REVIEW OF PROGRESS ON OXIDATIVE DETERIORATION IN FISH AND FISHERY PRODUCTS

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ABSTRACT

A review of the history and accomplishments of a cooperative research program, jointly sponsored by the University of California and the Bureau of Commercial Fisheries, is presented. Oxidative deterioration in (1) extracted fish oils and (2) in the tissues of fish, is being studied in terms of autoxidative and enzymatic mechanisms. Principal findings to date include (1) analyses of conditions affecting the efficacy of antioxidants, (2) catalytic effect of hematin compounds on the oxidation of and rancidification of fish oils in situ and after extraction, (3) effects of lack of reducing vitamins in the meat upon the oxidative discoloration of the meat during the canning process, (4) carbonyl-amine reaction and coupling plays a relatively minor part in the browning or "rusting" of fish and (5) comparative rates of oxidation of different fish meals were shown to be correlated directly with the relative contents of hematin compounds. A discussion of research now under way is also presented.

INTRODUCTION

A cooperative program concerned with the oxidative deterioration that occurs in fish and fishery products has been under way since September 1955 between the Food Technology Department and the Institute of Marine Resources, both of the University of California, and the Seattle Fishery Technological Laboratory of the U. S. Bureau of Commercial Fisheries (Brown 1956, Stansby 1957). The purpose of the present report is to review the accomplishments of this program and to outline briefly the nature of the current work.

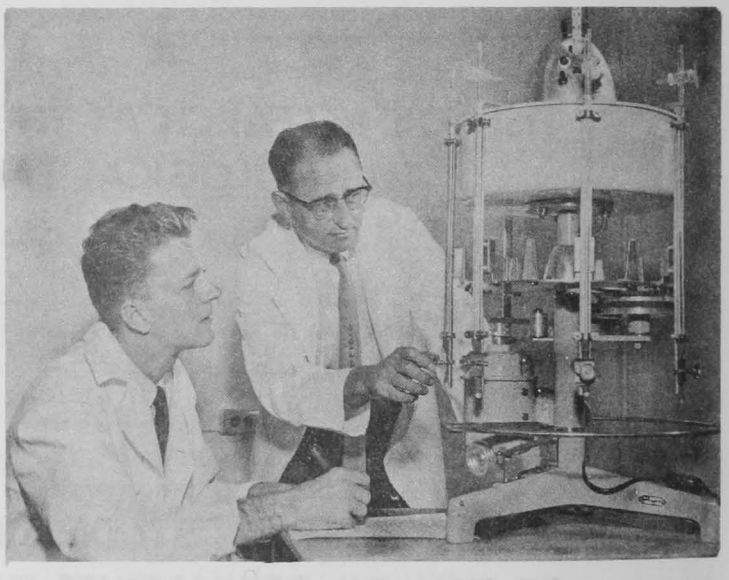

Fig. 1 - Operation of Warburg equipment for measurement of oxygen uptake by fish oils.

That portion of this program being carried out by the Institute of Marine Resources in the Food Technology Department at Berkeley is concerned primarily with the mechanism of oxidation in extracted fish oils. Dr. H. S. Olcott of the University of California is the project leader. He is assisted in the research by Dr. Edwin J. Kuta, Bureau of Commercial Fisheries chemist, and by two part-time Bureau of Commercial Fisheries physical science aides, Miss Esther Edery and Miss Carol DeJong.

That portion of the program at the Food Technology Department at Davis is concerned with oxidative deterioration occurring in the tissue of fish. Dr. A. L. Tappel of the University is the project leader. He is assisted by Dr. W. Duane Brown, Bureau chemist, and Mr. Michael Gumbmann, part-time chemist.

When the program first was started, in September 1955, it was carried out at Davis. The work began as a general preliminary survey of the mechanism of oxidation of the components (mainly oils and pigments) of fish tissue. Later, in July 1956, the presently-divided arrangement was made wherein the work on mechanism

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of oxidation in extracted oils was set up as a related but independent project for work under the then newly-established Institute of Marine Resources at Berkeley.

PRINCIPAL FINDINGS OF PROGRAM

Some of the most significant findings deal with the mechanism of oxidation in extracted fish oils. Important observations have been made (1) regarding conditions for applying antioxidants to such systems and (2) regarding the role of such factors as the amount of free fatty acid in the oil. Since these results are discussed in another report, no further mention of them will be made here.

HEMATIN CATALYSIS: The first outstanding finding from this program was the conclusive demonstration, previously shown for meat and other meat foods and now shown for fish, that the oxidation of oil in the meat, which results in rancidity, is very greatly accelerated by the presence of hematin compounds (Brown, Venolia, Tappel, and Stansby 1957). This catalytic effect was brought out strikingly in experiments in which the amount of oxygen adsorbed by cubes of dark meat and of light meat of fish was measured in a Warburg apparatus. The dark meat contains most of the hematin compounds. In these experiments, the light meat of such species of fish as salmon and tuna absorbed no measurable amount of oxygen, whereas the dark meat absorbed a very considerable amount.

These results were confirmed in other experiments in which model systems containing purified fatty acids and proteins found in fish were tested for adsorption of oxygen. Again, it was demonstrated clearly that the presence of hematin compounds increased the rate of oxygen adsorption many fold.

The content of hematin in a number of species of fish was measured. Halibut, ocean perch, trout, and cod contained between 0.1 and 0.4 x 10^{-5} M. Rockfish and lingcod contained 0.5 x 10^{-5} M; pilchard, 5.4 x 10^{-5} M; and tuna, 8.5 x 10^{-5} M. This order of arrangement of species is, in a general way, in the order of increasing susceptibility to oxidation, indicating that the content of hematin compounds is a major factor in determining the rate of development of rancidity in fish.

It was shown that during oxidation of the oil in the meat of fish, the hematin compounds decrease and are chemically altered. Thus, in one experiment (Brown et al., 1957), the concentration of hematin compound decreased during the course of the oxidation from 2.1 x 10^{-5} M to 1.0 x 10^{-5} M in the light meat, and from 67.2 x 10^{-5} M to 49.6 x 10^{-5} M in the dark meat. At the same time, the spectral adsorption curves of an aqueous extract of the samples before and after oxidation showed a change in maxima from 542 to 500 millimicrons in the one case, and from 578 to 630 millimicrons in the other, indicating a transformation of oxyhemoglobin or oxymyoglobin to methemoglobin or metmyoglobin.

TUNA PIGMENT CHANGES: The work on hematin catalysis of oil oxidation in fish tissue helped clarify an indirectly-related problem involving changes in hematin pigments in tuna, which affect color and marketability of canned tuna. Normally, the color of raw tuna changes during precooking from an indefinite gray to a light pink. This change in color is considered desirable. Some occasional batches of tuna, upon being precooked, do not become pink, however, but change to a variety of colors ranging from a greenish gray to shades of orange, tan, or brown. Such tuna are described as "green" tuna, and if the color is extreme, the fish are considered to be unmarketable. In some cases, the discoloration is accompanied by changes in texture, odor, and flavor. Before the present program was started, nothing was known about the chemistry of the changes in the pigments of tuna causing these changes in color.

Brown and Tappel (1957) now have shown that the pink color that normally develops in precooked tuna is due to hemochromes whose non-heme constitutent is either denatured globin or nicotinamide, or probably both. Brown, Tappel, and Ol-

cott (1958) have shown that the off-color (so-called "green" tuna) was caused by the presence of a hemichrome pigment that can be transformed back into the pink hemochrome by treatment with suitable reducing agents. A more rapid change occurs with the use of sodium hydrosulfite as reducing agent, and a slower change occurs with the use of ascorbic acid. In some lots of tuna, the restoration of the pink color is enhanced by the use of nicotinamide with the reducing agent. These reactions might be employed to ensure retention of proper color in the meat of fish during commercial canning of tuna.

FISH MEAL: In initial survey experiments, Brown and coworkers (1957) used samples of freeze-dried fish to simulate ideally-dried fish meal. They found that the rate of oxidation of such samples varied greatly, depending upon the part of the meat used and upon the species of fish. Thus the dark meat from freeze-dried pink salmon oxidized nearly 9 times as fast as did the light meat from this species, and a freeze-dried sample of pilchard oxidized over 300 times as fast as did one from cod.

Experiments in which antioxidants were added to commercial fish meals showed that use of BHA or BHT reduced the rate of oxidation to less than one-quarter the rate for untreated samples and that use of Santoquin (not yet approved by Food and Drug Administration) reduced the rate to less than one-eighth that of untreated samples.

PROTEIN-OIL REACTION: The mechanism of browning such as occurs in fish meat and in the rusting of frozen fish as a result of polymerization, oxidation, and the carbonyl-amine reaction has been investigated (Stansby 1957). The carbonyl-amine reaction has been found to play a minor role in such browning (Venolia and Tappel 1958).

CURRENT RESEARCH

The current research deals with (1) investigation of oxidative reaction mechanism in extracted fish oils, (2) investigation of enzymatic oxidation in the tissue of fish, and (3) further investigation of the alternation of pigment in tuna.

The investigation of oxidative reaction mechanism in extracted oils considers such factors as the effect upon oxidation rates of the presence of natural antioxidants and of contaminating heavy metals.

The investigation on enzymatic oxidation considers the enzymatic oxidation of unsaturated lipides, carbohydrate metabolism, and the tricarboxylic acid cycle in fish tissue. Each of these programs is discussed in separate reports, and accordingly, will not be discussed further here.

Additional investigation of green tuna is continuing as samples become available. Most samples of green tuna are of the type that can be reversibly changed back to the normal pink color by treatment with a reducing substance such as sodium hydrosulfite. Apparently, however, some lots of tuna are "green" because of some quite different pigment reaction, and these may not be reversibly reduced to the normal pink color. Samples of this type are rare; so far, only one has been found.

Even for those samples that can be reversibly altered back to the pink color, it is not entirely clear as to what factors in the handling of the fish determine whether use of standard canning procedures will result in green or in normal color in the precooked and canned product. In other words, do green tuna result from some factor before the fish are caught--for example, presence of some special feed--or do they result from some handling conditions, and if so, what are these conditions? Samples of tuna of known history are being obtained, and observations on effects on greening are being made. As sufficient samples for which handling history can be correlated with development of greening are obtained, the chemistry of pigment changes will be investigated with relation to this history.

DISCUSSION

The success of this program in obtaining results of practical value demonstrates the importance of a basic approach in undertaking research in the field of fishery technology. The initial planning in this program was based upon a rather theoretical investigation of factors influencing oxidative changes occurring in fish tissue. It was presumed that the first application of this basic research would be in the control of rancidity. The possibility of applying such findings to the problem of green tuna was not even considered when the program was started.

In a similar way, it is probable that work now at early stages of development on enzymatic changes and oxidative mechanisms in extracted oils may lead to applications not even conceived of at present.

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WATER PISTOL TECHNIQUE USED TO ANAESTHETIZE FISH

A new water pistol technique, developed by two American scientists, involves an anaesthetic called "M. S. 222." A sea-water solution of this drug is sprayed over a fish thrashing on the end of the line, stunning it within a minute. The technique was developed to capture sharks and rays for use in scientific studies, but these findings might also be useful to anglers who catch fish for food or exhibition. The drug has no harmful effect on the fish, and does not spoil the flavor.

The drug, an amethan-sulphonate compound, is sprayed over the fish by means of a water pistol, rubber bulb syringe, or small pump-type handsprayer. Within 15 seconds the M. S. 222 solution begins to take effect and, as a rule, even a 400-pound shark is anaesthetized in a minute or less. The fish return to consciousness in 5 to 30 minutes after being re-immersed in water, depending on their size and the amount of spray they received (Irish Fishing and Fish Trades Gazette, August 17, 1957).