

PRELIMINARY REPORT ON THE STATUS OF ELECTROPHORETIC STOCK IDENTIFICATION OF  
ATLANTIC BLUEFIN TUNA (*Thunnus thynnus*) FROM THE EASTERN AND WESTERN ATLANTIC OCEAN

by

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SUMMARY

A brief description of the experimental design for the electrophoretic stock identification research being conducted on eastern and western Atlantic bluefin tunas (*Thunnus thynnus*) at the National Marine Fisheries Service's Miami Laboratory is provided in this report. Accomplishments and observations of this research are discussed. These include the finding that liver tissue of 1-year-old western Atlantic bluefin tunas has four variable enzyme systems out of the 23 that were examined.

RESUME

Le présent document fournit une brève description du projet expérimental de recherche sur l'identification des stocks par l'électrophorétique, qui est actuellement en cours au laboratoire de Miami du "National Marine Fisheries Service", concernant le thon rouge (*Thunnus thynnus*) de l'Atlantique Est et Ouest. Ces recherches sont également évaluées et commentées. Il a été découvert, entre autres, que les tissus hépatiques des thons rouges de 1 an de l'Atlantique Ouest comptaient 4 systèmes enzymatiques variables sur les 23 qui ont été examinés.

RESUMEN

El documento presenta una breve descripción del proyecto experimental para identificación de stocks por medios electroforéticos, que se lleva a cabo sobre atunes (*Thunnus thynnus*) del Atlántico oriental y occidental en el Laboratorio de Miami del National Marine Fisheries Service. Se examinan los resultados y observaciones, que incluyen el hallazgo de que el tejido hepático de los atunes de 1 año de edad del Atlántico occidental tiene cuatro sistemas variables de enzimas de los 23 examinados.

## INTRODUCTION

One of the crucial points concerning the effective management of a fishery is to know how many populations comprise the total stock, because different management schemes may apply to different populations.

A biochemical technique which can be used to identify populations is polyacrylamide gel electrophoresis, a method of analyzing specific enzymes. By observing the absence or presence of isozymes of a certain variant enzyme system that has been adequately sampled with respect to groups of fish isolated by large geographic distances, one can determine whether these fish are from the same or from different breeding populations. This is the basic premise upon which this experiment is based. There have been successful applications of biochemical genetic methods for the identification of fish populations and these have demonstrated the usefulness of electrophoresis as a technique for fisheries management (de Ligny, 1969, 1972; Utter et al., 1974; Allendorf and Utter, in press).

The objective of this study is to determine if Atlantic bluefin tuna (*Thunnus thynnus*) from the eastern and western Atlantic Ocean constitute a single breeding population or two or more different populations.

## EXPERIMENTAL

Sixty 1-year-old bluefin tunas from a single location from both the eastern and western Atlantic Ocean constitute the samples for this study. These fish are preserved by freezing and held in this condition until they are analyzed. This study was designed to be accomplished in two phases. Phase I is a survey of 12 fish, six each from the eastern and western Atlantic Ocean. From each of the 12 fish, four tissues (heart, liver, white muscle, red muscle) are to be analyzed for 23 different enzymes in duplicate. These enzymes are: glucose-6-phosphate dehydrogenase, 6-phosphogluconate dehydrogenase, phosphohexose isomerase, phosphoglucomutase, tetrazolium oxidase, lactate dehydrogenase, esterase, carbonic anhydrase, alkaline phosphatase,  $\alpha$ -glycerophosphate dehydrogenase, glyceraldehyde-3-phosphate dehydrogenase, isocitrate dehydrogenase, leucine aminopeptidase, peroxidase, glutathione reductase, malate dehydrogenase, aminoaspartate transferase, acid phosphatase, glutamate dehydrogenase, xanthine dehydrogenase, malic enzyme, succinate dehydrogenase, and alcohol dehydrogenase.

The methodology used for the enzyme determinations is essentially that described by Brewer (1970). Modifications and adaptations of these methods were made to suit particular needs. Polyacrylamide gel electrophoresis was employed, rather than starch, simply because of polyacrylamide's superior resolution of bands.

The Phase I survey will provide the information necessary to make an appropriate selection as to which tissue(s) and enzyme(s) need to be more extensively studied for comparison of eastern and western Atlantic Ocean samples.

Phase II of the study will involve the analysis of the remaining 54 fish from both the eastern and western Atlantic Ocean. Emphasis will be on analysis of the particular enzyme(s) and tissue(s) indicated by data from Phase I.

## RESULTS AND DISCUSSION

For an enzyme system to be useful as a genetic marker in differentiating between populations of fish of the same species, it must be variable, i.e., display either or both different positioning or numbers of bands on the gel. If an enzyme does not show variability then its isozyme pattern can not be used for comparison and ultimate statistical analysis to that from another population. This is the reasoning behind conducting such a survey as in Phase I of this study.

During the summer of 1976, 65 1-year-old bluefin tunas were collected off Cape May, New Jersey, U.S.A. Analyses described previously have been completed on the 6 western Atlantic Ocean fish for Phase I. No samples were acquired from the eastern Atlantic Ocean in 1976. In August, 1977, 17 samples were obtained from off Casablanca, Morocco, Africa. Seventeen tunas from one location is not an adequate statistical sample; therefore, we must obtain a minimum of 60 fish from a single eastern Atlantic location for the data to be useful in the determination of breeding population segregation or intergration.

Every possible effort is being made to obtain more samples from the eastern Atlantic Ocean through contacts in both France and Spain. Phase I cannot be completed until acquisition and analysis of the eastern Atlantic Ocean samples has been completed.

The original objective has been temporarily delayed until a sufficient number of eastern Atlantic tunas are obtained. In the interim, we have an alternate objective which is to determine if any genetic polymorphism exists within the western Atlantic population.

From the 23 enzymes in each of the four different tissues that were surveyed in the 6 western Atlantic fish in Phase I, four enzymes appeared to display variation. With one exception, these four enzymes are all in the liver tissue. The enzyme, tetrazolium oxidase, was found to be variable in all four tissues examined. This enzyme is, however, the only enzyme from Atlantic bluefin tuna that has previously been reported to be a variant system. (Edmunds and Sammons, 1971, 1973). The other three enzymes we found to be variable in liver were malic enzyme, lactate dehydrogenase, and glucose-6-phosphate dehydrogenase. Enzyme analyses were performed on the remaining 59 western Atlantic fish for the four variable systems mentioned above and the data obtained confirmed our original observations that these systems are indeed variants. Although we have not statistically analyzed these data as yet, it appears that these four liver systems are polymorphic.

We have analyzed the 17 Moroccan tunas with respect to the four variant systems found in livers of the western Atlantic samples. As stated previously, 17 samples are not a sufficient quantity to enable drawing valid conclusions after comparison to western Atlantic data; however, we may be able to detect any trends that might be present. Our analyses of these tunas has shown that of the four systems analyzed none were variable in these 17 fish with the exception of tetrazolium oxidase. This is excellent for comparison to data derived from the variable western samples, but the sample size is too small to draw any conclusions.

Of the 65 fish analyzed from the western Atlantic, about half displayed six bands for the glucose-6-phosphate dehydrogenase system, whereas the other half displayed seven bands. The 17 eastern Atlantic fish all displayed the seven-band pattern. This is, perhaps, a trend which can only be verified with additional sampling.

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