

# IOWA STATE UNIVERSITY

## Digital Repository

---

Retrospective Theses and Dissertations

Iowa State University Capstones, Theses and  
Dissertations

---

1941

## Flavor development in unsalted butter

Theodore Isaac Hedrick

*Iowa State College*

Follow this and additional works at: <https://lib.dr.iastate.edu/rtd>



Part of the [Agriculture Commons](#), and the [Food Science Commons](#)

---

### Recommended Citation

Hedrick, Theodore Isaac, "Flavor development in unsalted butter" (1941). *Retrospective Theses and Dissertations*. 13956.  
<https://lib.dr.iastate.edu/rtd/13956>

This Dissertation is brought to you for free and open access by the Iowa State University Capstones, Theses and Dissertations at Iowa State University Digital Repository. It has been accepted for inclusion in Retrospective Theses and Dissertations by an authorized administrator of Iowa State University Digital Repository. For more information, please contact [digirep@iastate.edu](mailto:digirep@iastate.edu).



[www.manaraa.com](http://www.manaraa.com)

# **NOTE TO USERS**

This reproduction is the best copy available.





**FLAVOR DEVELOPMENT IN UNSALTED BUTTER**

by

**Theodore Isaac Hedrick**

**A Thesis Submitted to the Graduate Faculty  
for the Degree of**

**DOCTOR OF PHILOSOPHY**

**Major Subject Dairy Bacteriology**

**Approved:**

Signature was redacted for privacy.

**In charge of Major work**

Signature was redacted for privacy.

**Head of Major Department**

Signature was redacted for privacy.

**Dean of Graduate College**

**Iowa State College  
1941**

UMI Number: DP12749

## INFORMATION TO USERS

The quality of this reproduction is dependent upon the quality of the copy submitted. Broken or indistinct print, colored or poor quality illustrations and photographs, print bleed-through, substandard margins, and improper alignment can adversely affect reproduction.

In the unlikely event that the author did not send a complete manuscript and there are missing pages, these will be noted. Also, if unauthorized copyright material had to be removed, a note will indicate the deletion.



---

UMI Microform DP12749

Copyright 2005 by ProQuest Information and Learning Company.  
All rights reserved. This microform edition is protected against  
unauthorized copying under Title 17, United States Code.

ProQuest Information and Learning Company  
300 North Zeeb Road  
P.O. Box 1346  
Ann Arbor, MI 48106-1346

QR121  
H359 f

1126-89

- 2 -

TABLE OF CONTENTS

	Page
INTRODUCTION.....	4
HISTORICAL.....	7
Diacetyl and acetylmethylcarbinol as flavor constituents of butter.....	7
Formation of diacetyl and acetylmethylcarbinol.....	11
Unsalted butter.....	13
ANALYTICAL METHODS.....	15
EXPERIMENTAL.....	17
Part I. Preliminary Churnings.....	17
General procedure.....	17
Presentation of results.....	19
Effect of the percentage of acid developed in cream on the diacetyl and acetylmethylcarbinol contents.....	20
Effect of the addition of citric acid on diacetyl and acetylmethylcarbinol contents of ripened cream.....	27
Effect of agitation during ripening on diacetyl and acetylmethylcarbinol contents of cream.....	33
Effect of ripening temperatures on diacetyl and acetylmethylcarbinol contents of cream.....	37
Influence of different butter cultures on diacetyl and acetylmethylcarbinol contents of cream.....	40
Effect of various percentages of fat in cream or milk on diacetyl and acetylmethylcarbinol contents.....	47
Ratio of the diacetyl and acetylmethylcarbinol contents of ripened cream to those of the corresponding unsalted butter.....	57

T7207

	Page
Part II. Plant Churnings.....	59
General procedure.....	59
Effect of the percentage of acid developed in cream on the diacetyl and acetyl methylcarbinol contents.....	62
Effect of the addition of citric acid on diacetyl and acetyl methylcarbinol contents of ripened cream.....	65
Effect of agitation during ripening on diacetyl and acetyl methylcarbinol contents of cream.....	67
Effect of ripening temperatures on diacetyl and acetyl methylcarbinol contents of cream.....	69
Influence of different butter cultures on diacetyl and acetyl methylcarbinol contents of cream.....	72
Ratio of the diacetyl or acetyl methylcarbinol contents of ripened cream to those of the corresponding unsalted butter.....	75
Changes in diacetyl and acetyl methylcarbinol contents during holding of unsalted butter.....	77
Effect of various procedures during ripening of cream on the flavor of unsalted butter.....	85
DISCUSSION OF RESULTS.....	96
SUMMARY.....	101
Part I. Preliminary Churnings.....	101
Part II. Plant Churnings.....	103
APPENDIX.....	105
ACKNOWLEDGMENTS.....	143
LITERATURE CITED.....	144

#### INTRODUCTION

High-flavored unsalted butter has increased greatly in commercial importance during recent years, the principal areas of production being the mid-western states. This type of butter is sometimes called sweet butter and is made from either sweet or neutralized sour cream that has been ripened by the addition of butter culture. Distinct from this type of butter is the unsalted butter used in ice cream mixes and other products; this is made without butter culture from cream having a low acidity at the time of churning.

In the absence of salt it is necessary that the butter have a high flavor if it is to satisfy consumers. Accordingly, the problems with unsalted table butter differ from those of salted butter. The lack of salt permits acidities to be developed during cream ripening and holding of butter that would be prohibitive with salted butter because of the probability of fishiness developing; unsalted butter rarely becomes fishy. Development of acidity favors the production of flavor and also limits bacterial deterioration.

Since diacetyl is considered the most important compound contributing to the desirable flavor and aroma of butter, the amount of diacetyl was determined and used as an index of flavor development. The common assumption that diacetyl is formed by the biological oxidation of acetyl-methylecarbinol prompted the determination of acetyl-methylecarbinol also. There may be other materials, for example, volatile acids and esters of fatty acids, contributing to butter flavor, but they are of minor

importance.

The results reported herein cover a study of the effects of various factors on the contents of diacetyl and its probable precursor, acetyl-methyloarbinol, in ripened cream and in butter churned from the ripened cream; the effects of the factors on the flavor and keeping quality of butter also were considered.

Preliminary investigations were conducted under laboratory conditions on the following factors that might influence the contents of

diacetyl and acetyl-methyloarbinol in sweet or neutralized sour cream ripened to relatively high acidities and in the corresponding butter:

- (a) Effect of the percentage of acid developed in cream on the diacetyl and acetyl-methyloarbinol contents.
  - (b) Effect of the addition of citric acid on diacetyl and acetyl-methyloarbinol contents of ripened cream.
  - (c) Effect of agitation during ripening on diacetyl and acetyl-methyloarbinol contents of cream.
  - (d) Effect of ripening temperatures on diacetyl and acetyl-methyloarbinol contents of cream.
  - (e) Influence of different butter cultures on diacetyl and acetyl-methyloarbinol contents of cream.
  - (f) Effect of various percentages of fat in cream or milk on diacetyl and acetyl-methyloarbinol contents.
- These studies were then used as a basis for trials under commercial conditions. In addition to determining the diacetyl and acetyl-methyl-

carbinol contents of the cream just before churning and of the fresh butter, a sample of butter was analyzed for these compounds after holding 1 week at 36 to 40°F., 1 month at 36 to 40°F., and 3 days at 60°F. followed by 4 days at 36 to 40°F. Samples of butter were scored for flavor after holding 1 week at 36 to 40°F., after 1 month at 36 to 40°F., and after 6 months at -10 to 0°F.

## HISTORICAL

### Diacetyl and acetylmethylcarbinol as flavor constituents of butter

The importance of diacetyl as a flavor constituent of butter was indicated by the work of van Niel, Kluyver and Dierck (29), in 1929. Van Niel observed that specific strains of propionic acid bacteria, which apparently yielded acetylmethylcarbinol, produced an odor similar to that of butter with a high flavor. Samples of high-flavored butter gave positive reactions to acetylmethylcarbinol tests. However, purified acetylmethylcarbinol was odorless. It was concluded that diacetyl, oxidized from carbinol, gave the characteristic aroma to butter. Analyses revealed that fine butter contained from 0.0002 to 0.0004 per cent diacetyl and, by the addition of a similar amount to butter which lacked flavor, a desirable flavor was produced. Studies concerning the source of butter flavor indicated that cultures of certain organisms, used in the cream before manufacturing, produced a desirable flavor, but most organisms did not appear to have the ability to produce this flavor. This latter fact had previously been suggested by several investigators. Conn (6), as early as 1889, suggested that proper cream ripening was the result of the action of bacteria, several species being involved.

Boekhout and Ott de Vries (4), Hammer and Bailey (9) and Storch (26) announced, about the same time, that two types of bacteria are present in butter cultures, the lactic acid producers and another type which produced a desirable flavor and aroma. The formation of volatile acid

by the latter type was stressed by Hammer and Bailey. Hammer (8) later indicated the importance of citric acid as a source of volatile acid production by the two species of this type, which he named Streptococcus paracitrovorus and Streptococcus citrovorus.

Schmalfuss (23) noted the odor of diacetyl in a milk culture of Streptococcus acidi lactici and Streptococcus cremoris and confirmed its presence. He considered diacetyl to be an assimilation product of the organisms.

In studying butter cultures it was observed by Michaelian, Farmer, and Hammer (12) that most of the acetyl methyl carbinol plus diacetyl was produced in the latter stages of ripening. There seemed to be a definite pH limit of 4.0 to 4.3 for rapid production of flavor. It also appeared that the butter culture organisms might cause a destruction of acetyl-methylcarbinol plus diacetyl. High acidities helped to retard the destruction and neutralization accelerated the action.

It is generally conceded that any process which adds oxygen to a ripening butter culture will cause larger yields of the flavor constituents. Prill and Hammer (20) aerated cultures by shaking and obtained large increases in diacetyl on holding. Virtanen (30) observed that, if the ripening process were conducted in thin layers, the production of diacetyl and acetyl methyl carbinol was more active, while the opposite was true under more anaerobic conditions. He also reported that bubbling air through cream gave high-flavored butter, but the flavor diminished during storage. Brewer et al. (5) greatly increased the production of acetyl-

methylcarbinol and diacetyl by aeration of butter cultures under pressure.

Wiley et al. (33) reported investigations on the effect of temperature and starters on the diacetyl and flavor (taste) of ripened cream and the resulting butter. Starters with no "brightness" imparted as much flavor to butter as those having much "brightness". "Brightness" correlated well with the diacetyl content. However, the diacetyl content of cream did not correlate with the diacetyl content of the starter, and Streptococcus cremoris produced the same amount of diacetyl at 45°F. (7°C.) as at 70°F. (21°C.) but less acid at the lower temperature.

Mohr (16) considered the optimum temperature and acidity for growth of butter culture organisms was not the best for the formation of diacetyl. His data indicated one-fourth of the diacetyl and one-fifteenth of the acetyl methylcarbinol present in the cream were retained in butter, the washing operation eliminating much diacetyl and acetyl methylcarbinol. During storage of butter at 50°F. (10°C.) the diacetyl increased up to 4 days and after 10 to 12 days it decreased. At 1½ to 32°F. (-10 to 0°C.) the diacetyl of butter underwent no change. Mohr and Wellm (17) concluded that diacetyl seemed to behave the same in the storage of unsalted or salted butter. They reported that first-quality German butter showed little relationship between the diacetyl content and flavor.

Barniecoat (1) (2) (3), in reviewing the work of others, stated that the variation reported in literature for butter was from 0.05 to 5 p.p.m. in diacetyl content; sweet cream butter averaged about 0.05 p.p.m.,

mild starter butter from 0.3 to 0.4 p.p.m. and high-flavored starter butter 1.5 p.p.m. He reported that the percentage of diacetyl retained by butter in the manufacturing process varied from 0 to 33 per cent and averaged 15 per cent. In storing salted butter at 40°F. the diacetyl content increased for several days after manufacture. There was little change in butter stored at 14 to 17°F. for 6 months, except in butter containing large quantities and the loss then was considerable.

Prill and Hammer (21) emphasized that aeration of butter cultures increased the diacetyl content. There seemed to be a general relation between the diacetyl content of the cream before churning and that of the butter, but the relationship was not close.

From his studies Davies (7) concluded that high-flavored butter did not keep well. This was not because of the diacetyl content but because of other constituents or conditions of manufacture associated with this type of butter.

#### Formation of diacetyl and acetyl methylcarbinol

Attempts have been made by various investigators to explain the formation of diacetyl from citric acid in butter cultures. The most widely accepted idea is that diacetyl is formed from its precursor, acetyl methylcarbinol, by a biological oxidation. It is assumed that acetyl methylcarbinol is one of the products of the breakdown of citric acid in an acid medium.

Taperneux (27), in 1932, suggested the development of diacetyl as follows: Hexose sugar from lactose breaks down to yield methylglyoxal. A molecule of water is taken up to give methylglyoxal hydrate which forms lactic acid by hydrogen rearrangement. The lactic acid then breaks down to acetaldehyde, carbon dioxide, and hydrogen. A condensation of two molecules of acetaldehyde results in the formation of 2-hydroxy butaldehyde which changes into another compound having the same empirical formula, acetyl methylcarbinol. The acetyl methylcarbinol is either oxidized to diacetyl or reduced to 2,3-butylene glycol, depending upon the conditions.

Michaelian and Hammer (13) (14) showed that the production of diacetyl from acetyl methylcarbinol is not a simple chemical oxidation. Diacetyl was not formed after the citric acid fermenting organisms were killed with formalin, chloroform, or heat. Suspensions of dead organisms failed to produce diacetyl, although provided with the usual temperature and pH conditions. Purified acetyl methylcarbinol added to sterile skim milk which was adjusted for pH failed to yield diacetyl when oxygen was

bubbled through the milk.

Van Heynum and Pette (28) concluded from several studies on the decomposition of citric acid that Bacillus cereus produced acetic acid and ethyl alcohol in sweet milk and diacetyl, acetylmethylecarbinol, acetic acid, carbonic acid, and 2,3-butylen glycol in acidified milk.

The scheme proposed is as follows: (a) Citric acid breaks down to acetic acid, carbon dioxide, and pyruvic acid. (b) Pyruvic acid decarboxylates to carbon dioxide and acetaldehyde. (c) In a neutral medium the acetaldehyde undergoes a Cannizzaro reaction and yields acetic acid and ethyl alcohol, while in an acid medium there is a condensation to acetyl-methylecarbinol if conditions are anaerobic and diacetyl if conditions are aerobic. Both diacetyl and acetyl-methylecarbinol also may be reduced to 2,3-butylen glycol. In support of the scheme the authors could obtain no acetyl-methylecarbinol from a citric acid free medium, and the addition of synthetic acetyl-methylecarbinol to cultures of *Bacillus* failed to produce increases in diacetyl. They believe that diacetyl is not the result of a chemical or biological oxidation of acetyl-methylecarbinol but is formed from some intermediate product, probably acetaldehyde.

#### Unsalted butter

Investigations of unsalted butter usually have been confined to bacteriological deterioration and to keeping quality. Butter made without salt develops flavor defects from the action of microorganisms very rapidly at the common holding temperatures. Nelson and Hanner (18) obtained surprisingly large increases of butter culture organisms, by both the plate and microscopic counts, in unsalted butter held at 70°F. Organisms other than streptococci sometimes developed in large numbers. The bacterial count decreased in unsalted butter held at -4°F.

Hunsiker (11) stated that in the United States unsalted butter does not turn fishy, which he attributed to the absence of the solvent action of salt on protein and lecithin. Defects arising from bacterial action result in a limited keeping quality and unsalted butter must be held at cold storage temperatures. Stale, cheesy, rancid flavors, and mold growth may occur in succession. When made without salt, deterioration is more rapid in unripened than in ripened cream butter.

Wiley (32) concluded from his investigations that fat oxidation was greater in salted than unsalted butter. He (31) also reported that deterioration by bacterial action was not delayed by the presence of acid in unsalted butter held at either 65 or 70°F. At temperatures of 0 or 11°F. the greatest deterioration was observed in butter made from cream ripened to a pH of 5.0. Fat of ripened cream (pH 5.0) displayed noticeable oxidation after cold storage, but this was not true of the butterfat from cream acidified to the same pH. The pH had little effect

on the flavor of unsalted butter. Minster (15) regarded the presence of a few genuine lactic acid bacteria in butter as beneficial; they tended to retard the growth of objectionable organisms.

Slatter (24) and Slatter and Hammer (25) indicated that at suitable temperatures unripened cream butter made with culture increased in acetyl-methylocarbinol plus diacetyl and decreased markedly in pH. The increases in acetylmethylocarbinol plus diacetyl varied with different lots of butter held under the same conditions, but the largest increases usually occurred with long holding periods and low pH development. Some lots of butter decreased in diacetyl on holding and remained low, while others increased after an initial decrease. There also were samples that increased from the beginning of the holding. Although the time required was variable, all samples eventually decreased on holding.

Prill and Hammer (22) studied the changes in diacetyl and acetyl-methylocarbinol plus diacetyl contents in unsalted butter manufactured in a manner similar to that employed with salted butter in which culture is used. The unsalted butter was only partially worked, but it was not leaky. The changes in diacetyl and acetylmethylocarbinol plus diacetyl contents of unsalted butter manufactured from sweet or sour cream were negligible at -10 to 0°F. Significant changes in diacetyl and acetyl-methylocarbinol plus diacetyl occurred at 36 to 45°F. and at 70°F.

Changes in both compounds included increases, decreases and increases followed by decreases; increases occurred in most trials and the authors believe these increases were an important factor in flavor development.

#### ANALYTICAL METHODS

##### Acid determination

Nine grams of cream was weighed on a balance and titrated, without diluting, using 0.1 normal sodium hydroxide; phenolphthalein was used as an indicator. The results were calculated as lactic acid to the nearest 0.01 per cent.

##### pH determination

pH determinations on cream and on serum obtained from butter by melting and centrifuging were made with a Leeds and Northrup potentiometer, modified quinhydrone electrode and a calomel half-cell. From the voltage and temperature the pH values were calculated to the nearest 0.01 of a pH unit.

##### Fat determination

When necessary, the percentage of butterfat in the cream was determined according to the official Babcock method.

##### Diacetyl and acetyl methylcarbinol determination

Cream and butter were analyzed for diacetyl and acetyl methylcarbinol by the colorimetric method of Prill and Hammer (19), with a few minor modifications used by Hoecker (10). Fifty grams of butter was weighed into a 500 ml. flask for the diacetyl analysis and the air displaced with carbon dioxide by allowing a small stream to flow into the flask for 5 minutes. The sample was refluxed for 5 minutes by boiling with steam. The diacetyl

was distilled into a tube containing 1 ml. of hydroxylamine acetate solution (19), allowing 5 to 6 ml. of distillate to collect in about 15 minutes. The diacetyl in the distillate was converted into the ammono-ferrous salt, a rose-red colored compound, with ammonium hydroxide and ferrous sulphate and made up to 10 ml. with distilled water. The intensity of the color was measured with a Klett-Summerson photoelectric colorimeter. Readings were obtained with standard solutions of known strength and used to calculate the amount of diacetyl, which was expressed in parts per million (p.p.m.).

The acetylmethylcarbinol analysis was essentially the same. Twenty-five grams of butter was weighed into a 500 ml. flask. Thirteen ml. of a 40 per cent ferric chloride solution was added and the mixture refluxed for 10 minutes with steam to oxidize the acetylmethylcarbinol to diacetyl. Allowing approximately 15 minutes, the distillate was collected in 1 ml. of hydroxylamine acetate solution (19) until the volume was 10 ml. One ml. of the 10 ml. of distillate was transferred into another tube, converted into the ammono-ferrous salt with ammonium hydroxide and ferrous sulphate and made up to 10 ml. with distilled water. The intensity of the color was measured with the photoelectric colorimeter. The results were calculated as parts per million of acetylmethylcarbinol but actually represent acetylmethylcarbinol plus diacetyl. Since the amount of diacetyl was insignificant compared to the amounts of acetylmethylcarbinol, this procedure did not materially affect the acetylmethylcarbinol results.

## EXPERIMENTAL

### Part I. Preliminary Churnings

Preliminary studies were conducted in the laboratory on the factors influencing the formation of diacetyl and acetymethylcarbinol in cream ripened to relatively high acidities for the production of high-flavored unsalted butter. In these studies only small amounts of cream were used.

#### General procedure

Cream was pasteurized in a 300-gallon vat at 150°F. for 30 minutes in the butter laboratory. The butterfat content of the cream ranged from 32 to 37 per cent. The sweet cream had an original acidity of 0.20 per cent or less, and after pasteurisation it was neutralised (at 120°F.) to approximately 0.10 per cent acidity with sodium sesqui-carbonate. The sour cream had an original acidity of 0.40 to 0.50 per cent. Before pasteurisation this was reduced (at 90°F.) to 0.25 per cent with "Alkali Special" and after pasteurisation (at 120°F.) to 0.15 per cent with sodium sesqui-carbonate.

After cooling the cream to 36°F., a portion was removed to a steamed milk can. Five per cent culture by weight was added and 800-gram portions of the mixture were transferred to sterile quart glass jars with glass tops. The portions were warmed to the desired ripening temperatures and incubated until the proper acidities had developed. To avoid agitating the 800-gram portions during the ripening period and thus influencing the production of diacetyl by incorporating air, a 250-gram portion of

the mixture in a pint bottle was held at each of the ripening temperatures; the acidity of this portion was determined occasionally and used as an indication of the acidity of the 600-gram portion.

On completion of the ripening, 300 grams of cream was removed from each 600-gram portion and the acidity, pH, diacetyl, and acetylmethy-carbinol were determined immediately. The remainder of the cream was churned to large granules in the jar (at the ripening temperature), using a special experimental churn that agitated six jars at a time. The butter-milk was drained and the butter washed twice in cool distilled water by shaking for approximately 1 minute each time. The butter was worked in a small sterile dish by hand until all free moisture disappeared. It was held in parchment wrappers at 36 to 40°F. until the pH, diacetyl, and acetylmethy-carbinol could be determined (within 24 hours).

Except in the trials comparing different butter cultures, the cultures employed in ripening the cream were prepared by inoculating skim milk, pasteurised at 180°F. for 30 minutes and then cooled, with a selected mother culture. The incubation temperature was 70°F. When firmly coagulated the cultures were cooled; they were used the same evening. No citric acid was employed in preparing the cultures.

#### Presentation of results

In presenting results of the preliminary churings, the various factors studied were generally divided into two or more divisions depending upon the number of conditions that were varied.

Tables on each division were prepared and placed in the Appendix. The tables contain the ripening temperature, per cent solidity, pH, diacetyl, and acetyl methylcarbinol values of the ripened cream just before churning, and in most cases contain the pH, diacetyl, and acetyl-methylcarbinol values of the corresponding butter. In a few tables additional information is included. The values on the butter were determined as soon as possible after the completion of the churning process (within 2½ hours). This was necessary to prevent changes from taking place.

Many of the tables are long; therefore, a summary for each table has been prepared and is used in giving the results. The summaries include only the results on the ripened cream, pH, diacetyl, and acetyl-methylcarbinol contents of the unsalted butter will be discussed later in the paper under a separate division.

Effect of the percentage of acid developed in cream on  
the diacetyl and acetyl methylcarbinol contents

Acidity is one of the important factors in the production of large amounts of diacetyl and acetyl methylcarbinol in butter cultures. However, in ripening cream for the manufacture of unsalted butter, it is not feasible to develop as high acidities as commonly are developed in butter cultures. The data on the production of diacetyl and acetyl methylcarbinol in cream ripened to different acidities involve both sweet and sour cream; trials also were carried out with skim milk.

To study the effect of acidity on the production of diacetyl and acetyl methylcarbinol during the ripening of cream or skim milk, a sample was prepared for each acidity desired. The general procedure consisted of preparing three or four samples and thus obtaining a number of acidities by ripening various periods. The acidities usually were within the limits of 0.30 and 0.55 per cent. Occasionally, the cream plus culture also was held in ice water to prevent changes by the culture organisms; presumably, the diacetyl and acetyl methylcarbinol contents then represented the amounts added by the butter culture and quantities already present from fermentations prior to pasteurization.

A. Sweet cream ripened to various acidities

Results of 36 trials on sweet cream ripened to various acidities are recorded in Summary 1. The diacetyl increased with the development of a higher acidity in the cream in 29 (80.5 per cent) of the trials, while the acetyl methylcarbinol increased in 32 (88.9 per cent) of the trials.

Summary 1. Comparative diacetyl ( $\text{Ac}_2$ ) and acetylmethylcarbinol (Amc) contents of sweet cream ripened to various acidities

From Table 1

	$\text{Ac}_2$	Amc
Number of trials showing an increase in $\text{Ac}_2$ or Amc with an increase in acidity	29	32
Number of trials showing a decrease in $\text{Ac}_2$ or Amc with an increase in acidity	2	2
Number of trials showing an increase and then a decrease in $\text{Ac}_2$ or Amc with an increase in acidity	3	1
Number of trials showing an increase, a decrease and then an increase in $\text{Ac}_2$ or Amc with an increase in acidity	2	1
TOTAL	36	36

of the 29 diacetyl increases, 20 were significant, while nine increased less than 0.1 p.p.m. in diacetyl with a substantial decrease in pH (0.10 of a unit from a pH of 5.00 to 6.00). Most of the acetylmethylcarbinol increases were significant considering the decrease in pH.

The data in Table I indicate that generally the diacetyl increase was small in the pH range of 6.5 to 5.5, while at a lower pH the increase in diacetyl was much greater per unit decrease in pH. Acetylmethylcarbinol yields displayed similar trends under the same conditions.

The diacetyl decreased in two (5.5 per cent) of the trials with an increase in acidity, the first analysis being made at approximately 0.30 per cent acidity. One of these diacetyl decreases was slight, from 0.57 to 0.44 p.p.m. The other decrease was greater and presumably caused by a poor culture. The acetylmethylcarbinol, likewise, decreased in two trials (5.5 per cent) with an increase in acidity, the decrease being large in both cases. One of the decreases was in the trial that showed a decrease in diacetyl.

An increase in diacetyl followed by a decrease occurred in three (8.3 per cent) of the trials. One of the decreases was small. The remaining two decreases occurred at high acidities, one being especially high. In the three trials corresponding changes in acetylmethylcarbinol did not occur, but in another trial (2.8 per cent) the carbinol content increased and then decreased slightly, from 29.7 to 29.0 p.p.m.; the decrease probably is within the limits of experimental error.

There was an increase in diacetyl, then a decrease and finally an-

other increase in two trials (5.5 per cent) and a similar series of changes in carbinol in one trial (2.8 per cent). The decreases were small with the final increase being much larger than the first in all cases.

The diacetyl contents of the six lots of cream plus butter culture held during the normal ripening period at 32°F. ranged from 0.12 to 0.28 p.p.m. with an average of 0.22 p.p.m.; the acetyl methylcarbinol contents varied from 3.5 to 16.5 p.p.m. and averaged 10.6 p.p.m. The ratio of diacetyl to acetyl methylcarbinol ranged from 1:15.2 to 1:116.6.

#### B. Sour cream ripened to various acidities

Summary 2 gives the results of three trials on neutralized sour cream ripened to various acidities. The diacetyl increased in three (100.0 per cent) of the trials with the development of a higher acidity. These increases regularly were large considering the decrease in pH. The acetyl-methylcarbinol contents also increased in the three trials with an increase in acidity. One of the increases was small but the other two were large.

The amount of diacetyl or acetyl methylcarbinol in the neutralized sour cream plus starter held at 32°F. for the normal ripening period was generally greater than in sweet cream plus starter, the average diacetyl content being 0.42 p.p.m. (range from 0.35 to 0.55 p.p.m.) compared to 0.22 p.p.m. for the sweet cream. The average carbinol content was 22.1 p.p.m. (range from 20.9 to 23.3 p.p.m.) compared to 10.6 p.p.m. for the sweet cream. This relationship probably is explained by the development

**Summary 2. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc)  
contents of neutralized sour cream ripened to various acidities**

**From Table 2**

	$\text{Ac}_2$	Amc
Number of trials showing an increase in $\text{Ac}_2$ or Amc with an increase in acidity	3	3
Number of trials showing a decrease in $\text{Ac}_2$ or Amc with an increase in acidity	0	0
<b>TOTAL</b>	<b>3</b>	<b>3</b>

of the two compounds during the original scouring. The ratio of diacetyl to carbinol was from 1:38.0 to 1:66.6.

C. Sweet skim milk ripened to various acidities

Results of four trials on skim milk ripened to various acidities are given in Summary 3. Both diacetyl and acetyl methylcarbinol increased with an increase in acidity in the four trials (100.0 per cent). Table 3 shows that in most cases the increases in the compounds were comparatively small despite large increases in the acidities. The results suggest that the presence of butterfat may cause larger amounts of the two compounds to be produced.

**Summary 3. Comparative diacetyl ( $\text{Ac}_2$ ) and acethylmethylcarbinol (Ame) contents of skin milk ripened to various acidities**

From Table 3

	$\text{Ac}_2$	Ame
Number of trials showing an increase in $\text{Ac}_2$ or Ame with an increase in acidity	4	4
Number of trials showing a decrease in $\text{Ac}_2$ or Ame with an increase in acidity	0	0
<b>TOTAL</b>	<b>4</b>	<b>4</b>

Effect of the addition of citric acid on diacetyl and  
acetylmethyloxbinol contents of ripened cream

Citric acid is the principal source of the flavor constituents of unsalted butter. Accordingly, trials were conducted to determine the effect of adding small amounts on diacetyl and acetylmethyloxbinol contents of ripened cream. Citric acid was added to the cream at the same time as the butter culture, in the form of a 50 per cent sterilized aqueous solution, with agitation of the cream to avoid coagulation.

Since citric acid effects a small decrease in pH, it was necessary to ripen the controls longer than the cream with added citric acid when approximately the same pH was desired. Trials were carried out with sweet and sour cream; in some of the trials with sweet cream the ripening period was the same with and without the added citric acid so that there was a difference in pH.

A. Sweet cream ripened with added citric acid

Since sweet cream contains some citric acid or citrate, only 0.05 per cent of the acid was added to the cream. The cream with and without added citric acid was ripened to the same pH.

Results of 14 trials with sweet cream are given in Summary 4. In all the trials the production of diacetyl was greater with citric acid added. The acetylmethyloxbinol contents were greater with citric acid added in 12 (85.7 per cent) of the 14 trials. Of the increases in diacetyl that resulted from the addition of citric acid, half were rather large despite the fact that sweet cream contains some citric acid. Approximately one-

Summary 4. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methylcarbinol (Amc) contents of sweet cream, with and without added citric acid, ripened to the same pH

From Table 4

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc with added citric acid	14	12
Number of trials not showing a greater production of $\text{Ac}_2$ or Amc with added citric acid	0	2
TOTAL	14	14

half of the increases in acetyl methyl carbinols also were large.

B. Sour cream ripened with added citric acid

The trials with neutralized sour cream involved the addition of three amounts of citric acid since sour cream may be deficient in this compound as a result of bacterial action in it. In six trials the amounts used were 0.05, 0.10, and 0.15 per cent; in two trials only 0.10 per cent was employed. Samples within each trial were ripened to the same pH.

The results of eight trials with sour cream are presented in Summary 5. Samples in seven of the eight trials contained diacetyl contents that were equal or larger with increased amounts of added citric acid. In one trial the diacetyl content of the sample with 0.05 per cent added citric acid was slightly less than that of the sample with no added citric acid. However, this may have been due to the fact that the pH of the former was higher, 5.20 compared to 5.00. The remaining two samples in the same trial increased in diacetyl in the usual way. The acetyl-methyl carbinol contents were increased by the additions of citric acid in all the trials. The diacetyl increases were large in most of the trials; the carbinol increases were large in all cases.

C. Sweet cream ripened with and without added citric acid for equal periods

The trials consisted of ripening sweet cream with and without 0.05 per cent added citric acid for equal periods of time. The results of 12

Summary 5. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of neutralized sour cream, with various amounts of added citric acid, ripened to the same pH

From Table 5

	$\text{Ac}_2$	Amc
Number of trials showing an equal or greater production of $\text{Ac}_2$ or Amc with added citric acid	7	8
Number of trials not showing an equal or greater production of $\text{Ac}_2$ or Amc with added citric acid	1	0
<b>TOTAL</b>	<b>8</b>	<b>8</b>

trials are presented in Summary 6. In all trials the addition of citric acid resulted in a greater production of diacetyl. The increases in diacetyl generally were not large. In all the trials the samples containing added citric acid had larger yields of acetyl methyl carbinol. The amount of increase was generally small. The difference in production of diacetyl and acetyl methyl carbinol between the cream with added citric acid and that without added citric acid was not entirely the result of citric acid serving as a source of flavor substances. Citric acid also tended to decrease the pH to some extent which should slightly favor diacetyl and acetyl methyl carbinol production. Comparative pH values on the samples of ripened cream with and without the addition of 0.05 per cent citric acid were as follows: 5.10, 5.17; 4.90, 5.05; 4.78, 4.87; 5.57, 5.80; 5.43, 5.61; 5.36, 5.42; 5.51, 5.62; 5.42, 5.58; 5.61, 5.72; 5.29, 5.35; 5.23, 5.36; and 5.34, 5.65. Probably all the decrease in pH was not directly caused by the addition of 0.05 per cent citric acid; part may have been the result of increased activity of culture organisms.

Summary 6. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of sweet cream, with and without added citric acid, ripened for equal periods

From Table 6

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc with added citric acid	12	12
Number of trials not showing a greater production of $\text{Ac}_2$ or Amc with added citric acid	0	0
TOTAL	12	12

#### Effect of agitation during ripening on diacetyl and acetylmethylocarbinol contents of cream

Biological oxidation probably is one of the reactions involved in the break-down of citric acid to diacetyl. Therefore, various procedures have been used in attempting to increase the oxygen content of butter cultures and thus obtain a greater production of diacetyl.

Agitation of cream to incorporate oxygen was accomplished by shaking the glass jars vigorously for 10 seconds at regular intervals. Cream ripened at 70°F. had a short ripening period and consequently was agitated every hour for 3 to 4 hours. Cream ripened at 60°F. was agitated three times during the development of acid. Trials were conducted with sweet and sour cream and with cream containing added citric acid.

#### A. Effect of agitating sweet or neutralized sour cream during the ripening period

Approximately the same pH was developed in cream ripened with agitation and in the corresponding cream ripened without agitation. Of the nine trials, seven were with sweet cream and two with sour cream.

The data are presented in Summary 7. Diacetyl and acetylmethylocarbinol contents of the agitated cream were much larger than the contents of the unagitated cream in nine trials (100.0 per cent). Diacetyl contents generally doubled or tripled, while the acetylmethylocarbinol contents were frequently twice as large, indicating that the incorporation of air by agitation is definitely beneficial for the production of diacetyl and acetylmethylocarbinol.

**Summary 7. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methylcarbinol (Amc)  
contents of cream ripened with and without agitation**

From Table 7

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in agitated cream	9	9
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in unagitated cream	0	0
<b>TOTAL</b>	<b>9</b>	<b>9</b>

B. Effect of agitating cream plus citric acid during the ripening period

Trials were conducted to determine if the agitation of cream, to which citric acid had been added, would cause the production of large amounts of diacetyl and acetylmethylecarbinol; citric acid would provide the substrate and agitation would increase the air content for the oxidation of acetylmethylecarbinol to diacetyl. The agitated samples and the corresponding unagitated samples were ripened to approximately the same pH.

Four were sweet cream and two were sour cream trials.

Summary 8 shows the results of the six trials. Diacetyl and acetylmethylecarbinol contents of the agitated cream with added citric acid were much larger in all trials than the unagitated cream with added citric acid. Tables 7 and 8 show that the combination of agitation and the addition of citric acid seems slightly to increase the yields of diacetyl and acetylmethylecarbinol, as compared to the agitation of cream to which citric acid was not added.

**Summary 8. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methylcarbinol (Amc) contents of cream containing 0.1 per cent added citric acid ripened with and without agitation**

From Table 8

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in agitated cream plus citric acid	6	6
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in unagitated cream plus citric acid	0	0
<b>TOTAL</b>	<b>6</b>	<b>6</b>

Effect of ripening temperatures on diacetyl  
and acethylmethylecarbinol contents of cream

Since temperature affects the growth of butter culture organisms trials were conducted to determine the effect of the cream ripening temperature on the production of diacetyl and acethylmethylecarbinol.

The ripening temperatures used were 50°, 60°, and 70°F. Cream with 5 per cent culture was warmed to these temperatures and ripened to constant temperature incubators to approximately the same pH. The ripening time varied from 5 to 45 hours, depending on the temperature and the degree of acidity desired. Determinations of the diacetyl and acethylmethylecarbinol in the cream were made soon after the desired pH was reached. Sweet cream was used in all the trials.

The results of 18 trials are given in Summary 9. In 14 trials (77.7 per cent) the largest amount of diacetyl was produced in the cream ripened at 50°F., the next largest in the cream ripened at 60°F., and the lowest in the cream ripened at 70°F. In 11 trials (61.1 per cent) the acethylmethylecarbinol contents ranked in the same order (50°, 60°, and 70°F.); included in this ranking are equal contents between the cream ripened at 70°F. and at 60°F. in two cases and between 50°F. and 60°F. in one case. In two trials (11.1 per cent) the production of diacetyl was largest in the cream ripened at 50°F., next largest in cream ripened at 70°F., and lowest in the cream ripened at 60°F. In two trials (11.1 per cent) the acethylmethylecarbinol yields varied in the same order (50°, 70°, and 60°F.). In one trial (5.5 per cent) the yield of diacetyl was greatest at a

Summary 9. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of sweet cream ripened at 50°, 60°, and 70°F.

From Table 9

	$\text{Ac}_2$	Amc
Number of trials showing the highest production of $\text{Ac}_2$ or Amc in cream ripened at 50°F., and the lowest production at 70°F.	14	11
Number of trials showing the highest production of $\text{Ac}_2$ or Amc in cream ripened at 50°F., and the lowest production at 60°F.	2	2
Number of trials showing the highest production of $\text{Ac}_2$ or Amc in cream ripened at 60°F., and the lowest production at 70°F.	1	5
Number of trials showing the highest production of $\text{Ac}_2$ or Amc in cream ripened at 70°F., and the lowest production at 60°F.	1	0
TOTAL	18	18

ripening temperature of 60°F., was next greatest at 50°F., and lowest at 70°F. In five trials (27.7 per cent) the acetyl methyl carbinol yields varied in the same order (60°, 50°, and 70°F.). One trial (5.5 per cent) showed the largest diacetyl content in cream ripened at 70°F., the next largest content at 50°F., and the lowest content at 60°F. None of the trials showed acetyl methyl carbinol contents according to the same order (70°, 50°, and 60°F.). In nearly all trials the difference in diacetyl and acetyl methyl carbinol contents among the three ripening temperatures was very small.

Influence of different butter cultures on diacetyl  
and acetyl methylcarbinol contents of cream

The amount of diacetyl and acetyl methylcarbinol in a butter culture varies greatly. Trials were conducted to determine the influence of various butter cultures on the diacetyl and acetyl methylcarbinol contents of ripened cream.

Butter cultures from several creameries making unsalted butter were carried until active (two transfers) and then used in preparing bulk cultures for the trials. After being used three times in the preparation of bulk cultures they were discarded to eliminate the possibility that transferring under the same conditions would tend to cause uniform results. Sweet cream was employed in all trials.

A. Cream ripened with different butter cultures from the same source

A dozen cultures were obtained from Nebraska creameries, which were originally from the same commercial source. Three trials were conducted, of which two contained eight samples and one contained six samples. The samples of each trial were ripened to the same approximate pH with a different culture.

The ranges in diacetyl and acetyl methylcarbinol contents are presented in Summary 10 and the other diacetyl and acetyl methylcarbinol values are given in Table 10. In Trial 1 the diacetyl contents of the eight samples ranged from 0.41 to 0.80 p.p.m.

Three other diacetyl contents in this trial were 0.72, 0.76, and 0.78 p.p.m. and the remaining contents were 0.52, 0.56, and 0.45 p.p.m.

**Summary 10. Range in diacetyl ( $\text{Ac}_2$ ) and acetyl methylcarbinol (Ame) contents of cream ripened with butter cultures from the same mother culture**

From Table 10

Trial No.	No. of samples	Range in $\text{Ac}_2$ p.p.m.	Range in Ame p.p.m.
1	8	0.41 to 0.80	19.4 to 34.0
2	8	0.46 to 0.92	13.2 to 44.8
3	6	0.50 to 0.76	12.1 to 24.1

The carbinol contents varied from 19.4 to 34.0 p.p.m.; contents of the six samples within these extremes were 29.1, 26.0, 20.6, 20.5, 20.3, and 20.1 p.p.m. In Trial 2 the diacetyl contents were from 0.46 to 0.92 p.p.m. and the other contents were 0.88, 0.78, 0.78, 0.74, 0.69, and 0.55 p.p.m. The lowest acetylmethylcarbinol content was 13.2 p.p.m. and the largest 44.8 p.p.m. Contents between these extremes were 33.8, 31.2, 20.3, 18.4, 10.2, and 34.8 p.p.m. In Trial 3 the production varied from 0.50 to 0.76 p.p.m. and the remaining four samples had contents of 0.53, 0.62, 0.63, and 0.67 p.p.m. Acetylmethylcarbinol contents of this trial ranged from 12.1 to 24.1 p.p.m. and the other four contents were 14.1, 15.8, 20.8, and 22.4 p.p.m. The difference in diacetyl content between the highest and lowest samples in Trial 1 and Trial 2 is rather large while the difference in Trial 3 is rather small. However, the diacetyl contents of several of the samples within the extremes in all three trials were nearly the same. The extremes in acetylmethylcarbinol contents were rather large in all three trials, but some of the differences among the samples of each trial were very small.

#### B. Cream ripened with different butter cultures

Several cultures were obtained from various Iowa creameries for the trials, which were originally from mother cultures of several commercial concerns. Sweet cream was used in all trials and samples of each trial were ripened to the same approximate pH. Two trials contained six samples and one trial contained five samples.

Summary 11 gives the extremes of the diacetyl and acetylmethylcarbinol

Summary 11. Range in diacetyl ( $\text{Ac}_2$ ) and acetylmethylcarbinol (Amc) contents of cream ripened with different commercial butter cultures

From Table 11

Trial No.	No. of samples	Range in $\text{Ac}_2$ p.p.m.	Range in Amc p.p.m.
1	5	0.47 to 0.86	5.7 to 26.1
2	6	1.04 to 2.17	17.8 to 60.4
3	6	0.91 to 1.46	11.5 to 20.0

contents of three trials, and the other diacetyl and acetyl methylcarbinol values are given in Table II. In Trial 1 the range was from 0.47 to 0.86 p.p.m. The other three samples of cream in the trials contained 0.67, 0.73, and 0.75 p.p.m. The acetyl methylcarbinol contents varied from 5.7 to 26.1 p.p.m., while the remaining values were 11.3, 18.0, and 25.9 p.p.m. In Trial 2 diacetyl yields varied from 1.04 to 2.17 p.p.m. and the other values were 1.10, 1.16, 1.35, and 1.88 p.p.m. In the same trial the acetyl methylcarbinol yields were between 17.8 and 60.4 p.p.m. with the remaining four yields being 26.1, 28.2, 37.3, and 41.4 p.p.m. Trial 3 contained samples that varied in diacetyl content from 0.91 to 1.46 p.p.m. The diacetyl contents of the other samples were 1.03, 1.04, 1.14, and 1.40 p.p.m. Acetyl methylcarbinol contents of the same trial ranged from 11.5 to 20.0 p.p.m. The four remaining contents were 12.4, 14.2, 16.0, and 16.3 p.p.m. The difference in both diacetyl and acetyl methylcarbinol production between the highest sample and the lowest sample of all three trials was large. The difference in production of diacetyl and acetyl methylcarbinol among samples within the extremes was frequently small.

#### C. Cream ripened with different butter cultures carried under the same conditions

The trial consisted of ripening eight samples of sweet cream with different butter cultures regularly carried by the Dairy Bacteriology Laboratories of Iowa State College. An attempt was made to ripen all samples to the same pH. As these cultures were carried under the same

conditions, a difference in diacetyl and acetyl methylcarbinol production should be due to differences in the cultures.

Summary 12 gives the range of diacetyl and acetyl methylcarbinol production in one trial, while the other diacetyl and acetyl methylcarbinol values are given in Table 12. The range in diacetyl content of the ripened cream was from 0.71 to 0.93 p.p.m. The contents of five other samples were from 0.74 to 0.76 p.p.m. and the remaining content was 0.87 p.p.m. of diacetyl. The acetyl methylcarbinol contents of the same samples varied from 26.4 to 40.0 p.p.m. The other contents were 28.8, 29.1, 30.6, 30.7, 31.4, and 34.7 p.p.m. The difference in diacetyl content between any two of the eight samples was small. The difference in acetyl methylcarbinol contents of the same samples was not very large.

Summary 12. Range in diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of cream ripened with different butter cultures carried under the same conditions

From Table 12

Trial No.	No. of samples	Range in $\text{Ac}_2$ p.p.m.	Range in Amc p.p.m.
1	8	0.71 to 0.93	26.4 to 40.0

Effect of various percentages of fat in cream or milk on  
diacetyl and acetylmethylcarbinol contents

The results of previous trials on diacetyl and acetylmethylcarbinol contents of skim milk reported in this paper suggested that butterfat influenced diacetyl and acetylmethylcarbinol production. Skim milk and cream with various amounts of butterfat were ripened to approximately the same pH and then the diacetyl and acetylmethylcarbinol contents determined. Trials with skim milk containing various amounts of added mineral oil or added butter oil also were conducted.

A. Comparison of diacetyl and acetylmethylcarbinol contents of skim milk and cream

Cream and skim milk were obtained at the same time during the separation of fresh, high quality milk. Butterfat content of the cream was approximately 40 per cent and that of the skim milk was 0.02 per cent. The samples plus 5 per cent butter culture were ripened to approximately the same pH.

Summary 13 shows the results of 12 trials. In 12 trials (100.0 per cent) the diacetyl and acetylmethylcarbinol contents of the cream were larger than the contents of the skim milk. The diacetyl content was about 25 to 50 per cent higher in the cream and acetylmethylcarbinol contents were nearly as large, indicating that butterfat has a beneficial effect on the production of these two compounds.

**Summary 13. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Ame) contents of skim milk and cream ripened to the same pH**

From Table 13

	$\text{Ac}_2$	Ame
Number of trials showing a greater production of $\text{Ac}_2$ or Ame in cream	12	12
Number of trials showing a greater production of $\text{Ac}_2$ or Ame in skim milk	0	0
<b>TOTAL</b>	<b>12</b>	<b>12</b>

B. Comparison of diacetyl and acetylmethylecarbinol contents of skin milk  
and cream with various percentages of butterfat

Fresh skin milk and cream were used to prepare samples with various percentages of butterfat. Two trials consisted of comparisons on skin milk, 5, 10, 20, 30, and 40 per cent butterfats in the cream. Five per cent butter culture was added on the basis of total weight. In six trials the sample with 5 per cent butterfat was omitted and also the culture was added at the rate of 5 per cent of the serum weight (fat free). The samples of each trial were ripened to approximately the same pH.

Results of eight trials are recorded in Summary 14. The diacetyl and acetylmethylecarbinol contents increased with increasing percentages of butterfat in the samples of seven of the eight trials. In Trial 5 the sample with 40 per cent butterfat failed to yield as much diacetyl and also acetylmethylecarbinol as the sample with 30 per cent. However, the four other samples in this trial contained larger quantities of diacetyl and acetylmethylecarbinol with increased butterfat content. In Trial 6 the sample with 10 per cent butterfat yielded less acetylmethylecarbinol than the sample with 0 per cent fat. Table 14 indicates that each 10 per cent increase in butterfat causes about the same amount of increase in diacetyl and acetylmethylecarbinol production; the increases of the two compounds were small and gradual with each 10 per cent fat increase.

Summary 14. Comparative diacetyl ( $\text{Ac}_2$ ) and acetymethylcarbinol (Amc) contents of skim milk and cream with various percentages of butterfat ripened to the same pH

From Table 14

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in cream with an increase in butterfat content	7	6
Number of trials not showing a greater production of $\text{Ac}_2$ or Amc in cream with an increase in butterfat content	1	2
TOTAL	8	8

C. Comparison of diacetyl and acetyl methylcarbinol contents of skim milk and cream with various butterfat contents ripened under partial vacuum or under atmospheric pressure

Samples consisting of skim milk, 20, and 40 per cent cream were prepared in duplicate from freshly separated skim milk and cream. Culture was added at the rate of 5 per cent of the total weight. One set of samples was ripened at 70°F. under atmospheric pressure; corresponding samples were ripened to the same pH under 27 inches of vacuum.

The results of two trials are recorded in Summary 15. In two trials (100.0 per cent) the diacetyl and acetyl methylcarbinol yields increased in samples containing increased butterfat contents. The samples were ripened under partial vacuum. In all trials the diacetyl and acetyl-methylcarbinol contents likewise increased in duplicate samples ripened under atmospheric pressure. The increase in diacetyl and acetyl methylcarbinol among the samples ripened under partial vacuum was very small, while the increase among the duplicate samples ripened under atmospheric pressure was large. The difference in diacetyl and acetyl methylcarbinol contents between the samples ripened under the two pressures generally was greater as the cream had a higher fat content.

D. Comparison of diacetyl and acetyl methylcarbinol contents of skim milk with various amounts of added mineral oil

Pure mineral oil was added to skim milk in various amounts to determine the effect on diacetyl and acetyl methylcarbinol production. Two trials were conducted, each with 0, 10, 20, 30, and 40 per cent additions

Summary 15. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methylcarbinol (Amc) contents of skim milk and cream with various percentages of butterfat ripened under partial vacuum and under atmospheric pressure to the same pH

From Table 15

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in cream with an increased butterfat content ripened under partial vacuum or under atmospheric pressure	2	2
Number of trials not showing a greater production of $\text{Ac}_2$ or Amc in cream with an increased butterfat content ripened under partial vacuum or under atmospheric pressure	0	0
TOTAL	2	2

of mineral oil. The samples of each trial were ripened to approximately the same pH.

Results obtained from the two trials are presented in Summary 16.

In two trials (100.0 per cent) the diacetyl content increased in the samples with increased additions of mineral oil. In the same trials the acetylmethylecarbinol contents likewise showed a similar trend. Diacetyl increases were small for each 10 per cent increase in mineral oil added to the skin milk. However, the diacetyl content was nearly twice as large in the sample with 40 per cent mineral oil as it was in the sample with no mineral oil. The difference in acetylmethylecarbinol contents under the same conditions was much smaller.

E. Comparison of diacetyl and acetylmethylecarbinol contents of skin milk with various amounts of added butter oil  
To secure additional information on the nature of the effect of butterfat on diacetyl and acetylmethylecarbinol production, butter oil at 100°F. was added to skin milk in various amounts. The butter oil was prepared by melting sweet cream butter at 120°F. and by filtering through a high grade filter paper. Skin milk with 0, 10, 20, 30, and 40 per cent additions of butter oil was employed in the trial. The butter oil tended to rise and solidify at the top of the mixture. Consequently, some difficulty was encountered in obtaining a uniform portion of the mixture after the samples were ripened over night at 60°F. to approximately the same pH.

The results of one trial are given in Summary 17. The diacetyl

Summary 16. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methylcarbinol (Amc) contents of skin milk with various amounts of added mineral oil ripened to the same pH

From Table 16

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in skin milk with an increase in added mineral oil	2	2
Number of trials not showing a greater production of $\text{Ac}_2$ or Amc in skin milk with an increase in added mineral oil	0	0
TOTAL	2	2

Summary 17. Comparative diacetyl ( $\text{Ac}_2$ ) and acetylethylene-carbinol (Aec) contents of skim milk with various amounts of added butter oil ripened to the same pH

From Table 17

	$\text{Ac}_2$	Aec
--	---------------	-----

Number of trials showing a greater production of  $\text{Ac}_2$  or Aec in skim milk with an increase in added butter oil

1

1

Number of trials not showing a greater production of  $\text{Ac}_2$  or Aec in skim milk with an increase in added butter oil

0

0

TOTAL

1

1

content increased in the samples containing increased additions of butter oil. Acetylactyloarbinol contents of the same samples showed similar results. The dioctyl increases were fairly large for each 10 per cent increase in the amount of butter oil added to the sample, but the acetyl-methyloarbinol increases were comparatively smaller.

Ratio of the diacetyl and acetyl methylcarbinol contents  
of ripened cream to those of the  
corresponding unsalted butter

Unsalted butter was manufactured from the ripened cream in many of the trials, and the results of the pH, diacetyl, and acetyl methylcarbinol determinations on the butter were recorded in Tables 1, 2, and 4, to 12, inclusive, along with the data on the cream. As an indication of the retention of diacetyl or acetyl methylcarbinol in the butter, the ratio of the diacetyl or acetyl methylcarbinol content of the cream to the diacetyl or acetyl methylcarbinol content of the corresponding butter was calculated.

In the 309 comparisons of cream and butter, the ratios of the diacetyl content of the ripened cream to the diacetyl content of the butter varied from 1:0.111 to 1:2.060, the average ratio being 1:0.725. In the same comparisons the ratios of the acetyl methylcarbinol content of the cream to the acetyl methylcarbinol content of the butter varied from 1:0.054 to 1:0.961, and the average ratio was 1:0.339.

The ratios of diacetyl and acetyl methylcarbinol contents in ripened cream and in the corresponding butter were grouped according to the procedure employed in ripening the cream from which the butter was manufactured. In 107 comparisons in the studies on the effect of acidity of the cream, the average diacetyl ratio was 1:0.857 and the average acetyl-methylcarbinol ratio was 1:0.366. In 73 comparisons in the trials with added citric acid, the average diacetyl ratio was 1:0.642 and the

acetyl methylcarbinol ratio was 1:0.302. In 30 comparisons in which the effect of agitation during ripening was studied, the average diacetyl ratio was 1:0.636; the average acetyl methylcarbinol ratio was 1:0.262. There were 53 comparisons in the trials on the effect of various ripening temperatures for cream; in these the average diacetyl ratio was 1:0.700 and the average acetyl methylcarbinol ratio 1:0.323. In 47 comparisons in which the influence of different butter cultures was investigated, the average diacetyl ratio was 1:0.634, and the average acetyl methylcarbinol ratio was 1:0.375.

Data in Tables 1, 2, and 4 to 12, inclusive, show that in many of the trials the ripened cream with the highest content of diacetyl did not necessarily have the highest content of acetyl methylcarbinol. However, there was a rather general tendency for samples with high diacetyl contents also to have high acetyl methylcarbinol contents. The same general relationship applied to the diacetyl and acetyl methylcarbinol contents of the corresponding unsalted butter. The relationship of diacetyl to acetyl methylcarbinol in the ripened cream did not seem to affect the relationship in the butter.

#### Part II. Plant Churnings

The results obtained in the laboratory studies were used as a basis for larger scale trials under plant conditions. Five of the factors that affected the development of diacetyl and acetyl-methylearbinol in the laboratory trials were investigated. These were as follows: (a) effect of the percentage of acid developed in cream on the diacetyl and acetyl-methylearbinol contents; (b) effect of the addition of citric acid on diacetyl and acetyl-methylearbinol contents of ripened cream; (c) effect of agitation during ripening on diacetyl and acetyl-methylearbinol contents of cream; (d) effect of ripening temperatures on diacetyl and acetyl-methylearbinol contents of cream; and (e) influence of different butter cultures on diacetyl and acetyl-methylearbinol contents of cream.

#### General procedure

The cream employed for the churning either was pasteurised at 150°F. for 30 minutes in a 300-gallon vat or was run through a Vacreator using a temperature of 198°F. and a vacuum of 28 inches of mercury. The butter-fat content of the cream varied from 32 to 37 per cent. The original vat acidities of the sweet cream were 0.20 per cent or lower and those of the sour cream were 0.40 to 0.50 per cent. The sweet cream was neutralised (at 120°F.) after pasteurisation to 0.10 per cent acidity with sodium sesqui-carbonate. The sour cream was neutralised (at 90°F.) before pasteurisation to 0.25 per cent acidity with "Alkali Special" and then after pasteurisation (at 120°F.) to 0.15 per cent acidity with sodium sesqui-carbonate. The cream was cooled to 36°F. and held at that

temperature until it was used in the trials.

For each churning 150 pounds of cream was drawn into 10-gallon cans and dumped into a small vat. Butter culture was added at the rate of 5 per cent of the weight of the cream. Usually the mixture of cream and culture was heated to the ripening temperature, which generally was between 51° and 55°F., and held over night, but in the trials on the effect of temperature the ripening period was variable. The cream commonly would rise 10°F. in temperature during the ripening period because of the small size of the vats. When the ripening process was complete the temperature was lowered for churning to approximately 48°F., using ice water as a cooling medium. A sample of cream was taken from each vat for immediate pH, diacetyl, and acetylethylcarbinol determinations. Each lot of cream was then transferred to a Cherry Junior single roll Model 2B churn. The churn was of 50 pound capacity and was known to yield results comparable to those obtained with a larger churn. The cream was churned until the butter granules were about the size of a pea. The buttermilk then was drained and the granules were rinsed until the water ran from the drain without a milky appearance. Two washings with water about 50°F. colder than the buttermilk were employed. The free moisture was drained and an attempt was made to incorporate approximately 17.0 per cent moisture in the finished butter. No salt was used.

Several 1-pound samples of butter were taken from each churning and wrapped in parchment paper. These samples were used as follows: (a) one sample was analyzed immediately for pH, diacetyl, and acetylethylcarbinol;

(b) one sample was held for 7 days at 36 to 40°F. before scoring for flavor and analyzing for pH, diacetyl, and acetylmethylecarbinol; (c) one sample was held 3 days at 60°F. followed by 4 days at 36 to 40°F. and then the pH, diacetyl, and acetylmethylecarbinol were determined; (d) one sample was held 1 month at 36 to 40°F., scored for flavor and the pH, diacetyl, and acetylmethylecarbinol were determined; and (e) one sample was frozen immediately and held at approximately -10 to 0°F. for 6 months before scoring.

The results were presented in the same general manner as those obtained in the preliminary churings. The manner was described under the head, Presentation of results.

Effect of the percentage of acid developed in cream on the  
diacetyl and acetylmethylcarbinol contents

In studying the effect of the percentage of acid developed in cream on the diacetyl and acetylmethylcarbinol contents, care was taken to maintain as many of the conditions alike in each trial as was possible. An attempt was made to develop acidities between 0.30 and 0.45 per cent (in a few cases the acidity was higher) and to have a difference of at least 0.05 per cent between the acidities of each vat of cream in a comparison. Occasionally the pH, diacetyl, and acetylmethylcarbinol contents were determined on the pasteurized cream both before and after the butter culture had been added. Each of the first three trials consisted of three churning at different acidities, and each of the remaining seven trials consisted of two churning at different acidities. Sweet cream was used in seven trials and neutralized sour cream in three trials.

Results of the 10 trials are presented in Summary 18. The production of diacetyl was greater in nine (90.0 per cent) of the trials as the acidity increased. The increase was small in four of these trials, being 0.1 p.p.m. or less for 0.1 or more units decrease in pH. In one trial (10.0 per cent) the diacetyl content decreased as the acidity increased. Table 18 shows the decrease was from 1.88 to 1.49 p.p.m. with acidities of 0.37 and 0.49 per cent, respectively. In seven trials (70.0 per cent) the acetylmethylcarbinol contents increased as the acidity developed. One increase was small, from 11.8 to 11.9 p.p.m. In three trials (30.0

Summary 18. Comparative diacetyl ( $\text{Ac}_2$ ) and acetymethylcarbinol (Amc)  
contents of cream ripened to various acidities

From Table 18

	$\text{Ac}_2$	Amc
Number of trials showing an increase in $\text{Ac}_2$ and Amc with an increase in acidity	9	7
Number of trials showing a decrease in $\text{Ac}_2$ and Amc with an increase in acidity	1	3
TOTAL	10	10

per cent) there were rather large decreases in acetyl methyl carbinal contents as the acidity increased. Two of these trials involved sour cream with which there was a possibility of a depletion of citric acid and, consequently, a destruction of diacetyl and acetyl methyl carbinal by the citric acid fermenting organisms. This is substantiated by the fact that in these two trials the cream before the addition of culture had relatively high diacetyl and acetyl methyl carbinal contents; in one trial the cream showed a diacetyl content of 0.21 p.p.m. and an acetyl methyl carbinal content of 21.7 p.p.m. and in the other trial the values were 0.56 p.p.m. and 29.8 p.p.m., respectively. In producing high diacetyl and acetyl methyl carbinal contents during natural souring much of the citric acid content of the cream was probably utilized.

**Effect of the addition of citric acid on diacetyl and acetylmethylcarbinol contents of ripened cream**

In studying the effect of added citric acid on diacetyl and acetyl-methylcarbinol contents of cream, samples of cream were ripened with and without 0.05 per cent added citric acid. The desired amount of citric acid, as a 50 per cent aqueous solution, was diluted with sterile water and added to the cream along with the 5 per cent culture. The cream was agitated with the coils of the vat during the addition of the citric acid. Sweet cream was employed in four trials and neutralised sour cream in three trials.

Summary 19 gives the results of the seven trials. Diacetyl production was increased by the addition of citric acid in six of the seven trials. The increases in diacetyl varied from 0.12 to 2.12 p.p.m., but three increases were 0.20 p.p.m. or less. The diacetyl content was decreased 0.13 p.p.m. by the addition of citric acid in one trial (14.3 per cent). In three of the seven trials the acetylmethylcarbinol contents were increased by the addition of citric acid; the increases varied from 12.8 to 34.8 p.p.m. In one trial (14.3 per cent) the acetylmethylcarbinol values were the same in the cream with and without added citric acid and in three trials (42.8 per cent) the acetylmethylcarbinol contents in the cream with added citric acid were lower. The decrease in acetylmethylcarbinol ranged from 4.0 to 15.8 p.p.m.

Summary 19. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of cream, with and without added citric acid, ripened to the same pH

From Table 19

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc with added citric acid	6	3
Number of trials not showing a greater production of $\text{Ac}_2$ or Amc with added citric acid	1	4
TOTAL	7	7

Effect of agitation during ripening on diacetyl and  
acetyl methylcarbinol contents of cream

To study the effect of agitating cream during ripening on the production of diacetyl and acetyl methylcarbinol, cream plus butter culture was placed in vats late in the afternoon and ripened over night. The next morning approximately 3 hours before the ripening was complete the cream was agitated by revolving the vat coils for one-half hour. The vats were only one-third full; thus the agitation was more vigorous than if the vats had been full. The controls were not agitated but the other conditions were maintained as nearly alike as possible, including the pH. There were five trials of which three were with sweet cream and two with neutralized sour cream.

Summary 20 gives the results. The diacetyl yields were increased in all five trials by agitating the cream during ripening. The increases in the yields were large, frequently being 25 to 100 per cent. The acetyl-methylcarbinol yields likewise were greatly increased by agitation in all five trials. The agitated samples commonly contained twice as much acetyl-methylcarbinol as the unagitated samples.

**Summary 20. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of cream ripened with and without agitation**

From Table 20

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in agitated cream	5	5
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in unagitated cream	0	0
<b>TOTAL</b>	<b>5</b>	<b>5</b>

Effect of ripening temperatures on diacetyl and  
acetyl methyl carbinol contents of cream

Trials were conducted to study the diacetyl and acetyl methyl carbinol contents of cream ripened at various temperatures. Since it was impossible to control adequately the ripening temperature in the small vats, cream plus butter culture was warmed to the desired ripening temperature and then drawn into ten-gallon cans. The cans were placed in coolers, which were refrigerated to about the same temperature as that used for ripening and held until the desired pH was attained. A uniform temperature of 55°F. required too long a ripening period under the conditions used and was discontinued after one attempt. The comparisons consisted of cream ripened at 70°F. and at 61°F.; it was attempted to ripen the cream in each trial to the same pH. On reaching the desired pH, cream ripened at 70°F. was cooled by placing the cans in ice water and stirring. The temperature of the cream was lowered to 45°F. and then the cream was held over night in a cooler at 36 to 40°F. The next morning after warming the cream to 48°F. it was churned. The cream ripened at 61°F. reached the desired pH after ripening over night. The cans were placed in ice water, cooled to 48°F. and the churning process carried out as usual. Four trials were conducted of which three were with sweet cream and one with neutralized sour cream.

In three trials a special vat of cream was ripened along with the cream used for the temperature comparisons. The treatment of the special vat of cream consisted of adding 0.1 per cent citric acid and agitating

during the ripening over night to about 0.45 per cent acidity.

The results of the temperature comparisons are given in Summary 21. The diacetyl production in the cream ripened at 61°F. was larger in two trials (50.0 per cent), while in the other two trials it was larger in the cream ripened at 70°F. The difference generally was very slight in the comparisons. The acetyl methylcarbinol production showed similar results. In two trials (50.0 per cent) the contents were larger in the cream ripened at 61°F. and in two they were larger in the cream ripened at 70°F. The diacetyl and acetyl methylcarbinol contents of the cream from the special vats were much larger in all three trials than the diacetyl and acetyl methylcarbinol contents of the cream ripened either at 70°F. or at 61°F.

**Summary 21. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of cream ripened at 61°F. and at 70°F.**

From Table 21

	$\text{Ac}_2$	Amc
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in cream ripened at 70°F.	2	2
Number of trials showing a greater production of $\text{Ac}_2$ or Amc in cream ripened at 61°F.	2	2
<b>TOTAL</b>	<b>4</b>	<b>4</b>

### Influence of different butter cultures on diacetyl and acetyl methyl carbinol contents of cream

To study the influence of different butter cultures on diacetyl and acetyl methyl carbinol production, cultures were obtained from several creameries making commercial unsalted butter in Iowa. Originally the cultures came from various commercial sources. The cultures were all received the same day and transferred twice before using to inoculate bulk culture for cream ripening. The cultures were discarded after three successive transfers into milk for bulk cultures to eliminate a possibility of uniform results that might be caused by carrying under the same conditions. Each trial involved a comparison of three cultures.

The general conditions of ripening were kept as nearly alike as possible with the exception of the length of the ripening period. It was necessary to vary the ripening period in order to obtain approximately the same pH in the different vats of cream since the cultures varied in their ability to produce acid. Three of the trials were made with sweet cream and two with neutralized sour cream.

Summary 22 shows the range in diacetyl and acetyl methyl carbinol production among the three vats of cream in each of the five trials; the other diacetyl and acetyl methyl carbinol values are given in Table 22. Trial 2 showed the greatest difference in diacetyl contents among the three samples; it was 1.50 p.p.m. (3.40 to 4.90 p.p.m.) and is rather large. Trial 5 showed the smallest difference in diacetyl production among the three samples; it was only 0.04 p.p.m. (2.36 to 2.40 p.p.m.).

**Summary 22.** Range in diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc)  
contents of cream ripened with different butter cultures

From Table 22

Trial No.	No. of samples	Range in $\text{Ac}_2$ p.p.m.	Range in Amc p.p.m.
1	3	1.68 to 2.09	39.3 to 41.9
2	3	3.40 to 4.90	86.4 to 122.2
3	3	1.07 to 1.43	22.0 to 28.6
4	3	1.30 to 1.49	9.2 to 24.6
5	3	2.36 to 2.40	41.6 to 72.6

The remaining trials showed differences of 0.41, 0.36, and 0.19 p.p.m. between the highest and lowest values. Trial 2 also showed the largest difference in acetyl methylcarbinol contents among the three samples; it was 35.8 p.p.m. (86.4 to 122.2). The smallest difference in acetyl-methylcarbinol contents was 2.6 p.p.m. (39.3 to 41.9 p.p.m.) and occurred in Trial 1. In the other trials the differences in acetyl methylcarbinol contents were 6.6, 15.4, and 31.0 p.p.m.

Ratio of the diaetyl and acetylmethylecarbinol contents  
of ripened cream to those of the  
corresponding unsalted butter

The cream studied in the trials under plant conditions was manufactured into unsalted butter. The results of the pH, diaetyl, and acetylmethylecarbinol determinations on the butter are recorded in Tables 18 to 22, inclusive, together with the data on the cream. The retention of diaetyl or acetylmethylecarbinol in the butter is indicated by the figures representing the ratio of the content of the ripened cream to the content of the corresponding butter.

In the 74 comparisons of cream and butter, the ratios of the diaetyl contents of the cream to the contents of the butter varied from 1:0.142 to 1:0.708; the average ratio was 1:0.351. The ratios of the acetylmethylecarbinol contents in the same comparisons ranged from 1:0.059 to 1:0.679, the average ratio being 1:0.216.

The ratios of diaetyl and acetylmethylecarbinol contents in ripened cream and in the corresponding butter were grouped according to the procedure used in ripening the cream. In the studies on the effect of acidity of the cream, 23 comparisons of cream and butter were involved; the average of the ratios of the diaetyl contents of the cream to the contents of the butter was 1:0.389; the average acetylmethylecarbinol ratio was 1:0.215. In the trials on the effect of adding citric acid to the cream, there were 14 comparisons. The average diaetyl ratio was 1:0.327, and the average acetylmethylecarbinol ratio was 1:0.214. Ten comparisons were made in the

trials on the effect of agitation of cream. The average diacetyl ratio was 1:0.300 and the average acetymethylcarbinol ratio was 1:0.178. In trials on the effect of ripening temperature of cream the 12 comparisons showed an average diacetyl ratio of 1:0.387 and an average acetymethylcarbinol ratio of 1:0.243. In trials involving the influence of butter cultures, 15 comparisons had an average diacetyl ratio of 1:0.321; the average acetymethylcarbinol ratio was 1:0.227.

Tables 18 to 22, inclusive, show that there was a rather general relationship between the diacetyl and the acetymethylcarbinol contents of the ripened cream. However, in a few trials the samples with high diacetyl contents did not have high acetymethylcarbinol contents and vice versa. In the butter the relationship also was very general. The relationship of the diacetyl and acetymethylcarbinol contents in the ripened cream did not seem to influence the relationship in the corresponding butter.

Changes in diacetyl and acetyl methylcarbinol contents

during holding of unsalted butter

After churning the cream ripened with the various procedures, samples of the unsalted butter were obtained for studies on the changes in diacetyl and acetyl methylcarbinol contents during holding at different temperatures for various periods.

A. Changes in diacetyl and acetyl methylcarbinol contents of butter during holding for 1 week at 36 to 40°F.

Summary 23a gives the results on the butter in 56 trials; more detailed data are in Table 23. In 47 trials (83.9 per cent) there were increases in the diacetyl contents of the butter during 1 week at 36 to 40°F. The increases varied from 0.02 to 0.70 p.p.m. In 11 trials the increases were between 0.02 and 0.10 p.p.m.; in 9 between 0.11 and 0.20 p.p.m.; in 17 between 0.21 and 0.30 p.p.m.; and in 10 above 0.30 p.p.m. In two trials (3.5 per cent) the diacetyl contents were the same before and after holding. In seven trials (12.5 per cent) the diacetyl contents decreased during the holding, the decreases being small in several cases. One decrease was 0.01 p.p.m.; three were between 0.10 and 0.11 p.p.m.; and three, involving the same butter culture, were 0.55, 0.68, and 0.47 p.p.m. In 38 trials (67.8 per cent) the acetyl methylcarbinol contents increased during 1 week at 36 to 40°F. The range of increases was from 0.1 to 7.3 p.p.m. In seven trials the acetyl methylcarbinol contents increased less than 1.0 p.p.m.; in 21 between 1.1 and 3.0 p.p.m.; and in 10 between 3.1 and 7.3 p.p.m. In 18 trials (32.1 per cent) there were decreases in

Summary 23a. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of unsalted butter before and after holding 1 week at 36 to 40°F.

From Table 23

	$\text{Ac}_2$	Amc
Number of trials with a higher $\text{Ac}_2$ or Amc content in unsalted butter held 1 week at 36 to 40°F.	47	38
Number of trials with a higher $\text{Ac}_2$ or Amc content in unsalted butter before holding	7	18
Number of trials with no difference in $\text{Ac}_2$ or Amc content	2	0
TOTAL	56	56

acetylmethylcarbinol contents ranging from 0.3 to 4.7 p.p.m. during the holding. Five of the decreases were 1.0 p.p.m. or less; 11 were 1.1 to 3.0 p.p.m.; and 2 were larger than 3.0 p.p.m. In five of the seven trials in which there were decreases in diacetyl contents during the holding for 1 week at 36 to 40°F. there also were decreases in acetyl-methylcarbinol. In the other two trials the samples showing decreases in diacetyl gave very small increases in acetylmethylcarbinol, 0.9 and 0.1 p.p.m. Neither the addition of citric acid nor agitation of cream during ripening consistently prevented decreases in diacetyl and acetyl-methyl-carbinol in butter made from the cream.

B. Comparative changes in diacetyl and acetylmethylcarbinol contents of butter during holding for 1 week at 36 to 40°F. and for 3 days at 60°F. followed by 4 days at 36 to 40°F.

Summary 24b gives the results; more detailed data are given in Table 23. The diacetyl contents in 42 trials (75.0 per cent) were higher in butter held for 3 days at 60°F. and then 4 days at 36 to 40°F. than in the butter held 1 week at 36 to 40°F. The differences ranged from 0.01 to 0.69 p.p.m. In 19 trials the differences were between 0.01 and 0.10 p.p.m.; in ten between 0.11 and 0.20 p.p.m.; in eight between 0.21 and 0.30 p.p.m.; and in five above 0.30 p.p.m. In 13 trials (23.2 per cent) the butter had lower diacetyl contents after 3 days at 60°F. and 4 days at 36 to 40°F. than after 1 week at 36 to 40°F. The differences varied from 0.01 to 0.64 p.p.m. In seven trials the differences were 0.1 p.p.m. or less; in three between 0.11 and 0.20; and in three above 0.30 p.p.m.

Summary 23b. Comparative diacetyl ( $\text{Ac}_2$ ) and acetyl methyl carbinol (Amc) contents of unsalted butter held 1 week at 36 to 40°F. and held 3 days at 60°F. then 4 days at 36 to 40°F.

From Table 23

	$\text{Ac}_2$	Amc
Number of trials with a higher $\text{Ac}_2$ or Amc content in unsalted butter held 1 week at 36 to 40°F.	12	11
Number of trials with a higher $\text{Ac}_2$ or Amc content in unsalted butter held 3 days at 60°F. and then 4 days at 36 to 40°F.	43	40
Number of trials with no difference in $\text{Ac}_2$ or Amc content	1	4
TOTAL	56	55

In one trial (1.8 per cent) the diacetyl contents were the same after both holdings. The acetylmethylecarbinol contents in 40 trials (72.7 per cent) were higher in the butter held 3 days at 60°F. and then 4 days at 36 to 40°F. The differences were from 0.2 to 18.2 p.p.m. In 8 trials the differences were from 0.2 to 1.0 p.p.m.; in 12 trials from 1.1 to 3.0 p.p.m.; and in 20 trials from 3.0 to 18.2 p.p.m. In four trials (7.3 per cent) the acetylmethylecarbinol contents were the same with the different holding conditions.

In 11 trials (20.0 per cent) the acetylmethylecarbinol contents were lower in the butter held 3 days at 60°F. and 4 days at 36 to 40°F. The differences ranged from 1.6 to 12.8 p.p.m. Four of the differences were between 1.6 and 3.0 p.p.m.; three between 3.1 and 6.0 p.p.m.; and four between 6.1 and 12.8 p.p.m.

C. Changes in diacetyl and acetylmethylecarbinol contents of butter during holding for 1 month at 36 to 40°F.

Summary 230 presents the results and Table 23 gives more detailed data. Diacetyl contents in 39 trials (90.7 per cent) showed increases during 1 month at 36 to 40°F. The increases varied from 0.02 to 0.88 p.p.m. Six increases were between 0.02 and 0.10 p.p.m.; 13 between 0.11 and 0.30 p.p.m.; and the remaining 20 between 0.31 and 0.88 p.p.m. In four trials (9.3 per cent) the diacetyl contents decreased during holding. The decreases were 0.03, 0.11, 0.16, and 0.27 p.p.m. The acetylmethylcarbinol contents in 29 trials (65.9 per cent) showed increases during 1 month at 36 to 40°F. The increases varied from 0.1 to 10.6 p.p.m. Ten

Summary 23c. Comparative diacetyl ( $\text{Ac}_2$ ) and acetymethylecarbinol (Ame) contents of unsalted butter before and after holding 1 month at 36 to 40°F.

From Table 23

	$\text{Ac}_2$	Ame
Number of trials with a higher $\text{Ac}_2$ or Ame content in butter held 1 month at 36 to 40°F.	39	29
Number of trials with a higher $\text{Ac}_2$ or Ame content in unsalted butter before holding	4	14
Number of trials with no difference in $\text{Ac}_2$ or Ame content	0	1
TOTAL	43	44

increases ranged from 0.1 to 2.0 p.p.m.; 11 from 2.1 to 4.0 p.p.m.; and 8 from 4.1 to 10.6 p.p.m. In 14 trials (31.8 per cent) the butter decreased in acethylmethylcarbinol content during the holding. The decreases were from 0.2 to 6.0 p.p.m. In nine trials the increases were between 0.1 and 2.0 p.p.m.; in two between 2.1 and 4.0 p.p.m.; and in three between 4.1 and 6.0 p.p.m.

Data in Table 23 show that 72.0 per cent of the samples held 1 month at 36 to 40°F. had slightly higher diacetyl contents than corresponding samples held 1 week at 36 to 40°F. About 50 per cent of the acethylmethylcarbinol contents were higher in the butter held 1 month.

#### D. pH values of ripened cream and of the corresponding butter

pH values of the ripened cream and of the corresponding butter are given in Table 23. The pH of the butter immediately after churning generally was higher than the pH of the ripened cream from which it was churned. In 55 commercial trials 51 lots (90.9 per cent) of unsalted butter were higher in pH than the cream. The difference in pH was not uniform, varying from a few hundredths to several tenths of a unit. Four lots (7.3 per cent) of butter were lower in pH than the ripened cream, but the differences were very small, 0.04 of a unit or less. One lot (1.8 per cent) showed the same pH in the cream and in the butter. The decreases in pH of the butter during holding varied according to the temperature and time of holding, and with any of the three holding periods studied (1 week at 36 to 40°F., 1 month at 36 to 40°F., and 3 days at 60°F. and then 4 days at 36 to 40°F.) the final pH of the butter seemed to

correlate more closely with the pH of the fresh butter than with the pH of the ripened cream from which it was churned.

Effect of various procedures during ripening of cream  
on the flavor of unsalted butter

After churning the cream that was ripened with various procedures, samples of the butter were taken to determine the effect of these procedures on the flavor in the butter that was held at various temperatures for different periods. A sample of butter from each churning was scored for flavor after holding 1 week at 36 to 40°F., 1 month at the same temperature and 6 months in cold storage at -10 to 0°F. One or two experienced butter judges scored the unsalted butter without knowledge of its identity. It should be noted that the scoring of unsalted butter is not as well standardised as is the scoring of salted butter. The judges scored the unsalted butter to 0.25 of a point.

A. Unsalted butter from cream ripened to various acidities

Summary 244 shows the results on the butter in seven trials. When the butter was held 1 week at 36 to 40°F. that from the cream ripened to the higher acidity scored from 0.25 to 0.75 of a point higher than that from the cream ripened to the lower acidity in five (71.4 per cent) of the seven trials. There was no difference in score in one trial (14.3 per cent); the difference in the acidities of the two lots of cream was only 0.04 per cent (0.31 and 0.35 per cent). Both lots of butter were criticized as tallowy and this off-flavor probably tended to mask the desirable flavor. In one trial (14.3 per cent) both lots of butter were from cream relatively high in acidity and the butter from the cream with the higher acidity scored 0.25 of a point lower than that from the cream

Summary 2<sup>1/2</sup>a. Comparative scores on the flavor of unsalted butter  
from cream ripened to two acidities

From Table 2<sup>1/2</sup>

	Butter held 1 week at 36 to 40°F.	Butter held 1 month at 36 to 40°F.	Butter held 6 months at -10 to 0°F.
Number of higher scores in unsalted butter from cream that was ripened to the higher acidity	5	3	7
Number of higher scores in unsalted butter from cream that was not ripened to the higher acidity	1	2	0
Number of scores with no difference	1	2	0
TOTAL	7	7	7

with the lower acidity. The acidity spread in this trial was not large, from 0.48 to 0.51 per cent, and was the smallest in any of the trials. In the two trials in which the lots of cream had the largest differences in acidity, 0.08 per cent (0.38 to 0.46 per cent) and 0.12 per cent (0.37 to 0.49 per cent) the differences in the scores of the butter were the largest, being 0.75 of a point.

Butter from the same churning also was held 1 month at 36 to 40°F. In three trials (42.9 per cent) butter from the higher acid cream gave the higher score by 0.25 to 0.50 of a point. In two trials (28.6 per cent) butter from both acidities scored the same and in two trials (28.6 per cent) the butter from the cream ripened to the higher acidity scored 0.50 of a point lower.

Comparisons between butter held 1 week at 36 to 40°F. and 1 month at 36 to 40°F. from the same churning showed that the samples after 1 month had generally decreased in score from 0.25 to 1.50 points.

The butter from the same churning also was held approximately 6 months at -10 to 0°F. before scoring. Butter from cream ripened to higher acidity scored from 0.25 to 0.75 of a point more in all trials, indicating that the butter from the higher acid cream kept equally well or better than the butter from cream with the lower acidity when held frozen for 6 months.

The butter that was held frozen generally scored from 0.75 to 2.00 points less than the same butter held 1 week at 36 to 40°F., indicating a definite deterioration in flavor.

B. Unsalted butter from cream ripened with added citric acid

Summary 24b presents the results of seven trials on butter. After holding 1 week at 36 to 40°F. butter from cream to which citric acid had been added scored from 0.25 to 0.50 of a point higher than the butter from cream without added citric acid in four trials (57.1 per cent). In three trials (42.9 per cent) both samples of butter scored the same; in two of these trials sour cream was used and the desirable flavor was masked by tallowy and metallic off-flavors. In the other trial the flavor was slightly better in the butter from cream plus citric acid, but not enough to be given a higher score.

The butter was held 1 month at 36 to 40°F. and in six of the seven trials butter from cream plus citric acid scored 0.25 to 0.75 of a point more. In one trial (14.3 per cent) the butter scored the same.

Butter held 1 month usually decreased in score from 0.25 to 2.00 points compared to its score after holding 1 week at 36 to 40°F.

Butter in the trials held frozen (-10 to 0°F.) for 6 months, resulted in six of the seven samples of butter from cream having added citric acid showing higher scores than the butter from cream without added citric acid; the increase in score was from 0.25 to 0.75 of a point. In one trial (14.3 per cent) the score was 0.50 of a point lower in butter from cream with added citric acid.

Most of the butter from cream with added citric acid scored 0.50 to 2.25 points less after 6 months than after holding 1 week at 36 to 40°F.

Summary 24b. Comparative scores on the flavor of unsalted butter from cream ripened with and without 0.05 per cent added citric acid

From Table 24

	Butter held 1 week at 36 to 40°F.	Butter held 1 month at 36 to 40°F.	Butter held 6 months at -10 to 0°F.
Number of higher scores in unsalted butter from cream ripened with added citric acid	4	6	6
Number of higher scores in unsalted butter from cream ripened without added citric acid	0	0	1
Number of scores with no difference	3	1	0
TOTAL	7	7	7

C. Unsalted butter from cream ripened with agitation

Summary 21c presents the results on the butter in five trials. The butter was scored after holding 1 week at 36 to 40°F. The butter from cream that was agitated during ripening scored from 0.25 to 0.75 of a point higher in three of the five trials than the butter from unagitated cream. In the remaining two trials (40.0 per cent) the butter from both lots of cream scored the same; this was probably caused by pronounced off-flavors in the butter.

In comparisons held for 1 month at 36 to 40°F. the butter from the agitated cream showed higher scores in two of the five trials. The increases in score were from 0.50 to 0.75 of a point. In one trial (20.0 per cent) butter from cream that was agitated had the lower score by 0.25 of a point. In two trials (40.0 per cent) butter from both lots of cream scored the same. There seemed to be a tendency for some of the butter from agitated cream to have a slight oxidized flavor and thus to score lower in flavor.

The butter held 1 month at 36 to 40°F. generally deteriorated from 0.25 to 1.25 points in score, compared to the same butter scored after 1 week at 36 to 40°F.

The butter in five trials held 6 months at -10 to 0°F. resulted in that from the agitated cream scoring higher in four trials (60.0 per cent). Three of the scores were one point higher and the other score was 0.25 higher. In one trial (20.0 per cent) butter from both lots of cream showed equal scores.

**Summary 24. Comparative scores on the flavor of unsalted butter  
from cream ripened with and without agitation**

From Table 24

	Butter held 1 week at 36 to 40°F.	Butter held 1 month at 36 to 40°F.	Butter held 6 months at -10 to 0°F.
Number of higher scores in un-salted butter from cream ripened with agitation	3	2	4
Number of higher scores in un-salted butter from cream ripened without agitation	0	1	0
Number of scores with no difference	2	2	1
TOTAL	5	5	5

The butter from agitated cream declined from 0.25 to 2.00 points, compared to their scores after holding 1 week at 36 to 40°F.

D. Unsalted butter from cream ripened at various temperatures

Data on these trials are presented in Summary 24d. Butter in the four trials was held 1 week at 36 to 40°F. In two trials (50.0 per cent) the butter from cream ripened at 61°F. scored 0.25 and 0.75 of a point higher than butter from cream ripened at 70°F. In one trial (25.0 per cent) the butter from cream ripened at 70°F. scored 0.50 of a point higher and in one trial (25.0 per cent) butter from both lots of cream scored the same, although the flavor was very slightly better in the butter from cream ripened at 61°F.

Only three comparisons were conducted by holding the butter 1 month at 36 to 40°F. and in all three trials the score of the butter from the cream ripened at 61°F. was 0.50 to 0.75 of a point higher than butter from cream ripened at 70°F.

Most of the butter deteriorated 0.25 to 0.75 of a point in score during holding for 1 month at 36 to 40°F.

Butter in four trials was held frozen (-10 to 0°F.) for 6 months before scoring. In two trials (50.0 per cent) the butter from the cream ripened at 60°F. scored 0.50 higher and also in two trials (50.0 per cent) the butter from cream ripened at 70°F. scored 0.75 and 1.00 point higher.

The samples of butter deteriorated from 1.25 to 2.50 in score during the holding period of 6 months at -10 to 0°F. compared to their score at the end of holding 1 week at 36 to 40°F.

Summary 24d. Comparative scores on the flavor of unsalted butter  
from cream ripened at two temperatures

From Table 24

	Butter held 1 week at 36 to 40°F.	Butter held 1 month at 36 to 40°F.	Butter held 6 months at -10 to 0°F.
Number of higher scores in un- salted butter from cream ripened at 70°F.	1	0	2
Number of higher scores in un- salted butter from cream ripened at 61°F.	2	3	2
Number of scores with no difference	1	0	0
TOTAL	4	3	4

E. Unsalted butter from cream ripened with various butter cultures

Summary 24e presents the results of the trials. After holding 1 week at 36 to 40°F. the three samples of butter from cream ripened with different butter cultures did not score the same in any of the five trials. The difference in score was at least 0.25 of a point. In one of the five trials two of the samples scored the same and the third was three-fourths of a point lower. In the remaining four trials (80.0 per cent) the three samples all scored differently, ranging in difference from 0.25 to 0.75 of a point.

In the only trial the butter held 1 month at 36 to 40°F. showed two samples with the same score and the third sample with a score of 0.25 of a point less.

The butter in five trials was held for 6 months at -10 to 0°F. In one trial (20.0 per cent) the three samples of butter from cream ripened with different butter cultures scored the same; this may be explained by the development of a pronounced stale flavor. In four trials (80.0 per cent) the three samples of butter were each given a different score which did not vary more than one point in any of the four trials.

Nearly all samples of butter held 6 months at -10 to 0°F. decreased in score compared to their scores after 1 week at 36 to 40°F. The decrease in score varied from 0.50 to 2.25 points.

Summary 24e. Comparative scores on the flavor of unsalted butter  
from cream ripened with different butter cultures

From Table 24

	Butter held 1 week at 36 to 40°F.	Butter held 1 month at 36 to 40°F.	Butter held 6 months at -10 to 0°F.
Number of trials in which the three samples of unsalted butter from cream ripened with different cultures scored the same	0	0	1
Number of trials in which two of the three samples of unsalted butter from cream ripened with different cultures scored the same	1	1	0
Number of trials in which none of the three samples of un- salted butter from cream ripened with different cultures scored the same	4	0	4
TOTAL	5	1	5

#### DISCUSSION OF RESULTS

As with many other biological changes, the results obtained in the various trials showed pronounced tendencies but were not entirely consistent.

From the studies on butter culture by various investigators it would be expected that diacetyl and acetyl methyl carbinol contents would increase as acid developed during the ripening of cream. This was true in most trials under both preliminary and plant conditions. The range of acidities generally studied was from 0.25 to 0.55 per cent. Normally, the higher the acidity of the ripened cream (sweet or neutralized sour), the more effect each unit increase in acidity seemed to have on diacetyl and acetyl methyl carbinol production. In a few preliminary trials decreases in diacetyl and acetyl methyl carbinol contents occurred, or increases were followed by decreases, or (more rarely) increases, decreases, and then increases were observed. In a few plant trials decreases were observed. The results deviating from the usual trend may have been caused by a depletion of the citric acid that was available to the flavor producing organisms, with a consequent reduction of diacetyl and acetyl methyl carbinol to 2,3-butylene glycol. Neutralized sour cream frequently has a low citric acid content because of the amount utilized during the natural souring process. Very small changes in diacetyl and acetyl methyl carbinol contents may have been within the limits of experimental error. Defective cultures and the operation of unknown factors also may have contributed to the unusual trends.

Similarly to its effect in butter cultures, the addition of small amounts of citric acid to cream prior to ripening generally resulted in increased yields of diacetyl and acetylmethylecarbinol when the same acidity was developed as in the controls. This was true in both preliminary trials and plant trials. Besides serving as a substrate, the added citric acid probably caused a slight decrease in pH which tended to favor diacetyl and acetylmethylecarbinol production. This would apply particularly when comparative lots of cream were ripened equal periods rather than to the same acidity. The amount of citric acid added to cream probably should be limited to 0.1 per cent. Larger additions caused an objectionable curdy condition in the ripened cream, especially when high acidities were developed.

In the preliminary trials the large increases in diacetyl and acetyl-methylecarbinol contents of cream ripened with agitation (shaking) probably were caused by the incorporation of air; the additional oxygen supplied by the air would enhance biological oxidation of acetyl-methylecarbinol to diacetyl. In trials in which the cream had citric acid added as a substrate for the flavor producing organisms and also was agitated to increase oxidation, the results indicate that there was only a slightly increased yield of diacetyl and acetyl-methylecarbinol compared to samples that were agitated without added citric acid. Oxygen seemed to be much more important than citric acid in increasing the diacetyl and acetyl-methylecarbinol contents under the conditions studied. The diacetyl and acetyl-methylecarbinol contents of cream agitated (revolving the coils)

in the plant trials also increased, but the contents were not as large as in the preliminary trials. If the vats had been full probably the agitation by revolving the coils would have been less effective. In most plants there is no night shift and consequently agitating the cream would involve additional labor expense or automatic equipment.

The ripening temperature of cream had only a small effect on the amount of diacetyl and acetyl methyl carbinal produced. In preliminary trials the amounts were decreased as the temperatures increased from 50° to 60°F. and then to 70°F. It is known that the citric acid fermenting organisms grow well below 70°F. In a few trials one or more unknown factors caused a greater production of diacetyl and acetyl methyl carbinal at 60°F. or at 70°F. than at 50°F. In plant trials cream was ripened at 70°F. and at 61°F. The results were inconsistent, favoring each ripening temperature in approximately half of the comparisons. An explanation is the difference between the two procedures of handling the cream from the time of obtaining the desired acidity to the time of churning. With cream ripened at 70°F. the agitation during cooling and subsequent holding over night before churning probably incorporated air into the cream and resulted in slightly larger yields of diacetyl and acetyl methyl carbinal than would have occurred if the cream had been churned immediately. With cream ripened over night at 61°F. the desired acidity had developed by morning and the churning process was carried out at once.

As would be expected from the reports of numerous studies on butter cultures, the production of diacetyl and acetyl methyl carbinal in cream

ripened with different butter cultures frequently varied, especially with cultures that previously had been carried under different conditions. This was true of preliminary trials and of plant trials. Contamination with other organisms and unfavorable growth conditions which resulted in the partial elimination of the citric acid fermenting organisms are possible explanations.

In the preliminary trials increased amounts of butterfat resulted in larger yields of diacetyl and acetymethylecarbinol in the ripened cream. Mineral oil or butter oil added to skim milk caused similar results, indicating that the effect of fat may be physical, probably by the retention of air. Cream ripened under partial vacuum produced less diacetyl and acetymethylecarbinol than cream ripened under atmospheric pressure, undoubtedly because of the removal of air. The difference was greater with an increase in the butterfat content of the cream which may have been due to the tendency of the fat to retain the air.

Obviously, there is no advantage in developing large amounts of diacetyl and acetymethylecarbinol in ripened cream if the corresponding unsalted butter does not retain satisfactory amounts or develop satisfactory amounts on holding. For reasons not yet known the correlation between the diacetyl and acetymethylecarbinol contents of the ripened cream and the fresh butter was only very general in both preliminary and plant trials. Although large amounts of diacetyl and acetymethylecarbinol in the cream usually are necessary for large amounts in butter, relatively large amounts of the two compounds in ripened cream did not necessarily

mean that relatively large amounts would be retained in the butter.

In plant trials unsalted butter was held at different temperatures for various periods. The correlation in diacetyl and acetyl methylcarbinol contents between the ripened cream and the butter after holding seemed to be closer than when the butter was fresh. It is possible that if the determination of diacetyl and acetyl methylcarbinol could be made at the time of maximum production during the holding, the correlation would be very close. However, the length of time required for maximum production of diacetyl and acetyl methylcarbinol during holding varies with different cultures and also with the same culture under different conditions. The increases in diacetyl and acetyl methylcarbinol contents during holding of unsalted butter probably depend on temperature, period of holding and characteristics of the butter culture organisms.

SUMMARY

Part I. Preliminary Churnings

Diacetyl and acetylmethylcarbinol contents of sweet or neutralized sour cream and of skim milk generally increased as acid developed during the ripening. Infrequently, the diacetyl and acetylmethylcarbinol contents decreased or other trends occurred.

Diacetyl and acetylmethylcarbinol contents of ripened sweet or neutralized sour cream generally were slightly increased by adding small amounts of citric acid at the time of inoculation.

Diacetyl and acetylmethylcarbinol contents of sweet or neutralized sour cream were greatly increased by agitating (shaking) during the ripening.

In temperature comparisons the highest diacetyl and acetylmethylcarbinol contents generally developed in the cream ripened at 50°F.; the next highest in the cream ripened at 60°F.; and the lowest in the cream ripened at 70°F. There were a few variations from this order. Ordinarily, the differences among the three temperatures were small with both the diacetyl and acetylmethylcarbinol contents.

When different butter cultures were used to ripen cream, the differences between the extremes in diacetyl and acetylmethylcarbinol contents generally were rather large, but several of the values within the extremes were much the same.

In general, diacetyl and acetylmethylcarbinol contents of ripened cream increased with an increase in butterfat content. Similar results

were obtained in trials with various amounts of mineral oil or butter oil added to sweet skim milk.

Sweet cream ripened under partial vacuum produced less diacetyl and acetyl methyl carbinol than cream ripened under atmospheric pressure, and the higher the butterfat content of the cream the greater the difference.

In 309 churnings of unsalted butter, the retention of diacetyl as indicated by the ratio of the amount in cream to the amount in butter showed an average of 110.725. In the same samples the ratio of acetyl-methylcarbinol was 110.339.

Part II. Plant Churnings

Diacetyl and acethylmethyloarbinol contents of sweet or neutralized sour cream generally increased as acid developed during the ripening.

Diacetyl and acethylmethyloarbinol contents of ripened sweet or neutralized sour cream generally were slightly increased by adding small amounts of citric acid at the time of inoculation.

Diacetyl and acethylmethyloarbinol contents were increased in sweet or neutralized sour cream by agitating (revolving the coils) periodically during the ripening.

Diacetyl and acethylmethyloarbinol contents of sweet or neutralized sour cream ripened at 70°F. and at 61°F. showed only small differences.

The differences between the extremes in diacetyl and acethylmethyloarbinol contents of sweet or neutralized sour cream ripened with different butter cultures were large in some trials and very small in others.

The ratios of diacetyl to acethylmethyloarbinol in the 74 samples of cream ripened with different procedures varied extensively. The ratios of diacetyl to acethylmethyloarbinol in the corresponding butter also varied, but the variation was smaller than occurred in the cream.

In 74 samples the average ratio of the diacetyl in the cream to the diacetyl in the corresponding unsalted butter was 1:0.351 and the average acethylmethyloarbinol ratio in the same samples was 1:0.216.

Diacetyl and acethylmethyloarbinol contents of most samples of butter were higher after holding 1 week at 36 to 40°F. or 1 month at

36 to 40°F. than before holding. Holding 3 days at 60°F. and the remainder of the week at 36 to 40°F. was superior to holding 1 week at 36 to 40°F. from the standpoint of the numbers of increases in diacetyl and acetymethylcarbinol contents.

After holding 1 week at 36 to 40°F., 1 month at 36 to 40°F., or 6 months at -10 to 0°F., butter manufactured from cream ripened to higher acidities (0.03 to 0.12 per cent higher), from cream with citric acid added previous to ripening or from cream agitated during ripening generally scored higher than the control churning. There were some variations from these relationships. The scores on the butter from cream ripened at 70°F. and at 61°F. differed very little. Using different butter cultures to ripen cream resulted in the corresponding butter differing in score as much as 0.75 of a point. Unsalted butter that was held 1 month at 36 to 40°F. generally decreased in flavor score from 0.5 to 1.5 points, and unsalted butter that was held 6 months at -10 to 0°F. decreased in flavor score from 1.0 to 2.5 points.

APPENDIX

TABLE 1. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Amo) CONTENTS OF SWEET CREAM, RIPEMED TO VARIOUS ACIDITIES, AND OF THE CORRESPONDING UNSALTED BUTTER

Laboratory Trials

Trial No.	Ripening temp.	Cream			Butter		
		Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amo p.p.m.	pH	$\text{Ac}_2$ p.p.m.
1	70° F.	0.37	5.13	0.84	35.6	4.95	0.76
	70	.44	4.89	1.49	42.9	4.73	1.54
	70	.51	4.67	2.28	52.4	4.54	.96
2	70	.33	5.54	.46	30.9	5.46	.71
	70	.38	5.43	.59	34.1	5.28	.83
	70	.44	5.14	.74	40.7	5.09	.54
3	70	.35	5.60	.27	24.8	5.60	.18
	70	.38	5.50	.34	25.9	5.53	.24
	70	.42	5.45	.39	27.2	5.45	.24
4	70	.34	5.71	.52	39.6	5.69	.40
	70	.40	5.45	.60	41.3	5.46	.59
	70	.44	5.35	.63	41.7	5.38	.45
5	70	.33	5.63	.48	37.6	5.68	.20
	70	.39	5.49	.50	38.5	5.49	.36
	70	.44	5.34	.61	42.2	5.34	.26
6	70	.35	5.24	.75	34.8	4.94	.98
	70	.42	5.18	1.05	37.7	4.80	.81
	70	.49	4.48	1.67	50.4	4.57	1.23
7	70	.41	5.40	.31	25.9	5.03	.12
	70	.50	5.12	.37	28.0	5.02	.66
	70	.55	4.95	.44	29.0	4.92	.60
8	70	.35	5.54	.52	33.8	5.36	1.06
	70	.40	5.32	.57	38.3	5.06	1.13
	70	.44	5.09	.72	45.4	4.88	1.30

TABLE 1. Continued

Trial No.	Cream					Butter		
	Ripening temp.	Per cent acid	pH	Ac <sub>2</sub> p.p.m.	Ame p.p.m.	pH	Ac <sub>2</sub> p.p.m.	Ame p.p.m.
9	65° F.	.42	5.02	0.90	36.8	4.76	1.46	27.4
	65	.47	4.81	1.05	39.4	4.80	1.33	27.1
	65	.51	4.77	1.05	42.4	4.77	1.13	20.4
	65	.56	4.73	1.75	55.4	4.75	1.81	27.4
10	65	.30	5.47	.71	19.4	5.40	.42	6.7
	65	.36	5.23	.74	17.3	5.30	.74	7.5
	65	.40	5.15	.72	12.0	5.15	.68	7.4
	65	.44	5.00	.75	12.0	5.07	.80	10.9
11	65	.46	4.77	1.14	60.0	4.64	1.77	34.6
	65	.50	4.75	1.30	61.2	4.63	1.35	24.5
	65	.53	4.71	2.78	68.6	4.63	1.50	18.5
	65	.59	4.67	2.32	85.8	4.62	1.46	26.2
12	60	.34	5.61	.40	23.7	5.73	.20	3.9
	60	.38	5.55	.47	25.9	5.59	.20	5.2
	60	.42	5.50	.32	27.3	5.53	.29	5.4
13	60	.37	5.57	.57	26.8	5.58	.59	12.9
	60	.49	5.12	.56	29.8	5.12	.56	9.8
	60	.54	4.98	.44	29.0	4.95	.54	10.1
14	60	.33	5.78	.58	43.8	5.87	.22	5.6
	60	.39	5.45	.59	44.9	5.56	.23	6.4
	60	.45	5.28	.60	47.3	5.39	.25	7.2
15	60	.38	5.22	1.40	34.9	4.98	1.07	20.1
	60	.44	5.05	1.45	38.1	4.94	1.06	10.4
	60	.49	4.84	1.70	52.4	4.78	1.39	30.6
16	60	.38	5.12	1.02	36.3	4.91	.98	16.2
	60	.46	4.89	1.52	41.4	4.81	.99	17.7
	60	.52	4.70	1.77	52.5	4.72	.55	14.6

TABLE 1. Continued

Trial No.	Cream					Butter		
	Ripening temp.	Per cent acid	pH	Ac <sub>2</sub> p.p.m.	Ame p.p.m.	pH	Ac <sub>2</sub> p.p.m.	Ame p.p.m.
17	60°F.	0.35	5.48	0.62	28.0	5.42	0.72	10.2
	60	.41	5.29	.84	28.5	5.14	.98	14.4
	60	.46	5.01	.98	32.6	5.03	.84	15.0
18	60	.36	5.42	.84	14.4	5.38	1.04	21.9
	60	.40	5.25	.85	14.6	5.06	.99	23.6
	60	.45	5.09	.97	18.4	5.00	1.05	18.9
19	60	.32	5.78	.66	14.4	5.82	.20	5.8
	60	.39	5.52	.70	15.4	5.67	.27	6.3
	60	.44	5.38	.72	52.1	5.58	.30	5.2
20	32	.14	6.46	.28	16.5	5.43	.33	10.6
	55	.28	5.49	.56	19.9	5.30	.35	6.6
	55	.34	5.34	.82	13.1	5.14	.38	4.4
	55	.39	5.21	.86	15.2	5.08	.52	7.4
	55	.44	5.09	.60	15.5			
21	32	.19	5.62	.27	10.0	4.81	1.80	30.2
	55	.41	5.11	.87	31.4	4.84	1.84	29.8
	55	.44	5.05	.97	31.4	4.85	1.62	26.4
	55	.51	4.86	1.03	39.5	4.84	1.69	27.6
	55	.54	4.80	1.17	40.8			
22	55	.44	4.91	1.44	59.1	4.63	1.58	24.5
	55	.49	4.79	1.54	60.6	4.65	1.42	24.0
	55	.53	4.77	1.73	65.7	4.61	1.57	21.4
	55	.58	4.73	2.06	74.1	4.62	1.53	23.6
23	55	.27	5.65	.59	21.6			
	55	.35	5.33	.69	24.9			
	55	.40	5.03	.72	26.9			
	55	.44	4.82	.84	34.0			

TABLE 1. Continued

Trial No.	Cream					Butter		
	Ripening temp.	Per cent acid	pH	Ac <sub>2</sub> p.p.m.	Amc p.p.m.	pH	Ac <sub>2</sub> p.p.m.	Amc p.p.m.
24	32° F.	0.18	6.43	0.20	9.9			
	55	.29	5.53	.62	29.2			
	55	.35	5.38	.71	31.2			
	55	.43	5.21	.75	31.2			
	55	.48	4.92	.84	32.4			
25	32	.10	6.54	.23	3.5			
	55	.36	5.06	.90	34.5			
	55	.42	4.97	1.11	37.7			
	55	.48	4.84	1.20	46.7			
	55	.52	4.78	1.84	52.3			
26	32	.20	6.39	.21	9.4			
	55	.41	5.15	1.13	39.9			
	55	.50	4.85	1.72	51.3			
	55	.55	4.75	2.02	69.6			
	55	.60	4.69	3.86	81.0			
27	32	.18	6.57	.12	14.1			
	55	.30	5.85	.46	2.5			
	55	.34	5.81	.27	2.5			
	55	.42	5.67	.24	2.3			
	55	.45	5.56	.19	2.2			
28	55	.28	5.80	.59	25.9			
	55	.33	5.61	.65	25.9			
	55	.36	5.48	.72	30.6			
	55	.43	5.19	.74	35.0			
29	55	.35	5.17	.57	21.9	5.26	1.04	17.0
	55	.45	5.05	.67	22.4	5.07	1.22	11.3
	55	.51	4.87	.56	23.9	4.97	1.10	12.1
	55	.56	4.74	1.12	35.8	4.80	.99	10.0

TABLE I. Continued

Trial No.	Cream					Butter		
	Ripening temp.	Per cent acid	pH	Ac <sub>2</sub> p.p.m.	Amo p.p.m.	pH	Ac <sub>2</sub> p.p.m.	Amo p.p.m.
30	55° F.	0.35	5.72	0.56	45.4	5.55	0.44	12.9
	55	.39	5.56	.59	46.0	5.22	.51	13.5
	55	.43	5.35	.60	47.0	5.10	.43	10.8
	55	.45	5.36	.61	50.4	5.12	.34	7.6
31	50	.33	5.62	.89	42.9	5.76	.27	12.0
	50	.39	5.42	.90	44.0	5.65	.34	7.3
	50	.46	5.17	1.12	45.7	5.20	.47	7.6
32	50	.40	5.36	.60	27.4	5.34	.54	10.2
	50	.51	5.04	.82	32.2	5.06	.41	5.4
	50	.58	4.90	1.04	35.2	4.92	.69	12.6
33	50	.33	5.66	.79	40.2	5.76	.20	6.1
	50	.40	5.40	.85	42.7	5.51	.32	6.1
	50	.48	5.20	1.00	44.8	5.21	.52	8.9
34	50	.37	5.13	1.12	35.6	5.03	1.08	14.4
	50	.45	4.99	1.14	38.6	4.80	1.45	21.6
	50	.50	4.85	1.85	58.8	4.77	1.94	56.4
35	50	.33	5.68	.55	25.8	5.73	.31	4.8
	50	.39	5.52	.62	27.9	5.77	.33	4.8
	50	.43	5.43	.64	30.5	5.66	.33	2.6
36	50	.40	4.96	1.32	37.2	4.83	1.23	14.1
	50	.44	4.84	1.63	46.6	4.70	1.72	18.8
	50	.50	4.72	2.03	53.7			

TABLE 2. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Amc) CONTENTS OF NEUTRALIZED SOUR CREAM, RIPENED TO VARIOUS ACIDITIES, AND OF THE CORRESPONDING UNSALTED BUTTER

Trial No.	Laboratory Trials							
	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.
1	32° F.	.15	6.89	0.37	22.2	5.16	0.48	5.6
	62	.34	5.23	1.35	27.2			
	62	.36	5.10	1.85	39.8			
	62	.39	5.00	2.11	55.6			
	62	.42	4.95	2.57	75.3			
2	32	.14	6.75	.35	23.3	5.69	.44	5.4
	60	.32	5.70	1.41	43.1			
	60	.37	5.14	1.81	44.2			
	60	.42	4.85	1.90	50.6			
3	32	.14	6.69	.55	20.9	5.24	.44	3.9
	58	.31	5.36	1.40	23.8			
	58	.40	4.99	1.73	24.0			
	58	.44	4.79	2.11	24.3			

TABLE 3. DIACETYL ( $\Delta\text{O}_2$ ) AND ACETILMETHYLCARBINOL (Ame) CONTENTS OF SKIM MILK RIPENED TO VARIOUS ACIDITIES

Laboratory Trials					
Trial No.	Skin milk				
	Ripening temp.	Per cent acid	pH	$\Delta\text{O}_2$ p.p.m.	Ame p.p.m.
1	55°F.	0.41	5.54	0.38	18.0
	55	.49	5.30	.49	18.6
	55	.61	5.05	.57	23.6
	55	.71	4.82	.57	26.3
2	55	.36	5.75	.38	18.1
	55	.40	5.49	.38	19.2
	55	.50	5.36	.46	20.0
	55	.65	4.92	.50	23.5
3	55	.34	5.75	.30	15.9
	55	.42	5.56	.35	16.8
	55	.53	5.14	.42	17.6
	55	.64	4.92	.45	20.8
4	55	.30	5.75	.37	10.5
	55	.40	5.40	.40	11.3
	55	.58	5.05	.45	12.9
	55	.66	4.75	.51	18.1

TABLE 4. DIACETYL ( $\text{Ac}_2$ ) AND ACETYL METHYL CARBINOL (Amc) CONTENTS OF SWEET CREAM, RIPENED WITH AND WITHOUT ADDED CITRIC ACID TO THE SAME pH, AND OF THE CORRESPONDING UNSALTED BUTTER

Trial No.	Laboratory Trials									
	Cream						Butter			
	Per cent citric acid added	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	
1	0	55 F.	0.36	5.06	0.90	34.5		1.20	30.7	
	0.05	55	.35	5.15	1.01	32.6		1.61	30.6	
2	0	55	.42	4.97	1.11	37.7		1.30	31.2	
	0.05	55	.40	5.02	1.45	36.5		2.10	26.9	
3	0	55	.48	4.85	1.20	46.7		1.80	42.1	
	0.05	55	.46	4.91	1.49	49.2		1.30	28.4	
4	0	55	.52	4.78	1.84	52.3		1.20	27.8	
	0.05	55	.50	4.81	2.52	55.2		1.40	27.6	
5	0	55	.30	5.85	.47	12.5	5.90	.09	.7	
	0.05	55	.32	5.73	.98	18.5	5.68	.13	.9	
6	0	55	.34	5.81	.27	2.5	5.80	.07	.7	
	0.05	55	.35	5.64	.98	13.1	5.64	.07	.7	
7	0	55	.36	5.67	.24	2.4	5.67	.06	.7	
	0.05	55	.36	5.61	.63	5.2	5.58	.07	.6	
8	0	55	.37	5.56	.18	2.2	5.55	.06	.6	
	0.05	55	.37	5.51	.56	2.7	5.52	.10	.8	
9	0	55	.56	4.74	1.19	35.8	4.80	1.00	10.0	
	0.05	55	.57	4.72	1.27	44.4	4.78	.99	12.2	
10	0	55	.39	5.19	.75	35.0				
	0.05	55	.39	5.19	.78	58.6				
11	0	54	.30	5.69	.20	10.2	5.49	.25	4.3	
	0.05	54	.31	5.65	.25	12.1	5.55	.27	4.0	

TABLE 4. Continued

Trial No.	Cream						Butter		
	Per cent citric acid added	Ripening temp.	Per cent acid	pH	Ac <sub>2</sub> p.p.m.	Ame p.p.m.	pH	Ac <sub>2</sub> p.p.m.	Ame p.p.m.
12	0	54° F.	.34	5.52	0.19	10.8	5.46	0.16	3.2
	0.05	54	.34	5.55	.20	12.5	5.43	.20	2.8
13	0	55	.35	5.56	.59	46.0	5.22	.51	13.5
	0.05	55	.34	5.58	.66	54.4	5.43	.37	9.0
14	0	60	.42	5.02	.87	37.9	4.80	.70	9.3
	0.10	60	.41	5.01	1.09	45.1	4.83	.67	10.7

Laboratory Trials									
	Per cent of water		Ripening Per cent add		p.p.m.		p.p.m.		No.
	Per cent added temp.		add		p.p.m.		p.p.m.		
1	0.05	60	.36	5.14	1.80	111.2	1.97	0.63	7.8
	.10	60	.38	5.09	1.80	118.0	1.97	.82	25.0
	.15	60	.39	4.99	2.02	56.3	1.93	.82	25.0
2	0.05	60	.11	1.75	1.81	37.4	.64	8.1	
	.10	60	.15	1.76	2.01	56.2	.69	8.2	12.0
	.15	60	.17	1.74	2.75	72.2	.63	1.02	17.2
3	0.05	58	.10	1.73	1.73	38.3	.50	4.6	
	.10	58	.13	1.79	2.00	50.5	.58	5.0	20.2
	.15	58	.15	1.79	2.05	57.3	.59	5.0	10.2
4	0.05	58	.11	1.70	1.70	58.0	.79	7.2	
	.10	58	.15	1.75	2.68	111.9	.80	7.2	7.3
	.15	58	.15	1.74	3.09	115.3	.76	7.2	7.3
5	0.10	63	.13	1.73	2.59	33.2	.74	9.5	
	.15	63	.13	1.71	2.66	33.2	.74	9.5	25.6
6	0.10	63	.17	1.67	3.90	33.9	.65	7.1	
	.15	63	.17	1.69	3.63	3.76	.65	7.1	21.8
7	0.05	62	.12	1.98	1.11	32.1	.33	6.6	
	.10	62	.12	1.98	1.11	32.1	.33	6.6	7.6
8	0.05	62	.12	1.98	1.11	32.1	.33	6.6	12.2

TABLE 5. DIAGETIC ( $\text{Ag}_2$ ) AND AGTITMERICARBOYL ( $\text{Ag}_2$ ) CONTENTS OF  
MUTRALIZED SOY GRAN, RIPENED WITH VARIOUS AMOUNTS OF  
ADDED CITRIC ACID TO THE SAME pH, AND OF THE  
CORRESPONDING UNSALTED BUTTER

TABLE 6. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Amc) CONTENTS OF SWEET CREAM, RIPENED WITH AND WITHOUT ADDED CITRIC ACID FOR EQUAL PERIODS, AND OF THE CORRESPONDING UNSALTED BUTTER

Trial No.	Laboratory Trials									
	Cream			Butter				pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.
	Per cent citric acid added	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.				
1	0	55°F.	0.35	5.17	0.57	21.9	5.26	1.04	17.0	
	0.05	55	.36	5.10	.71	29.9				
2	0	55	.39	5.05	.67	22.4	5.07	1.22	11.3	
	0.05	55	.43	4.90	.92	30.0				
3	0	55	.51	4.87	.56	23.9	4.97	1.10	12.1	
	0.05	55	.54	4.78	.99	37.4				
4	0	55	.28	5.80	.58	25.9				
	0.05	55	.31	5.57	.59	32.5				
5	0	55	.33	5.61	.65	25.9				
	0.05	55	.35	5.43	.66	36.6				
6	0	55	.36	5.42	.72	30.6				
	0.05	55	.37	5.36	.74	48.2				
7	0	55	.35	5.62	.24	10.4	5.46	.22	3.6	
	0.05	55	.37	5.51	.27	12.0				
8	0	55	.36	5.58	.25	10.4	5.41	.20	3.2	
	0.05	55	.38	5.42	.37	11.8				
9	0	56	.33	5.72	.55	45.4	5.55	.44	12.9	
	0.05	56	.34	5.61	.65	50.8				
10	0	56	.36	5.35	.61	47.0	5.10	.44	10.8	
	0.05	56	.37	5.29	.69	59.0				
11	0	56	.36	5.36	.61	50.4	5.12	.34	7.6	
	0.05	56	.38	5.23	.69	55.2				
12	0	70	.32	5.65	.52	31.3	5.57	.36	7.2	
	0.10	70	.34	5.34	.77	39.6				

No.	Laboratory Trials								
	Type	Hypersoln	Per cent	pH	$\text{Al}_2\text{O}_3$	Alum	p.p.m.	p.p.m.	p.p.m.
Treatment of oream temp. cold p.p.m. p.p.m. p.p.m.									
1	Control	Swt.	60°p.	0.25	6.31	0.53	12.7	5.59	0.62
2	Control	Swt.	60	.26	6.13	.84	106.6	5.57	1.63
3	Control	Swt.	60	.55	1.67	1.10	51.0	6.57	1.58
4	Control	Swt.	70	.36	6.01	1.13	28.9	5.80	1.10
5	Control	Swt.	70	.35	5.65	.52	31.3	5.57	.36
6	Control	Swt.	60	.27	5.32	.76	30.6	5.18	.59
7	Control	Swt.	60	.17	5.02	.87	37.9	4.80	.70
8	Control	Swt.	63	.13	4.73	1.59	33.2	4.40	.74
9	Control	Swt.	63	.17	4.67	1.78	33.9	3.90	.74

TABLE 7. DIACETYL ( $\text{Ac}_2$ ) AND ACETILMERICARBOYL ( $\text{Ac}_2$ ) CONCENTRATIONS OF CREAM, HYPERSOLN WITH AND WITHOUT AGITATION, AND OF THE CORRESPONDING UNASALTED BUTTER

Laboratory Trials

Butter

Cream

Trial No.	Laboratory Trials									
	Type	Repeating	Per cent	pH	$\text{As}_2$	Amo	pH	$\text{As}_2$	Amo	Amo p.p.m.
1	Control	Swt.	70°F.	0.38	5.34	0.77	39.6	5.50	0.50	11.7
	Control	Swt.	70	.37	5.19	1.16	79.5	5.31	.92	21.6
	Control	Swt.	70	.37	5.19	1.16	79.5	5.31	.92	21.6
2	Control	Swt.	70	.45	4.98	.91	18.7	5.22	.19	10.2
	Control	Swt.	70	.47	5.11	1.30	96.9	5.04	.90	20.7
	Control	Swt.	70	.47	5.11	1.30	96.9	5.04	.90	20.2
3	Control	Swt.	60	.41	5.01	1.09	15.1	4.83	.67	10.7
	Control	Swt.	60	.40	5.02	1.57	61.0	4.91	.65	20.1
	Control	Swt.	60	.40	5.02	1.57	61.0	4.83	.67	20.7
4	Control	Swt.	60	.49	4.74	1.73	65.3	4.65	.90	23.9
	Control	Swt.	60	.46	4.72	2.66	73.2	4.08	1.28	15.6
	Control	Swt.	60	.47	4.72	2.66	73.2	4.08	1.28	18.1
5	Control	Swt.	63	.47	4.76	4.72	3.50	83.9		
	Control	Swt.	63	.47	4.76	4.72	3.50	83.9		
6	Control	Swt.	63	.50	4.63	3.27	77.4	1.99	11.6	20.8
	Control	Swt.	63	.50	4.63	3.27	77.4	1.99	11.6	20.8

TABLE 8. DIGESTS (As<sub>2</sub>) AND ACETYLIMINOCARBONOL (Amo) COUNTS OF OREAN,  
CONTAINING 0.1 PER CENT ADDS CITRIC ACID AND RIPPLED WITH AND WITHOUT  
AGITATION, AND OF THE CORRESPONDING UNAGITATED ROTTEN

TABLE 9. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Ame) CONTENTS OF SWEET CREAM, RIPENED AT DIFFERENT TEMPERATURES, AND OF THE CORRESPONDING UNSALTED BUTTER

Laboratory Trials								
Trial No.	Ripening temp.	Cream				Butter		
		Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Ame p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Ame p.p.m.
1	70° F.	.33	5.63	.48	37.5	5.68	0.20	7.9
	60	.32	5.78	.66	44.4	5.82	.20	5.8
	50	.33	5.62	.89	42.9	5.76	.27	12.0
2	70	.39	5.49	.49	38.5	5.49	.36	9.9
	60	.39	5.52	.70	45.0	5.67	.27	6.3
	50	.39	5.42	.89	45.0	5.65	.34	7.3
3	70	.44	5.34	.61	42.2	5.34	.26	7.3
	60	.44	5.38	.72	52.1	5.58	.30	5.2
	50	.46	5.17	1.12	45.7	5.20	.47	7.6
4	70	.54	5.71	.52	39.6	5.69	.40	9.0
	60	.33	5.78	.58	43.8	5.87	.22	5.6
	50	.33	5.66	.79	40.2	5.76	.20	6.1
5	70	.40	5.45	.60	41.3	5.46	.59	14.1
	60	.39	5.45	.58	44.9	5.56	.23	6.4
	50	.40	5.40	.85	42.7	5.51	.32	6.1
6	70	.44	5.35	.63	41.7	5.38	.45	8.4
	60	.45	5.28	.60	47.3	5.39	.26	7.2
	50	.47	5.20	1.00	44.8	5.21	.53	8.9
7	70	.40	5.40	.31	26.0	5.43	.12	18.9
	60	.37	5.57	.57	26.8	5.58	.59	12.9
	50	.40	5.36	.60	27.4	5.34	.55	10.2
8	70	.50	5.12	.37	28.0	5.02	.67	18.6
	60	.49	5.12	.56	29.8	5.12	.56	9.8
	50	.51	5.04	.82	32.2	5.06	.41	5.4

TABLE 9. Continued

Trial No.	Cream					Butter		
	Ripening temp.	Per cent acid	pH	$\Delta e_2$ p.p.m.	Ame p.p.m.	pH	$\Delta e_2$ p.p.m.	Ame p.p.m.
9	70° F.	0.55	4.95	0.44	29.0	4.92	0.60	12.0
	60	.54	4.98	.44	29.0	4.95	.55	10.1
	50	.58	4.90	1.04	35.2	4.92	.70	12.6
10	70	.35	5.60	.28	24.8	5.60	.18	5.2
	60	.34	5.61	.40	23.7	5.73	.21	3.9
	50	.33	5.68	.55	25.8	5.73	.32	4.8
11	70	.38	5.50	.34	25.9	5.53	.24	6.9
	60	.38	5.55	.47	25.9	5.59	.20	5.2
	50	.39	5.52	.62	27.9	5.77	.33	4.8
12	70	.42	5.45	.39	27.2	5.45	.24	5.0
	60	.42	5.43	.39	27.3	5.53	.29	5.4
	50	.43	5.43	.64	30.5	5.66	.33	2.6
13	70	.37	5.13	.84	35.6	4.95	.76	12.4
	60	.38	5.12	1.02	36.3	4.91	.98	16.2
	50	.40	4.96	1.32	37.3	4.83	1.23	14.1
14	70	.44	4.89	1.49	43.0	4.73	1.55	13.8
	60	.46	4.89	1.52	41.4	4.81	.99	17.7
	50	.44	4.84	1.63	46.6			
15	70	.51	4.67	2.28	52.4	4.54	.97	27.5
	60	.52	4.70	1.77	52.5	4.72	.55	14.8
	50	.51	4.72	2.03	53.7	4.70	1.72	18.8
16	70	.35	5.24	.75	34.8	4.94	.98	16.6
	60	.36	5.22	1.40	34.9	4.98	1.07	20.1
	50	.37	5.13	1.12	35.6	5.03	1.08	14.4

TABLE 9. Continued

Trial No.	Ripening temp.	Cream				Butter		
		Per cent acid	pH	Ac <sub>2</sub> p.p.m.	Amc p.p.m.	pH	Ac <sub>2</sub> p.p.m.	Amc p.p.m.
17	70°F.	.42	5.18	1.05	37.6	4.80	0.82	26.7
	60	.44	5.05	1.45	38.1	4.94	1.06	10.4
	50	.45	4.99	1.45	38.6	4.80	1.45	21.6
18	70	.49	4.84	1.67	50.4	4.57	1.23	25.6
	60	.49	4.84	1.70	52.4	4.78	1.39	30.6
	50	.50	4.85	1.85	58.8	4.77	1.95	56.4

TABLE 10. DIAGETES ( $Ad_2$ ) AND ADULTILABYRINTHOCYTOMES ( $Am_2$ ) CONTRASTS OF CHINA, RIMMED WITH BOTTER CULTURES FROM THE SAME MOTHER CULTURE, AND OF THE CORRESPONDING UNRIMED BUTTER.

TABLE II. DIACTORIAL ( $\text{AO}_2$ ) AND AGENTILIALEHYLICARBOYLIC ( $\text{AO}_3$ ) COMPOUNDS OF OMEGA, RIPENED WITH DIFFERENT COMMERCIAL BUTTER CULTURES.

TABLE 12. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Ame) CONTENTS OF CREAM, RIPENED WITH DIFFERENT BUTTER CULTURES CARRIED UNDER THE SAME CONDITIONS, AND OF THE CORRESPONDING UNSALTED BUTTER

Laboratory Trials										
Trial No.	Culture No.	Ripening temp.	Cream				Butter			
			acid	pH	$\text{Ac}_2$ p.p.m.	Ame p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Ame p.p.m.	
1	1	62 F.	0.41	5.00	0.74	30.7	4.94	0.60	9.0	
	2	62	.45	5.06	.87	29.1	5.00	.39	7.4	
	3	62	.40	5.10	.76	30.6	5.22	.32	8.2	
	4	62	.42	4.89	.71	26.4	5.03	.30	6.3	
	5	62	.44	4.97	.74	34.7	4.90	.60	12.1	
	6	62	.45	4.94	.75	40.0	4.92	.53	9.3	
	7	62	.43	4.87	.93	31.4	4.78	.51	9.3	
	8	62	.45	4.87	.76	28.8	4.78	.55	9.8	

TABLE 13. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Amc) CONTENTS OF SKIM MILK AND CREAM RIPENERED TO THE SAME pH

Laboratory Trials										
Trial No.	Cream					Skim milk				
	Ripening temp.	Per cent acid	pH	As <sub>2</sub> p.p.m.	Ame p.p.m.	Per cent acid	pH	As <sub>2</sub> p.p.m.	Ame p.p.m.	
1	55° F.	.29	5.53	0.62	29.2	0.36	5.75	0.39	18.1	
2	55	.35	5.38	.71	31.2	.40	5.49	.37	19.2	
3	55	.43	5.21	.75	26.5	.50	5.36	.46	19.3	
4	55	.48	4.92	.84	32.4	.65	4.92	.50	23.5	
5	55	.29	5.80	.59	25.9	.34	5.75	.31	15.9	
6	55	.34	5.61	.65	25.9	.41	5.56	.35	16.8	
7	55	.37	5.42	.72	30.6	.53	5.14	.42	17.6	
8	55	.43	5.19	.75	35.0	.64	4.92	.43	20.8	
9	55	.27	5.65	.59	21.6	.30	5.75	.37	10.5	
10	55	.34	5.33	.69	24.9	.40	5.40	.40	11.3	
11	55	.39	5.03	.72	26.9	.58	5.05	.45	12.3	
12	55	.44	4.82	.84	34.0	.66	4.75	.52	18.1	

TABLE 14. DIACETYL ( $\text{Ac}_2$ ) AND ACETYL METHYL CARBINOL (Amc) CONTENTS OF SKIM MILK OR CREAM WITH VARIOUS PERCENTAGES OF BUTTERFAT RIPENED TO THE SAME pH

Laboratory Trials							
Trial No.	Per cent fat	Per cent culture	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.
1	0	5	70°F.	.50	5.27	0.28	16.6
	5	5	70	.49	5.24	.38	19.7
	10	5	70	.47	5.26	.39	20.3
	20	5	70	.43	5.31	.41	21.6
	30	5	70	.39	5.26	.47	25.3
	40	5	70	.35	5.20	.55	31.7
2	0	5	60	.55	5.00	.52	21.7
	5	5	60	.48	5.12	.52	21.7
	10	5	60	.47	5.19	.52	21.8
	20	5	60	.45	5.20	.62	22.5
	30	5	60	.43	5.16	.82	33.2
	40	5	60	.40	5.08	.92	40.5
3	0	5	60	.45	5.40	.41	22.4
	10	4.5	60	.43	5.40	.64	26.9
	20	4	60	.40	5.45	.69	33.1
	30	3.5	60	.36	5.39	.80	37.4
	40	3	60	.34	5.37	.99	47.2
	0	5	60	.45	5.41	.42	22.2
4	10	4.5	60	.43	5.42	.53	22.4
	20	4	60	.41	5.47	.63	27.3
	30	3.5	60	.38	5.46	.80	29.0
	40	3	60	.35	5.42	.88	33.5
	0	5	61	.59	4.83	.61	25.6
5	10	4.5	61	.46	5.21	.69	30.8
	20	4	61	.43	5.28	.89	31.7
	30	3.5	61	.42	5.13	1.00	32.7
	40	3	61	.41	4.92	.85	28.8
	0	5	61	.59	4.83	.60	25.3
6	10	4.5	61	.46	5.30	.63	21.6
	20	4	61	.45	5.26	.70	27.6
	30	3.5	61	.49	5.00	.88	28.2

TABLE 14. Continued

Trial No.	Per cent fat	Per cent culture	Ripening temp.	Per cent acid	pH	Ac <sub>2</sub> p.p.m.	Ame p.p.m.
7	0	5	70° F.	0.50	4.91	0.52	27.5
	10	4.5	70	.47	5.04	.61	29.2
	20	4	70	.45	5.02	.92	38.9
	30	3.5	70	.39	5.07	.99	40.8
	40	3	70	.40	5.02	1.04	41.7
8	0	5	70	.50	5.43	.40	25.7
	10	4.5	70	.48	5.50	.57	30.6
	20	4	70	.40	5.53	.61	36.9
	30	3.5	70	.36	5.52	.80	41.5
	40	3	70	.36	5.11	1.01	54.1

TABLE 15. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Amc) CONTENTS  
OF SKIM MILK OR CREAM WITH VARIOUS PERCENTAGES OF BUTTERFAT  
RIPENED UNDER PARTIAL VACUUM AND UNDER  
ATMOSPHERIC PRESSURE TO THE SAME pH

Laboratory Trials

Trial No.	Treatment	Per cent fat	Per cent culture	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.
1	None	0	5	70°F.	0.49	5.43	0.40	25.7
	Vacuum	0	5	70	.49	5.42	.37	22.6
	None	20	5	70	.40	5.53	.60	36.9
	Vacuum	20	5	70	.42	5.37	.38	27.0
	None	40	5	70	.39	5.11	1.01	54.1
	Vacuum	40	5	70	.44	4.83	.63	27.8
2	None	0	5	70	.57	4.91	.52	27.3
	Vacuum	0	5	70	.50	5.23	.38	10.1
	None	20	5	70	.45	5.02	.92	38.9
	Vacuum	20	5	70	.43	5.04	.42	17.8
	None	40	5	70	.40	5.02	1.04	41.6
	Vacuum	40	5	70	.45	4.86	.51	18.6

Trial No.	Laboratory Trials								
	Per cent Mineral oil	Per cent Ripening agent	Per cent P.P.M.	Per cent Acetone	Per cent Sodium p.p.m.				
1	0	5	0.57	70.91	0.42	27.3	32.5	33.9	36.5
	10	5	70	.57	70	.42	70	.42	70
	20	5	70	.45	70	.42	70	.42	70
	30	5	70	.42	70	.42	70	.42	70
	40	5	70	.38	70	.38	70	.38	70
2	0	5	70	.37	70	.37	70	.37	70
	10	5	70	.35	70	.35	70	.35	70
	20	5	70	.35	70	.35	70	.35	70
	30	5	70	.35	70	.35	70	.35	70
	40	5	70	.35	70	.35	70	.35	70
	50	5	70	.35	70	.35	70	.35	70
	60	5	70	.35	70	.35	70	.35	70
	70	5	70	.35	70	.35	70	.35	70
	80	5	70	.35	70	.35	70	.35	70
	90	5	70	.35	70	.35	70	.35	70
	100	5	70	.35	70	.35	70	.35	70
	110	5	70	.35	70	.35	70	.35	70
	120	5	70	.35	70	.35	70	.35	70
	130	5	70	.35	70	.35	70	.35	70
	140	5	70	.35	70	.35	70	.35	70
	150	5	70	.35	70	.35	70	.35	70
	160	5	70	.35	70	.35	70	.35	70
	170	5	70	.35	70	.35	70	.35	70
	180	5	70	.35	70	.35	70	.35	70
	190	5	70	.35	70	.35	70	.35	70
	200	5	70	.35	70	.35	70	.35	70
	210	5	70	.35	70	.35	70	.35	70
	220	5	70	.35	70	.35	70	.35	70
	230	5	70	.35	70	.35	70	.35	70
	240	5	70	.35	70	.35	70	.35	70
	250	5	70	.35	70	.35	70	.35	70
	260	5	70	.35	70	.35	70	.35	70
	270	5	70	.35	70	.35	70	.35	70
	280	5	70	.35	70	.35	70	.35	70
	290	5	70	.35	70	.35	70	.35	70
	300	5	70	.35	70	.35	70	.35	70
	310	5	70	.35	70	.35	70	.35	70
	320	5	70	.35	70	.35	70	.35	70
	330	5	70	.35	70	.35	70	.35	70
	340	5	70	.35	70	.35	70	.35	70
	350	5	70	.35	70	.35	70	.35	70
	360	5	70	.35	70	.35	70	.35	70
	370	5	70	.35	70	.35	70	.35	70
	380	5	70	.35	70	.35	70	.35	70
	390	5	70	.35	70	.35	70	.35	70
	400	5	70	.35	70	.35	70	.35	70
	410	5	70	.35	70	.35	70	.35	70
	420	5	70	.35	70	.35	70	.35	70
	430	5	70	.35	70	.35	70	.35	70
	440	5	70	.35	70	.35	70	.35	70
	450	5	70	.35	70	.35	70	.35	70
	460	5	70	.35	70	.35	70	.35	70
	470	5	70	.35	70	.35	70	.35	70
	480	5	70	.35	70	.35	70	.35	70
	490	5	70	.35	70	.35	70	.35	70
	500	5	70	.35	70	.35	70	.35	70
	510	5	70	.35	70	.35	70	.35	70
	520	5	70	.35	70	.35	70	.35	70
	530	5	70	.35	70	.35	70	.35	70
	540	5	70	.35	70	.35	70	.35	70
	550	5	70	.35	70	.35	70	.35	70
	560	5	70	.35	70	.35	70	.35	70
	570	5	70	.35	70	.35	70	.35	70
	580	5	70	.35	70	.35	70	.35	70
	590	5	70	.35	70	.35	70	.35	70
	600	5	70	.35	70	.35	70	.35	70
	610	5	70	.35	70	.35	70	.35	70
	620	5	70	.35	70	.35	70	.35	70
	630	5	70	.35	70	.35	70	.35	70
	640	5	70	.35	70	.35	70	.35	70
	650	5	70	.35	70	.35	70	.35	70
	660	5	70	.35	70	.35	70	.35	70
	670	5	70	.35	70	.35	70	.35	70
	680	5	70	.35	70	.35	70	.35	70
	690	5	70	.35	70	.35	70	.35	70
	700	5	70	.35	70	.35	70	.35	70
	710	5	70	.35	70	.35	70	.35	70
	720	5	70	.35	70	.35	70	.35	70
	730	5	70	.35	70	.35	70	.35	70
	740	5	70	.35	70	.35	70	.35	70
	750	5	70	.35	70	.35	70	.35	70
	760	5	70	.35	70	.35	70	.35	70
	770	5	70	.35	70	.35	70	.35	70
	780	5	70	.35	70	.35	70	.35	70
	790	5	70	.35	70	.35	70	.35	70
	800	5	70	.35	70	.35	70	.35	70
	810	5	70	.35	70	.35	70	.35	70
	820	5	70	.35	70	.35	70	.35	70
	830	5	70	.35	70	.35	70	.35	70
	840	5	70	.35	70	.35	70	.35	70
	850	5	70	.35	70	.35	70	.35	70
	860	5	70	.35	70	.35	70	.35	70
	870	5	70	.35	70	.35	70	.35	70
	880	5	70	.35	70	.35	70	.35	70
	890	5	70	.35	70	.35	70	.35	70
	900	5	70	.35	70	.35	70	.35	70
	910	5	70	.35	70	.35	70	.35	70
	920	5	70	.35	70	.35	70	.35	70
	930	5	70	.35	70	.35	70	.35	70
	940	5	70	.35	70	.35	70	.35	70
	950	5	70	.35	70	.35	70	.35	70
	960	5	70	.35	70	.35	70	.35	70
	970	5	70	.35	70	.35	70	.35	70
	980	5	70	.35	70	.35	70	.35	70
	990	5	70	.35	70	.35	70	.35	70
	1000	5	70	.35	70	.35	70	.35	70

TABLE 16. DIGESTIVE (AO<sub>2</sub>) AND ACETYLCHOLYLACETONIC (AMC) CONTENTS

OF SKIN MILK RIPENED WITH VARIOUS AMOUNTS OF ADDITIVE

MINIMAL OIL TO THE SAME PD

TABLE 17. DIACETYL ( $\text{Ac}_2$ ) AND ACETYL METHYL CARBINOL (Ame) CONTENTS OF SKIM MILK RIPENED WITH VARIOUS AMOUNTS OF ADDED BUTTER OIL TO THE SAME pH

Laboratory Trials							
Trial No.	Per cent butter oil	Per cent culture	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Ame p.p.m.
1	0	5	61°F.	.49	5.00	0.41	31.7
	10	4.5	61	.47	4.81	.97	43.2
	20	4	61	.45	4.89	1.18	44.9
	30	3.5	61	.45	4.73	1.66	47.4
	40	3	61	.40	4.90	1.74	49.9

TABLE 18. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Ame) CONTENTS OF CREAM, RIPENED TO VARIOUS ACIDITIES, AND OF THE CORRESPONDING UNSALTED BUTTER

Trial No.	Plant Trials								
	Cream			Butter					
	Type of cream	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Ame p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Ame p.p.m.
1	Swt.	53° F.	0.36	5.39	0.14	11.8	5.70	0.23	3.7
	Swt.	53	.42	5.23	.48	11.9	5.40	.18	3.1
	Swt.	53	.49	4.91	.54	15.3	5.21	.33	2.9
2	Swt.	53	.35	5.34	.43	17.9	5.61	.16	6.9
	Swt.	53	.40	5.26	.60	24.0	5.39	.28	6.1
	Swt.	53	.45	5.17	.72	38.5	5.23	.38	9.0
3	Swt.	53	.35	5.33	.46	20.4	5.64	.30	6.2
	Swt.	53	.42	5.12	.68	25.7	5.38	.38	6.8
	Swt.	53	.48	4.95	.97	30.1	5.29	.34	7.0
4	Swt.	50	.48	4.79	4.23	37.4	4.79	1.14	9.3
	Swt.	50	.52	4.73	4.80	101.4	4.77	.99	6.6
5	Swt.	52	.48	4.90	2.94	87.4	5.02	.71	8.2
	Swt.	52	.51	4.83	3.78	62.2	5.14	.62	6.5
6	Swt.	54	.35	5.33	1.19	28.9	5.54	.31	3.6
	Swt.	54	.40	5.15	1.23	32.5	5.23	.57	9.0
7	Swt.	55	.38	4.98	1.24	23.8	5.42	.49	6.0
	Swt.	55	.46	4.85	2.15	32.8	5.55	.60	7.0
8	Sr.	51	.31	5.35	1.55	62.0	5.44	.72	13.6
	Sr.	51	.35	5.26	1.95	72.4	5.83	.56	11.7
9	Sr.	After past.	.23	6.52	.21	21.7			
	Sr.	Plus cult.	.26	6.47	.28	34.0			
	Sr.	54	.32	5.92	.67	34.1	5.88	.35	8.0
	Sr.	54	.38	5.69	.76	31.7	5.72	.30	5.6
10	Sr.	After past.	.17	6.58	.56	29.8			
	Sr.	Plus cult.	.18	6.52	.62	36.0			
	Sr.	53	.37	5.29	1.88	64.7	5.32	.56	9.4
	Sr.	53	.49	4.98	1.49	28.8	5.31	.41	5.7

TABLE 19. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Amc) CONTENTS OF CREAM, RIPENED WITH AND WITHOUT ADDED CITRIC ACID TO THE SAME pH, AND OF THE CORRESPONDING UNSALTED BUTTER

Trial No.	Plant Trials									
	Cream						Butter			
	Type of cream	Per cent citric acid added	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.
1	Swt.	0	52°F.	.48	4.76	3.28	76.1	4.90	0.88	12.6
	Swt.	0.05	52	.51	4.70	4.44	108.7	5.04	.92	16.2
2	Swt.	0	50	.48	4.79	4.23	37.4	4.79	1.14	9.3
	Swt.	0.05	50	.53	4.71	4.10	21.6	4.87	1.27	13.5
3	Swt.	0	53	.42	5.29	1.50	46.2	5.32	.82	9.8
	Swt.	0.05	53	.44	5.24	2.28	59.0	5.42	.69	9.0
4	Swt.	0	51	.51	4.83	3.78	62.2	5.14	.62	6.5
	Swt.	0.05	51	.53	4.77	5.90	97.0	4.95	.84	10.6
5	Sr.	0	51	.31	5.35	1.55	62.0	5.44	.72	13.6
	Sr.	0.05	51	.34	5.20	1.67	62.0	5.72	.56	10.3
6	Sr.	0	54	.32	5.92	.67	34.0	5.88	.35	8.0
	Sr.	0.05	54	.35	5.55	.82	30.0	5.53	.35	6.8
7	Sr.	0	53	.37	5.34	1.40	39.3	5.62	.48	8.1
	Sr.	0.05	53	.37	5.31	1.60	33.2	5.53	.46	6.1

TABLE 20. DIACETYL ( $\text{Ac}_2$ ) AND ACETILMETHYLCARBINOL ( $\text{Amc}$ ) CONTENTS OF CREAM, RIPENED WITH AND WITHOUT AGITATION, AND OF THE CORRESPONDING UNSALTED BUTTER

Trial No.	Treatment of cream	Type temp.	Ripening time	Cream			Butter		
				Per cent solid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.
1	Control	Swt.	52° F.	0.48	4.76	2.28	76.1	4.90	0.88
	Agitated	Swt.	52	.48	4.80	5.90	164.0	4.99	1.43
2	Control	Swt.	54	.35	5.33	2.19	28.9	5.54	.31
	Agitated	Swt.	54	.33	5.35	2.30	67.6	5.39	.38
3	Control	Swt.	55	.38	4.98	1.24	23.8	5.42	.49
	Agitated	Swt.	55	.37	4.97	1.91	55.8	5.49	.70
4	Control	Sr.	55	.37	5.34	1.40	39.3	5.62	.48
	Agitated	Sr.	55	.36	5.41	2.12	78.0	5.63	.66
5	Control	Sr.	53	.49	4.98	1.50	28.8	5.31	.41
	Agitated	Sr.	53	.45	5.15	1.63	39.1	5.56	.62

TABLE 21. DIACETYL ( $\text{Ac}_2$ ) AND ACETILMETHYLCARBINOL (Amc) CONTENTS OF CREAM, RIPENED AT DIFFERENT TEMPERATURES, AND OF THE CORRESPONDING UNSALTED BUTTER

Plant Trials

Trial No.	Cream						Butter		
	Treatment	Type of cream	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.
1	Ripened at 70°F.	Swt.	.57	4.72	2.99	45.8	4.94	1.06	8.9
	Ripened at 62	Swt.	.55	4.85	2.74	42.6	4.89	.91	8.9
	Ripened at 55	Swt.	.55	4.95	2.57	36.8	5.09	.53	5.4
2	Ripened at 70	Swt.	.52	4.73	1.92	33.0	4.73	.93	11.1
	Ripened at 62	Swt.	.55	4.65	1.95	42.9	4.69	.51	7.9
	Special	Swt.	.42	4.94	2.34	114.6	4.98	1.46	24.0
3	Ripened at 70	Swt.	.36	4.95	.79	8.2	5.60	.56	5.6
	Ripened at 61	Swt.	.34	4.99	1.68	36.2	5.38	.60	7.9
	Special	Swt.	.36	5.18	2.39	82.2	5.76	.76	12.1
4	Ripened at 70	Sr.	.51	4.75	2.13	46.2	4.90	.70	11.1
	Ripened at 62	Sr.	.50	4.83	2.06	45.1	4.99	.71	11.3
	Special	Sr.	.50	4.79	4.10	190.0	5.25	1.37	20.3

TABLE 22. DIACETYL ( $\text{Ac}_2$ ) AND ACETYLMETHYLCARBINOL (Amc) CONTENTS OF CREAM, RIPENED WITH DIFFERENT BUTTER CULTURES, AND OF THE CORRESPONDING UNSALTED BUTTER

Plant Trials										
Trial No.	Cream							Butter		
	Culture No.	Type of cream	Ripening temp.	Per cent acid	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.
1	46	Swt.	55°F.	.41	5.17	1.68	40.0	5.16	0.58	7.2
	27	Swt.	55	.43	4.97	2.09	39.3	5.24	.74	8.9
	12	Swt.	55	.41	5.02	1.86	41.9	5.44	.67	9.5
2	46	Swt.	54	.49	4.64	3.40	101.2	4.74	.98	16.9
	27	Swt.	54	.49	4.62	4.22	86.4	4.69	1.06	15.5
	1B	Swt.	54	.50	4.55	4.90	122.2	4.59	1.03	18.6
3	15/1	Swt.	56	.36	4.89	1.07	28.6	5.35	.52	8.9
	14	Swt.	56	.36	4.84	1.43	28.5	5.23	.44	6.8
	12	Swt.	56	.30	5.34	1.27	22.0	5.90	.42	5.8
4	103	Sr.	53	.45	4.95	1.30	9.2	5.09	.45	3.6
	14	Sr.	53	.46	4.86	1.49	24.6	5.07	.65	6.8
	12	Sr.	53	.37	5.30	1.42	17.8	5.60	.39	4.3
5	46	Sr.	53	.46	4.83	2.38	72.6	5.15	.72	13.0
	27	Sr.	53	.46	4.82	2.36	49.1	5.09	.68	9.0
	1BP	Sr.	53	.45	4.84	2.40	41.6	5.23	.59	7.8

TABLE 23. DIACETYL ( $\text{Ac}_2$ ) AND ACETYL METHYL CARBINOL (Amc) CONTENTS OF RIPENED CREAM AND OF UNSALTED BUTTER HELD FOR VARIOUS PERIODS AT DIFFERENT TEMPERATURES

Trial No.	Plant Trials														
	Cream			Butter											
	Not held		Held 1 week at 36 to 40°F.		Held 3 days at 60°F. and 4 days at 36 to 40°F.		Held 30 days at 36 to 40°F.								
pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.	pH	$\text{Ac}_2$ p.p.m.	Amc p.p.m.				
1	5.29	1.50	46.2	5.32	0.82	9.8	5.28	0.81	10.7	5.12	0.84	11.8	5.25	1.14	13.1
2	5.24	2.28	59.0	5.42	.69	9.0	5.39	.94	16.3	5.10	.59	19.7	5.20	1.28	13.7
3	4.76	3.28	76.1	4.99	.88	12.6	4.99	1.10	14.7	4.92	1.19	15.5	5.13	.95	13.9
4	4.70	4.44	108.7	5.04	.92	16.2	4.82	1.02	18.4	4.68	1.38	21.5	4.82	1.37	19.4
5	4.80	5.90	164.0	4.99	1.43	23.1	4.94	1.86	25.2	4.78	1.89	27.8	4.88	1.80	25.0
6	4.79	4.23	37.4	4.79	1.14	9.3	4.79	1.41	11.5	4.76	1.36	11.7	4.81	1.39	10.9
7	4.71	4.10	21.6	4.87	1.27	13.5	4.85	1.42	13.1	4.77	1.42	15.2	4.85	1.50	13.5
8	4.73	4.80	101.4	4.77	.99	6.6	4.75	1.15	7.5	4.61	1.14	8.7	4.74	1.08	8.9
9	5.35	1.55	62.0	5.44	.72	13.6	5.52	.75	13.7	5.38	1.01	16.3	5.36	.79	12.1
10	5.20	1.67	62.0	5.72	.56	10.3	5.63	.56	9.8	5.43	.57	9.8	5.57	.65	8.9
11	5.26	1.95	72.4	5.83	.56	11.7	5.74	.60	10.1	5.73	.62	11.6	5.59	.70	10.0
12	4.90	2.94	87.4	5.02	.71	8.2	4.99	.16	5.6	4.60	.29	2.0	4.96	.44	4.0

TABLE 23. Continued

Trial No.	Cream						Butter					
	Not held			Held 1 week at 36 to 40°F.			Held 3 days at 60°F. and 1 day at 36 to 40°F.			Held 30 days at 36 to 40°F.		
	pH	$\text{AO}_2$	Amo	pH	$\text{AO}_2$	Amo	pH	$\text{AO}_2$	Amo	pH	$\text{AO}_2$	Amo
13	4.77	5.98	97.0	4.95	0.84	10.6	4.90	0.16	9.2	4.60	0.06	2.8
14	4.83	3.78	62.2	5.14	.62	6.5	5.09	.15	3.8	4.75	.04	2.2
15	5.35	1.19	28.9	5.54	.31	3.6	5.52	.54	5.6	5.33	.64	8.9
16	5.15	1.23	32.5	5.23	.57	9.0	5.25	.86	11.6	5.15	.96	13.8
17	5.35	2.30	67.8	5.39	.38	1.0	5.26	.60	5.9	5.12	.77	9.0
18	5.92	.67	34.0	5.88	.35	8.0	5.77	.66	13.6	5.49	.71	22.2
19	5.55	.82	30.0	5.53	.35	6.8	6.65	.59	9.1	5.60	.61	16.2
20	5.69	.71	31.7	5.72	.30	5.6	5.66	.58	9.6	5.20	.87	18.6
21	4.98	1.24	23.8	5.42	.49	6.0	5.28	.71	7.7	4.97	.87	16.2
22	4.85	2.15	32.8	5.55	.60	7.0	5.03	.84	9.5	4.77	1.05	18.2
23	4.97	1.91	55.8	5.49	.70	10.9	5.29	1.10	15.1	4.91	1.16	22.5
24	4.72	2.99	15.8	4.94	1.06	8.9	4.87	1.32	10.5	4.70	1.69	19.6
25	4.85	2.71	42.6	4.89	.91	8.9	4.81	1.26	10.8	4.65	1.39	13.3

TABLE 23. Continued

Trial No.	Cream						Butter					
	Not held			Held 1 week at 36 to 40°F.			Held 3 days at 60°F. and 1 day at 36 to 40°F.			Held 30 days at 36 to 40°F.		
	pH	As <sub>2</sub>	As <sub>2</sub> p.p.m.	pH	As <sub>2</sub>	As <sub>2</sub> p.p.m.	pH	As <sub>2</sub>	As <sub>2</sub> p.p.m.	pH	As <sub>2</sub>	As <sub>2</sub> p.p.m.
26	4.95	2.57	36.8	5.09	0.53	5.4	5.04	0.94	8.4	6.70	1.21	15.8
27	5.34	1.40	39.3	5.62	.48	8.1	5.47	.92	10.2	5.04	1.04	10.4
28	5.31	1.64	33.2	5.53	.46	6.1	5.56	.69	9.6	5.03	.61	4.0
29	5.41	2.12	78.0	5.83	.66	11.4	5.63	.99	11.3	5.10	1.14	11.8
30	4.75	1.93	30.0	4.73	.93	11.1	4.73	1.16	12.3	4.45	.83	4.0
31	4.65	1.95	12.9	4.69	.51	7.9	4.68	.93	11.7	4.49	1.26	15.3
32	4.94	2.34	114.6	4.98	1.46	24.0	4.91	2.16	19.3	4.77	2.14	20.4
33	5.29	1.88	64.7	5.32	.56	9.5	5.31	.60	9.8	4.93	1.20	27.1
34	4.98	1.49	28.8	5.31	.41	5.7	5.31	.56	5.2	5.18	.74	12.4
35	5.15	1.63	39.1	5.56	.62	11.6	5.19	.85	15.4	5.18	.87	26.2
36	4.95	.79	8.2	5.60	.60	5.6	5.34	.69	6.8	5.04	.93	12.3
37	1.68	36.0	5.38	.60	7.9	5.14	1.08	11.0	4.94	1.19	14.3	4.88
38	5.18	2.39	82.2	5.76	.76	12.1	5.68	.83	11.8	5.26	.93	5.27

TABLE 23. Continued

Trial No.	Cream						Butter					
				Held 1 week at 36 to 40°F.			Held 3 days at 60°F., and 4 days at 36 to 40°F.			Held 30 days at 36 to 40°F.		
	pH	Ao <sub>2</sub> p.p.m.	Amo p.p.m.	pH	Ao <sub>2</sub> p.p.m.	Amo p.p.m.	pH	Ao <sub>2</sub> p.p.m.	Amo p.p.m.	pH	Ao <sub>2</sub> p.p.m.	Amo p.p.m.
39	5.17	1.68	39.0	5.16	0.58	7.2	5.16	0.61	9.0	5.02	0.70	9.0
40	4.97	2.09	39.3	5.24	.74	8.9	5.19	.82	8.3	5.06	1.01	8.6
41	5.02	1.86	41.9	5.44	.67	9.5	5.31	.89	11.4	5.21	.98	13.5
42	4.83	2.38	72.6	5.15	.72	13.0	5.08	.95	17.8	5.01	1.64	36.0
43	4.82	2.36	49.1	5.09	.68	9.0	5.14	.85	11.1	4.95	1.22	18.3
44	4.84	2.10	41.6	5.23	.59	7.8	5.21	.79	8.2	4.94	.80	8.2
45	4.64	3.40	101.2	4.74	.98	16.9	4.74	1.12	20.1	4.49	1.38	22.4
46	4.52	4.22	86.4	4.59	1.06	15.5	4.57	1.24	11.1	4.39	.60	4.0
47	4.55	4.90	122.2	4.59	1.03	18.6	4.54	1.24	21.0	4.46	1.12	8.2
48	4.95	1.30	9.2	5.09	.45	3.6	5.18	.34	2.0	5.00	.32	2.0
49	4.86	1.49	24.6	5.07	.65	6.8	5.07	.54	6.9	4.85	.61	4.0
50	5.30	1.42	17.8	5.60	.39	4.3	5.70	.41	3.2	5.58	.29	1.6
51	4.75	2.13	46.2	4.90	.70	11.1	4.79	.84	9.9	4.66	1.08	10.7

TABLE 23. Continued

Trial No.	Cream						Butter					
	Not held			Held 1 week at 36 to 40°F.			Held 3 days at 60°F., and 4 days at 36 to 40°F.			Held 30 days at 36 to 40°F.		
	pH	As <sub>2</sub>	Ams	pH	As <sub>2</sub>	Ams	pH	As <sub>2</sub>	Ams	pH	As <sub>2</sub>	Ams
52	4.83	2.06	45.1	4.99	0.71	11.3	4.89	0.76	8.9	4.72	0.77	3.8
53	4.79	4.10	190.0	5.25	1.37	20.3	5.10	1.39	20.7	5.06	1.34	21.5
54	4.89	1.07	28.6	5.35	.52	8.9	5.32	.42	6.7	5.26	.50	9.6
55	4.84	1.43	28.5	5.23	.44	6.8	5.22	.55	5.4	5.14	.60	5.7
56	5.34	1.27	22.0	5.90	.42	5.8	5.95	.42	2.8	5.65	.62	3.6

TABLE 24. FLAVOR AND KEEPING QUALITY OF UNSALTED BUTTER FROM CREAM RIPENED WITH VARIOUS PROCEDURES

Plant Trials

Trial No.	Cream ripening procedure	Type of cream	Butter held several days at 36 to 40°F.			Butter held several weeks at 36 to 40°F.			Butter held several months at -10 to 0°F.		
			Days	Score	Days	Score	Days	Score	Days	Score	
1	Control	Srt.	10	92.50	32	91.00	217	90.50			
	Plus 0.05 per cent citric acid	Srt.	10	93.00	32	91.25	217	91.00			
2	Control	Srt.	8	92.50	30	91.00*	215	91.25			
	Agitated	Srt.	8	92.50	30	91.00*	215	92.25			
3	Plus 0.05 per cent citric acid	Srt.	8	93.00	30	91.00	215	92.00			
	Ripened to higher acidity	Srt.	5	92.25	35	91.75	212	91.25			
4	Plus 0.05 per cent citric acid	Srt.	5	92.50	35	92.25	212	91.75			
	Ripened to higher acidity	Sr.	12	90.50	32	90.00	239	91.25			
5	Plus 0.05 per cent citric acid	Sr.	12	90.50	32	90.00	239	91.50			
	Ripened to higher acidity	Srt.	10	91.75	36	91.50	237	91.50			
6	Plus 0.05 per cent citric acid	Srt.	10	91.50	36	91.75	237	92.00			
	Ripened to higher acidity	Srt.	10	91.75	36	92.25	237	92.25			
7	Agitated	Srt.	8	92.75	32	92.25	227	91.50			
	Ripened to higher acidity	Sr.	9	92.50	34	91.50	223	89.75			
	Plus 0.05 per cent citric acid	Sr.	9	92.50	34	92.25	223	90.50			

TABLE 24. Continued

Trial No.	Cream ripening procedure	Type of cream	Butter held several days at 36 to 40°F.			Butter held several weeks at 36 to 40°F.			Butter held several months at -10 to 0°F.		
			Days	Score	Days	Score	Days	Score	Days	Score	
8	Control	Srt.	6	92.25	36	92.00	220	90.50			
		Srt.	6	93.00	36	91.50	220	91.00			
		Srt.	6	92.50	36	91.75	220	91.50			
9	Cream ripened at 70°F. Cream ripened at 62 Cream ripened at 55	Srt.	19	91.75	37	91.50	218	89.00			
		Srt.	19	92.00	37	92.00	218	89.50			
		Srt.	19	91.25	37	91.75	218	91.50			
10	Control Plus 0.05 per cent citric acid Agitated	Sr.	6	91.00	35	91.25	216	90.00			
		Sr.	6	91.00	35	91.75	216	89.50			
		Sr.	6	91.00	35	91.75	216	90.25			
11	Cream ripened at 70°F. Cream ripened at 62 Special	Srt.	6	93.00	32	92.25	213	92.00			
		Srt.	6	92.50	32	92.75	213	91.00			
		Srt.	6	93.00	32	93.25	213	91.25			
12	Control Ripened to higher acidity Agitated	Sr.	9	91.25	31	90.75	211	89.75			
		Sr.	9	92.00	31	91.25	211	90.00			
		Sr.	9	91.75	31	91.50	211	90.75			
13	Cream ripened at 70°F. Cream ripened at 61 Special	Srt.	7	91.25	29	91.00	209	90.25			
		Srt.	7	92.00	29	91.75	209	90.75			
		Srt.	7	91.75	29	91.75	209	91.25			
14	Culture No. 27 used Culture No. 46 used Culture No. 12 used	Srt.	9	92.50	24	91.50	206	91.50			
		Srt.	9	92.25	24	91.50	206	91.25			
		Srt.	9	92.00	24	91.25	206	92.00			

TABLE 24. Continued

Trial No.	Cream ripening procedure	Type of cream	Butter held several days at 36 to 40°F.			Butter held several weeks at 36 to 40°F.			Butter held several months at -10 to 0°F.		
			Days	Score	Days	Score	Days	Score	Days	Score	Days
15	Culture No. 46 used	Sr.	8	92.75			202	92.00			
	Culture No. 27 used	Sr.	8	92.50			202	91.50			
	Culture No. HP used	Sr.	8	92.00			202	91.00			
16	Culture No. 27 used	Srt.	8	92.50			199	92.00			
	Culture No. 46 used	Srt.	8	92.50			199	91.75			
	Culture No. N used	Srt.	8	91.75			199	91.25			
17	Culture No. 12 used	Sr.	6	91.75			197	90.00			
	Culture No. 11 used	Sr.	6	91.50			197	90.00			
	Culture No. 103 used	Sr.	6	91.25			197	90.00			
18	Cream ripened at 70°F.	Sr.	7	92.00			192	91.00			
	Cream ripened at 61°F.	Sr.	7	92.00			192	90.25			
	Special	Sr.	7	91.50			192	91.50			
19	Culture No. 12 used	Srt.	7	93.00			188	90.75			
	Culture No. 14 used	Srt.	7	92.25			188	91.50			
	Culture No. 15/1 used	Srt.	7	92.75			188	91.00			

ACKNOWLEDGMENTS

The author sincerely appreciates the advice and valuable direction of Dr. B. W. Hammer in conducting these studies and his helpful suggestions in writing the manuscript; the facilities and opportunities provided by Dr. N. E. Fabricius for conducting the commercial churning; and many helpful suggestions from colleagues in the Department of Dairy Industry at Iowa State College.

LITERATURE CITED

1. Barniccoat, C. R.  
1935. Diacetyl in cold-stored butters.  
*Jour. Dairy Res.*, 6:397-406.
2. Barniccoat, C. R.  
1935. Determination of diacetyl and acetyl methyl carbinol.  
*Analyst*, 60:653-662.
3. Barniccoat, C. R.  
1937. Diacetyl in cold-stored butters. II.  
*Jour. Dairy Res.*, 8:15-30.
4. Boekhout, F. W. J. and Ott de Vries, J. J.  
1919. Aromabildner bei der Rahmsäuerung. *Centbl. Bakt. 2 Abt.*  
49:373-382.
5. Brewer, C. R., Werkman, C. H., Michaelian, M. B., and Hammer, B. W.  
1938. Effect of aeration under pressure on diacetyl production  
in butter culture. *Iowa Agr. Exp. Sta., Res. Bul.* 233.
6. Conn, H. W.  
1889. Bacteria in milk, cream, and butter.  
*Conn. (Storrs) Agr. Exp. Sta., Second Annual Report*, 1889:62.
7. Davies, W. L.  
1937. The development of aroma in butter.  
*XI. Milchwirtschaftlicher Weltkongress, Berlin, Wiss. Ber.*  
2:78-80.
8. Hammer, B. W.  
1920. Volatile acid production of *S. lacticus* and the organisms  
associated with it in starters. *Iowa Agr. Exp. Sta., Res. Bul.*  
63.
9. Hammer, B. W. and Bailey, D. E.  
1919. The volatile acid production of starters and of  
organisms isolated from them. *Iowa Agr. Exp. Sta., Res. Bul.*  
55.
10. Hoecker, W. H.  
1938. Diacetyl and acetyl methyl carbinol contents of butter  
made from cream ripened with various Streptococci.  
*Thesis, Iowa State College*.

11. Munsiker, O. F.  
1927. The butter industry, page 234.  
Published by the author, La Grange, Illinois.
12. Michaelian, M. B., Farmer, R. S., and Hammer, B. W.  
1935. The relationship of acetyl methyl carbinol and diacetyl to butter cultures. Iowa Agr. Exp. Sta., Res. Bul. 155.
13. Michaelian, M. B. and Hammer, B. W.  
1935. Studies on acetyl methyl carbinol and diacetyl in dairy products. Iowa Agr. Exp. Sta., Res. Bul. 179.
14. Michaelian, M. B. and Hammer, B. W.  
1936. The oxidation of acetyl methyl carbinol to diacetyl in butter cultures. Iowa Agr. Exp. Sta., Res. Bul. 205.
15. Minster, J. T.  
1932. Tests for the keeping quality of unsalted butter. Analyst, 57:615-621.
16. Mohr, W.  
1938. De l'arome du beurre. Lait, 18:743-758.
17. Mohr, W. and Wellm, J.  
1937. Der Diacetylgehalt in deutscher Butter und Einfluss des Herstellungverfahrens auf den Diacetylgehalt der Butter. XI. Milchwirtschaftlicher Weltkongress, Berlin, Wiss. Ber. 2:89-97.
18. Nelson, J. A. and Hammer, B. W.  
1935. Studies on butter culture organisms in butter. Jour. Dairy Sci., 16:375-385.
19. Prill, E. A. and Hammer, B. W.  
1938. A colorimetric method for the microdetermination of diacetyl. Iowa State College Jour. of Sci., 12:385-395.
20. Prill, E. A. and Hammer, B. W.  
1939. Production of diacetyl from citric acid in butter cultures. Jour. Dairy Sci., 22:67-77.
21. Prill, E. A. and Hammer, B. W.  
1939. Changes in the diacetyl and acetyl methyl carbinol contents during the manufacture of butter. Jour. Dairy Sci., 22:79-88.

22. Prill, E. A. and Hammer, B. W.  
1940. Changes in diacetyl and acetylmethylcarbinol contents of butter at various temperatures. Jour. Dairy Sci., 23:159-168.
23. Schmalfuss, H.  
1928. Diacetyl ein Stoffwechselprodukt. Ztschr. Angew. Chem., 41:847.
24. Slatter, W. L.  
1936. Changes in the acetylmethylcarbinol plus diacetyl content of butter. Nat. Butter and Cheese Jour., No. 20:20-24; No. 21:18-26.
25. Slatter, W. L. and Hammer, B. W.  
1937. Changes in the acetylmethylcarbinol plus diacetyl content of butter. Iowa Agr. Exp. Sta., Res. Bul. 211.
26. Storch, V.  
1919. Fortsatte undersøgelser over fremstillingen af syrevaekkere. K. Veterinær-og landbohøjskole, Landøkonomiske forsøgs laboratorium, Copenhagen. Beretning 102.
27. Taperneoux, A.  
1932. Le diacyle, parfum du beurre et de la margarine. Lait, 12:1043-1055.
28. van Beynum, J. and Pette, J. W.  
1939. The decomposition of citric acid by Betacoccus cremeris. Jour. Dairy Res., 10:250-266.
29. van Niel, C. B., Kluyver, A. J., and Derx, H. G.  
1929. Über das Butteraroma. Biochem. Ztschr., 210:234-251.
30. Virtanen, A.  
1937. The influence of oxygen on the formation of butter aroma. XI. Milchwirtschaftlicher Weltkongress, Berlin, Wiss. Ber. 2:121-123.
31. Wiley, W. J.  
1937. Effect of acidity on keeping quality of butter. Jour. Council Sci. Ind. Res., Australia, 10:327-332.
32. Wiley, W. J.  
1939. The oxidation of the fat of butter during cold-storage. Jour. Dairy Res., 10:300-309.

33. Wiley, W. J., Cox, G. A., and Whitehead, H. R.  
1939. The formation of diacetyl by starter cultures.  
Jour. Council Sci. Ind. Res., Australia, 12:232-238.