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Effect of water depth and water velocity upon the surfacing frequency of the bimodally respiring freshwater turtle, *Rheodytes leukops*

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Summary

This study examines the effect of increasing water depth and water velocity upon the surfacing behaviour of the bimodally respiring turtle, *Rheodytes leukops*. Surfacing frequency was recorded for *R. leukops* at varying water depths (50, 100, 150 cm) and water velocities (5, 15, 30 cm s^{-1}) during independent trials to provide an indirect cost–benefit analysis of aquatic *versus* pulmonary respiration. With increasing water velocity, *R. leukops* decreased its surfacing frequency twentyfold, thus suggesting a heightened reliance upon aquatic gas exchange. An elevated reliance upon aquatic respiration, which presumably translates into a decreased air-breathing frequency, may be metabolically more efficient for *R. leukops* compared to the expenditure (i.e. time and energy) associated with air-breathing within fast-flowing riffle zones. Additionally, *R. leukops* at higher water velocities preferentially selected low-velocity microhabitats, presumably to avoid the metabolic expenditure associated with high water flow. Alternatively, increasing water depth had no effect upon the surfacing frequency of *R. leukops*, suggesting little to no change in the respiratory partitioning of the species across treatment settings. Routinely long dives (>90 min) recorded for *R. leukops* indicate a high reliance upon aquatic O_2 uptake regardless of water depth. Moreover, metabolic and temporal costs attributed to pulmonary gas exchange within a pool-like environment were likely minimal for *R. leukops*, irrespective of water depth.

Key words: turtle, *Rheodytes leukops*, diving, bimodal respiration, aquatic respiration.

Introduction

Principles of resource ecology predict that as the travel cost (e.g. time and energy) to the site of resource gain increases, the relative value of the resource diminishes (Andersson, 1978). Although primarily applied in behavioural investigations concerned with optimal foraging (see Pyke, 1984; Stephens and Krebs, 1986; Kamil et al., 1987), resource maximisation models also pertain to the acquisition of oxygen from water *versus* air in bimodally respiring vertebrates (Kramer, 1988; Boutilier, 1990). The 'theory of optimal breathing' (Kramer, 1988) predicts that, as the cost of travel for pulmonary gas exchange increases, the proportion of total V_{O_2} accounted for by atmospheric O_2 uptake decreases relative to aquatic V_{O_2}.

Habitat selection by aquatic vertebrates affects metabolic and temporal cost associated with travel to and from the surface for pulmonary respiration (Kramer and McClure, 1981; Feder and Moran, 1985; Kramer, 1983; Bevan and Kramer, 1987). As water depth increases, select air-breathing fish and amphibian species offset the heightened transit cost associated with aerial respiration by increasing their reliance upon aquatic respiration, as reflected by a decreased surfacing frequency or an increased aquatic ventilation rate (Kramer and McClure, 1981; Feder and Moran, 1985; Bevan and Kramer, 1986, 1987; Shannon and Kramer, 1988). However, the interaction of water depth, surfacing behaviour and respiratory partitioning in aquatically respiring turtles has primarily been ignored (Hua and Wang, 1993), despite the fact that diving respiratory investigations on chelonians have generally been conducted under unnaturally shallow conditions (i.e. <50 cm). An additional factor that has been overlooked amongst bimodally respiring vertebrates is water velocity. Aquatically respiring turtles inhabiting areas of high water flow would probably experience difficulty in reaching the surface for pulmonary gas exchange, thus leaving the animal with the choice of increasing its dependence upon aquatic O_2 uptake or moving into areas of slower flow (i.e. pools). Therefore, the aim of this study was to investigate the effect of increasing water depth and water velocity upon the surfacing behaviour of a bimodally respiring freshwater turtle in order to providing an indirect cost–benefit analysis of aquatic *versus* pulmonary respiration.

The Fitzroy turtle *Rheodytes leukops* is a short-neck Australian chelid whose preferred habitat is described as shallow, fast-flowing riffle zones characterised by well-oxygenated water (Legler and Cann, 1980; Cann, 1998; Tucker et al., 2001). Compared to other bimodally respiring turtles, *R.*

leukops has '*seemingly achieved the greatest emancipation from air-breathing of any aquatic chelonian*' (Legler and Georges, 1993). *R. leukops* obtains up to 70% of its total V_{O_2} *via* aquatic routes, with the turtle's highly modified cloacal bursae accounting for the majority of aquatic O_2 uptake (Priest, 1997; Franklin, 2000). The bursae sacs of *R. leukops* are ventilated *via* two antagonistic muscle groups (Legler, 1993), with aquatic gas exchange facilitated by highly vascularised, branching papillae that align the mucosal lining of the bursae walls and effectively increase the surface area 16-fold (Legler and Georges, 1993; Priest, 1997). The high reliance of *R. leukops* upon aquatic respiration translates into significantly longer dives compared to primarily air-breathing turtles (Priest and Franklin, 2002; Gordos et al., 2003a), with aerobic dives of up to 12 h being recorded (Gordos et al., 2004).

Materials and methods

Animal capture and husbandry

Rheodytes leukops (Legler and Cann) were captured within a stretch of the Connors River (22°12.525″S, 149°01.311″E) in central Queensland, Australia. Upon capture, mass, straight carapace length (SCL), sex and maturity status were recorded (Table 1). All *R. leukops* were considered mature as determined by size, with sex determination being based upon tail-length measurements and cloacal vent location (Legler and Cann, 1980). Turtles were transported to The University of Queensland where they were housed within two 1000 l holding tanks (2.7 m×1.2 m×0.4 m) for a 3-week period prior to experimentation. Tanks were located in a constant temperature room (25°C) with a photoperiod set at 12 h:12 h L:D. Turtles were fed prawns and meal worms twice weekly to satiation, with water changes occurring the following day using aged water.

Table 1. *Summary of biological data for* Rheodytes leukops *tested in water depth and water velocity trials*

Experiment	*N*	Mass (kg)	SCL (cm)
Water depth			
Males	8	1.31±0.54	23.9±0.2
Females	8	1.36±0.62	23.6±0.3
Water velocity (cm s^{-1})			
5			
Males	6	1.35±0.10	24.4±0.5
Females	6	1.41±0.96	24.1±0.5
15			
Males	6	1.27±0.70	23.7±0.4
Females	6	1.34±0.73	23.8±0.4
30			
Males	6	1.29±0.82	23.7±0.3
Females	6	1.40±0.86	23.6±0.4

SCL, straight carapace length.
Values are means ± S.E.M.

Water depth

Trials investigating the effect of water depth upon the surfacing frequency of *R. leukops* were carried out within a purpose-built dive tank (2.0 m×2.0 m×2.0 m) fitted with a dual biological/sand filter system (Model S166T High-Rate sand filter; Hayward Pool Products Inc.; Elizabeth, NJ, USA). Two large acrylic windows fitted on the front wall of the tank facilitated videotaping of turtle behaviour during experimentation (e.g. active periods and surfacing episodes). Early observations indicated that *R. leukops* had difficulty ascending to and remaining near the water's surface due to the negatively buoyant nature of the species. Therefore, net structures composed of plastic mesh (5.0 cm×5.0 cm) stretched over a PVC pipe frame were placed against each wall of the tank to provide adequate footing for the turtles to climb. During experimentation, *R. leukops* used the net structures exclusively to ascend to the surface rather than swimming. Additionally within the tank, four refuges (halved 30 cm diameter PVC pipe) provided a darkened environment for turtles to reside in. Water temperature was held constant throughout experimentation at 25.0±0.1°C using a counteractive heating (Compu-heat Pool heater; 6 kw; Gold Coast; Australia) and cooling system (air conditioner).

Time-depth recorders (TDRs; 55 mm×16 mm; 1 g in water; Model LTD_10, Lotek Marine Technologies Inc., Newfoundland, Canada) were attached to two male and two female *R. leukops* (Gordos and Franklin, 2002) prior to the turtles' introduction into the experimental tank. For each 1-week trial, water depth within the dive tank was randomly pre-set to one of three depths: 50 cm, 100 cm and 150 cm. Aquatic P_{O_2} levels were recorded at the beginning and end of each depth trial (YSI Model 55 Dissolved oxygen/temperature system, Yellow Springs, OH, USA), with dissolved oxygen levels remaining near saturation (143.0±1.1 mmHg) throughout experimentation. Three days after being introduced into the dive tank, the light cycle was changed from a 12 h:12 h L:D to a 24 h:0 h L:D regime in order to facilitate videotaping of the frequently long dives (>12 h) observed for *R. leukops*, as well as to limit the effect of photoperiod upon the activity levels (and hence surfacing frequency) of the turtles (Gordos et al., 2003b). Additionally, faeces were removed from the bottom of the tank on the third day, whereupon access to the room was prohibited for the remainder of the trial. Continuous TDR sampling and closed circuit videotaping commenced at midnight of the start of the fifth day and proceeded for the following 3 days, with enabled TDRs logging water pressure (±4.0 cm) every 4 s and water temperature (±0.1°C) every 5 min. At the completion of each 1-week trial, turtles were removed from the dive tank and placed back into their original holding troughs where a 12 h:12 h L:D cycle and normal feeding regime were resumed. Each group of four turtles (2 male; 2 female) was tested at each of the three experimental depths, with one week's rest occurring between successive trials.

Videotaped trials were analysed for surfacing and dive duration for individual turtles using a time-lapse video recorder. Assignment of surfacing episodes to specific turtles was facilitated through the analysis of a turtle's TDR dive profile. For each turtle, median dive time, maximum dive time, median surfacing time and surfacings h^{-1} were determined at all three depths. Additionally within each trial, ten surfacing events were randomly selected for each turtle to calculate mean ascent time from the floor of the dive tank to the surface. Finally, vertical displacement day^{-1} was determined for individual turtles from TDR records. Differences in diving performance and surfacing behaviour between genders and among water depth treatments were investigated using a two-way repeated measures analysis of variance (RM-ANOVA) on one factor (P<0.05). Following a significant finding, a *post hoc* Tukey's test was used to elucidate specific differences between treatment groups. In cases where assumptions of equal variance or normality failed, data were ranked prior to analysis.

Distributions of dive times and surfacing duration were produced for all three depths, with histogram intervals selected based upon results from previous investigations (Gordos and Franklin, 2002; Gordos et al., 2003a) and from an initial pilot study. Differences among depth treatments in the frequency of dives or surfacings within a specific interval period (e.g. dives <15 min) were analysed using a two-way RM-ANOVA (water depth and gender) on one factor (P<0.05). To achieve normality, proportions were square-root and arcsine transformed ($p' = \arcsine\sqrt{p}$) prior to statistical analysis. Following a significant finding, a *post hoc* Tukey's test was used to elucidate specific differences between treatment groups. For correlation analyses between dive duration and subsequent surfacing intervals, results from all *R. leukops* were combined within each experimental depth, whereupon Pearson's product moment was determined.

Water velocity

Trials investigating the effect of increasing water velocity upon the surfacing frequency of *R. leukops* were conducted in a custom-built fiberglass flume (4.0 m×1.6 m×0.65 m). Water depth within the flume was maintained at 50 cm, while water temperature was held constant by a counteractive heating/cooling system (Julabo heater; John Morris Scientific Pty, Ltd., Seelbach, Germany). Two Minn Kota electric motors (Racine, WI, USA) propelled the water around the oval-shaped flume at one of three experimental velocities: 5, 15 and 30 cm s^{-1}. Experimental water velocities were selected based upon field recordings of water flow in pools and riffle sections of the species' type locality (Legler and Cann, 1980; Gordos and Franklin, 2002). The study area (1.5 m×0.60 m×0.65 m) was positioned on the opposite side of the flume with respect to the motors, being separated by a 30 cm wide fibreglass partition. Horizontally laid PVC pipes (5 cm diameter; 0.40 m×0.60 m×0.65 m) enclosed the study area both at the front and back while simultaneously promoting laminar flow. Observations at all three experimental water velocities indicated that the PVC barriers also provided a suitable substrate for *R. leukops* to climb to the water's surface. Finally, a one-way mirrored glass wall stretching the length of the study area facilitated videotaping of flume trials.

Initial observations indicated that at higher water velocities (i.e. 30 cm s^{-1}), *R. leukops* had difficulty maintaining its position on the flume floor due to a lack of footholds. Therefore, a perspex floor with raised strips (1 cm high) placed perpendicular to the flow of water provided a substrate that turtles could get their claws under. Furthermore, field observations of *R. leukops* indicated that turtles in riffle zones often reside behind or under rocks and submerged logs (Legler and Cann, 1980; M. A. Gordos, unpublished observation), presumably to avoid direct exposure to high water velocity flows. Thus, in an attempt to mimic field conditions as well as to provide a behavioural investigation into habitat preference, a ramped acrylic deflector (10 cm high) that spanned the width of the observation area was fixed onto the perspex floor midway along the length of the study area (see Fig. 1). Water velocity profiles (Flo-mate Model 2000, Marsh-McBirney, Inc., Frederick, MD, USA) recorded at three depths (5, 25, 45 cm) at each of five lengths (5, 37.5, 75, 112.5, 145 cm; Fig. 1) along the study area show that water velocity directly behind the deflector was reduced considerably at 15 cm s^{-1} and 30 cm s^{-1}, indicating that the deflector effectively split the study area into two experimental velocity treatments (i.e. high and low water velocity) for the two fastest water velocity settings. Water velocity above the height of the deflector (i.e. at 25 cm depth) remained at or above the initial trial setting for the length of the study area (see Fig. 1).

Trial order regarding water velocity and sex was randomised prior to the start of the study. *R. leukops* were placed into the darkened flume at 18:00 h on the day preceding experimentation to allow the turtle to become accustomed to the flume. In order to provide a directional cue to the turtles (i.e. upstream *versus* downstream), water velocity was initially set at 2 cm s^{-1}. The following morning, timed lights switched on at 06:00 h. At 08:00 h, water temperature and aquatic P_{O_2} level were recorded (YSI Model 55 Dissolved oxygen/temperature system) prior to setting the experimental water velocity. Initial observations of turtle behaviour indicated that *R. leukops* adjusted to the set water velocity within 2 h; therefore, this period of time was excluded from data analysis. A closed circuit time-lapse video system recorded turtle behaviour over 8 h from 10:00–18:00 h. After the completion of the trial, turtles were removed from the flume and water temperature and aquatic P_{O_2} level were again recorded. Water temperature remained constant throughout experimentation (25.3±0.02°C), while aquatic P_{O_2} level remained near saturation (146.4±0.4 mmHg) for the duration of the study. A total of 36 *R. leukops* were used during the flume trials, with six male and six female turtles being run at each of the three water velocity settings (Table 1).

Videotapes were analysed for the number of surfacing episodes per trial, as well as for the percentage of bottom time spent in front of the deflector. Due to the lack of surfacing events at 30 cm s^{-1}, other parameters such as dive duration and

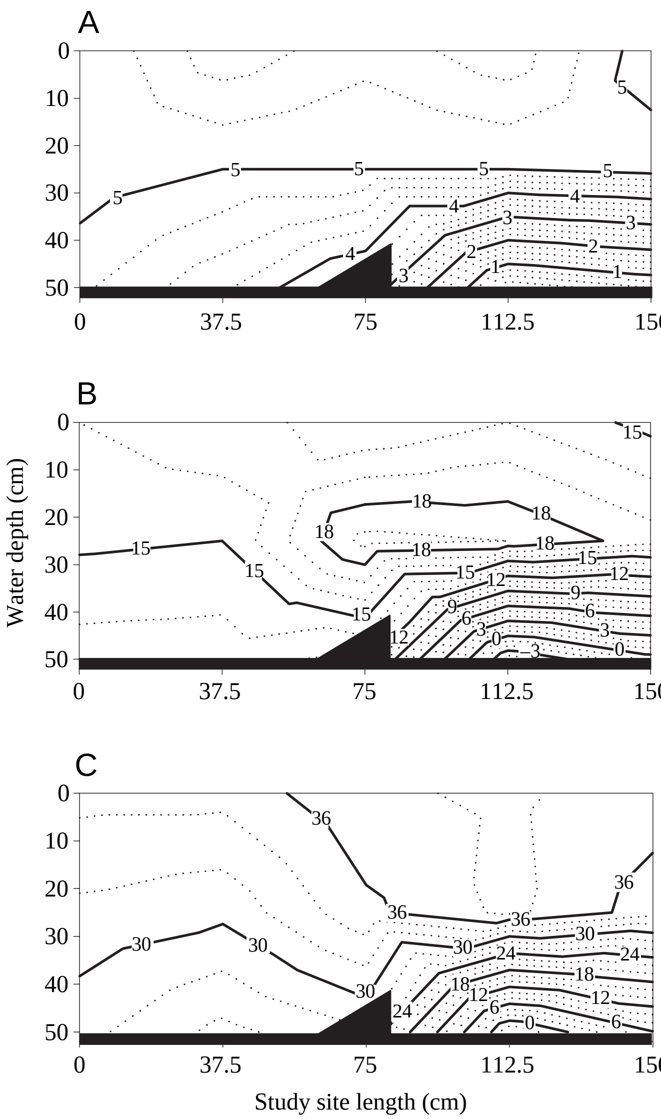

Fig. 1. Water velocity profiles throughout the study area at trial settings of (A) 5 cm s^{-1}, (B) 15 cm s^{-1} and (C) 30 cm s^{-1}. Major (solid lines) and minor (dotted lines) water velocity contours (cm s^{-1}) were computer generated based upon repeated water velocity recording at three depths (5, 25 and 45 cm) at each of five positions along the flume (5, 37.5, 75, 112.5 and 145 cm). A false floor with an inclined water deflector (10 cm high) was positioned on the bottom of the flume.

surfacing time could not be calculated. To achieve normality, the percentage of time spent in front of the deflector was square-root and arcsine transformed ($p'=\text{arcsine}\sqrt{p}$) prior to statistical analysis. Individual values for surfacing events per trial and proportion of time spent in front of the deflector were averaged together within each gender for each treatment group for analysis using a two-way ANOVA ($P<0.05$). Following a significant finding, a *post hoc* Tukey's test was used to investigate specific differences among groups. Where assumptions of normality or equal variance failed, data were ranked prior to analysis.

For all diving and surfacing parameters recorded during the two investigations, no effect was observed regarding the turtle's gender. Additionally, no statistical difference was observed in *R. leukops*' mass or SCL between genders for both studies (Table 1). Therefore, results for male and female *R. leukops* were combined and presented as a single figure. However, due to the small sample size within each experiment, the power of the performed tests with respect to gender was low and is acknowledged as a possible limiting factor. Unless specified, all results hereafter are presented as means ± S.E.M.

Results

Water depth

In general, *R. leukops* spent the majority of time lying motionless on the bottom of the tank, often within the refuges provided. Occasionally though, individual *R. leukops* would display burst periods of activity characterised by multiple surfacing episodes lasting from several minutes to a few hours regardless of water depth. No difference was observed in median dive duration ($F_{[2,28]}$=1.518; P=0.237), maximum dive duration ($F_{[2,28]}$=0.987; P=0.385) and surfacings h^{-1} ($F_{[2,28]}$=0.565; P=0.574) among the water depth treatments for *R. leukops* (Table 2). Increasing water depth did, however, influence the distribution of dives undertaken by *R. leukops* ($F_{[2,28]}$=3.902; P=0.032; Fig. 2). Significantly fewer short dives (<15 min) were recorded for the species at 150 cm water depth (13.0±4.9%) than at 50 cm (25.8±5.7%) and 100 cm (24.4±7.2%; P<0.05; Fig. 2). Excluding short dives (<15 min), modal dive duration for *R. leukops* ranged from 30 min to 75 min for the three depth treatments, while dives >150 min accounted for up to 41.9±7.0% (150 cm) of logged immersions (Fig. 2).

Ascent time increased significantly with depth ($F_{[2,28]}$=37.752; P<0.001; Table 2), with turtles at 150 cm taking seven times longer to reach the surface than *R. leukops* at 50 cm depth (P<0.001). Water depth also influenced the surfacing interval, with the percentage of emersions <60 s decreasing significantly ($F_{[2,28]}$=9.180; P<0.001; Fig. 3) for *R. leukops* at 150 cm depth (22.6±5.4%; P<0.01) compared to turtles at 50 cm (42.4±7.1%) and 100 cm (36.2±5.1%). Additionally, *R. leukops* proportionately logged three times the number of long surfacing episodes (>600 s) at 150 cm depth (11.8±3.1%) than at 50 cm (3.7±1.4%) and 100 cm depth (3.2±1.1%; Fig. 3). Such changes in surfacing behaviour for *R. leukops* among water depths resulted in a significantly longer median surfacing time at 150 cm compared to the other two treatments ($F_{[2,28]}$=13.623; P<0.001; Table 2). At all three depths, surfacing time represented less than 4.0% of the total trial duration (range 2.7±0% to 3.6±0.9%). No correlation was observed between dive times and subsequent surfacing intervals for the three water depth treatments (range: r=–0.0854–0.0374; Fig. 4), with increased surfacing times recorded for *R. leukops* at 150 cm depth being evenly distributed across the observed diving intervals when compared to surfacing times at 50 cm

Table 2. *Summary of diving and surfacing behaviour for* Rheodytes leukops *at three water depth treatments*

Water depth (cm)	Median dive duration (min)	Maximum dive duration (h)	Surfacings h^{-1}	Ascent time (s)	Median surfacing duration (s)
50	106.9±18.4	9.68±1.20	0.52±0.08	13.0±1.1	91.8±12.6
100	95.5±23.0	9.24±1.49	0.51±0.07	58.0±5.1	102.0±12.7
150	133.2±20.4	7.82±0.80	0.43±0.06	98.4±13.1	176.8±27.5

Values are means ± S.E.M. (*N*=16).

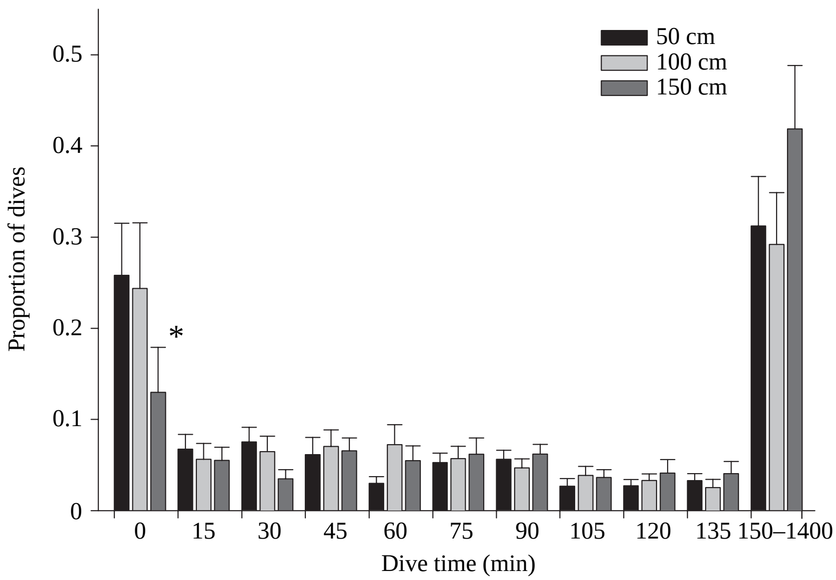

Fig. 2. Comparison of the distribution of dive times undertaken by *Rheodytes leukops* at three water depths. Histograms are divided into 15 min intervals. Values are means ± S.E.M. (*N*=16); asterisk indicates a significant difference between water depths within a surfacing interval ($P<0.05$).

and 100 cm water depth (Fig. 4). Finally, vertical displacement day^{-1} increased significantly ($F_{[2,28]}$=16.546; $P<0.001$) from *R. leukops* at 50 cm (9.1±1.8 m day^{-1}) compared to turtles at 100 cm (31.2±5.8 m day^{-1}) and 150 cm (35.5±5.5 m day^{-1}), with no difference being recorded between the later two depths (P=0.665).

Water velocity

Qualitative differences were observed in the behaviour of *R. leukops* among the three water velocity treatments. At 5 cm s^{-1}, *R. leukops* appeared more active and moved more frequently between the front and the back of the study area. With increasing water velocity, movement was reduced, with *R. leukops* spending significantly less time in front of the water deflector at 30 cm s^{-1} (2.1±2.1%; $F_{[2,30]}$=4.562; $P<0.05$; Fig. 5A) than at 15 cm s^{-1} (25.4±11.2%) and 5 cm s^{-1} (35.1±11.4%). Additionally, *R. leukops* surfaced significantly fewer times per trial as water velocity increased ($F_{[2,30]}$=19.926; $P<0.001$; Fig. 5B), with surfacing frequency decreasing from 8.3±1.6 surfacings $trial^{-1}$ at 5 cm s^{-1} (range 1–15 surfacings $trial^{-1}$) to 0.4±0.3 surfacings $trial^{-1}$ at 30 cm s^{-1} (range 0–3 surfacings $trial^{-1}$; $P<0.001$). Ten of the twelve *R. leukops* tested at 30 cm s^{-1} did not surface during their respective 8 h trial.

Discussion

Bimodal respiration offers organisms the flexibility to alter their respiratory partitioning to changing environmental conditions for the purpose of optimising O_2 uptake relative to the cost associated with respiration (Boutilier, 1990). With increasing distance to the surface, the theory of optimal breathing (Kramer, 1988) predicts that bimodally respiring vertebrates will shift their reliance towards aquatic routes of O_2 extraction in order to reduce the travel cost (i.e. time and energy) associated with pulmonary gas exchange. Previous investigations on bimodally respiring fish and amphibians observed diminished air-breathing frequencies and increased aquatic ventilation rates with increasing water depth (Feder and Moran, 1985; Bevan and Kramer, 1986, 1987; Shannon and Kramer, 1988), indicating an elevated reliance upon aquatic O_2 uptake (Graham et al., 1977; Burggren, 1979; Johnston et al., 1983). Similar findings were observed for the soft-shelled turtle, *Trionyx sinensis*, where a marked decrease in surfacing frequency and a concomitant increase in the ventilation of the aquatic respiratory organ (i.e. the buccal-pharynx) resulted when water depth increased from 15 cm to 45 cm (Hua and Wang, 1993). However, no difference was observed in the surfacing frequency of *R. leukops* as water depth increased, suggesting little to no change in the respiratory partitioning of the species.

The relationship between water depth and surfacing frequency in bimodally respiring vertebrates is dependent upon the assumption that increased travel costs associated with pulmonary respiration are biologically significant. Results from this investigation, however, suggest that the costs attributed to pulmonary gas exchange in *R. leukops* were probably minimal irrespective of water depth due to the infrequent surfacing bouts recorded for the species. At the deepest depth, *R. leukops* traveled only 35 m in the vertical

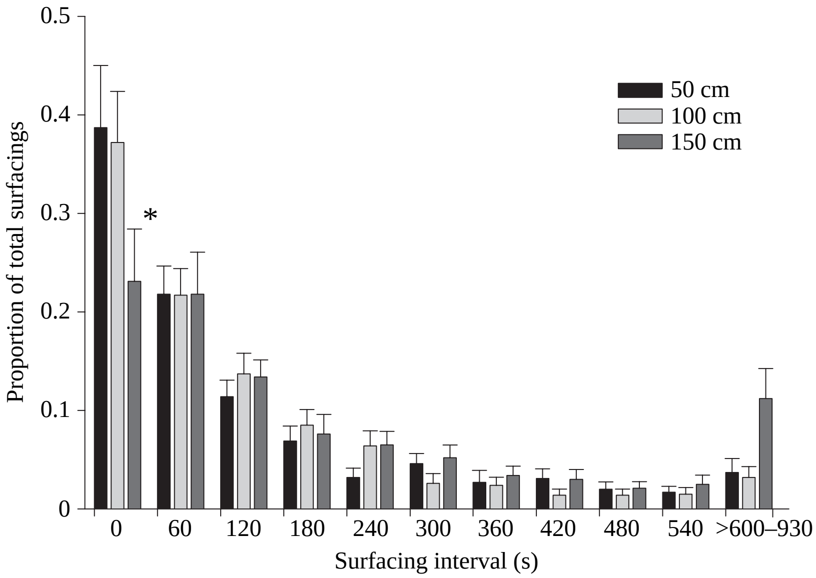

Fig. 3. Distributions of surfacing time for *Rheodytes leukops* during water depth trials. Histograms are divided into 60 s intervals. Values are means ± S.E.M. (N=16); asterisk indicates a significant difference among water depths within a dive duration interval (P<0.05).

direction per day, thus raising doubts about the metabolic expenditure associated with transit for atmospheric respiration. Additional support comes from the temporal cost of pulmonary respiration, where surfacing accounted for less than 4% of the total time budget of *R. leukops* at all three water depths.

A further consideration as to why water depth failed to influence the surfacing frequency of *R. leukops* is the biomechanical cost of aquatic *versus* pulmonary respiration. The energetic expenditure of respiring water is considerably greater than for pulmonary ventilation, given the high density and viscosity of water *versus* air and the relatively low concentration and diffusional coefficient of oxygen in water (Boutilier, 1990; Dejours, 1994; Schmidt-Nielsen, 1997). Gill ventilation in fish accounts for more than 10% of the animal's metabolic cost (Jones and Schwarzfeld, 1974; Holeton, 1980), while the oxidative expenditure of pulmonary respiration in the freshwater turtle, *Chrysemys picta bellii*, amounts to only 1% of the total energy budget (Jackson et al., 1991). Moreover, compared to gill ventilation or cutaneous gas exchange, ventilation of the bursae sacs in *R. leukops* is presumably more expensive due to the continual change in the kinetic energy of water associated with tidal ventilation (Schmidt-Nielsen, 1997). Therefore, the metabolic expenditure associated with increasing the aquatic O_2 extraction rate of *R. leukops* may outweigh the cost saved by reducing the turtles' air-breathing frequency with increasing water depth. However, in spite of a presumably high cost associated with cloacal ventilation in *R. leukops*, the routinely long dives recorded for the species suggest a high reliance upon aquatic gas exchange regardless of water depth.

Although water depth did not affect the diving performance of *R. leukops* (i.e. median and maximum dive time), water depth did influence the distribution of dives undertaken by the species. *R. leukops* generally displayed two dive types, short submergences (<15 min) that were characterised by active 'exploratory' behaviour and longer resting dives with a modal duration of 30–75 min. As water depth increased, *R. leukops* logged significantly fewer short dives, suggesting a behaviour change to less active dives. However, given that no qualitative difference was observed in the activity level of *R. leukops* among depth treatments, an alternative consideration is that turtles in shallower water were more likely to inadvertently breach the surface during active periods due to their relatively close proximity to the surface. Irrespective of the reason, overall surfacing frequency for *R. leukops* was unaffected across the treatment depths.

The theory of optimal breathing also predicts that with increasing water depth, bout times at the surface will increase due to an increased loading of oxygen into the lungs (Kramer and McClure, 1981; Kramer, 1988). Although longer surfacing times were observed for *R. leukops* with increasing water depth, increased loading of oxygen into the lungs would presumably translate into longer dives. Given that no difference was observed in the diving performance of *R. leukops* among depth treatments, longer emersions are instead attributed to an increased depletion of oxygen stores at the end of the dive during the turtle's ascent to the surface. Previous TDR diving investigations on marine chelonians have also suggested that extended surfacing bouts may be indicative of lactate oxidation resulting from anaerobic glycolysis (van Dam and Diez, 1996; Southwood et al., 1999; Hays et al., 2000);

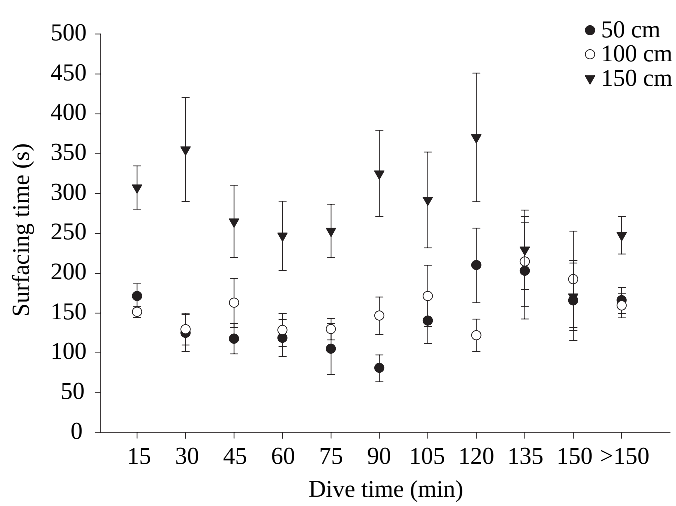

Fig. 4. Comparison of mean surfacing time per dive duration interval for *Rheodytes leukops* at the three water depth treatments. Values are means ± S.E.M. (N=16).

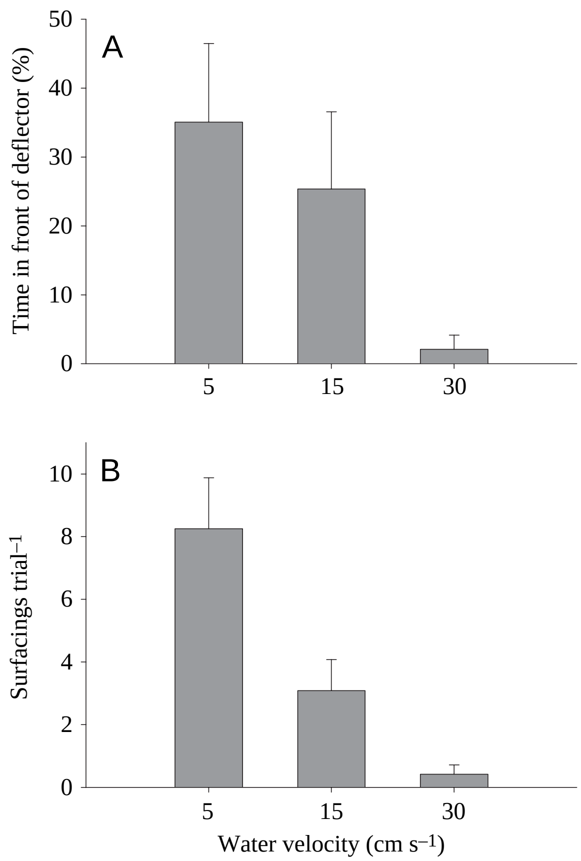

Fig. 5. The effect of water velocity on (A) the percent of bottom time where *Rheodytes leukops* resided in front of the deflector, and (B) the mean number of surfacings per trial for *R. leukops*. Values are means ± S.E.M. (*N*=12).

however, no correlation was observed between dive times and subsequent surfacing durations at either of the three water depths, thus suggesting that turtles remained aerobic when submerged.

Unlike water depth, increasing water velocity significantly impeded the surfacing frequency of *R. leukops*, thus supporting the argument that travel to and from the surface represents a significant cost for *R. leukops* residing within fast-flowing riffle zones. An elevated reliance upon aquatic respiration, which presumably translates into a decreased surfacing frequency, may be metabolically more efficient for *R. leukops* compared to the expenditure (i.e. time and energy) associated with air-breathing at higher water velocities. Additionally, surfacing within riffle zones may pose a threat to *R. leukops*' safety, with risk including injury from suspended debris and current displacement. Support for these assumptions comes from field-based observations of diving behaviour for *R. leukops* following pulses of high water flow. During a flood event in Marlborough Creek, a tributary of the Fitzroy River, consecutive dives of 3.8, 2.2 and 2.8 days were recorded for *R. leukops*, with subsequently short surfacing intervals (<5 min) suggesting that the turtle remained aerobic (M. A. Gordos, unpublished observations).

Unfortunately, this study does not provide a direct assessment of respiratory partitioning between varying water velocity treatments. Given that surfacing frequency is only a rough estimate of a turtle's dependence upon pulmonary respiration, decreased air-breathing frequencies recorded at higher water velocities cannot conclusively be attributed to an increased reliance upon aquatic respiration. Alternative strategies including an increased depletion of lung oxygen stores and a depressed metabolic rate while diving must be considered. However, such alternative strategies alone cannot account for the observed 20-fold decrease in surfacing frequency observed for *R. leukops* at higher water velocities. In addition, although turtles possess a high potential for anaerobic metabolism (Burggren and Shelton, 1979; Ultsch and Jackson, 1982), blood chemistry analysis indicates that *R. leukops* avoids the development of a metabolic and respiratory acidosis during voluntary dives of up to 12 h (Gordos et al., 2004).

Compared to the slow flowing pools, areas of fast-flowing water exert a considerable physical force against animals attempting to maintain their position within riffle zones (Facey and Grossman, 1992). For stream-dwelling fish, the metabolic cost associated with maintaining a fixed position increased with water velocity, regardless of whether the fish was a column dwelling or benthic species (Brett, 1964; Webb, 1971; Facey and Grossman, 1990). Assuming a similar relationship for freshwater turtles, results from this study suggest that *R. leukops* preferentially selected low-velocity microhabitats to minimise the metabolic cost required to hold a position with increasing water flow. Support for this assertion comes from investigations on stream-dwelling salmonids, where the distribution and abundance of fish in fast-flowing reaches is determined by the presence of low-velocity cover (Fausch, 1984; Cunjak and Power, 1987; Moore and Gregory, 1988; Facey and Grossman, 1992; Meyer and Griffith, 1997; Vehanen et al., 2000). Moreover, McLaughlin and Noakes (1998) calculated that utilisation of current–velocity refuges by brook trout (*Salvelinus fontinalis*) decreased the fishes metabolic expenditure by 10%.

The results of the present study suggest that aquatic respiration allows *R. leukops* to inhabit and exploit fast-flowing riffle zones, a niche from which primarily air-breathing turtles are generally excluded (Ernst and Barbour, 1982; Cann, 1998). Benefits associated with riffle zones include an abundant food supply of aquatic macroinvertebrates, which are foraged upon almost exclusively by *R. leukops* captured within fast-flowing waterways (Legler and Cann, 1980; Cann, 1998), and possible reduced predation risk (e.g. by *Crocodylus porosus*). Turtle species attempting to exploit such an environment, however, have to contend with problems associated with high water velocity, including current displacement during surfacing and maintenance of position on the river bed. Aquatic respiration allows *R. leukops* to exploit the high levels of dissolved oxygen characteristic of riffle zones, which translates into a reduced surfacing frequency and increased time available for foraging, mating and resting. Furthermore, negative buoyancy presumably reduces the metabolic effort required for *R. leukops* to remain on the river bed, while simultaneously limiting the problems associated with current displacement experienced by positively buoyant species when surfacing. Finally, *R. leukops* at higher water velocities select sheltered habitats behind or under obstructions (e.g. rocks and organic debris; Legler and Cann, 1980; Cann, 1998), presumably to avoid the increased metabolic cost associated with maintaining their position on the river bed when exposed to the direct flow of water.

The findings of the present study further demonstrate the interaction between changing environmental conditions and the respiratory physiology of bimodally respiring vertebrates. For aquatically respiring turtles, water temperature and aquatic P_{O_2} directly affect the animal's respiratory partitioning strategy due to fluctuations in the demand and efficiency of O_2 uptake (Gatten, 1980; Herbert and Jackson, 1985; Ultsch, 1985; Stone et al., 1992; King and Heatwole, 1994; Priest and Franklin, 2002). In addition, ecological processes such as predation risk and food availability are also known to influence the surfacing patterns of air-breathing fish (Kramer and Braun, 1983; Kramer et al., 1983). Here we demonstrate that water velocity significantly alters the surfacing behaviour and habitat preference of *R. leukops*, suggesting an increased reliance upon aquatic O_2 uptake due to elevated costs associated with pulmonary gas exchange.

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