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Injuries to Barramundi *Lates calcarifer* Resulting from **Lip-Gripping Devices in the Laboratory**

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Abstract.—The barramundi Lates calcarifer (also known as barramundi perch) is a renowned recreational sport fish that is the target of considerable catch-and-release fishing effort. Mechanical lip-gripping devices (lip grippers) are often used to handle barramundi because they allow the angler to easily and safely lift their catch, remove the hooks, and return it to the water while minimizing contact between the fish and other surfaces. The effects of these devices on the fish are largely unknown and to date have only been specifically quantified for bonefish Albula spp., which suffered serious injuries as a result of being handled with lip grippers. To investigate the effects of these grips on barramundi of a range of sizes, we held 21 fish (53-102 cm total length) above the water using lip grippers-10 without any additional support, and 11 with a hand holding them in a horizontal position-and compared survival and sublethal effects with those of 10 fish that had been held in landing nets for the same period of time (20 s). Two fish (7%) died 10 d posttreatment, but death could not be attributed directly to any handling treatment. All fish held with lip grippers and receiving no additional support had small holes in the membranes of the lower jaw, compared with 81% of fish that were held with lip grippers and supported by a hand. X-rays showed that lifting fish with lip grippers altered the alignment of the vertebrae, which did not return to normal after 3 weeks.

Barramundi Lates calcarifer (also known as barramundi perch) is an iconic sport fish of northern Australia. It grows to around 150 cm and 55 kg (Grant 1982), and is valued for its fighting and eating qualities. An estimated 761,763 barramundi were caught by recreational fishers in Australia in 12 months (2000 to 2001), of which 545,816 (72%) were returned to the water (Henry and Lyle 2003). Many fish are released because of maximum and minimum size restrictions and bag limits (Souter et al. 2008), while others are released in the hope of maintaining stocks for future fishing experiences (Arlinghaus et al. 2007; Cooke and Schramm 2007). To achieve this goal, fish should be handled in a manner that minimizes injuries and increases their chances of survival.

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After capture, angled fish are handled to remove the hooks and, on occasion, to be measured and photographed. Handling may be done with landing nets, gaffs, soft cradles, gloves, or mechanical lip-gripping devices (lip grippers; Danylchuk et al. 2008). Landing nets are effective handling tools that are commonly used for barramundi, although fin damage and scale loss can occur, even in knotless landing nets (Barthel et al. 2003; de Lestang et al. 2009). Gaffs are generally unsuitable for catch and release because they can cause fatal injuries to fish, and soft cradles are large and cumbersome to use on small boats (Danylchuk et al. 2008). Barramundi have spines on the opercula and fins that often cut or stab anglers who handle them with bare hands, and the presence of saltwater crocodiles Crocodylus porosus in many waterways makes it dangerous to place hands in the water near struggling fish (de Lestang et al. 2009).

Lip grippers are intended to overcome the shortcomings of other fish handling tools. These devices use the weight of the fish to force two opposing metal plates together, on either side of the lower jaw. This allows anglers to lift and handle their catch with one hand, leaving the other to hold a rod, remove the hook, or take a photograph. They also limit contact between the fish and other surfaces, reducing scale and slime loss (Danylchuk et al. 2008).

Given that large numbers of barramundi are released after being caught on line and that lip grippers are commonly used to handle barramundi, some knowledge of the impacts of these devices on the fish is required for management of this species. Danylchuk et al. (2008) found that lip grippers caused considerable injuries to bonefish Albula spp., namely large tears to jaw membranes and even broken jaws, but none of these injuries appeared to kill captive fish within 48 h. To date, no formal assessment has been conducted on how lip grippers affect the longer-term survival of barramundi, or other fish species. Similarly, very little is known about what internal injuries could result from the use of these devices, although Danylchuk et al. (2008) suggested that hanging fish by the jaw may increase the separation between vertebrae. Our objectives were to assess the extent of injuries to barramundi caused by mechanical lip-gripping devices, especially

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TABLE 1.—Water quality parameters for barramundi used in experimental treatments. Salinity was intentionally varied to help manage parasites (see text for details).

Variable	Mean	Minimum	Maximum
Temperature (°C)	27.5	24.0	32.0
Dissolved oxygen (mg/L)	5.8	4.2	6.8
pH	7.8	5.9	8.2
Salinity (‰)	30.7	0.0	37

to the lower jaw and the vertebrae, and to determine whether the resulting injuries are likely to lead to direct or indirect mortality.

Methods

Experimental subjects.—Thirty-five barramundi were caught between 20 and 28 March 2008, from Darwin Harbor and nearby Shoal Bay, using 15.2-, 17.8-, and 20.3-cm (stretch mesh) gill nets. The subjects were collected using gillnets to minimize jaw injuries that could confound effects of experimental treatments. Barramundi were quickly and gently cut from the net and placed into highly flushed (1 L/s) and well-oxygenated tubs for transport to the Darwin Aquaculture Centre (DAC).

At DAC, the total length (TL) of each barramundi was measured (range = 53–102 cm) and a Hallprint dart tag was inserted in the area flanking the first dorsal fin ray (Davis and Reid 1982). Fish were then transferred by hand to one of four circular fiberglass tanks. Each 5,000-L tank had a diameter of 3.8 m, a water depth of 70 cm, and contained no more than 10 fish of similar size (to prevent cannibalism). Tanks were continuously flushed (2 L/s) with seawater that was nominally filtered to 5 μm , and water parameters are summarized in Table 1.

Fish were fed striped mullet *Mugil cephalus* three times a week, and tanks were cleaned every day. After 1 week, the tank water was gradually turned from salt to fresh overnight, then returned to seawater 2 weeks later. The aim was to reduce parasite loads on these essentially catadromous fish. Fish took 1 to 2 weeks to commence feeding. Four fish (60–97-cm TL) died before the experiment commenced.

Experimental design.—Each of the 31 fish used was preassigned to a treatment using a stratified random process that ensured each treatment had a similar size distribution (Table 2). Fish were subjected to treatments on 26 May (six fish), 28 May (19 fish), or 29 May 2008 (6 fish), and each treatment was imposed on at least two fish each day.

In preparation for treatment, the water level of the holding tank was lowered to between 20 and 30 cm. This facilitated capture and prevented the barramundi

TABLE 2.—Numbers and sizes of barramundi used in the three treatments described in this paper.

Size-class (TL [cm])	Lip grippers only	Lip grippers + support	Landing net
50-55	1	1	1
56-65	5	7	6
66-75	1	0	0
76-85	1	1	1
86-95	1	1	1
96-105	1	1	1
	10	11	10

from jumping out. Individual fish were then slowly corralled into a knotless, flat-bottomed landing net, similarly to de Lestang et al. (2009). Captured fish were then placed individually into another tank, where the water level was kept at 30 cm. Each fish was then exercised by quickly walking behind it and tapping its tail with a foot until the fish appeared lethargic or rolled onto its side. We exhausted fish in this manner to simulate the effect of being line caught without inflicting any jaw damage (Danylchuk et al. 2008).

The exercised fish was quickly recaptured using a knotless, flat-bottomed net, lifted from the water, and subjected to one of three treatments: (1) placing lip grippers on the lower jaw, lifting the fish out of the water, and holding it vertically for 20 s (lip-grippersonly treatment); (2) placing lip grippers on lower jaw and, while supporting the midsection with a hand holding the fish horizontally, lifting and holding the subject out of the water for 20 s (supported treatment); or (3) lifting fish out of the water for 20 s using a knotless, flat-bottomed net (net treatment), which has been shown to cause minimal external injuries to barramundi and does not directly kill them (de Lestang et al. 2009).

Immediately after a treatment was imposed, fish were placed on a wet towel on a flat surface, photographed, and quickly examined for obvious injuries. Fish were then placed into a deeper tank (70 cm) of seawater and monitored for mortality or obvious signs of stress until 3 July 2008 (35–38 d posttreatment).

Jaws.—Three days after treatment, two randomly selected fish from each treatment were anesthetized by placing them in a 100-L tub containing a 4 μ L/L solution of Aqui-S (Aqui-S, New Zealand) in seawater. After the jaws were relaxed, fish were removed and placed on a solid surface that was covered with a wet towel. Jaws were inspected, visually and tactically, for any abnormal movements or lesions. Fish were then revived by moving them through water without anesthetic, in a 5,000-L tank. All fish were anesthetized and examined similarly on 19 June 2008, 21–24 d after treatments were imposed. The same observations were

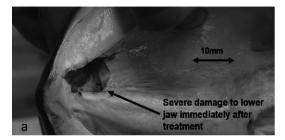




FIGURE 1.—Damage to the lower jaw membrane of a barramundi that had been held by lip grippers (a) immediately afterwards and (b) 22 d later, when this injury had healed.

made at this time. Lower jaws were photographed, and an independent observer used these photographs to give each fish a score for inflammation (0 = no inflammation, 3 = worst cases that we observed: obvious scar tissue, an area of redness >5 cm², or both). Figure 1b shows the lower jaw damage of a barramundi that was given an inflammation score of 3. Inflammation scores were compared between treatments with the commonly used Pearson's χ^2 test (Conover 1999), and the presence or absence of holes was compared between treatments using binomial generalized models.

Internal injuries.—The two largest and two smallest fish from the lip-grippers-only and supported treatments were X-rayed when the treatments were imposed (i.e., when the fish were lifted) and then again when fish were lying on a flat surface immediately afterwards. The largest and smallest fish from the net treatment were only X-rayed while lying on a flat surface immediately after being held in the net. Fish lying on a flat surface were the same distance from the machine as fish that were being held in fish grips, and the same exposure was used. Film was developed immediately after each fish in a makeshift darkroom. To investigate long-term impacts such as arthritic degeneration, all fish were X-rayed (same exposure) while laying down on a flat surface on 19 June 2008 (21–24 d posttreatment).

It was apparent from the X-ray images that the natural curvature of the first four vertebrae (Figure 2)

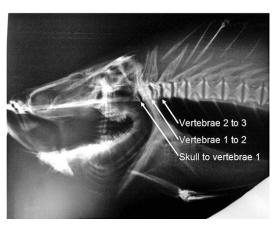


FIGURE 2.—X-ray of a barramundi showing the natural curvature of the first four vertebrae and the locations where intervertebral distances were measured

was lost, and this area of the spine became essentially straight when barramundi were held using lip grippers. To quantify any changes in this curvature, intervertebral distances were measured at the dorsal edge of the joint, in the center of the joint, and on the ventral edge for the first four vertebrae, and between the skull and the first vertebrae whenever possible.

Data analysis.—Total length of each fish, the time taken for fish to become exhausted (roll onto their side or become obviously lethargic), and total air exposure time (all as continuous variables) were compared between treatments using separate analyses of variance (ANOVAs), normality of residuals and constancy of variance checked graphically. Fish length was \log_e transformed before analysis.

Fish survival was compared across treatments (fish length and treatment-length interactions serving as covariates) using parametric survival analyses. The timing of the fish deaths suggested that a constant-hazard model was appropriate, which assumes that the instantaneous risk of a fish dying was constant throughout the time fish were monitored. This hazard model is a "remarkably robust assumption in many applications" (Crawley 2002).

The presence of holes in the lower jaw immediately after treatments were imposed were compared between treatments using binomial generalized linear models. The first model compared the proportion of fish with holes across all three treatments, fish length serving as a covariate (also investigating the treatment–length interaction). To compare the effects of supporting fish while using lip grippers, the same model was run again excluding landing net treatment. Inflammation scores (21–24 d after treatment) were compared between

treatments with the commonly used Pearson's χ^2 test (Conover 1999).

Intervertebral distances were initially investigated graphically to quantify any stretching of the spine or dislocation of vertebrae. Of the distances that could be measured accurately and consistently from the X-rays, the distance between the second and third vertebrae on the ventral side was clearly the most affected by treatments and were analyzed further. These distances were divided by fish TL and investigated further in a repeated-measures (mixed-effects) linear model. Fixed effects were treatment, time, and treatment-time interaction, and fish tag number was the random variable (Crawley 2002). To further investigate how well fish recovered from the treatments, an additional ANOVA was used to investigate this intervertebral distance from X-rays taken 21-24 d posttreatment divided by TL.

Preliminary analyses.—Fish TL did not vary significantly between treatments ($F_{2,\ 28}=0.03$; P=0.97). Fish took an average of 115 s (SE = 7) to lose equilibrium or become clearly lethargic when being chased in a shallow tank, and this did not vary significantly between treatments ($F_{2,\ 28}=0.7$; P=0.5). Total air exposure time averaged 95 s (SE = 4), and again this did not vary significantly between treatments ($F_{2,\ 27}=1.5$, P=0.3; note: one fish was not timed accurately and was therefore excluded from analysis).

Results and Discussion

Survival

We found no evidence that lifting fish by the lower jaw killed any fish directly. Only two barramundi died, both 10 d after treatments were imposed: one fish was from the lip-grippers-only treatment, and one was from the support treatment. Both fish that died had also been X-rayed during and immediately after treatments were imposed. Treatments did not significantly affect survival over the 21–24 d barramundi were held in captivity posthandling ($\chi^2 = 1.6$; df = 2; P = 0.4). The fact that both fish died 10 d into a 35–38-d trial suggests that our assumption of a constant risk of death used in the survival analyses is reasonable.

Fish deaths were most likely caused by parasitic infections. The two fish that died were in the same tank, did not feed when prompted, and had red eyes prior their deaths, symptoms consistent with an infection by the protozoon *Cryptocaryon irritans* (Schipp et al. 2007). Other fish in the same tank also displayed similar symptoms. Sudden and large outbreaks of *C. irritans* often kill captive fish, especially when stressed (Colorni and Burgess 1997). Tank water was at cool temperatures (24–25°C) for 14 consecutive days prior to the infestation becoming apparent, and

such temperatures can stress captive barramundi (Katersky and Carter 2007). This suggests that water temperature may have contributed to fish stress, although the effects of treatment may also be apparent. The cypocarium infection was treated by changing tank water from salt to fresh over several days, and all other fish in this tank recovered.

We found that larger fish were less likely to survive in our experiment overall ($\chi^2=8$; df = 1; P=0.004), and this is also reflected in fish survival before we imposed any experimental treatments. Before treatments were imposed, three large fish (90, 96, and 97 cm), and only one small fish (60 cm) died. Interestingly, survival of radio-tracked fish that had been caught and released showed that fish size had little effect on survival (de Lestang et al. 2004).

Although we found that fish survival decreased with an increase in overall size, our experimental treatments affected all fish equally (i.e., the length-treatment interaction clearly did not affect survival; $\chi^2 < 0.001$, df = 2, P = 0.99). This appears to conflict with anecdotal evidence suggesting that large barramundi were more susceptible to mortality after being lifted by the lower jaw than were small barramundi. In a small, unpublished trial, three large (roughly 95-cm TL) and three smaller barramundi were lifted by the lower jaw while anesthetized. The three large barramundi died 7-10 d after the trial, while the three smaller fish survived. A further 10 large anesthetized barramundi were then weighed in a wet sling, and all survived. This trial was conducted after previously noticing that all broodstock barramundi lifted by the lower jaw without support died (A. Hogan, Department of Primary Industries and Fisheries, personal communication). It should be noted that we only had three fish longer than 95 cm in our experiment; thus, we may have found a stronger treatment-length interaction if a greater number of very large fish were used. Alternatively, the anesthetic may affect fishes' response to being suspended by the lower jaw.

Jaws

All barramundi that were lifted with lip grippers alone had holes in the lower jaw, whereas 81% of fish that were lifted and supported had holes, and none of the fish handled in nets had such holes (Table 3). There was a significant difference between all three treatments in this regard ($\chi^2=31$; df = 2; P<0.0001). However, there was no significant difference between treatment when fish from the landing net treatment were excluded from the analysis ($\chi^2=2.8$; df = 1; P=0.1). The presence of holes did not vary significantly with fish length ($\chi^2=0.7$; df = 1; P=0.3), and the

TABLE 3.—Damage caused to barramundi that were held by lip grippers only, compared with that of fish that were held by lip grippers while supported horizontally and fish held in a landing net for the same period of time.

Variable	Lip grippers only	Lip grippers + support	Landing net
Number of fish with holes in jaw immediately after treatment Number of fish with some inflammation on lower jaw after 3 weeks Average score for jaw inflammation (0-3), after 3 weeks	10 (N = 10) 5 (N = 9) 1.1	9 (N = 11) 2 (N = 10) 0.5	0 (N = 10) 1 (N = 10) 0.6

length–treatment interaction was not significant (χ^2 < 0.001; df = 2; P = 0.99).

No gross jaw deformities were noticed on any fish examined under anesthetic, and all jaws appeared to move normally. All fish returned to feeding within 3 to 5 d of being subjected to a treatment, and all holes appeared to have healed within 3 weeks (Figure 1).

In a similar study, Danylchuk et al. (2008) recorded wounds to all bonefish Albula spp. that were lifted from the water by lip grippers without any support, and 80% of fish had wounds when held horizontally in the water with lip grippers. Such similarities in injury rates across two morphologically different fish species, suggests that lip grippers may affect other species similarly in some respects. On the other hand, fish in our experiment sustained minor external injuries to the lower jaw from lip grippers (e.g., holes in the lower jaws of four fish measured between 2 and 15 mm across immediately after being held in lip grippers, which is comparable to hook injuries). Nearly all barramundi with similar holes in the lower jaw membranes survived for at least 1 week (de Lestang et al. 2004), suggesting that such injuries are unlikely to be directly fatal. In comparison, bonefish handled with lip grippers had large holes, tears, and even broken mandibles (Danylchuk et al. 2008), which were more likely to result in postrelease stress and perhaps mortality. Barramundi lower jaws consist of robust bones and thin membranes that contain no obvious muscle tissue, whereas bonefish jaws appear to have relatively delicate bones and more muscle. It may therefore be possible to predict the effects that lip grippers would have on jaws of a particular fish species by visually assessing jaw structure, but studies on additional species are required to confirm this theory.

Inflammation scores did not show any strong relationship to treatment ($\chi^2 = 5.0$; df = 6; P = 0.5) 3 weeks after treatments were imposed, and likewise, the presence or absence of obvious inflammation was not strongly correlated to treatment ($\chi^2 = 5.3$; df = 2; P = 0.07). Two fish from the landing net treatment had some minor inflammation on the lower jaw, suggesting that other factors may confound treatment effects.

Internal Injuries

The most striking parameter measured from the X-ray images was the separation between the second and third vertebrae, especially on the ventral side, which would be consistent with either the spine being elongated or the head being tilted back (dorsally), or both (Figure 2). This intervertebral distance varied significantly between treatments overall ($F_{2,27} = 11$; P = 0.0004), fish held in lip grippers alone having vertebrae stretched further than other fish (Figure 3). Barramundi vertebrae did not fully recover from this stretching (compared with net treatment; $F_{2,14} = 1.5$, P = 0.3 for the treatment–time interaction in the repeated-measures model).

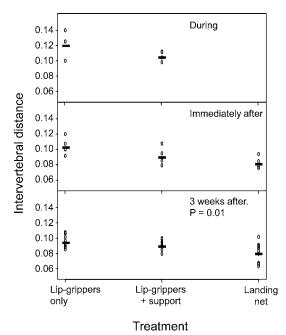


FIGURE 3.—Distances (mm) between the second and third vertebrae of barramundi on the ventral side divided by fish TL (cm) while fish were being lifted (top panel), immediately after lifting (middle panel), and 21–24 d after being subjected to one of three treatments: held with lip grippers, held with lip grippers and supported in a horizontal position, and held in a knotless landing net (bottom panel). Circles represent raw data, horizontal lines represent means.

Even after 21–24 d, vertebral alignment still differed between fish from the three treatments (ANOVA: $F_{2/25}$ = 5.1, P = 0.01; Figure 3), fish from the lip-grippersonly treatment having more disrupted spinal alignment than fish that were held with lip grippers while supported, which, in turn, appeared more disrupted than fish that were held in nets only (Figure 3). We can conclude that lifting barramundi by the lower jaw causes dislocation or separation of vertebrae, from which fish did not fully recover after 3 weeks. The severity of the intervertebral separation when fish were handled using lip grippers (both treatments) was consistent with sprain injury to the intervertebral joint capsules and associated ligaments (J. Humphrey, Department of Regional Development, Primary Industry, Fisheries, and Resources, personal communication). X-rays taken three weeks after fish were handled showed no evidence of arthritic degeneration in any articulations.

Implications for Barramundi

The holes in the lower jaw membranes caused by using lip grippers, although small, may impede the ability of barramundi to capture prey. Barramundi feed by rapidly expanding the buccal cavity, which effectively draws the immediate water and prey into the large, open mouth (Reynolds 1978; Davis 1985). Any hole in the thin membranes of the lower jaw may decrease the effectiveness of the sucking action, which could reduce predatory success, although this would be difficult to quantify directly.

Lifting barramundi by the jaw can cause some longer-term internal injury, but we cannot conclude what impacts these injuries would have on the survival of released fish. Fish with severe spinal injuries resulting from electrofishing tended to have impaired growth, but such injuries could rarely be directly attributed to fish death (Dalbey et al. 1996; see review by Snyder 2003). Compared with many of these fish, the spinal injuries of the barramundi in our study were minor, which may suggest these injuries may have little impact on the survival of released barramundi. Spinal injury to fish held vertically by the lower jaw is certainly caused largely by gravity; therefore, such injuries are unlikely to occur if the fish is handled with lip grippers while still in the water, which would be the recommended method for using such devices in areas without crocodiles. Reducing exposure to air results in increased postrelease survival of many fish species (Bartholomew and Bohnsack 2005), another reason to leave fish in the water whenever it is safe for the angler to do so.

Not enough is known about barramundi to comment on whether internal or external injuries caused by lip grippers are likely to affect their survival or fitness. Similarly we cannot state whether injuries sustained from lip grippers are more likely to affect barramundi survival compared with injuries from lifting barramundi in landing nets (de Lestang et al. 2009). Comparisons of damage to epithelia and scales between fish handled with lip grippers and landing nets would be useful for developing science-based, best-practice guidelines for handling fish that will be released.

It should be noted that barramundi used in this trial were exercised until they lost equilibrium or stopped responding to being tapped, which took an average of 115 s, while line-caught barramundi are generally landed more quickly than this, while still active (e.g., 36-66 s for experienced anglers; de Lestang et al. 2004). Many line-caught barramundi may therefore thrash more while being held by lip grippers than the fish used in this trial did, which could increase the incidence and severity of any injuries. The need for investigations into the effects of lip grippers on fish that have been exercised to various levels of exhaustion has already been noted (Danylchuk et al. 2008). Importantly, we used one commonly used model of lip grippers in this trial, and we do not endorse this or any other particular lip gripper device.

Conclusion and Recommendations

In conclusion, we did not find any strong evidence to suggest that lifting barramundi using lip grippers kills the fish directly, either for large or small barramundi. We did find, however, that lip grippers can cause internal injuries from which the fish may never recover fully. External injuries to barramundi from the lip grippers were considered minor and healed within 3 weeks, but still had potential to disrupt normal feeding until healed. If barramundi are to be lifted from the water using lip grippers, we recommend that the fish is also supported with one hand so that it is in a horizontal position as this caused less disruption to the alignment of their spine, and put fewer holes in the jaw membranes. Wherever it is safe to do so, such devices could be used to hold fish in the water while hooks are removed.

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