Review of the Scientific program and information relevant to the International Dolphin Conservation Program Act (IDCPA) held at the Southwest Fisheries Science Center, La Jolla, California, 8 to 11 March, 1999.

By

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As directed by the International Dolphin Conservation Program Act (IDCPA, 1997), the Southwest Fisheries Science Center of the National Marine Fisheries Service (NMFS) has undertaken a program of research to address the question of whether intentional deployment on or encirclement of dolphins with purse seine nets is having a significant adverse impact on any depleted dolphin stock in the eastern tropical Pacific Ocean. The mandate specifies that an Initial Finding be submitted to Congress by March 31, 1999, and that the document(s) be subjected to peer review. This document constitutes a portion of that review and represents the views of the author based on examination of prepared materials, and presentations and interviews conducted March 8-11, 1999.

The investigation by NMFS has focused on the three dolphin stocks recognized as depleted under the Marine Mammal Protection Act (MMPA): the northeastern offshore spotted dolphin (*Stenella attenuata*), the eastern spinner dolphin (*S. longirostris*), and the coastal spotted dolphin (*S. attenuata graffmani*). The evidence concerning the depleted status of these stocks relative to historical levels is sound and the efforts by NMFS to examine population trends is appropriate.

NMFS has addressed its mandate in this issue by formulating a series of questions, as described in "Decision Framework for Assessing the Status of the Eastern Tropical Pacific Dolphin Stocks" prepared by D. Goodman. The phrasing of these questions, and their application in guiding the investigation prescribed by the IDCPA, is logical and appropriate. They clarify the objectives and specify criteria that would be used to make a determination of whether the growth rates of the populations in question were within acceptable limits. These threshold criteria for acceptable risks of extinction, exceeding potential biological removal (PBR), and delayed recovery, respectively, appear to be a sound basis for evaluating the information derived from stock assessments and population models.

To address the question of whether there has been a failure to recover in any of the identified stocks, NMFS has implemented abundance surveys from dedicated vessels in 1998. The findings were reviewed at a January 21, 1999, meeting and presented in the draft report titled "Preliminary Estimates of the 1998 Abundance of Four Dolphin Stocks in the Eastern Tropical Pacific" by T. Gerrodette. A critique of the survey and analytical methodologies is beyond the background and expertise of this reviewer.

The next phase of the investigation, following the decision framework established in Goodman's report, was to model the populations to make the determination of rate of recovery (or decline). The approach used is presented in "Description of the Population Analysis" by P. Wade. Data were derived from various sources, including the abundance estimates determined by Gerrodette, fisheries mortality statistics, and the tuna vessel observer program. The finding was made that none of the stocks met the established criteria for acceptable risk relative to recovery rate. Aspects of the modeling approach, data selection and appropriateness, and robustness of the conclusions are addressed by other members of the review panel.

To explain the finding that the stocks in question are not growing at the expected rate, NMFS has considered two possible causes: environmental variability and unobserved mortality resulting from stress or injury associated with fishery activities. Examination of environmental conditions was presented in the report titled "Eastern Tropical Pacific Dolphin Habitat Variability" by P. Fielder. It was concluded that inter-annual variability during recent years has not been anomalous, and that shifts in dolphin distribution that might have biased abundance estimates probably have not occurred. The analytical methodology supporting this conclusion is addressed in more detail by other members of the review panel.

Stress Studies

My comments focus on efforts to address the other possible cause under consideration to account for the failure to grow at the expected rate. It has been hypothesized that encirclement during fishing activities is a stressor that produces insidious physiological changes that compromise dolphin health, fecundity, or fitness. NMFS was directed by the Act to review available literature on the stress response in mammals and to identify plausible mechanisms through which stress might impair the ability of the dolphin population to recover. The findings were presented in the report titled "Stress in Mammals: The Potential Influence of Fishery-Induced Stress on Dolphins in the Eastern Tropical Pacific Ocean" by B. Curry. The stated objective of the report was "... to provide a context for future scientific findings by describing what is known about physiological and behavioral responses to stress in mammals and relating that information to the chase and encirclement of dolphins in the ETP fishery."

The Abstract and Introduction indicate that four general areas of study were reviewed, and that these are outlined in Section 1 of the report. The categories are somewhat ambiguous and arbitrary. For instance, it is unclear what "biomedical laboratory research" means, and how it differs from "research on domestic animals." "Research on free-ranging mammal populations" would more appropriately be termed "research on free-ranging mammals", reflecting an emphasis on physiological consequences on the individual rather than population level effects which are inferred but not directly studied. Nevertheless, the literature reviewed is relevant to the mandated task.

To evaluate whether the review has accomplished its stated objective, I pose a series of questions which I would expect to see addressed in the

document. This is done in part because the organization of the manuscript makes it difficult to examine in sequence the events and associated information base from the scientific literature. For example, consideration of the immediate physiological effects related to the chase (Section II.B.3) appears after a discussion of the effects of isolation and restraint following capture (Section II.B.2.b). The following represent, in my view, the questions fundamental to the issue.

Could any activities associated with tuna fishing be considered as potential stressors to dolphins?

The techniques used to encircle dolphins (and associated tuna) are clearly described, and compared with similar activities in terrestrial mammals. For example, the report notes that disturbance caused by helicopter overflights has documented effects on the behavior of bighorn sheep. Pursuit prior to kill produces measurable changes in blood constituents in red deer. Other examples are scattered throughout the review, and together are sufficient to support the conclusion that chase and confinement are recognized stressors in mammals. The review also appropriately considers psychosocial issues that might compound the stress of encirclement, namely crowding, separation, novelty, and isolation. Evidence that such conditions constitute stressors in other mammals is also adequately presented.

Some consideration is given to the possibility that habituation might occur in animals repeatedly encircled in the fishery. Captive dolphins can become accustomed to performing behaviors that allow blood collection (a potentially stressful procedure), yielding samples that are considered to reflect an unstressed state (St. Aubin *et al. 1996*). However, such behaviors typically require lengthy training and consistent positive reinforcement, conditions unlikely to be associated with encirclement in the wild. It is conceivable, however, that experienced dolphins might show a diminished stress response to repeated encounters with nets, such as is presumed to occur with regularly captured bottlenose dolphins in Sarasota Bay. It is not reasonable to expect that the stress response would be eliminated.

Do similar stressors produce physiological changes known as the stress response in other mammals?

Considerable information is presented to demonstrate how stressors comparable to those identified with tuna fishing operations can influence physiological systems in mammals. These physiological and endocrinological perturbations are generically termed the stress response, but as the review acknowledges, there are pitfalls associated with attempts to describe a common response to a wide variety of stressful stimuli. It has also proven difficult to demonstrate direct correlations between either the duration or intensity of a particular stressor and the measured physiological effect. Nevertheless, some gradation of response exists, but the review takes few opportunities to develop this point, often referring generically to stressors such as restraint, isolation, and electric shock with little information on the duration and intensity of the stressor.

Such information would be useful to allow evaluation of what degree of stress is necessary to produce specific changes. For instance, it is recognized by veterinary practitioners that the duration of chase can influence the likelihood of developing capture myopathy. This point is particularly relevant to the pursuit of dolphins in the ETP.

The review should recognize apparently fundamental differences in an individual's response to a stressor that it can avoid compared with one that it cannot escape. This point might also be developed to include possible differences between physical restraint (which can be intensely stressful and physically traumatic) and confinement within a relatively broad space (a relatively mild stressor). With respect to the latter, one might expect that confinement of any form would be stressful to animals such as offshore dolphins habituated to an environment without boundary. Still, one might expect that the experience would be qualitatively different from physical constraints on body movements.

What is the nature of the stress response in other mammals?

The recognized physiological and biochemical features of the mammalian stress response are adequately presented. The report relies on a combination of primary works and important reviews. It was not the mission of this undertaking to resolve controversies within the literature regarding aspects of glucocorticoid physiology, for example. Sufficient recognition is given those points in which are apparently conflicting (e.g. lactation may be impaired under stressful conditions yet prolactin, a hormone that promotes lactation, may be elevated as part of the stress response) to demonstrate that the measurement of stress can be problematic. The report does recognize that short-term, adaptive responses are unlikely to have appreciable effects at the population level whereas chronic (sustained or repeated) stress responses could.

As one external reviewer of the first draft of the manuscript suggested, consideration of the role of catecholamines should form a larger part of the report. It is relevant to later discussion of myocardial lesions, and deserves more extensive treatment here than it was given.

What is the nature of the stress response in cetaceans?

The literature on the stress response in cetaceans is sparse compared with other mammals. There are very few studies specifically investigating this issue, and the review includes the most relevant. However, this section (I.C.4.f) might have been expanded to include information that appears later in the document in order to provide a comprehensive account of the cetacean stress response. The reader should not be expected to bring these points together.

Section I.C.4.f.i. suggests that there are several notable aspects of the adrenocortical response to stress in cetaceans but presents only two, which are in fact the only two currently recognized. This leaves the reader wondering what the others might be. Some attention is given to the modest levels of cortisol in comparison with stressed terrestrial animals, and to the participation of aldosterone (though the mention of studies on phocid seals at this point is not particularly relevant). Beyond that, all other evidence which should appear in this

section supports the conclusion that mechanisms of stress physiology are fundamentally the same in cetaceans as they are in other mammals.

It was also suggested in an earlier review that, wherever possible, information should be provided about the duration of the perturbations constituting to the stress response. Both from a comparative standpoint and to allow postulation regarding the lingering physiological effects of encirclement, it is important to consider the time course of changes to various physiological systems. Such detail is not consistently presented.

An additional source of information on the stress response of cetaceans not considered in this report is that derived from studies on animals captured for exhibit and from mass stranded whales and dolphins. The former usually exhibit transient changes in blood constituents, reinforcing the information gained from the directed studies. The latter group often typifies the extreme expression of the cetacean stress response, providing insight into the condition of distress. Under such circumstances, constituents such as cortisol may reach excessively high levels due to impaired hepatic function. It is recognized that much of this information does not appear in the primary literature, but some does and would augment the review. However, omission of this information does not detract from the general impression imparted by the studies considered in the report.

What studies have been undertaken to examine the response of cetaceans to stressors such as might be encountered during fishing-related activities?

The report considers literature relating to the effects of activities similar to or otherwise relevant to the stressors encountered by dolphins during tuna fishing operations. Specifically, chase, confinement and physical manipulations have been examined and found to elicit changes in circulating levels of a variety of blood constituents considered to be indicative of a stress response in other mammals. Still, descriptions of the relevant studies typically do not include information on the duration of the stressful event. For example, the first paragraph of Section 2.B.1 describes a suite of changes in bottlenose dolphins following capture. It is not specified, but important to know, how the animal was captured, including how long it was pursued. This is germane to relating the observations to those presented in the ensuing paragraph, which refers to prolonged chase preceding capture, and to the time frame described earlier for the chase and encirclement activities in the tuna fishery.

The literature on cetaceans contains no directed investigations of the physiological response associated specifically with the psychogenic aspects of the stress response. It is therefore difficult to address the question of whether encirclement followed by release might be stressful by itself, notwithstanding the exertion of the chase or other aspects of crowding and sociopsychological factors detailed in the report. However, the cetacean literature does contain some information relevant to this issue. Captive beluga whales showed anticipatory changes in circulating cortisol concentrations in response to lowering water levels in their holding tank, without any other superimposed stressors (St. Aubin and Geraci 1992). This observation simply confirms that even in the absence of

handling, measurable changes in circulating constituents indicative of a stress response are evident.

What evidence is there of stress and/or associated pathologies in cetaceans examined from the tuna fishery?

Previous efforts to identify morphological changes in dolphins killed during tuna fishing operations have failed to produce evidence of capture myopathy or other conditions that might contribute to delayed mortality. Yet the review still suggests that capture myopathy is likely in some proportion of the encircled dolphins. Four variations of the capture myopathy syndrome are recognized, one of which is the delayed-peracute form. Unfortunately, the condition is not sufficiently emphasized in the report as a plausible mechanism through which delayed mortality might occur. Terrestrial mammals exhibiting the delayed-peracute form of capture myopathy may not succumb until a day or more after initial capture, usually in response to a secondary stress. Damage resulting from the first insult may be subtle and not developed within the time frame represented by chase, encirclement and death in the net for those individuals examined in earlier studies. Thus, while at least one form of capture myopathy remains a plausible outcome of dolphin chase and entrapment, the evidence for its possible occurrence is not presented in sufficient detail.

What effects might be expected, including those not yet observed?

In addition to the possibility of delayed capture myopathy previously described, other potential effects on immune function, reproductive physiology, growth and metabolism are identified in the report and adequately supported with appropriate literature. Chronic dysfunction in any of these physiological processes could reasonably account for reduced fitness of the population, and the presumed failure to recover at the expected rate. The key question is whether the stress associated with encirclement is sufficient to impact any or all of these systems. There is sufficient empirical evidence provided from other species to support the conclusion that it can, and therefore deserves continued investigation.

Comments of other reviewers

In the preparation of the document submitted as part of this peer review, NMFS sought comments from a number of experts and agencies. Many of the comments received were incorporated into the current version, or adequately addressed in a document titled "Responses to comments on the draft literature review ..." Some additional observations on points raised by the earlier reviewers, and on the response by NMFS, are warranted here.

The IATTC noted that there may be quantitative differences in the intensity of stress experienced by dolphins in the ETP and those studied in the literature cited in the review. As discussed above, this point would be more effectively addressed in the review if the author were to include more information about the types and duration of stressors imposed during the experiments. The reader would then be able to better judge the comparability of the conditions in the ETP and in the experiments.

The IATTC also felt that the recognition of peculiarities of the cetacean stress response should temper statements about their reaction to encirclement. NMFS responded appropriately by noting that cetaceans exhibit the basic mammalian response to stress. Lower peak levels of circulating cortisol in stressed cetaceans do not signify a fundamental difference in the function of the hypothalamic-pituitary axis, only that cortisol concentrations are not as useful as a diagnostic indicator of stress in dolphins as it is in other mammals. However, several other constituents do show cortisol-induced changes, demonstrating that this hormone does play a role in the cetacean stress response. By making this point more clearly in the literature review as stated above, NMFS will avoid further confusion on this question.

One reviewer introduced the concept of encirclement as a sub-clinical stressor, costing resources that might be needed for a subsequent response to other stressors, such as infection, or for successful reproduction. The reviewer suggests that no clinical signs of such a condition could be detected. The concept is therefore more academic than useful for the present review, and need not be addressed further.

Conclusions

NMFS has provided sufficient scientific information to establish that tuna fishing activities are potentially stressful to dolphins and that the stress response as determined in other cetaceans could compromise fitness and productivity. Nevertheless, the review should be more cautious in how the conclusions are presented. For example, the Abstract states that as "...it seems likely that reproduction for some proportion of female dolphins will be disrupted..." and "(i)t is therefore plausible that stress ... is having a population level effect ..." Such statements should be rephrased, in the absence of data, to indicate that reproduction could be disrupted and that it is plausible that stress could have a population level effect, based on our understanding of stress and its effects on other mammals. This basic revision would not invalidate the stated objective nor the mandated task, and would relieve concerns that the review is biased towards a finding of negative effect.

The review provides an adequate framework from which to conduct further studies on the stress response in dolphins subjected to tuna fishing operations, as directed by the IDCPA. The proposed studies currently include necropsies of dolphins killed in tuna nets and an experiment involving the capture, release and recapture of dolphins to evaluate the residual effects of stress. These investigations may provide some of the information absent from the literature reviewed in Curry's report. Some comments on the status and objectives of those studies are provided below.

Necropsy Study

The Act stipulates that a 3-year program of necropsies is to be conducted to address the question of stress-associated pathology in dolphins encircled and accidentally killed in seine nets. This effort was to begin in 1998, but it was not until September 1998 that one tuna-fishing nation, Mexico, agreed to work with

NMFS on this project. A training session was held by the SWFSC in January, 1999, but as of the time of this review, no fishing vessels have been made available. Consequently, there has been no progress towards collection of the specimens required by the Act. In the time remaining before a final opinion is due, it will be difficult for NMFS to achieve the intensity of sampling without substantially increasing the sampling effort. It remains to be determined whether this element of the program can yield an appropriately large sample size.

The report from the training session was made available for review. It contains sample data sheets and instructions provided to the technicians charged with conducting the sampling program. Two levels of sampling are described, one representing a minimum series of specimens and a more extensive second level sampling to be undertaken as time and conditions at sea allow. Information requirements were minimized to ensure completeness. While the need to maintain a streamlined approach to data collection is recognized, it is unfortunate that certain potentially useful measures were not included in the protocol. Specifically, blubber thickness and body weight are not required. Such information, when combined with body length and girth, can be used to establish condition indices that might be used as an indirect measure of habitat quality, and thereby help to address the question of environmental variability. It was suggested to NMFS that blubber samples collected for toxicological analysis could serve as a proxy source of data. Assuming that samples are collected in the standardized fashion prescribed in the protocol, thickness measurements could be taken by the laboratory charged with conducting the toxicological analyses.

At the time of this review, NMFS had not established a detailed list of the specific analyses that would be performed, other than generic histopathology, immunohistology, toxicology and genetics. Life history data would be derived from teeth and reproductive tracts, presumably analyzed in-house. Potential collaborators were identified for histopathology and immunological studies. It was suggested that if the sampling program was as successful as originally designed (estimated 150 dolphins in each of 3 years) it might be expedient to selectively analyze those cases for which level 2 sampling had been performed, with a view to equalizing the number of specimens in various age/sex classes to allow statistical comparisons. Delays in the initiation of the necropsy program and uncertainties concerning the extent of cooperation that will be received from the tuna fleet may preclude any need to subset the specimens.

Handling Study

NMFS was instructed to develop an experiment involving the repeated capture of spinner or spotted dolphins to chart the physiological responses to such handling. The expectation is that data derived from such a study would allow a more informed assessment of the potential for chronic or cumulative effects resulting from fisheries-related activities, and thereby address the question of whether such activities might reduce individual fitness or reproductive success. A workshop was convened in July, 1997, to develop strategies for the study, while examining whether there was a reasonable expectation that samples could be obtained and analyses performed to answer this question (Curry, 1998).

The workshop concluded that this would be possible, and NMFS has proceeded with preliminary discussions of logistic requirements to complete the experiment. No further details of the project or specifics of the analyses had been developed for the purposes of this review. NMFS suggested that it was likely that a panel of experts would be convened to establish a sampling regimen and identify specific analyses that would provide meaningful information on this issue. The study is expected to variably include blood samples, tissue biopsies, imaging techniques (ultrasound, infrared thermography), and physiological sensors (pulse oxymetry, core and muscle thermometry).

One potential limitation of the study design is that it will not be possible to determine recovery for those blood constituents that are expected to be acutely responsive to the stress of recapture. Thus, while it may be important to recognize that cortisol levels remain elevated because of continued secretion or delayed clearance, this effect may be lost within the elevations associated with recapture. In addition, it will be difficult to establish baseline data, unaffected by capture stress, for many of the constituents of interest. Studies on captive animals benefit from the ability to sample animals voluntarily providing access for venipuncture. Such opportunities will not be available to researchers in the proposed studies.