in the marine environment).

Based on the frequency ranges recorded for the vocalisations of *C. oblonga*, it might be reasonable to suggest that their aquatic hearing frequency range extends to these higher frequencies. The auditory sensitivity of turtle hearing is considered to fall away above 1-2 kHz (Wever, 1978; Legler, 1993). However, the higher frequencies in some of their harmonically structured calls and given the work by Wever (1978) on the effects of temperature on auditory sensitivity; turtles may well have greater sensitivity at the higher frequencies than previously thought – particularly during the summer months. It is also possible that alternative structures could be involved in sound reception (e.g. Lenhardt, 1982; Ketten *et al.,*1999). Neither the anatomical site at which sound production occurred nor the mechanism is known, however given the structural complexity seen in calls, the process probably involves the movement of air across specific structures using some form of circular air movement.

Despite the assertions of a number of earlier researchers (e.g. Carr, 1952; Mrosovsky, 1972; Gans and Maderson, 1973) who suggested hearing and sound production was an insignificant sense in many reptiles, it is not considered to be the case here given the extent of vocal categories recorded and the results of the preliminary play-back studies. While this thesis is largely a descriptive investigation into the underwater vocalisations of *C. oblonga* and many speculations have been offered in this thesis as to the functional classification of these sounds - sound production has no doubt conferred some evolutionary advantage for these animals and in-depth investigations are required to fully determine the role of sound in the ecology and behaviour of these animals. Whereas the senses of vision, olfactory and vibratory sensitivities are likely to be dominant senses at close range - particularly for prey detection, sound would be a useful adjunct to their other sensory abilities enabling turtles to fully exploit their complex aquatic environments, providing information at distances beyond the acuity of their other senses.

The earlier investigations of ambient noise in wetlands has revealed that sound production is not only restricted to the freshwater turtles, but is also produced by the invertebrates existing within these inland freshwater systems. These animals used frequencies from around 3 kHz extending to as high as 14.5 kHz (although higher frequencies were observed at other times of the year outside the summer 2003 ambient sound study). Calls presented here are only those within the audible range due to limitations imposed by the recording and analysing capabilities of the equipment. A whole new sound field may exist at ultrasonic frequencies. While identification of sound producing organisms was not determined in this study (apart from the turtles), the percussive displays were of types similarly produced by invertebrates in the terrestrial environment, with the exception of the distinctive 'Bird-like-song' recorded at Lake Leschenaultia and Glen Brook Dam.

Due to the paucity of literature on ambient sound fields in freshwater systems comparisons were drawn with the marine environment. A number of similarities between freshwater and marine systems were evident, such as the diel calling cycles – in particular, the chorusing activity from dusk through to midnight. However, differences between the marine and freshwater environments were also evident with the energy in the freshwater choruses higher than typical marine chorus activity. Moreover, the influence of wind at Beaufort Wind Scales of 1 and 2 was negligible in these wetlands compared to the influence of wind at some marine locations at similar wind speeds (see Cato, 1976). It is biological noise which dominates in the freshwater environment at these wind speeds. Based on the three wetlands studied, results suggest that inland waters will likely reveal their own discrete ambient sound fields intrinsic to their own diel periodicities according to the distribution, diversity and abundances of organisms present as well as the influence of seasonal changes. This study forms the basis upon which further research on the sound fields in inland freshwater systems can continue.

In summary, the major findings of this research include the following:

- Only the in-air auditory sensitivities for a single turtle from the family Chelidae has been determined, indicating a high sensitivity in the range from 60-600 Hz extending up to around 1500 Hz (Wever, 1978). No underwater sensitivities have been determined for *C. oblonga.* Most of the literature on vocalisations by chelonians has focused on those sounds associated with breeding activities (e.g. Bogert, 1960; Auffenberg, 1964; Jackson and Awbrey, 1978; McKeown *et al.,* 1990; Sacchi *et al.,* 2003 and Galeotti *et al.,* 2004), with no known studies on the underwater vocalisations of any chelonian.

- *Chelodina oblonga* has an extensive underwater repertoire of at least 17 calls, as well in-air surface calls. Frequency use ranged from around 100 Hz up to 3.5 kHz, with 'clicks' extending beyond the 20 kHz upper limit of the recording equipment. Most calls were considered to be relatively close to medium range calls, but the percussive calls containing dominant low frequency elements, were potentially for propagation over relatively longer distances.

- *Chelodina oblonga* is capable of sustained vocalisations lasting up to at least nine minutes. These 'pulse-bouts' are hypothesised to be an acoustic advertisement display possibly related to breeding activities. This was composed of two distinct phases with the first phase consisting of stereotypical pulses either singly or in a series; and the second phase consisting of a series of complex rapid pulses in doublets, roll-down triplets and quadruplets known as the 'vibrato'.

- The 'Chirp' calls are hypothesised to be *C. oblonga's* main contact calls and playback of these calls has revealed that turtles respond to these by adopting an 'alert posture'.

- Summer ambient sound recordings occurred in three clear-water wetlands and revealed temporal and spatial differences in the macroinvertebrate distributions and the biologic calling activity. Seven distinct calls were recognised with noise levels greatest at dusk and to a lesser extent at midnight with chorusing only evident at the most enriched wetland. Biologics used frequencies ranging from 3 kHz up to around 14 kHz with the exception of the 'bird-like song' which extended from 500 Hz up to around 10 kHz. There was negligible sound contribution to ambient noise at low wind speeds of Beaufort Wind Scale

8.2 Further Research Questions

Further investigations are needed on many different aspects of sound and its use by *C. oblonga* as well as the use of and importance of sound for other freshwater fauna.

8.2.1 Turtles

Future research should now be focused on investigating the importance of sound to *Chelodina oblonga* by: (1) examining the behavioural significance of all turtle vocalisations, (2) the biological function of the turtle calls and (3) their sound-producing mechanism. The results of such investigations may have repercussions for turtles existing in degraded wetlands where acoustic properties are being altered due to human influences.

Additional investigations should explore the possibility that dialect differences may occur between geographically remote populations. This was suggested in the differences noted between vocalisations produced by the Masons Gardens turtles and those produced by the Pinwernying Dam turtles. For example, 'Chirp' calls from the Pinweryning Dam males were often more richly harmonic and many appeared to be of longer duration with more frequency modulation than vocalisations produced by the Masons Gardens turtles. Differences in acoustic output (amplitude, duration or complexity), can also be related to size of the animal such as heard in the Tokay gecko (Tang *et al.,* 2001) which needs to be investigated for *C. oblonga.*

Future research will also need to examine issues of acoustic subtleties such as rate of frequency modulation or call rate and what these may mean for freshwater turtles, particularly in mate selection (see Sacchi *et al.,* 2003; Galleotti *et al.,*2004). Differences in structure of calls can occur with changes in temperature (Pough *et al.,* 1998; King, 1999a) and these differences also need to be investigated further with turtle calls produced under winter vs summer sound conditions.

A further question is to examine how widespread underwater sound communication is in the other Australian chelid turtles. Underwater sound production in chelonians may be more widespread than realised and future research should be extended to include all freshwater turtle groups including the marine turtles. Based on the in-air vocalisations reported for marine turtles (Carr, 1952; Mrosovsky, 1972) and the use of sound to call these animals by some indigenous peoples (Kuchling, pers. comm.); the possibility exists that marine turtles may use an underwater acoustic communication system also.

8.2.2 Propagation Characteristics within the Wetland Environment

Future research needs to address source levels of turtle sounds and propagation characteristics in different wetlands. Investigations are needed to understand the propagation of sound energy which would help understand how far turtle calls would travel and the distances over which these animals are communicating within these very complex and shallow freshwater environments. Investigations would need to include; both summer and winter sound-speed profiles (Jensen and Kuperman, 1982) as well as the influence of the gas content of the sediments (Anderson and Hampton, 1980a,b). This information would be considered important in communication (see Myrberg *et al.,* 1978) and have some relevance for aquatic communication in wetlands so that researchers can more fully understand the factors affecting transmission of turtle vocalisations in the varied settings presented in their habitat. Importantly, it would give some understanding to researchers and to environmental managers as to what effects human activities might have on the acoustical characteristics within these environments and the implications for the turtle populations utilising them. For example: what acoustical changes might occur with increased sediment loading as a result of land clearing for development adjacent to the wetland; or what impacts there might be to breeding activities if deeper water depths are unavailable as a result of draw-down from local groundwater bores if turtles need to call over long distances for a mate.

8.2.3 Anthropogenic Noise Contributions

While the wetlands of the Swan Coastal Plain are generally used as areas of passive recreation such as: walking and bird-watching and as picnic sites; with activities such as boating or swimming usually prohibited; the dense urbanisation around these areas suggest that urban noise contributions and impacts on the organisms within a wetland are likely to be high. The impacts of urban noise, particularly those which produce persistent noise such as water aerators and what effects these may have on the food webs within these ecosystems needs to be more fully investigated.

8.2.4 Ambient Noise

This research has also raised the prospect of examining the role and importance of sound for the aquatic invertebrate communities utilising inland wetlands. In a broader geographical context; Tucker and Gazey (1966) believe animals from colder and temperate waters may be less noisier than those animals from the tropical and warmer waters. To test for this in freshwater environments, comparisons could be made between a range of wetlands – from tropical to temperate to colder climates, including those wetlands that contain water less permanently e.g. ephemeral wetlands in the Goldfields; with the prospect of utilising sound as a bio-monitoring tool for wetland assessments. Also of interest is the sound field in inland saline systems and even investigating the possible use of sound by the stygofauna in karst systems. Further investigations could include comparisons with the diel periodicity in the sound cycles between a range of inland systems and the marine environment e.g. Lake Eyre.

8.3 Limitations

There were many logistical constraints encountered in this research. For example, the need for battery recharge and tape length limited recording times, and also the ease at which wetlands on the Swan Coastal Plain could be accessed by the general public meant that equipment could not be left unattended. These constraints meant that 24-hour recordings could not be undertaken and so representation of the sound field could only be determined for short blocks of time and so there may have been calls that were missed and remain unrepresented in this study.

Difficulties also arose trying to observe turtles in a natural setting. *Chelodina oblonga* are cryptic and mobile animals, and even when they were observed underwater in a natural setting, our presence disturbed them and they swam away and hid amongst the submerged vegetation. Despite the attempts to recreate a 'natural setting' within the artificial ponds, it is recognised that any observed behaviour in an artificial setting may not necessarily reflect what occurs in the wild. For example, high densities in artificial ponds may have produced exaggerated responses such as aggressive interactions.

Although recordings were made from turtles constrained within an artificial environment, it was still difficult to determine which animal actually produced the call. This difficulty was exacerbated by the lack of obvious signs of sound production in *C. oblonga*. Some of the physical limitations associated with use of an artificial pond included reverberation from complex habitat features and the likely distortion of acoustic signals from reflective interfaces (Hawkins and Myrberg, 1983). Although a significant proportion of the tapes were utilised in data analysis, the selected portion could only provide a general representation of the sound. It was not possible to utilise all recorded data and perhaps in doing this, some important calls or subtle differences may have been over-looked – in particular, the 'chirp' calls which had a polymorphic nature.

Although the research described in this thesis was undertaken as a scientific study, it also required a considerable amount of time to be devoted to issues of turtle husbandry. A significant amount of time was dedicated to cleaning the artificial ponds and their filtration systems. Turtles that became sick needed special attention. The subsequent death of some turtles, which appeared to be related to undetermined factors associated with captivity, also caused much emotional anguish.

In conclusion, the major limitation of this research project was considered to be one of time. Research such as this can only give a ‘snapshot’ view into the vocal world of these animals. With every new result, more questions arose and many still remain unanswered.

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APPENDIX. 1.

Comparing Wind Speed Data at N S W E sites

Wind speed data was analysed using one-way analysis of variance for each wind speed using recordings made at all sites (NSWE) (Tables. 1,2,3). As there was no significant difference between the recordings of NSWE, the data was then averaged to give a mean frequency spectrum for each Beaufort Wind Scale speed of 0,1 & 2. Only Blue Gum Lake wind speed data was compared for all three wind speeds and if was found that there was no significant difference between Beaufort Wind Scale speeds of 0,1 or 2 (Table. 4). Spectra were also examined as overlays, but no obvious deviations were evident at Beaufort Wind Scale speeds of 0, 1 or 2.

Table.1. One-way analysis of variance at Beaufort Wind Scale 0 using sites N S W E at each wetland.

WIND SPEED **Beaufort Scale 0**				
Wetland	*F-ratio*	*Degrees of freedom*	*p-value*	*Pooled Std Dev*
Glen Brook Dam	0.01	3, 92	0.998	6.689
Lake Leschenaultia	0.01	3, 92	0.999	6.246
Blue Gum Lake	0.02	3, 92	0.997	6.348

Table.2. One-way analysis of variance at Beaufort Wind Scale 1 using sites N S W E at each wetland.

WIND SPEED **Beaufort Scale 1**				
Wetland	*F-ratio*	*Degrees of freedom*	*p-value*	*Pooled Std Dev*
Glen Brook Dam	0.00	3, 92	1.0	6.079
Lake Leschenaultia	0.39	3, 92	0.763	6.768
Blue Gum Lake	0.08	3, 92	0.972	6.157

Table.3. One-way analysis of variance at Beaufort Wind Scale 2 using sites N S W E at each wetland.

WIND SPEED
Beaufort Scale 2

Wetland	*F-ratio*	*Degrees of freedom*	*p-value*	*Pooled Std Dev*
Glen Brook Dam	0.01	3, 92	0.999	6.635
Lake Leschenaultia	not available	not available	not available	not available
Blue Gum Lake	0.02	3, 92	0.997	6.346

Table.4. One-way analysis of variance at Beaufort Wind Scale 0,1 & 2 at Blue Gum Lake.

AVERAGED WIND SPEED
Beaufort Scale 0,1 & 2

Wetland	*F-ratio*	*Degrees of freedom*	*p-value*
Blue Gum Lake	0.02	3, 71	0.977

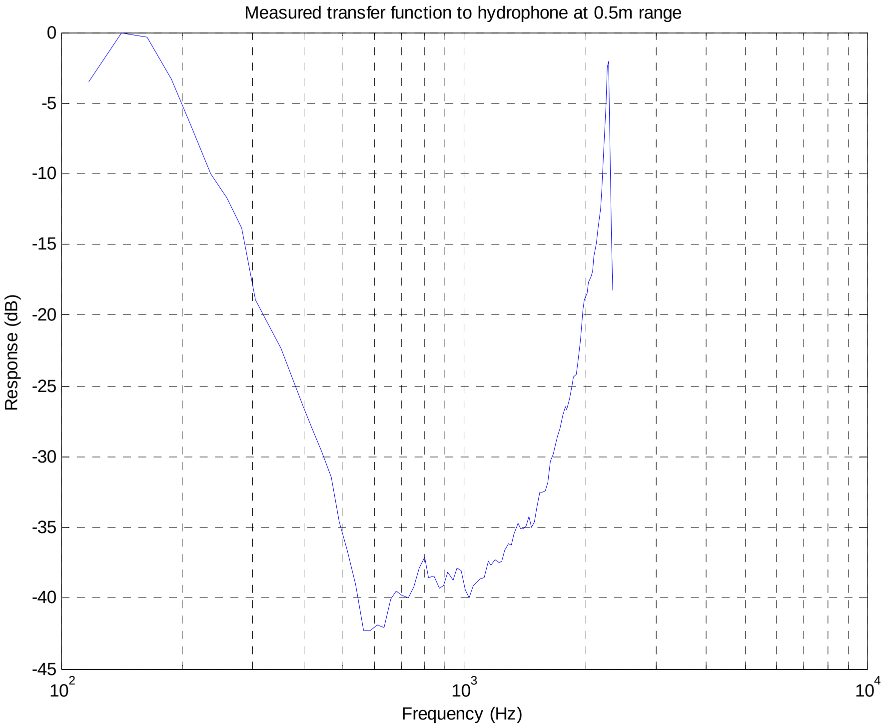
APPENDIX. 2.

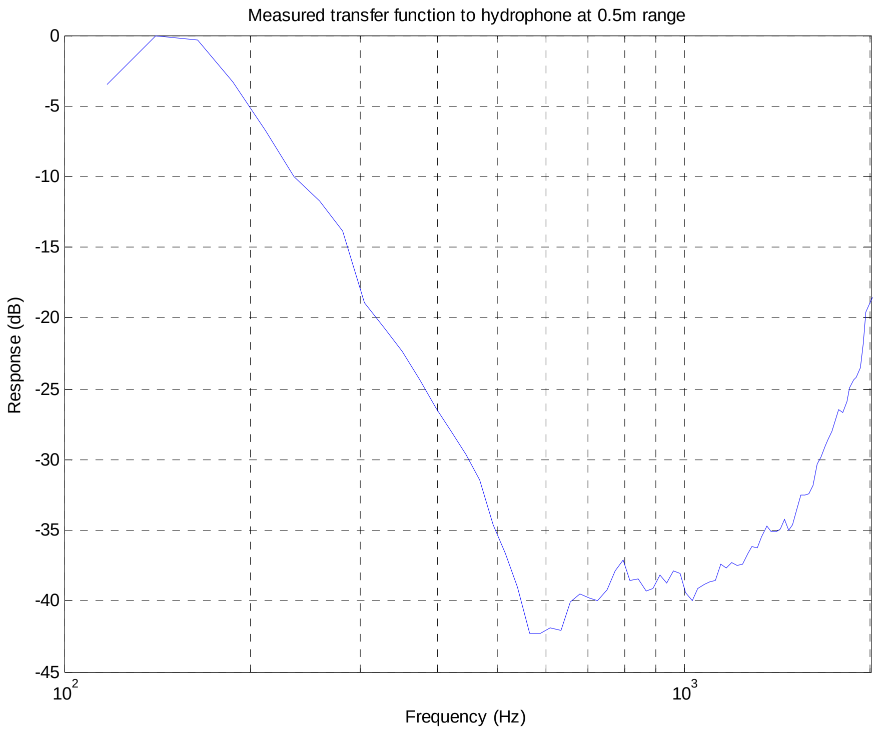
Inverse filtering procedure

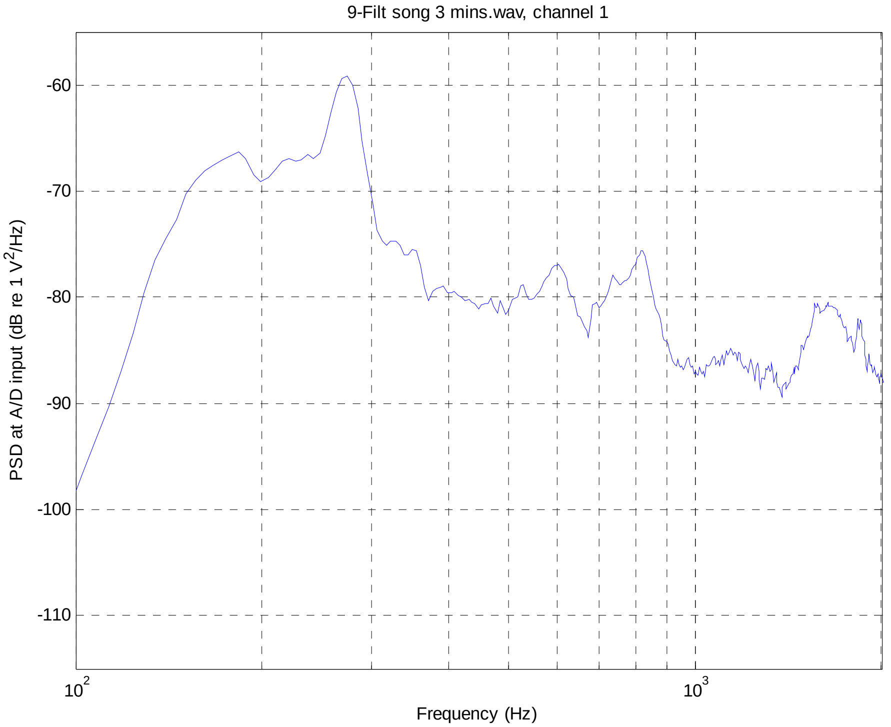
Produced by Dr Alec Duncan
CMST, Curtin University, Perth WA.

The transfer function was measured by applying a slow frequency sweep signal to the acoustic source and recording the sound level at a range of 0.5m using a hydrophone. The ratio of the recording system output amplitude to the driving signal amplitude then gives the transfer function amplitude as a function of frequency.

To compensate any recorded response for this (or any other) transfer function the following procedure was carried out:
Fourier transform the signal to obtain its frequency spectrum.
Divide the signal spectrum by the transfer function amplitude at each frequency.
Inverse Fourier transform to obtain the filtered time series.
(There are some subtleties here – the Fourier transform gives results for both positive and negative frequencies. The transfer function therefore has to be halved in amplitude and copied to negative frequencies before multiplying by the signal spectrum.)

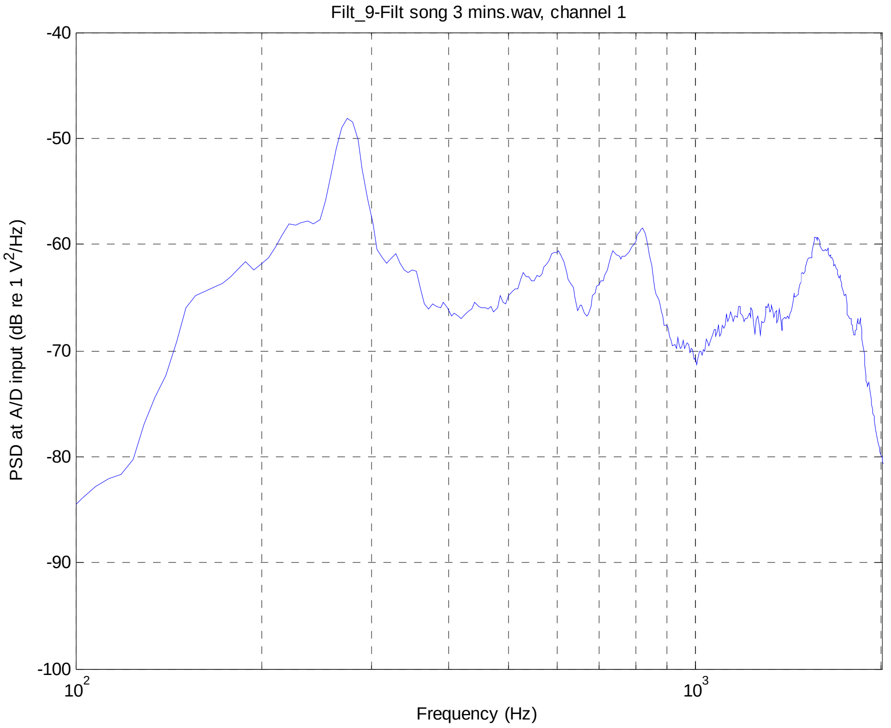




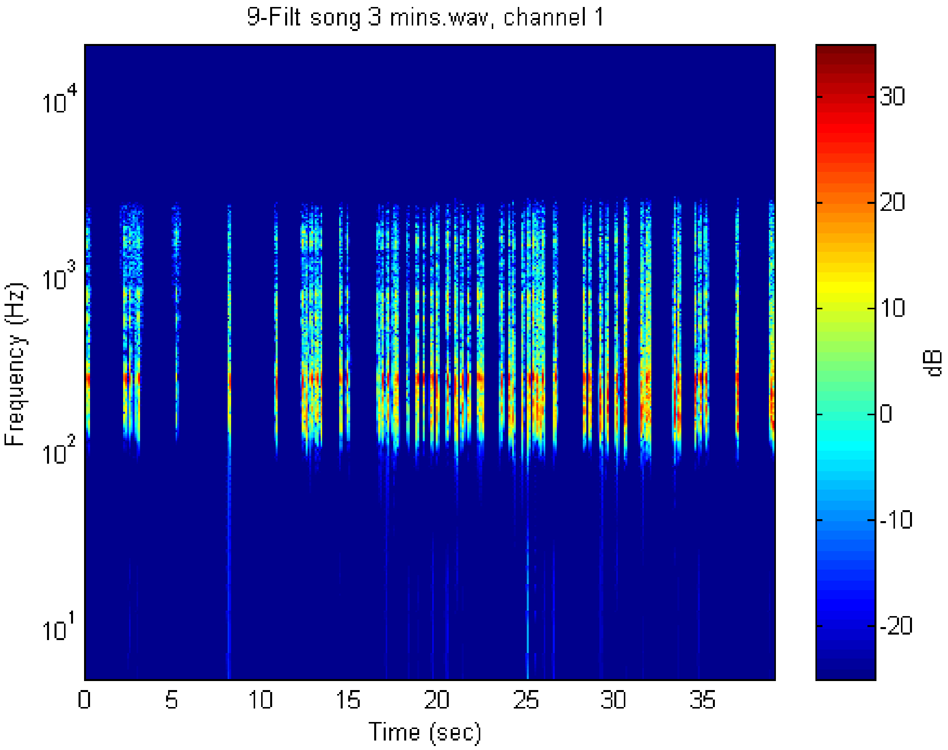


No inverse filtering

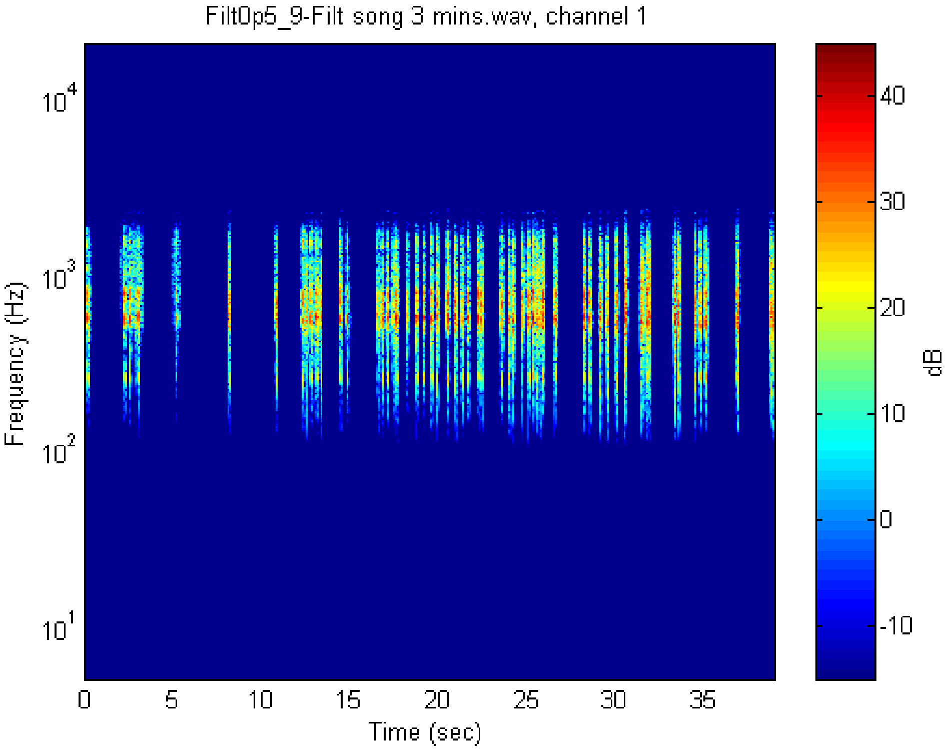
Inverse filtered using transfer function derived from sweep in tank @0.5m range



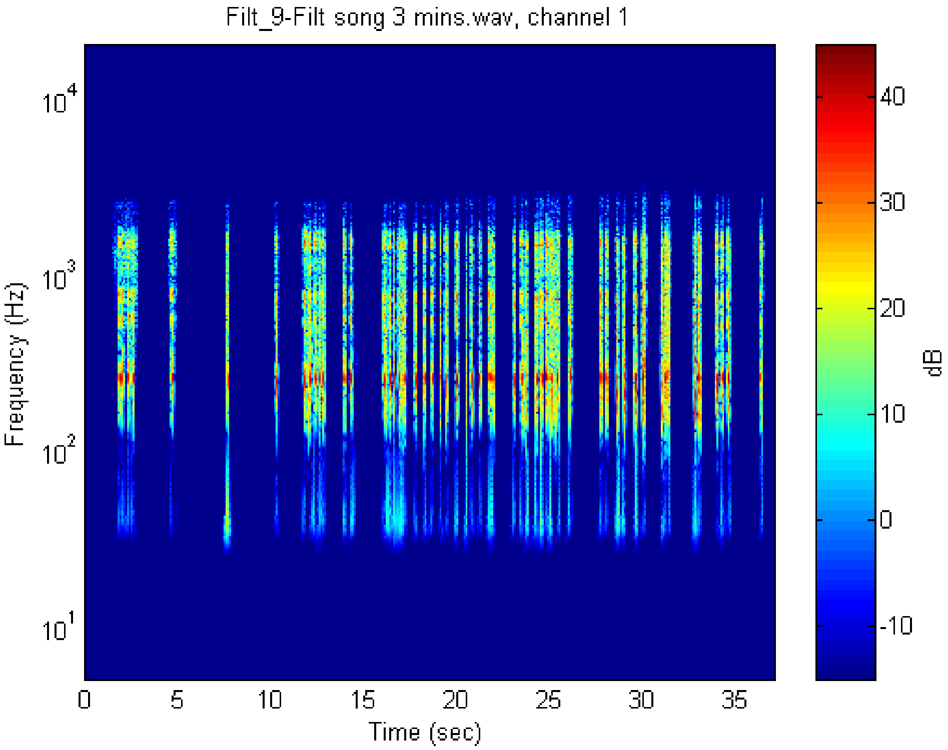
Inverse filtered using source free-field response



First 40 sec. Unfiltered.



Inverse filtered using transfer function derived from sweep in tank @0.5m range



Inverse filtered using transducer free-field response