The autoxidative behavior of milk lipid fractions in systems of controlled composition
by Shun Ku

A thesis submittted to the Graduate Faculty in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in Agricultural Products Utilization
Montana State University

Abstract:
Fractions of milk lipids representing the phospholipid protein complex (fat globule membrane material from buttermilk (FGM I), and from butter serum (FGM II), the true fat (butteroil), and the intact milk fat globules (dialyzed cream) were allowed to autoxidize in systems composed of distillated water or milk serum or occasionally pH 6.60 phosptiate buffer. The effect of the following factors was investigated: (1) copper ion alone, ascorbic acid alone and copper ion plus ascorbic acid (2) sul-phydryl groups (L-cysteine), whole casein and K-casein (3) synthetic and naturally occurring antioxidants (NDGA and α-tocopherol).

The autoxidative behavior of these milk lipid fractions was dependent on the particular fraction used and the nature and composition of the aqueous system into which it was dispersed. Copper ion alone and ascorbic acid alone, catalyzed oxidized flavor development by the fat globule membrane and dialyzed cream. Copper ion plus ascorbic acid gave the highest oxidized flavor intensity with fat globule membrane material. Copper ion from various salts gave variable catalytic power indicating that the catalytic power of copper ion in milk serum is dependent on the dominant kinds of cuprous salt complexes. The lowest oxidation intensities were given by the butteroil.

The fat globule membrane from buttermilk (FGM I)? differed very significantly in its oxidative behavior in comparison to that from butter serum (FGM II). Generally FGM II gave much higher oxidation rates and intensities than FGM I.

L-cysteine, whole casein, K-casein, NDGA and α-tocopherol behaved either as antioxidatants or pro-oxidants depending on the milk lipid fractions in question and the aqueous medium used. The reason for such behavior is discussed.

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ABSTRACT

Fractions of milk lipids representing the phospholipid protein complex (fat globule membrane material from buttermilk (FGM I), and from butter serum (FGM II), the true fat (butteroil), and the intact milk fat globules (dialyzed cream) were allowed to autoxidize in systems composed of distilled water or milk serum or occasionally pH 6.60 phosphate buffer. The effect of the following factors was investigated: (1) copper ion alone, ascorbic acid alone and copper ion plus ascorbic acid (2) sulphydryl groups (L-cysteine), whole casein and K-casein (3) synthetic and naturally occurring antioxidants (NDGA and α-tocopherol).

The autoxidative behavior of these milk lipid fractions was dependent on the particular fraction used and the nature and composition of the aqueous system into which it was dispersed. Copper ion alone and ascorbic acid alone, catalyzed oxidized flavor development by the fat globule membrane and dialyzed cream. Copper ion plus ascorbic acid gave the highest oxidized flavor intensity with fat globule membrane material. Copper ion from various salts gave variable catalytic power indicating that the catalytic power of copper ion in milk serum is dependent on the dominant kinds of cuprous salt complexes. The lowest oxidation intensities were given by the butteroil.

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L-cysteine, whole casein, K-casein, NDGA and α-tocopherol behaved either as antioxidatants or pro-oxidants depending on the milk lipid fractions in question and the aqueous medium used. The reason for such behavior is discussed.

INTRODUCTION

Off-flavors resulting from lipid oxidation are among the most common flavor defects occurring in milk and its products. These off-flavors are often described as: cardboard, oxidized, metallic, oily, tallowy, or fishy depending on the intensity of the defect. This flavor defect causes millions of dollars of losses every year both to the milk producers and processors.

The bulk of the fat in milk is in the form of small fat globules averaging 2-5 u in diameter. The surface of each fat globule is covered with a layer of material commonly known as the fat globule membrane (FGM). This membrane is a complex made up mainly of phospholipids and proteins. From the standpoint of autoxidation, it is of importance to realize that these globules exhibit a large surface area which is rich in oxidizable phospholipids. The most important factors that tend to accelerate oxidized flavor development are metal catalysis and the presence of ascorbic acid which provide a properly poised hydrogen accepting and donating system in the reduced-oxidized ascorbic acid relationship.

Research on oxidized flavor development in milk is complicated by the fact that different milks show extreme variation in resistance and susceptibility. Some milks are objectionably oxidized within a few hours from milking, while in others, this effect cannot be induced even with large additions of copper. There is little doubt that the complexity and variability of milk composition are contributing factors to the confusing results sometimes obtained in oxidized flavor research. In the present investigation, model systems of known composition were used in an effort to minimize the variables. Various fractions of milk lipid were incorporated in these systems with the objective of determining the source of "off-flavors" and the compositional conditions that modify their intensities. Among the lipid fractions used were: fat globule membrane materials both from buttermilk, and butter serum; whole fat globules as dialyzed cream; butteroil prepared by skimming (devoid of phospholipids), and butteroil prepared by complete dehydration (rich in phospholipids). Comparative analyses were made to investigate the following:

(1) The development and intensity of the flavor in various media such as milk serum, phosphate buffer and salt-free medium (water).

(2) The effect of variation in the concentration of some of the major and minor milk constituents on the course and intensity of flavor development.

(3) The role of copper ions and ascorbic acid in promoting or inhibiting oxidized flavor development in the various milk lipid fractions.

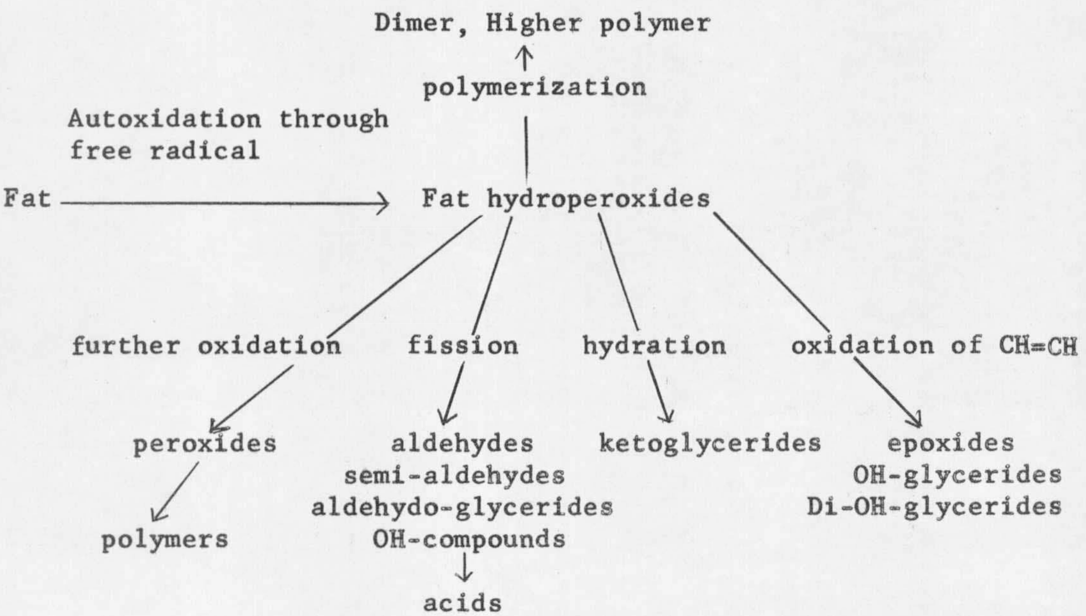
(4) The effect of some antioxidants and the conditions leading to their relative effectiveness.

REVIEW OF LITERATURE

I. THE NATURE OF AUTOXIDATION

A. The Origin of Oxidized Flavor

Oxidized flavor was first recognized as due to autoxidation of milk lipids by Rogers and Gray (1909). Greenbank (1940) believed this flavor was due to a group of unstable intermediate compounds which could be further oxidized to flavorless substances. The compounds resulting from lipid autoxidation represent a complex system, and the general routes of the reactions taking place were reported by Lea (1962) as follows:



El-Negoumy *et al.* (1962a) indicated that the polyunsaturated fatty acids, especially linolec and linolenic acid are the precursors for flavor compounds. According to Farmer and Sutton (1943b) and Farmer and Sundralingam (1943a), lipid autoxidation is initiated through a free radical mechanism as follows: