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in

Bridges C.R. (ed.), García A. (ed.), Gordin H. (ed.).
Domestication of the bluefin tuna *Thunnus thynnus thynnus*

Zaragoza : CIHEAM
Cahiers Options Méditerranéennes; n. 60

2003
pages 37-39

Article available on line / Article disponible en ligne à l'adresse :

<http://om.ciheam.org/article.php?IDPDF=3600087>

To cite this article / Pour citer cet article

Brill R.W. **Handling and manipulating tunas in captivity: A physiologist's perspective.** In : Bridges C.R. (ed.), García A. (ed.), Gordin H. (ed.). *Domestication of the bluefin tuna *Thunnus thynnus thynnus**. Zaragoza : CIHEAM, 2003. p. 37-39 (Cahiers Options Méditerranéennes; n. 60)



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Handling and manipulating tunas in captivity: A physiologist's perspective

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SUMMARY – The Kewalo Research Facility (National Marine Fisheries Service, Honolulu Laboratory) has been routinely maintaining juvenile (1-3 kg) yellowfin and skipjack tunas in shoreside tanks for more than 40 years. Research conducted at this laboratory has shown that these high energy demand fishes are not particularly tolerant of acute reductions of ambient oxygen. Therefore, when maintaining tunas in pens for aquaculture, every effort should be made to ensure clean and well oxygenated conditions. If for whatever reasons this can not be done, then efforts should be directed at not increasing the fishes metabolic rates (for example by not feeding them) until water quality problems can be rectified. Also, manipulation of fish should be avoided if possible. If required, anaesthesia is probably not necessary for procedures that can be accomplished within a minute or two, as fish quickly removed from the water will generally remain completely quiescent for this period. If anaesthesia is required, benzocaine is preferable over MS222 as the former does not make the water acidic. Injectable anaesthetics are generally not recommended as they tend to leak from the injection site, and the exact dosage given is often indeterminate.

Key words: Tuna, anaesthesia, stress, aquaculture, husbandry.

RESUME – "Conduite et manipulation des thons en captivité : Le point de vue du physiologue". La station de recherches de Kewalo (Service National de Pêches Marines, Laboratoire de Honolulu) a maintenu en routine des juvéniles d'albacores et listaos (1-3 kg) de thons yellowfin et skipjack dans des bassins côtiers pendant plus de 40 années. Les recherches menées dans ce laboratoire ont montré que ces poissons à forte demande énergétique ne sont pas particulièrement tolérants à des réductions aiguës de l'oxygène ambiant. Cependant, lorsque l'on maintient les thons en stabulation pour l'aquaculture, il faudrait tout faire pour assurer des conditions de propreté et bonne oxygénation. Si pour une quelconque raison ce n'est pas possible, les efforts devraient viser à ne pas augmenter les taux métaboliques des poissons (par exemple en ne les alimentant pas) jusqu'à ce que les problèmes de qualité de l'eau soient corrigés. De même la manipulation des poissons devrait être évitée, si possible. Si nécessaire, l'anesthésie n'est probablement pas nécessaire pour les manipulations pouvant être effectuées en une ou deux minutes, car les poissons sortis subitement de l'eau resteront généralement tout à fait tranquilles pendant ce temps. Si l'anesthésie s'avère nécessaire, la benzocaïne est préférable à MS222 car la première ne rend pas l'eau acide. Les anesthésiants injectables ne sont pas recommandés en général car ils tendent à s'écouler hors du site d'injection, et la dose exacte administrée est souvent imprécise.

Mots-clés : Thon, anesthésie, stress, aquaculture, élevage.

Introduction

The Kewalo Research Facility (National Marine Fisheries Service, Honolulu Laboratory) pioneered the practices required to acquire and maintain live tunas in shore-side tanks more than 40 years ago (e.g. Nakamura, 1962). Fish are purchased from local commercial fishermen, whose boats dock literally at the laboratory's front door. The fishery employs a "live bait" technique, and tunas are returned in the vessels' 6000 l bait wells. Fish are offloaded using chamois-lined dip nets and are moved to their holding tanks in a 2700 l oval-shaped transfer tank. These procedures are intended to minimize skin and fin damage, as well as general trauma to the fish. In spite of these efforts, about 50% of tunas delivered to the laboratory die within 48 hours, the majority on the second day after arrival (Bourke *et al.*, 1987). Fish experience up to approximately a 50% decrease in blood protein concentrations immediately following arrival in captivity (Bourke *et al.*, 1987), and for many years it was thought that a generalized hemodilution was occurring. Being marine fish, however, tunas would be expected to show hemoconcentration because the inevitable skin damage and general stress resulting from capture and transport should result in a net decrease in total body water (i.e. a decrease in blood volume and an increase in plasma protein concentrations). Moreover, tunas recently arrived in captivity have body masses up to 10% below those calculated from length-weight equations, implying that a general loss of body water is indeed occurring.

Recent experiments have begun to shed light on this apparent paradox. It is now known tunas have capillaries "tighter" (i.e. less permeable to plasma proteins) than other fishes, except for individuals that are newly arrived in captivity whose capillaries are far "leakier" (Jones *et al.*, 2002). Therefore, the decreases in plasma protein levels (and probably high levels of mortality) observed in recently delivered fish are most likely due to plasma proteins moving into the intercellular space at rates higher than normal. Unfortunately, procedures to minimize or to actively counteract this problem remain to be developed.

In spite of the difficulties of performing physiological experiments on live tunas, extensive research has been conducted on the energetics and performance of cardio-respiratory system. Experiments using juvenile (1-3 kg) skipjack and yellowfin tunas have elucidated the ability of tunas' cardio-respiratory system to deliver oxygen to the tissues under various reduced ambient temperature and oxygen conditions (reviewed in Brill, 1994). Not surprisingly, tunas are not particularly tolerant of reductions in ambient oxygen. This problem is especially acute under circumstances where metabolic rates are elevated (e.g. during bouts of increased in swimming activity, immediately after exhaustive exercise, or following feeding). Tunas' ability to transfer oxygen from the water to the tissues is likewise impaired during exposure to particulate material in the water or any toxic algae. Under these conditions, the gills either produce excessive mucous or there is generalized swelling of the gill tissue, both of which impair oxygen transfer from the water to the blood. Therefore, the following is recommended. Tuna pens should be placed such that none is immediately "downstream" of other pen(s) to preclude any fish being exposed to significant reductions in ambient oxygen. Likewise, care should be taken to ensure there is never excessive flocculent material in the water nor significant algal blooms. Should any of these circumstances arise, it is strongly recommended that nothing be done that increases the fishes' metabolic rates, including feeding. Healthy tunas can withstand at least several weeks of starvation. Withholding food is clearly not desirable in pen rearing operations, but under circumstances where the fishes' cardio-respiratory function is likely to be impaired, this may at least help prevent excessive mortalities.

Handling and anaesthesia

At first glance it may appear that manipulating tunas for biopsies, implanting sonic transmitters or archival tags, attaching physiological sensors, or other procedures should involve anaesthesia. Extensive experience, both at sea and in the laboratory, has shown that tunas quickly removed from the water exhibit complete immobility and insensitivity to touch or manipulation for approximately two minutes or more (e.g. Holland *et al.*, 1990; Musyl *et al.*, 2002). During this period, carefully planned and quickly executed procedures can be accomplished. Moreover, induction of anaesthesia is fraught with problems as it is difficult to accomplish without physical trauma to the fish (e.g. skin or fin damage), and possibly irreversible blood acidosis. Recovery from anaesthesia is likewise problematic, as tunas regain their ability to struggle before they regain their ability to swim. Because they are obligate ram ventilators and negatively buoyant, tunas that cannot swim sink and rapidly suffocate. Anaesthetizing tunas is, therefore, not generally recommended unless procedures cannot be accomplished without it.

If anaesthesia is necessary, the following procedures are recommended. Because tunas must be kept in large tanks, adding anaesthetics directly to the holding tanks is not practical. At the Kewalo Research Facility, fish (1-3 kg body mass) to be anaesthetized are guided into a plastic bag containing 5-10 l of seawater with anaesthetic. The bag is then sealed and gently rocked back and forth for about two minutes to flush anaesthetic solution over the fish's gills. Following this, fish are immediately moved into the laboratory and force ventilated with a dilute anaesthetic solution. With these techniques, extensive surgery and instrumentation are routinely performed (e.g. Bushnell and Brill, 1992). During experiments, fish are sedated and kept from swimming by blocking spinal motor nerves with an injection of 0.1-0.3 ml local anaesthetic (xylocaine) directly into the neural canal immediately behind the skull. Fish are also placed in front of a pipe delivering seawater at a velocity roughly equivalent to the fish's normal swimming speeds. With this arrangement, tunas are able to set their own ventilation volumes and respond normally to changes in ambient temperature and oxygen conditions.

Of the two most commonly used fish anaesthetics, benzocaine (ethyl aminobenzoate) may be preferable to tricaine methanesulfonate (MS222, Fiquel) because it is significantly cheaper yet equally effective and safe. More important, benzocaine does not make the water acidic even at the high concentrations required for the initial rapid anaesthetization of tunas. Benzocaine does have one slight disadvantage. Because it is insoluble in water, it must be dissolved in ethyl alcohol first. For initial anaesthesia, 1 g benzocaine per litre of seawater has been found to be safe, but also rapidly to induce

immobility. A concentration 0.03 to 0.1 g/l has been found sufficient for maintenance of anaesthesia. An acceptable plane of anaesthesia is difficult to achieve in tunas, however, and overdosing (resulting cardiorespiratory collapse and death) is a constant threat. Therefore, during any extended anaesthesia, the fish's heart rate should be monitored via ECG leads placed near the heart. As soon as heart rate becomes irregular, the concentration of anaesthetic in the sea water reservoir being used to ventilate the fish's gills should be reduced. Likewise, if the fish is responsive to touch, or showing overt signs of movement, the anaesthetic concentration should be increased.

At first glance, injectable anaesthetics might seem a way to circumvent the problems associated with water soluble anaesthetics, but in actuality their use is even more difficult. Direct intravascular (IV) injection is difficult to safely achieve, especially in large fish. With intramuscular (IM) injection, the actual dose reaching the blood stream is unknown, as a variable amount of anaesthetic solution inevitably leaks from the injection site. Of the injectable anaesthetics, Saffan (a steroid anaesthetic sold by Schering-Plough) appears to be good choice (Oswald, 1978). A dose of about 0.5 ml/kg of stock solution can induce anaesthesia without hyperactivity. Nembutal (sodium pentobarbital) is not recommended. It generally has such a slow induction that multiple doses can be mistakenly given eventually resulting in overdose (i.e. lethal levels being reached). Ketamine hydrochloride appears to have some promise for use with tunas where only short periods of immobility and insensitivity are required. Both induction and recovery are rapid. Unfortunately, its use in tunas has been minimal and appropriate dosage regimes remain to be defined.

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