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Reducing Wastewater from Cucumber Pickling Process by Controlled Culture Fermentation

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REDUCING WASTEWATER FROM CUCUMBER PICKLING PROCESS BY CONTROLLED CULTURE FERMENTATION

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FOREWORD

When energy and material resources are extracted, processed, converted, and used, the related pollutional impacts on our environment and even on our health often require that new and increasingly more efficient pollution control methods be used. The Industrial Environmental Research Laboratory - Cincinnati (IERL-Ci) assists in developing and demonstrating new and improved methodologies that will meet these needs both efficiently and economically.

This report presents an evaluation of a modified cucumber pickling process using controlled culture fermentation as compared to the conventional natural fermentation process. At commercial scale the modified process produced a product equal to or exceeding that of the natural fermentation with a significant reduction in the quantity of salt used during fermentation. Further information on the subject can be obtained by contacting the Food and Wood Products Branch, Industrial Environmental Research Laboratory-Cincinnati.

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ABSTRACT

On a demonstration scale, the controlled culture fermentation process (CCF) developed by the U. S. Food Fermentation Laboratory was compared with the conventional natural fermentation process (NF) in regard to product quality and yield and volume and concentration of wastewaters. The experiments were conducted at the Perfect Packed Products Company, Inc., Henderson, North Carolina. Nine 800-gal. tanks were brined. In each case, weight of cucumbers, volume of water, and amounts of additives were recorded. pH, acidity, salinity, and temperature were closely monitored. After brining, brinestock quality was evaluated by a panel of experts from the US Food Fermentation Laboratory and the Heinz Company. The brinestock was then processed; spent brines and processing waters were collected. Volume and wastewater characteristics (salinity, BOD, N and P forms, residues) were determined for the waters and weight of brinestock was determined. The cucumbers were then packed using a conventional finishing procedure for whole dill pickles and hamburger dill chips. Yield of final product was determined. Acceptability of the finished products was evaluated by a panel.

Analysis of data indicates that the CCF produces a product of quality equal to or exceeding that of NF; that a reduction of the total dissolved solids load in the wastewaters was achieved; and that fermentation occurs more rapidly and predictably. Under the carefully controlled conditions of the experiment, the NF procedure produced brinestock of better quality than that usually achieved by this process, indicating that higher yields could be obtained with existing tankyard procedure if better control over tank condition and over salting schedules was maintained.

Experiments on recycling of spent brines indicate that in the coagulation-precipitation reconditioning procedures inactivation of undesirable enzymes is due to denaturation of the enzymes at high pH rather than to physical removal of the enzymes by flocculation. A combination of lime and sodium hydroxide at pH's above 10 produced a clear brine with little or no enzyme activity. Lower pH levels, or use of alum and polymers could achieve clarification of the brine but enzyme activity remained. Brine recycling studies also indicate that CCF spent brine cannot be reused directly unless the brine pH is raised above the brine's characteristic pH 3.2 - 3.5, which is too low for the desirable Lactobacilli.

NOTE: This report follows the prevailing canning industry practice of using the international system of units in the laboratory and U.S. units in the manufacturing operations. A table of conversion factors is included.

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ABBREVIATIONS AND SYMBOLS

BOD, BOD_5	5-day 20°C biochemical oxygen demand
°C	Degrees Celsius
CCF	Controlled culture fermentation
Cl^-	Chloride
COD	Chemical oxygen demand
g	Gram
gal	Gallon
i.d.	Inside dimension
JTU	Jackson Turbidity Unit
kg	Kilogram
l	Liter
lb	Pound
mg	Milligram
min	Minute
ml	Milliliter
N	Nitrogen
NF	Natural fermentation
o.d.	Outside dimension
P	Phosphorus
ppm	Parts per million
°S	Degrees salometer
SS	Suspended solids
TDS	Total dissolved solids
TKN	Total Kjeldahl nitrogen
TOC	Total organic carbon
TSS	Total suspended solids

CONVERSION FACTORS AND METRIC PREFIXES[a]

CONVERSION FACTORS

To convert from	to	Multiply by
Degree Fahrenheit (^{o}F)	Degree Celsius (^{o}C)	$t^{o}_{C} = 0.56\ (t^{o}_{F}-32)$
inch (in)	metre (m)	2.54×10^{-2}
foot (ft)	metre (m)	3.048×10^{-1}
gallon (gal)	metre^3 (m^3)	3.784×10^{-3}
bushel (bu)	metre^3 (m^3)	3.524×10^{-2}
3.8 gal/bu	0.408 m^3/m^3	1.0
grain (gr)	kilogram (kg)	6.48×10^{-5}
ounce (oz)	kilogram (kg)	3.11×10^{-2}
pound (lb)	kilogram (kg)	4.536×10^{-1}
gallons per minute (gal/min)	metre^3/second (m^3/s)	6.308×10^{-5}
ounces per gallons (oz/gal)	kilogram/metre^3 (kg/m^3)	8.218
pounds per ton (lb/ton)	kilogram/kilokilogram (kg/kkg)	4.643×10^{-1}
pounds per 1000 pounds (lb/1000 lb)	kilogram/kilokilogram (kg/kkg)	1.0
tons per year (ton/yr)	kilokilogram/year (kkg/yr)	9.074×10^{-1}
gallons per ton (gal/ton)	metre^3/kilokilogram (m^3/kkg)	4.17×10^{-3}

To convert from	to	Multiply by
pounds per gallon (lb/gal)	kilogram/metre^3 (kg/m^3)	1.1984×10^2
pounds per 1000 gallons (lb/1000 lb)	kilogram/metre^3 (kg/m^3)	1.1984×10^{-1}
cost per pound ($/lb)	cost/kilogram ($/kg)	4.536×10^{-1}
cost per gallon ($/gal)	cost/metre^3 ($/$m^3$)	2.642×10^2
cost per 1000 gallon ($/ton)	cost/metre^3 ($/kkg)	2.642×10^{-1}

METRIC PREFIXES

Prefix	Symbol	Multiplication factor	Example
kilo	k	10^3	2 kg = 2×10^3 grams
centi	c	10^{-2}	2 cm = 2×10^{-2} metre

[a]Standard for Metric Practice. ANSI/ASTM Designation: E 380-76$^{\epsilon}$, IEEE Std 268-1976, American Society for Testing and Materials, Philadelphia, Pennsylvania, February 1976. 37 pp.

ACKNOWLEDGMENTS

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Mr. Thomas A. Bell and Dr. Henry Fleming, U. S. Food Fermentation Laboratory, Raleigh, N. C., who participated in the development of the controlled culture fermentation process, assisted in the project in experimental design, evaluation of brinestock and product quality, and enzyme analysis. Their participation in this project was in great measure responsible for its successful completion.

Special thanks is extended to Mr. Lloyd Hontz, Vice-President, Mount Olive Pickle Company, for supplying cucumbers for laboratory scale experiments. Appreciation is also due Miles Laboratories Inc. which provided some of the cultures used in the project.

Ms. Mablelene Smith, Program Coordinator, gave freely of her time and contributed exceptional skills in the preparation of this manuscript for which we are deeply grateful.

SECTION 1

INTRODUCTION

GENERAL

The cucumber pickle has long been a popular item in the American diet, and its popularity is increasing as shown by the following per capita consumption figures from the USDA Statistical Reporting Service:

Year	lb/capita
1940	2.88
1950	4.62
1960	5.19
1970	7.60
1976	8.26

In terms of standard 24/303 cases, in 1976 74 million cases were sold for a total retail sales value of $593,982,000.

TYPES OF PACKS

There are two major types of cucumber pickle pack--cured and freshpack. Cured pickles undergo natural fermentation and storage in salt brines. During the storage period the salt concentration of the brine is very high (45-65% saturation) so the brined cucumbers must be partially desalted before being packed in vinegar solution. Fresh-pack pickles, on the other hand, are prepared from uncured, unfermented cucumbers, packed directly in vinegar solution, and heat-sterilized. Currently about 40% of the annual crop is made into freshpack products.

PICKLE PRODUCTION AND THE WASTEWATERS GENERATED

Fresh packing of cucumbers creates relatively small wastewater loads. The two major sources of wastewater are washwaters and pasteurizer cooling waters. These streams are typically low in BOD, COD, nutrients, and chlorides. Due to the seasonal availability of fresh cucumbers, these fresh pack products can only be made during the "green season."

On the other hand, cucumbers preserved by the ancient practice of pickling can be stored up to three years before final packing. The three major operations in production of cured pickles are (1) brining, (2) "processing" or freshening (desalting), and (3) finishing. In the first stage the green cucumbers, usually unwashed, are placed in a salt brine maintained at about 25-30 ^{o}S.* This salt concentration favors growth of indigenous lactic acid bacteria which convert the sugars diffusing out of the cucumbers into lactic acid, lowering the pH and further suppressing the growth of undesirable organisms. When the fermentation is complete the pH will be about 3.4 to 3.6 and most of the sugar will have been converted to lactic acid. After the active fermentation period, the level is raised to 45-65 ^{o}S to further suppress bacterial activity. In the low pH, high salt environment the cucumber tissue cures, i.e., it loses its opaque white appearance and becomes translucent. When the brinestock is ready for packing, it is removed from the spent brine and processed in fresh water to lower the salt content. The spent brine and desalting (or processing) waters are characterized by their low pH, high salt content, and high oxygen demand. The spent brine and desalting waters usually become wastewaters. Before or after processing the brinestock may be subjected to various treatments such as slicing, chipping, and dicing. Wastewater from these operations is also characterized by high salt concentration, low pH, and high organic load.

A detailed characterization of the pollutional characteristics of spent brines and other pickling wastewaters was conducted in a previous study (Little, Lamb, and Horney, 1976). A typical spent brine has the following characteristics:

Total organic carbon, mg/l	3,400
Suspended solids, mg/l	330
Total Kjeldahl nitrogen, mg/l	732
Total phosphorus, mg/l	87
Chlorides, g/l	111
pH	3.4

Spent brines represent a major source of the total load of wastewaters generated in pickle packing. They are the major source of the salt content.

*In the pickle industry brine strengths are expressed in terms of degrees salometer, measured with a hydrometer calibrated in percent saturation with respect to sodium chloride. A saturated salt solution would read 100^{o}S.

APPROACHES TO REDUCTION OF WASTES

Because of the difficulty and expense of removing salt from wastewaters, a number of approaches have been proposed for reducing the quantity of salt discharged in the final plant effluent. One approach is regeneration and reuse of the salt brines. Regeneration may be accomplished with chemical treatment (Popper, 1967; Geisman and Henne, 1973; Palnitkar and McFeeters, 1974; Little et al., 1976), heat treatment (McFeeters et al., 1978) or physical treatment such as reverse osmosis or molecular filtration (Little et al.,1976).

Another approach is adoption of lower salometer salt storage for the brinestock. Despite the heavy use of salt, natural fermentations have been described as "unrestricted, heterogeneous, highly complex, and variable," often leading to production of defective brinestock (Etchells et al.,1973). Lowered brinestock quality is especially apt to result from growth of yeasts or coliform-type bacteria in the brines.

Because of the problems and unpredictability of natural fermentations, the U.S. Food Fermentation Laboratory has developed the controlled culture fermentation (CCF) process (Etchells et al.,1973). This process was designed to minimize such brinestock defects as bloaters, poor texture, and off-tastes.

In brief, the CCF process involves removal or suppression of indigenous bacteria, followed by heavy inoculation with the desired lactic acid bacteria. During the resulting rapid fermentation, the sugars are rapidly consumed and high acidity is achieved. Because of the high acidity and the absence of undesirable organisms, the brinestock can be stored at 25 °S, requiring about half or less of the salt commonly used. The following calculations indicate the potential salt savings with this procedure:

At 65 °S, 16.25 lb of salt is required for each bushel of cucumbers;

At 25 °S, only 6.18 lb/bushel is required, a reduction of 10.07 lb/bushel (62% reduction)

Additionally, the lower salt concentration in the brinestock means that less salt will have to be removed by processing.

The CCF process had been extensively tested in the laboratory and in relatively small-scale tankyard studies. However, studies on a large scale, accompanied by assessment of the potential for reducing pollutional loads, had not been conducted.

This project was initiated in 1975. Three tankyard experiments were conducted during the 1976 green season. In each case, both CCF and NF (natural fermentation) tanks were set up. The raw product was weighed before brining and records of all additions of salt and other materials were kept. Fermentation progress was closely monitored, After completion of brining, quality and quantity of brinestock, spent brines, processing water, and finished products were compared.

OBJECTIVES OF PROJECT

This project addressed the following:

1. Demonstrate on a commercial scale the potential for reducing the salt and water required for brining and processing cucumbers by substitution of the controlled culture fermentation procedure for the current natural fermentation procedure.

2. Compare product quantity and quality of processed pickles produced by controlled culture fermentation with those produced in the conventional natural fermentation procedure.

3. Compare waste streams (volume, oxygen demand, residues, chlorides, N and P forms) under conventional and demonstration conditions.

4. Investigate the possibility of recycling CCF and NF brines both directly and after treatment.

5. Collect data from both natural and controlled fermentation on labor, chemical costs, capital costs, water use, wastewater characteristics, and product quality to enable an economic analysis of the financial feasibility of CCF.

6. Collect data for computation of a mass balance of salt (i.e., how much salt is retained in the product and sold, how much is required in brining, and how much salt is wasted).

SECTION 2

CONCLUSIONS

1. Controlled culture fermentations (CCF) proceed more rapidly and result in higher levels of acidity than do natural fermentations (NF). CCF fermentations were also more reproducible and produced consistently good brinestock.

2. Under the conditions tested, the volume of wastewater produced per unit weight of raw product was approximately the same. However, desalting of CCF brinestock required less time.

3. Products made from CCF were equal to or superior to those produced by NF. Whole dills were equal in taste and superior in appearance and texture to those from NF. Hamburger chips from brinestock from the two processes were similar.

4. Under the careful experimental procedures employed in this project, the brinestock and finished products from NF test tanks were of higher quality than those generally obtained on the tankyard, indicating that more attention to salting schedules, control of leaks, etc., could substantially reduce brinestock damage in the NF procedure.

5. Spent brine from the CCF tanks is substantially higher in acidity than that from NF tanks. It cannot be recycled directly because the low pH depresses growth of the desired lactobacilli. Adjustment to near neutral should be the only pretreatment necessary.

6. In the high pH procedure for regenerating spent brines, the removal of pectinase activity is due to denaturation of the enzyme by the elevated pH rather than to removal of the enzyme by coagulation-precipitation. Clarification of the brines by coagulation at neutral pH's failed to eliminate pectinase activity.

7. As expected, substantially less salt was required for CCF than for NF brinings; thus the salt concentration in the spent brines was lower.

SECTION 3

RECOMMENDATIONS

1. Adoption of the CCF procedure by pickle companies offers the following advantages:

   (1) Reduction of salt requirements

   (2) Less salt to be discharged as a waste

   (3) More rapid and consistent fermentation

   (4) Quicker desalting of brinestock per volume of processing water (or if desired, lower volume of processing water per a given desalting time)

   (5) Consistent production of brinestock equal or superior to that from NF

   (6) Less loss of brinestock due to bloater formation

2. If CCF is not adopted, the NF process can be managed to produce a much better quality and quantity of brinestock than is usually the case by careful attention to salting schedules and improved housekeeping practices.

3. CCF brines should be recycled after appropriate treatment to adjust the acidity and pH.

SECTION 4

EXPERIMENTAL DESIGN

APPROACH

The work plan for this project was directed to three major areas:

1. Large-scale demonstration of controlled culture fermentation (CCF) and comparison with conventional natural fermentation (NF).

2. Characterization of wastewaters generated.

3. Laboratory studies on recycling spent brines.

The large-scale demonstration was conducted at Perfect Packed Products, Inc., (PPP) Henderson, N. C., using 800-gallon tanks. Size 3 cucumbers, the size most commonly used for hamburger chips and whole dills, were used. PPP supplied cucumbers, water, salt, and acetic acid, as well as labor for tanking, heading, routine monitoring of fermentation, untanking, processing, and packing. Three experiments were conducted. In each experiment, half the tanks were brined conventionally and the other half according to the CCF process. Progress of fermentations was closely monitored by PPP, North Carolina Agricultural and Technical State University (A&T), and University of North Carolina at Chapel Hill (UNC-CH) personnel.

After brining, brinestock quality and quantity were determined in accordance with procedures developed by the U.S. Food Fermentation Laboratory. The brinestock was then processed into hamburger dill chips and whole dills.

Spent brines and desalting waters were collected for determination of quantity and quality. A&T University personnel determined the concentration of BOD_5, COD, TDS, TSS, TKN, TP, and other wastewater parameters. These brines were also used in brine recycling experiments.

After packing and storage, the quality of the finished products was evaluated by a taste panel.

MATERIALS

The raw materials used in this project were, where possible, those in general use on the commercial scale, since a major goal of the project was

to compare feasibility of natural and controlled culture fermentation processes under actual tankyard conditions.

Cucumbers

Fresh cucumbers were obtained from those received by Perfect Packed Products, Inc. They were graded by PPP routinely into three to four major size categories. Size 3 (1½-2 inches in diameter) cucumbers were used for all experiments as it was felt that the large size would be the most likely to bloat and would thus provide the most stringent test of the CCF procedure.

Salt

The salt used was that in common use at the plant. It was rock salt of a grade suitable for brining food products.

Cultures

Cultures of Lactobacillus plantarum were employed in controlled culture fermentation. L. plantarum, an exceptionally acid-tolerant organism capable of rapid fermentation, has been recommended for the CCF procedure (Etchells et. al., 1973). Starter cultures were obtained from Chr. Hansen's Laboratory, Inc., Milwaukee, Wisconsin, and from Miles Laboratories, Inc., Madison, Wisconsin. L. plantarum is homofermentative, producing primarily lactic acid and a small amount of CO_2. It tolerates salt levels as high as 26-28 °S and pH as low as pH 3.2. Cultures were shipped in dry ice and stored at -70 °C.

Inert Gas

Compressed nitrogen gas (Air Products Co.) was used to purge controlled culture tanks.

Tanks

Tanks were fiberglass coated wooden tanks of 800-gallon capacity, 80"-83" I. D. and 37 3/4" tall. They were fitted with stainless steel sample ports and drains. The tanks were headed using plastic netting and widely spaced boards drilled with holes to facilitate gas escape.

Sparging Apparatus for CCF Tanks

Each CCF tank was provided with equipment through which nitrogen gas could be introduced to sweep CO_2 out of the brine. The sparging equipment was located at the bottom of the tank and consisted of a single coil (3.5' in diameter) of perforated plastic tubing (polyvinyl chloride tubing, Hi-Mol, Carlon Products, Wilton, Conn.), 1.3 cm O. D. In each coil, 12 equally spaced perforations (1/64" I. D.) were made with a special drill and bit. The two ends of the coil were attached to a tube through which compressed nitrogen gas was supplied. The coil was attached to a plywood board which was weighted down with bricks coated in parafin. The whole unit was placed in the bottom of the tank prior to filling. The gas flow rate was controlled with

a rotameter. Initial flow rate during the early rapid fermentation was 2400 ml/min (3 ml/min/gal). Flow rate was decreased when CO_2 levels remained consistently low (<15 mg CO_2/100 ml) for two consecutive readings. Flow rate was decreased stepwise (2000 ml/min, 1600 ml/min), then maintained at 800 ml/min until fermentation was completed.

METHODS FOR BRINE AND WASTEWATER ANALYSES

Acidity

Determination of percent titratable acidity (expressed as lactic acid) was performed as follows: (1) pipet 10 ml of brine sample into titration flask, (2) add two drops of phenolphthalein indicator, (3) titrate with 0.1N NaOH to a permanent pink end point, (4) multiply the ml of titrant required by the factor 0.09 to give percent lactic acid by volume.

$$\frac{\text{ml 0.1N NaOH x 0.09 x 100}}{\text{10 ml brine sample}} = \text{percent lactic acid by volume}$$

This determination was specified by Mr. T. A. Bell of the U.S. Food Fermentation Laboratory and is in common use in the pickle industry.

pH

For the plant studies, pH of samples was measured electrometrically with a Fisher Accumet pH Meter standardized with two buffers at pH's bracketing the pH of the test sample (APHA et al., 1976, Method 424). For laboratory recycling studies, Leeds and Northrup or Beckman pH meters were employed.

Salometer

In the pickle industry brine strengths are expressed in terms of degrees salometer (^{o}S), as measured with a hydrometer calibrated in percent saturation 100 ^{o}S. For this project, salometer readings were performed with a common salometer routinely used on the tankyard. A salt solution conversion chart is shown in Table 1 .

Chloride

Chloride determinations were performed with a specific ion probe (Orion) and a Fisher Accumet pH meter, according to the directions provided by Orion.

Turbidity

In laboratory recycling studies, turbidity was determined with the aid of a commercial turbidimeter (Hach, Model 2100) using directions supplied by the manufacturer. This procedure was consistent with Method 214A (APHA et al., 1976).

TABLE 1. SALT (NaCl) SOLUTION CONVERSION CHART*

Degrees Salometer	Percent salt by weight	Grams of salt per 100 cc. water by vol.	Grams of salt per 100 cc. of solution	Specific gravity $20^{o}/4^{o}$	Ounces of salt per gallon of water	Pounds of salt per gallon of water	Ounces of salt per gallon of solution	Pounds of salt per gallon of solution
0	0.00	0.00	0.00	0.998	0.00	0.000	0.00	0.000
5	1.32	1.34	1.33	1.007	1.78	0.111	1.77	0.111
10	2.64	2.71	2.68	1.017	3.61	0.226	3.57	0.223
15	3.96	4.12	4.06	1.026	5.49	0.342	5.41	0.338
20	5.28	5.58	5.47	1.036	7.44	0.465	7.29	0.455
25	6.61	7.08	6.91	1.046	9.44	0.590	9.20	0.575
30	7.93	8.62	8.36	1.055	11.5	0.717	11.12	0.696
35	9.25	10.2	9.85	1.065	13.6	0.850	13.11	0.820
40	10.6	11.8	11.4	1.074	15.7	0.984	15.2	0.950
45	11.9	13.5	12.9	1.084	18.0	1.12	17.2	1.07
50	13.2	15.2	14.5	1.094	20.2	1.26	19.3	1.21
55	14.5	17.0	16.0	1.104	22.6	1.41	21.3	1.33
60	15.8	18.8	17.6	1.14	25.1	1.56	23.4	1.46
65	17.2	20.7	19.3	1.125	27.6	1.73	25.7	1.61
70	18.5	22.7	21.0	1.135	30.3	1.89	28.0	1.75
75	19.8	24.7	22.7	1.145	32.9	2.06	30.2	1.89
80	21.1	26.8	24.4	1.156	35.7	2.23	32.5	2.03
85	22.2	28.5	25.9	1.166	37.9	2.37	34.5	2.16
90	23.8	31.2	28.1	1.179	41.6	2.60	37.4	2.34
95	25.1	33.5	29.9	1.191	44.6	2.79	39.8	2.49
100	26.4	35.9	31.7	1.202	47.8	2.99	42.2	2.64

*From USDA Food Fermentation Laboratory

Metals

In laboratory recycling studies, metals were measured by atomic absorption spectrophotometry. Brine samples were collected in acid-washed glassware, diluted with deionized water, and acidified with concentrated nitric acid. Analyses were performed by the UNC-CH Limnology Laboratory using a Perkin-Elmer Model 303 atomic absorption spectrophotometer according to the manufacturer's specifications as described in the manual "Analytical Methods for Atomic Absorption Spectrophotometry."

Chemical Oxygen Demand (COD)

COD was measured by the dichromate reflux method (APHA et al., 1976, Method 508) using appropriate addition of mecuric sulfate to compensate for chloride interference.

Biochemical Oxygen Demand (BOD)

Five-day BOD was determined according to APHA et al. (1976), Method 507. To ensure a sufficient concentration of microorganisms in the sample, the normal flora in the sample were supplemented by addition of seed from a municipal wastewater source.

Residue

Residues were determined according to APHA et al. (1976). Total suspended solids (SS) were determined by Gooch crucible filtration followed by drying at 103 ^{o}C and weighing of the residue (Method 208D). Total dissolved solids (TDS) were measured gravimetrically after evaporation at 103 ^{o}C (Method 208C).

Carbon Dioxide

Carbon dioxide was estimated by a modification of the Harleco procedure (Fleming et al., 1974), using the Harleco CO_2 Apparatus.

Phosphorus

Total phosphorus (TP) measurements were performed according to the automated ascorbic acid method (EPA, 1974), using a Technicon Autoanalyzer. All standards and samples were run in duplicate. Prior to analysis, brines were digested by addition of ammonium persulfate and concentrated sulfuric acid, followed by autoclaving.

Nitrogen

Total Kjeldahl nitrogen determinations were performed according to EPA procedures, (EPA, 1975) using an automated phenolate procedure to determine ammonia. Prior to analysis, samples were digested by addition of a mercuric sulfate - sulfuric acid - potassium sulfate mixture followed by block digestion at 370 ^{o}C.

EVALUATION OF QUALITY OF BRINESTOCK AND FINISHED PRODUCTS

Sample Procedure for Brinestock

Representative samples of brinestock were obtained by passing a lucite cylinder (1 ft I. D.) to the bottom of the tank and removing all the brinestock therein by netting. Two cores were removed from each tank.

Evaluation of Brinestock for Bloater Damage

Brinestock cucumbers were cut longitudinally and examined for balloon, honeycomb, lens, and other types of bloaters (Etchells et al., 1968). Subjective evaluations were based on type and severity of the defect (Fleming et al., 1973a).

Brinestock Texture

For each core, firmness of 20 pickles was measured by the USDA Fruit Pressure Tester with 5/16 inch tip. Firmness is expressed in terms of pounds resistance of the pickle. A rating of 18 and above indicates a very firm pickle; 14-17, firm; 11-13, inferior; 5-10, soft; and below 5, mushy. A firm pickle is considered desirable.

Evaluation of Finished Products

Experimental brinestock was processed and packed as hamburger dill chips or whole dills in 5-gal plastic pails. After storage, selected pails were coded and sampled for evaluation by a taste panel consisting of project personnel, industry personnel, and U.S. Food Fermentation Laboratory personnel. The products were rated on a 10-point scale for appearance, taste, and texture.

PROCEDURES FOR FERMENTING CUCUMBERS

Controlled Culture Fermentation Procedure

The CCF procedure was that suggested by Etchells et al. (1973). It is outlined in Table 2. In-tank shrinking was employed. Chlorination was achieved by addition of calcium hypochlorite (Lo-Bax®, Olin Corporation). The only exception to the published procedure was omission of the second chlorination 10-12 hours before inoculation. It was assumed that sufficient sanitizing was provided by the washing and initial chlorination and that further chlorination after the cucumbers had been soaking in the brine might cause production of undesirable chlorinated organic compounds.

Natural Fermentation Procedure

The NF procedure was that commonly used by the pickle company. It is outlined in Table 3.

TABLE 2. CONTROLLED CULTURE FERMENTATION PROCEDURE
(Adapted from Etchells et al., 1973)

1.	Raw product receiving	-	Visual inspection. Grade out moldy and diseased cucumbers.
2.	Washing	-	Remove field soil with reel-type washer.
3.	In-tank	-	Place in clean sanitized tank containing a 10-12 inch deep cushion of 25 °S brine containing about 20 mg/liter Cl_2. Use a 65:35 (wt/wt) ratio of cucumbers to brine.
4.	Coverbrine	-	Add chlorinated 25 °S brine to a level about 4-6 inches above the headboard. Circulate tank to allow shrinkage of cucumbers.
5.	Covering and heading	-	Head with plastic netting and widely spaced boards with holes for gas escape.
6.	Acidification and circulation	-	Acidify with acetic acid (vinegar) at the rate of 6 ml of glacial acetic acid per gallon of brined material.
7.	Salt additions	-	Add required salt to equalize at 25 °S.
8.	Acetate addition	-	Add 0.5% sodium acetate (18.8 g/gal) about 2-3 hours before culture addition.
9.	Culture addition	-	Add lactic acid bacteria, Lactobacillus plantarum, 2-3 hours after acetate addition.
10.	Purging action	-	As soon as the tank is headed, brined, and acidified, purge the dissolved CO_2 from the brine with N_2 gas. Test brine for CO_2, sugar, and acidity. Discontinue purging when fermentation is complete.

(continued)

TABLE 2 (continued)

11. Maintaining brine strength	-	Hold at 25 °S by additions of salt as necessary.
12. Quality of stock	-	Examine brinestock for firmness, bloaters, color, and cure.

Figure 1. Photograph of experimental tanks.

TABLE 3 . NATURAL FERMENTATION PROCEDURE

1.	Raw product receiving	-	Visual inspection. Grade out moldy and diseased cucumbers.
2.	In-tank	-	Place cucumbers in clean tank containing a 12" cushion of 30 °S brine. Use a 65:35 (wt) ratio of cucumbers to brine.
3.	Covering and heading	-	Add 30 °S cover brine. Circulate to allow cucumbers to shrink. Head with plastic netting and widely spaced boards with holes for gas escape.
4.	Salt additions	-	Add 1.25 lb. salt per bushel of cucumbers on head of tank after covering with 30 °S brine. On the second morning, check salt readings at bottom and top of tank. If readings are under 25 °S, add second day salt addition of 1 lb./bu. If readings are $\geq$ 25 °S, recheck salometer at end of day. If reading is $<$ 25 °S, add required salt for second addition. On the third morning, check salt readings at top and bottom of tanks. If $<$25 °S, add 0.5 lb. salt/bu. If >25 °S, recheck in afternoon and adjust. Check on a daily basis until fermentation is complete. Adjust as necessary to maintain 25 °S.
5.	Increasing brine strength	-	When 0.6% lactic acid acidity has been achieved, start feeder salt. Raise tanks ~3 °S weekly by addition on the head of 0.6 lb. salt/bu. Continue addition to 40 °S, or to 45 °S if tanks will be held over the winter.
6.	Quality of stock	-	Examine brinestock for firmness, bloaters, color, and cure.

EXPERIMENTS ON BRINE RECYCLING

A major consideration in brine recycling is the possibility of carryover of pectinases, the enzymes responsible for softening of brinestock. A secondary consideration is the carryover of suspended solids which represent microorganisms and debris. Recycling studies were chiefly addressed to removal of pectinases and turbidity from the brines. An additional study involved actual recycling, on a bench-scale, of brine collected in the demonstration phase of the project. Much of the work on recycling has been presented as a master's report (Wendle, 1977).

The laboratory investigations involved three major experimental procedures: (1) jar testing to determine feasibility of enzyme and turbidity removal by coagulation-sedimentation processes, (2) analysis of enzyme activity in treated and untreated brines to determine the extent of the enzyme problem and to evaluate success of treatment, (3) acidity titrations with real and synthesized brines to predict the amount of alkali needed for pH adjustment and to determine fate of metals during brine clarification.

Jar Test Methods

Jar tests were conducted with the aid of a six-paddle gang stirrer, using 200 ml aliquots of brine in 250 ml beakers. After addition of coagulants or coagulant aids, the samples were flash-mixed at 100 rpm for 30 seconds. The samples were then flocculated at 30 rpm for 30 minutes. While jar tests generally allow for settling of 30 min to 1 hr, settling time was not considered critical in these experiments since full-scale treatment of brine at pickle plants is likely to be a batch operation. Consequently, sedimentation was allowed to continue until floc had fully settled (a minimum of 30 min) or for a maximum of 24 hr. Supernatant was removed with a pipette. In most cases, jar tests were preceded by preliminary tests in which visual analysis was sufficient to determine the potential of chemical additions to provide clarification at a reasonable dosage and thus indicate if further investigation was warranted.

A slight variation of the above procedure was used when evaluating coagulation by pH adjustment. Because the spent brines contained such metal species as aluminum, calcium, and magnesium, the corresponding metal precipitates formed at pH levels above pH 7. In investigations of this effect, pH was adjusted with sodium hydroxide (NaOH) or with a lime slurry (CaO and water) during rapid mixing on a magnetic stirrer. When the sample reached the desired pH, it was placed on the gang stirrer, flocculated at 30 rpm for 30 min, and allowed to settle for approximately an hour.

Sample brines were obtained from actual spent brines collected from the tankyard of Perfect Packed Products. Spent brines from brining Number 2 size cucumbers were used, since this size is fairly small and thus more likely to be associated with pectinase enzyme. However, in most cases the brines did not contain significant pectinase enzyme activity. Therefore, for the purposes of experimentation, the brines were "spiked" with Pectinol, a commercial polygalacturonase available from Rohm & Haas. The product, supplied in powder form, was preserved by storage in a freezer. For

experiments, a stock solution was prepared by dissolving the powder in distilled water; the solution was preserved by refrigeration and addition of toluene.

A variety of coagulants and coagulant aids were tested:

(1) Sodium aluminate. Both reagent grade sodium aluminate and a commercial liquid sodium aluminate preparation (Nalco 2) were evaluated. Stock solutions of each were prepared by dilution with distilled water.

(2) Aluminum sulfate. A stock solution was prepared from $Al_2(SO_4)_3 \cdot 18\ H_2O$ to give a concentration of 100 g/l.

(3) Clay. Nalco 8151, a slightly anionic bentonite supplied in solution, was added directly as obtained.

(4) Polymers. Polymers evaluated included Nalco 7144 A and Dow A-21, high molecular weight anionic polymers, and Dow C-31, a high molecular weight cationic polymer.

(5) Sodium hydroxide. Stock solutions were prepared from reagent grade NaOH and distilled water.

(6) Lime. Lime was added as a slurry prepared from reagent grade CaO and distilled water.

Enzyme Activity Analysis

Enzyme analyses were performed according to U.S. Agricultural Research Service recommendations (Bell Etchells, and Jones, 1955). This procedure is based on indirect determination of enzyme activity via measurement of the change in viscosity of a 1.2% sodium polypectate solution. In the presence of pectinase enzyme, the viscosity is decreased in proportion to the amount of enzyme activity present.

Samples for enzyme analysis were collected by pipette, placed in 20 ml screw-top test tubes, preserved by addition of 1-3 drops of toluene, and refrigerated until dialysis.

Dialysis was performed to reduce interference from chlorides, which cause gelling of pectate solution and thus interfere with viscosity changes (Bell, Etchells, and Jones, 1955). Dialyses were performed with seamless cellulose dialysis tubing (Fisher 8-667C 1974). Samples were immersed for 3 hr in a continuous-flow tapwater bath. The bath was then drained and refilled with distilled water, in which the samples were immersed for a minimum of an hour. Samples were then transferred to screw-top test tubes and refrigerated until analysis.

Enzyme activity was measured in an Ostwald-Fenske viscometer as specified. Loss of viscosity was calculated and pectinase enzyme activity was determined as specified.

The recommended procedure was modified in that only 20 hr viscosity determinations were made, whereas, the protocol states that if a 50% loss in viscosity does not occur within the first 20 hr, then a 44 hr determination should be made. Within the scope of this project, it was felt that the additional time and effort needed for 44 hr tests was unwarranted, since preliminary tests indicated that the 20 hr tests were somewhat conservative and yielded the same or slightly higher readings than the 44 hr tests.

Acidity Titrations

To determine acidity and buffer capacity, acidity titrations were performed using 50 ml sample aliquots of spent brines. Titrations were generally completed at pH 11, the pH generally recommended for coagulation with bases (Little et al., 1976). Further titrations were conducted on synthetic brines which were prepared to simulate spent brines in terms of acidity and aluminum, magnesium, and salt concentration. Titration curves generated with these synthetic brines were compared with those from actual brines to help identify the processes responsible for precipitation. Sodium hydroxide solution (0.1 N, Fisher Scientific Co.) was used for all acidity titrations. Synthetic brines were prepared with distilled water and the following reagent grade chemicals: sodium chloride, concentrated lactic acid, aluminum sulfate, magnesium chloride. The resulting brine was 35 °S with an acidity of 0.1 M as lactic acid, containing 70 mg and 500 mg $Al_2(SO_4)_3 \cdot 18\ H_2O$ per liter.

Analytical Methods

Turbidity was measured with the aid of a Hach turbidimeter, Model 2100. pH was determined electrometrically with a Leeds & Northrup or Beckman pH meter.

For metal analyses, samples were collected in acid-washed glassware, diluted with deionized water, and acidified with concentrated nitric acid. Analyses were performed with the aid of a Perkin-Elmer atomic absorption spectrophotometer, Model 303, according to the manufacturer's instructions as described in "Analytical Methods for Atomic Absorption Spectrophotometry."

Bench-scale Recycling Studies in 5-Gal Pails

Limited recycling studies with spent brines were conducted. Fresh cucumbers (no. 3) were obtained from a nearby commercial pickling plant. They were packed into 5-gal plastic pails and "headed" with a rigid plastic grid. The brinings were conducted in the laboratory at 19-22 °C; throughout the test the pails were held under constant ultraviolet lighting from a bank of germicidal lamps, in order to minimize surface growths. Six pails were brined according to the NF procedure (Table 3). Two pails (1 and 2) served as controls and received the usual new brine (30 °S). Pails 3 and 4 were brined with NF spent brine (Tank 6, Expt. 3) diluted to 30 °S; the brine for pail 3 was treated with the high pH method before recycle, while pail 4 received untreated brine. Pails 5 and 6 were brined with undiluted, untreated brine from CCF fermentation (Tank 5, Expt. 3); initial salometer was 25 °S. Temperature, salometer, acidity, and sugar content were monitored; brinestock was evaluated at completion of the study. Sugar in brines was measured with a kit(Diastix®).

SECTION 5

RESULTS AND DISCUSSION

COMPARISON OF NATURAL AND CONTROLLED CULTURE FERMENTATIONS

During this project, three plant-scale experiments were conducted. In experiment I, initiated June 23, 1976, two tanks were brined, one by natural fermentation (NF) and one by controlled culture fermentation (CCF). In experiment II, initiated July 10, 1976, four tanks were brined, two by NF and two by CCF; in experiment III, August 3, 1976, one by NF and two by CCF.

Day to day measurements on the brines are shown in the Appendix in tabular and graphic form. Figure 2 shows the progress of fermentation in the tanks as indicated by the production of acidity. In the case of duplicate tanks, the average value is shown. The median afternoon brine temperatures were 80.5 ^{o}F (expt. I); 84.2 ^{o}F (expt. II); and 84.8 ^{o}F (expt. III). The initial acidity was higher in the CCF tanks due to addition of acetic acid, as directed in the CCF procedure. Note that rate of acid production was generally more rapid in CCF tanks and that higher final acidities were attained in these tanks. This indicates the desired high activity of the bacteria used for the inoculum, and it also indicates that fermentation time can be shortened by use of the CCF procedure.

In each case, the brinestock produced in the vats was evaluated for texture and for defects, especially those due to bloating. Overall acceptability and determination of usable brinestock was computed by two different numerical systems, one developed by Fleming et al. (1977) and one developed by S. D. Rubin of Perfect Packed Products. A summary of the evaluation is shown in Table 4, which indicates that CCF brinestock typically had a lower bloater index. It is also apparent that the quality of the CCF brinestock was much more consistent and predictable than that of NF brinestock, despite the fact that the NF experimental tanks received much more attention than would a tank in the typical tankyard.

The brinestock from the experimental tanks was processed into hamburger dill chips and into whole dill pickles. Sample packs from the first two experiments were evaluated by a six-member panel made up of USDA, Perfect Packed Products, and A & T personnel. Figure 3 indicates that in terms of appearance, taste, and texture, chips prepared by NF and CCF brinings were similar in quality and compared well with those produced commercially. Figure 4 indicates that appearance and texture of whole dills prepared from CCF were better than those from NF, while taste was similar.

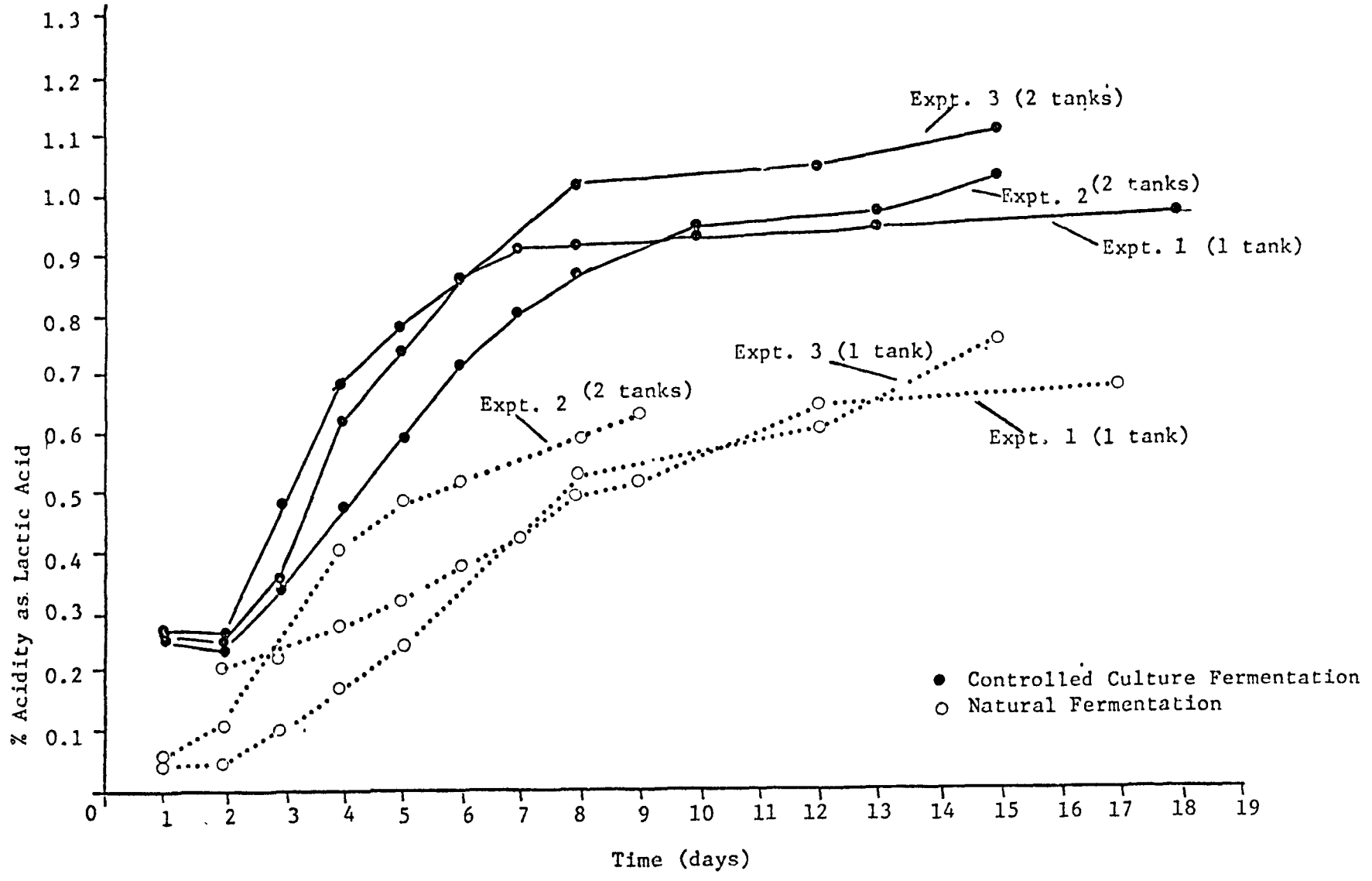


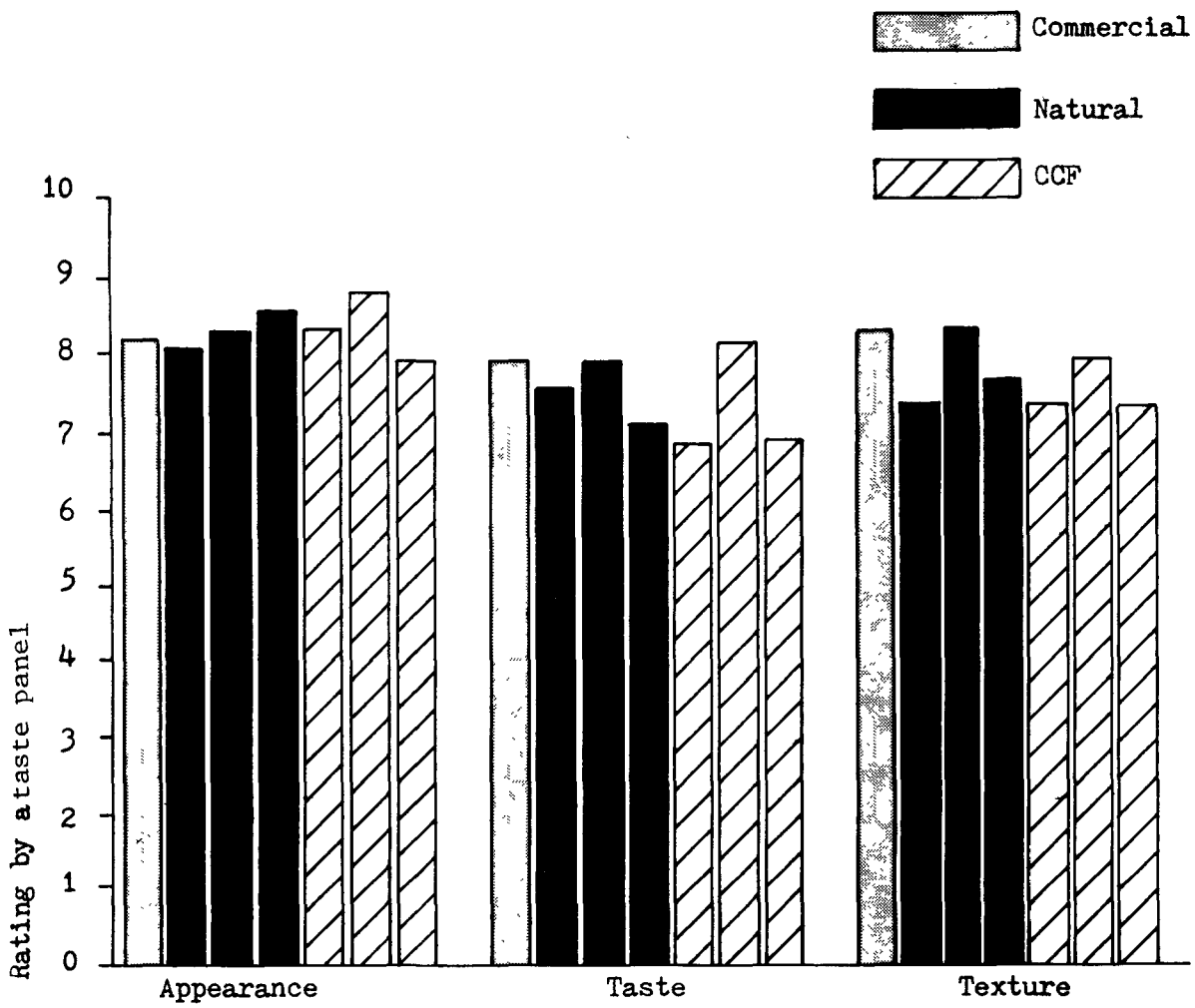
Figure 2. Comparison of lactic acid production in controlled culture and natural fermentations (three experiments).

TABLE 4. EVALUATION OF BRINESTOCK FROM NATURAL FERMENTATIONS AND FROM CONTROLLED CULTURE FERMENTATIONS

Expt.	Pressure test		Examination for freedom from defects		
	#Tested	Average (lb)	# Examined	Estimated yield (%)[1]	Bloater index[2]
Natural fermentations					
I	40	16.7	200	82.4	13.8
II	40	20.0	200	97.0	1.1
	40	17.0	200	96.0	1.6
III	40	20.5	160	78.0	17.2
Average		18.6		88.4	8.4
Controlled culture fermentations					
I	40	17.8	200	97.5	2.1
II	40	19.5	200	93.0	1.9
	40	18.5	190	92.5	1.2
III	40	21.5	200	97.5	1.0
	40	22.0	200	95.5	2.3
Average		19.9		95.2	1.7

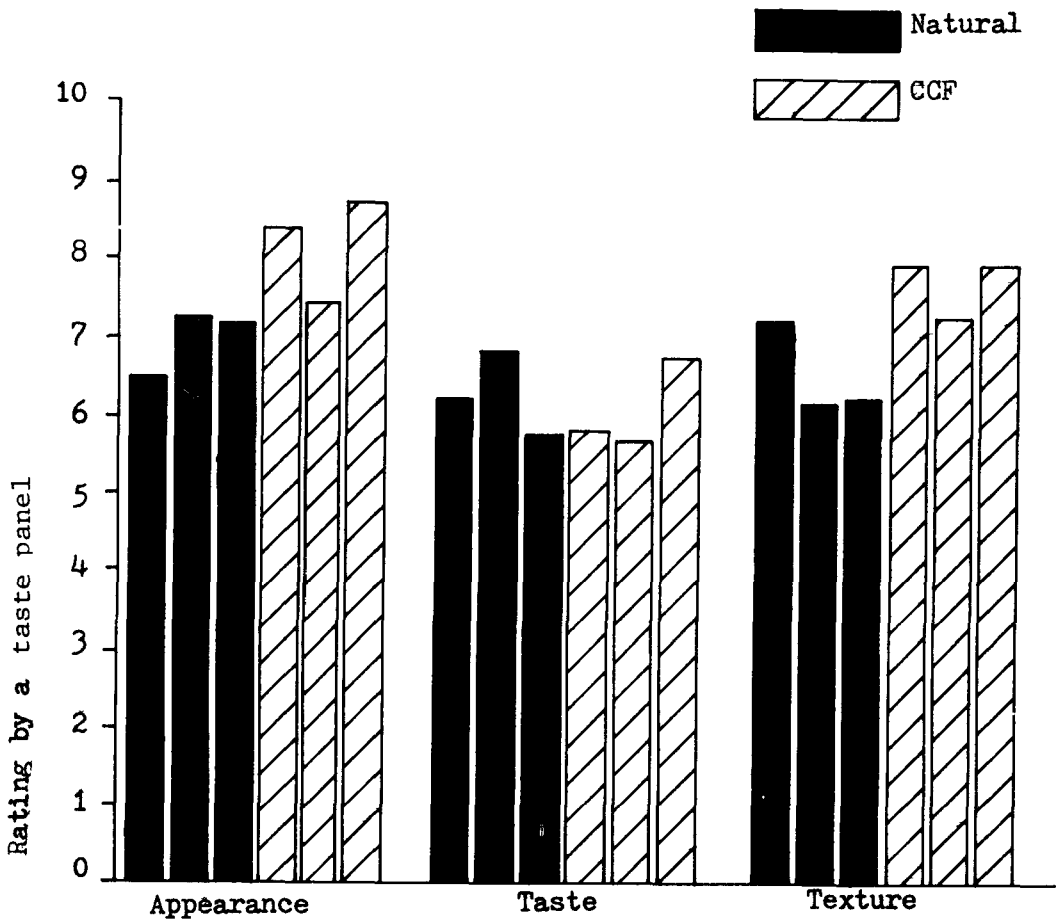
[1]Using system devised by S. D. Rubin, Perfect Packed Products, Inc.

[2]Fleming et al. (1977)



Average ratings:	Appearance	Taste	Texture
NF	8.2	7.4	7.6
CCF	8.3	7.2	7.4

FIGURE 3. Evaluation of hamburger chips prepared by natural fermentation (Including a commercial product) and by controlled culture fermentation.



Average ratings:	Appearance	Taste	Texture
NF	7.0	6.3	6.9
CCF	8.2	6.1	7.8

FIGURE 4. Evaluation of whole dill pickles prepared by natural fermentation and by controlled culture fermentation.

Taste tests were also conducted on the products prepared from brinestock in experiment III. In this case, a five-member panel judged the hamburger chips and a four-member panel judged the whole dill pickles. As shown in Table 5, CCF chips were markedly superior in appearance and somewhat superior in texture and taste. CCF whole dills were similar in appearance to the NF product and markedly superior in texture, taste, and overall acceptability. The NF tank in this experiment was the most difficult to manage of all the NF tanks in the project. Difficulties were encountered in keeping down yeast growth. The tank showed a more noticeable lag before production of lactic acid (Figure 2). Softening enzyme activity was the highest encountered in the experiments.

These results and observations served to further confirm the predictability of the CCF procedure, in which the brine is initially "swamped" with a high concentration of the desired organisms, compared to the NF procedure in which one must depend to a large extent on luck in getting a sufficient number of acidformers to initiate the fermentation.

Experimental results of tests of product quality from the three experiments were pooled and subjected to statistical analysis to determine if noted differences could be considered significant based on the available amount of information. Results are summarized in Table 6. Overall, CCF products were significantly superior to NF products in terms of texture and acceptability.

Quantity and composition of brines from NF and CCF fermentations were extensively monitored. Results are presented in Table 7 (expt. I), Table 8 (expt. II), and Table 9 (expt. III). They are summarized in Table 10, which indicates the similarity of the processes in terms of amount of spent brine generated, amount of process water generated, and BOD, COD, TKN, and TP loads in wastewaters. As expected, TDS and TSS levels were significantly greater in the NF brinings (Table 6). Unexpectedly, the amount of wastewater generated in the two desalting processes did not differ significantly (Table 6). This can be partly explained by failure to sufficiently desalt the NF brinestock in the first experiment. In addition, it was observed that plant personnel tended to vary duration of desalting, rather than volume of desalting water, in response to the amount of salt left in the brinestock. Based on plant priorities, the CCF procedure could offer reduced desalting time per given volume of water or reduced volume of water for a given desalting time (Figure 5).

The large significant difference in salt loading in the wastewaters from NF and CCF brinings would have been even greater if the tanks were held for longer periods and the salt addition to NF tanks was continued, according to the usual practice, to a final level of 45-65 ^{o}S.

An examination of the data collected during the active fermentation periods indicates that the improvement in brinestock quality noted in CCF tanks might not be due simply to the presence of inoculum but might also be in part due to the use of sparging. Fleming et al. (1973), noting that bloater damage causes "serious economic losses to the pickle industry," reported that dissolved CO_2 concentration in brines is directly related to

TABLE 5. TASTE TESTS, EXPERIMENT 3 - (2 CORES PER TANK TESTED)*

Type	CCF A	CCF B	NF
Whole			
Appearance	7.5	7.9	7.9
Texture	7.6	7.9	6.0
Taste	5.9	7.0	5.4
Overall	6.9	7.4	5.4
Chips			
Appearance	8.2	8.2	7.7
Texture	8.1	8.1	7.9
Taste	6.2	6.6	6.0
Overall	7.0	7.7	7.2

*A five-member taste panel evaluated chips; a four-member panel evaluated whole pickles.

TABLE 6 . STATISTICAL ANALYSIS OF EXPERIMENTAL RESULTS,
ANALYSIS: ONE-WAY ANALYSIS OF VARIANCE, DIFFERENT SAMPLE SIZES

Parameter	Units	Mean Value		α	Result
		NF	CCF		
1) Waste waters (spent brines and desalting waters)					
a) Salt	kg/ton of raw product	105	66	0.005	NF ≠ CCF
b) Total suspended solids	g/ton of raw product	665	481	0.05	NF ≠ CCF
c) Volume of wastewaters	gal/ton of raw product	279	267	0.05	NF = CCF
2) Product quality					
a) Pressure test of brine-stock	lb pressure	18.6	19.9	0.05	NF = CCF
b) Texture of whole finished product	1-10 scale	6.6	7.8	0.005	NF ≠ CCF
c) Appearance of whole finished product	1-10 scale	7.2	8.0	0.05	NF = CCF
d) Overall acceptability of whole finished product	1-10 scale	6.0	7.1	0.01	NF ≠ CCF

TABLE 7 . COMPARISON OF QUANTITY AND COMPOSITION OF BRINES FROM NATURAL AND CONTROLLED FERMENTATIONS (EXPT. 1)

Parameters	NF	CCF
No. tanks	1	1
Cucumbers tanked, bushels	72	74
Cucumbers tanked, tons	1.80	1.85
Final Salometer	45	29
Spent brine		
Volume, gal.	364	284
BOD_5, mg/l	5,000	7,800
COD, mg/l	11,160	16,000
COD:BOD	2.2	2.1
TSS, mg/l	580	685
TDS, g/l	67.5	48.3
TKN, mg/l	425	418
NH_3-N, mg/l	70	45
TP, mg/l	93	84
pH	3.5	3.8
Turbidity, ppm as SiO_2	600	400
Process (Desalting) Water		
Volume, gal.	182	178
BOD_5, mg/l	2,700	3,300
COD, mg/l	6,475	8,330

(continued)

TABLE 7 (continued)

Parameters	NF	CCF
COD:BOD	2.4	2.5
TSS, mg/l	42	49
TDS, g/l	48.2	27.4
TKN, mg/l	200	230
NH_3-N, mg/l	26	9
TP, mg/l	32	30
pH	3.2	3.2
Turbidity, ppm as SiO_2	425	750
Gal. spent brine/ton tanked	202	154
Gal. spent brine/bu. tanked	5.05	3.84
Gal. process water/ton tanked	101	96
Gal. process water/bu. tanked	2.53	2.41
BOD, g/ton tanked	4,800	5,600
BOD, g/bu. tanked	120	140
COD, g/ton tanked	11,040	12,320
COD, g/bu. tanked	276	308
TDS, g/ton tanked	70,000	37,600
TDS, g/bu. tanked	1,750	940
TSS, g/ton tanked	600	560
TSS, g/bu. tanked	15	14
TKN, g/ton tanked	404	328
TKN, g/bu. tanked	10.1	8.2
TP, g/ton tanked	80	60
TP, g/bu. tanked	2.0	1.5

TABLE 8. COMPARISON OF QUANTITY AND COMPOSITION OF BRINES FROM NATURAL AND CONTROLLED FERMENTATION (EXPT. 2)

Parameters	NF-1	NF-2	CCF-1	CCF-2
Cucumbers tanked, bu.	80	80	80	80
Cucumbers tanked, tons	2.0	2.0	2.0	2.0
Final Salometer	45	47	30	28
Spent brine				
Vol., gal	297	234	275	288
BOD_5, mg/l	7,200	6,600	8,800	8,800
TSS, mg/l	400	550	355	380
TDS, g/l	152	138	92	121
TKN, mg/l	555	750	450	855
TP, mg/l	122	103	105	108
pH	3.8	3.5	3.8	3.8
Turbidity, ppm as SiO_2	450	475	500	500
First process water				
Vol., gal	52	52	364	295
BOD_5, mg/l	7,200	2,600	3,000	3,200
COD, mg/l	3,820	-	-	-
TSS, mg/l	195	125	415	630
TDS, g/l	63	170	39	39

(continued)

TABLE 8 (continued)

Parameters	NF-1	NF-2	CCF-1	CCF-2
TKN, mg/l	245	223	300	238
TP, mg/l	41	36	29	30
pH	3.4	3.3	3.6	3.5
Turbidity, ppm as SiO_2	350	150	475	525
Second process water				
Vol. gal	241	245	-	-
BOD_5, mg/l	4,000	3,200		
TSS, mg/l	760	780		
TDS, g/l	66	76		
TKN, mg/l	293	298		
TP, mg/l	45	42		
pH	3.4	3.3		
Turbidity, ppm as SiO_2	475	500		
Gal. spent brine/ton tanked	148	117	138	144
Gal. spent brine/bu. tanked	3.7	2.9	3.4	3.6
Gal. process water/ton tanked	146	148	182	148
Gal. process water/bu. tanked	3.6	3.7	4.6	3.7
BOD, g/ton tanked	7,280	10,160	6,800	6,600
BOD, g/bu. tanked	182	127	170	165
TDS, g/ton tanked	120,000	110,000	75,200	84,800
TDS, g/bu. tanked	3,000	2,750	1,880	2,120
TSS, g/ton tanked	600	600	480	560
TSS, g/bu. tanked	15	15	12	14

(continued)

TABLE 8 (continued)

Parameters	NF-1	NF-2	CCF-1	CCF-2
TKN, g/ton tanked	472	488	440	600
TKN, g/bu. tanked	11.8	12.2	11.0	15.0
TP, g/ton tanked	93	69	75	76
TP, g/bu. tanked	2.3	1.7	1.9	1.9

TABLE 9. COMPARISON OF QUANTITY AND COMPOSITION OF BRINES FROM NATURAL AND CONTROLLED FERMENTATIONS (EXPT. 3)

Parameters	NF	CCF	CCF
Cucumbers tanked, bu.	80	80	80
Cucumbers tanked, tons	2	2	2
Final Salometer	37	33	24
Spent brine			
Vol., gal	312	273	270
BOD_5, mg/l	8,400	11,100	9,400
COD, mg/l	16,630	20,115	18,650
COD:BOD	2.0	1.8	2.0
TSS, mg/l	410	335	320
TDS, g/l	120	102	80
TKN, mg/l	495	495	453
TP, mg/l	118	110	105
pH	3.3	3.4	3.5
Turbidity, as ppm SiO_2	250	250	300
Process water			
Vol., gal	194	196	204
BOD_5, mg/l	3,400	6,100	4,900
COD, mg/l	10,925	13,225	11,625
COD:BOD	3.2	2.2	2.4
TSS, mg/l	1,680	740	500

(continued)

TABLE 9 (continued)

Parameters	NF	CCF	CCF
TDS, g/l	31	84	50
TKN, mg/l	328	290	280
pH	2.8	2.9	3.0
Turbidity, as ppm SiO_2	400	1,000	625
Gal. spent brine/ton tanked	156	136	135
Gal. spent brine/bu. tanked	3.9	3.4	3.4
Gal. process water/ton tanked	97	96	102
Gal. process water/bu. tanked	2.4	2.4	2.6
BOD, g/ton tanked	6,200	8,000	6,640
BOD, g/bu. tanked	155	200	166
COD, g/ton tanked	13,600	15,200	8,800
COD, g/bu. tanked	340	380	220
TDS, g/ton tanked	66,000	80,000	48,000
TDS, g/bu. tanked	1,650	2,000	1,200
TSS, g/ton tanked	860	448	355
TSS, g/bu. tanked	21.5	11.2	8.9
TKN, g/ton tanked	413	364	337
TKN, g/bu. tanked	10.3	9.1	8.4
TP, g/ton tanked	82	84	79
TP, g/bu. tanked	2.0	2.1	2.0

TABLE 10 . SUMMARY COMPARISON OF QUANTITY AND COMPOSITION OF BRINES FROM NATURAL AND CONTROLLED FERMENTATIONS

Parameters	NF	CCF
No. tanks	4	5
Spent brine, gal/ton	156	141
Process water, gal/ton	123	125
Total water, gal/ton	279	267
BOD, g/ton	7,110	6,730
BOD:N:P	100:6:1	100:6:1
COD, g/ton	13,320*	12,110*
TDS, g/ton	91,500	65,120
TSS, g/ton	665	480
TSS, lb/ton	1.46	1.06
TKN, g/ton	444	414
TP, g/ton	81	75
BOD, lb/ton	15.6	14.7
Salt, 10^3 g/ton	105	66

*CODs were not available for experiment 2. Value represents average of 2 NF tanks and 3 CCF tanks.

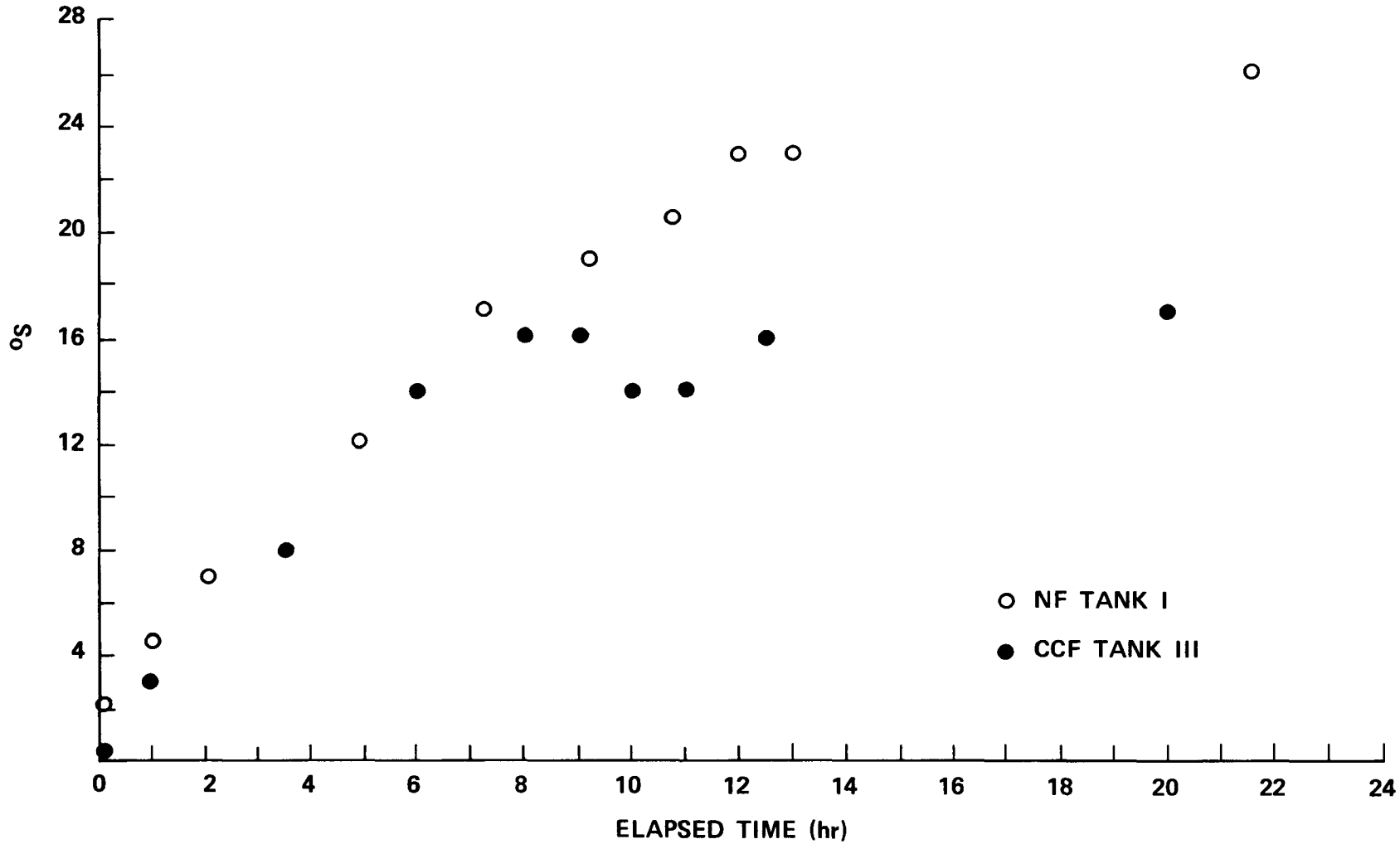


Figure 5. Progress of desalting in experiment 1: salt concentration in processing water as a function of time.

bloating of cucumbers and that even with a CCF procedure substantial concentrations of CO_2 build up unless sparging is practiced. They noted visible bloater damage in No. 3 cucumbers when the CO_2 concentration was $\geq$ 60 mg/100 ml brine. During the course of our experiments, CO_2 was monitored in both the NF and CCF tanks (Figures 6, 7, and 8). As a comparison, CO_2 levels in tanks in the PPP tankyard were also monitored (Table 11). Note that in all the experimental NF tanks, the CO_2 concentration exceeded 60 mg/100 ml during the active fermentation period, despite the relatively large surface-to-volume ratio in these tanks. In the 4 large commercial brinings, the CO_2 concentrations were excessive throughout the entire first six days in which they were monitored. In contrast, because of the nitrogen-sparging, the CCF tanks maintained low CO_2 levels ($< \sim$ 32 mg/100 ml). Obviously, further studies must be made of the relative contributions to brinestock quality of culture addition and nitrogen sparging, since the experimental design of this project does not permit this distinction.

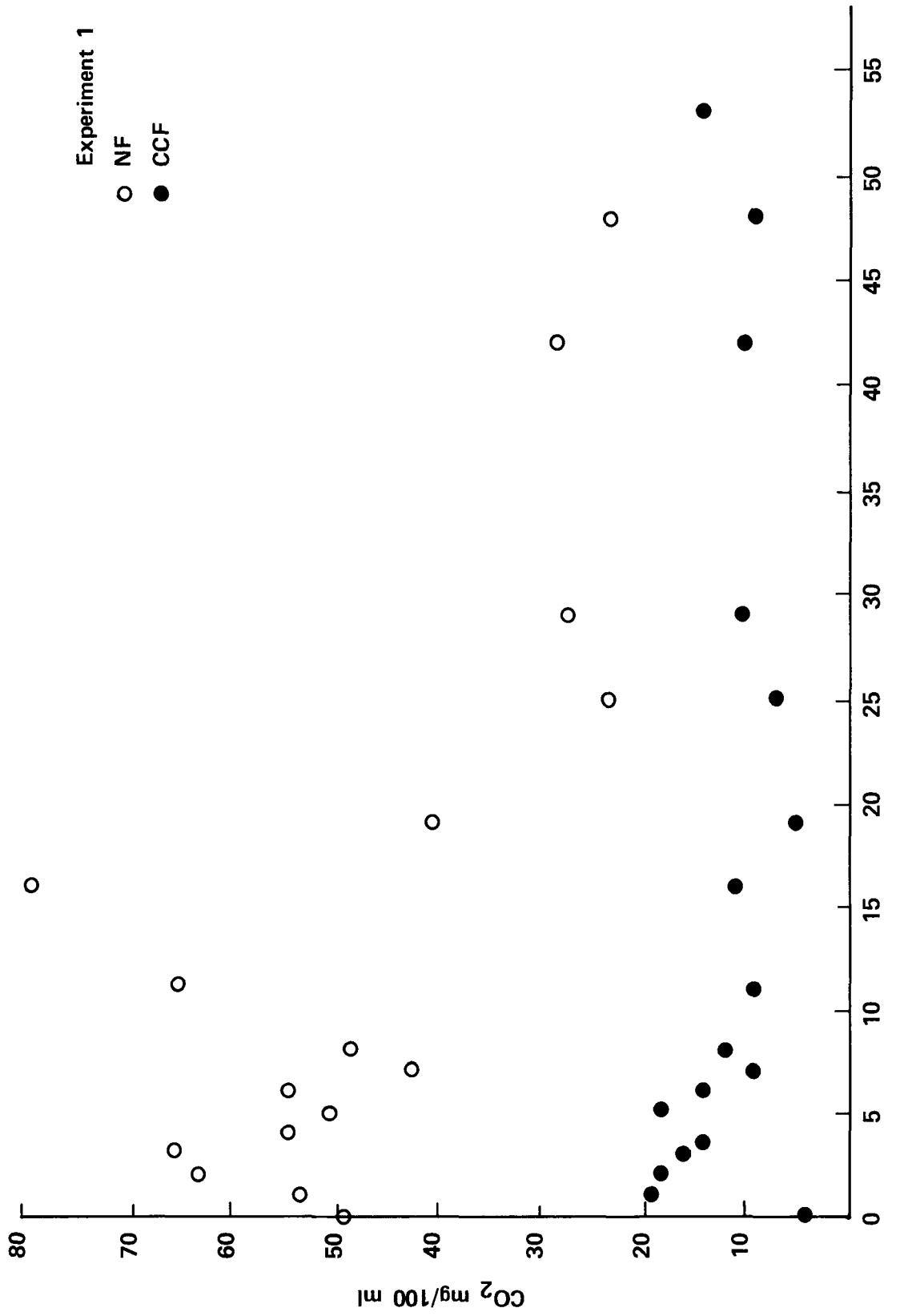


Figure 6. Carbon dioxide accumulation in experimental tanks, experiment 1.

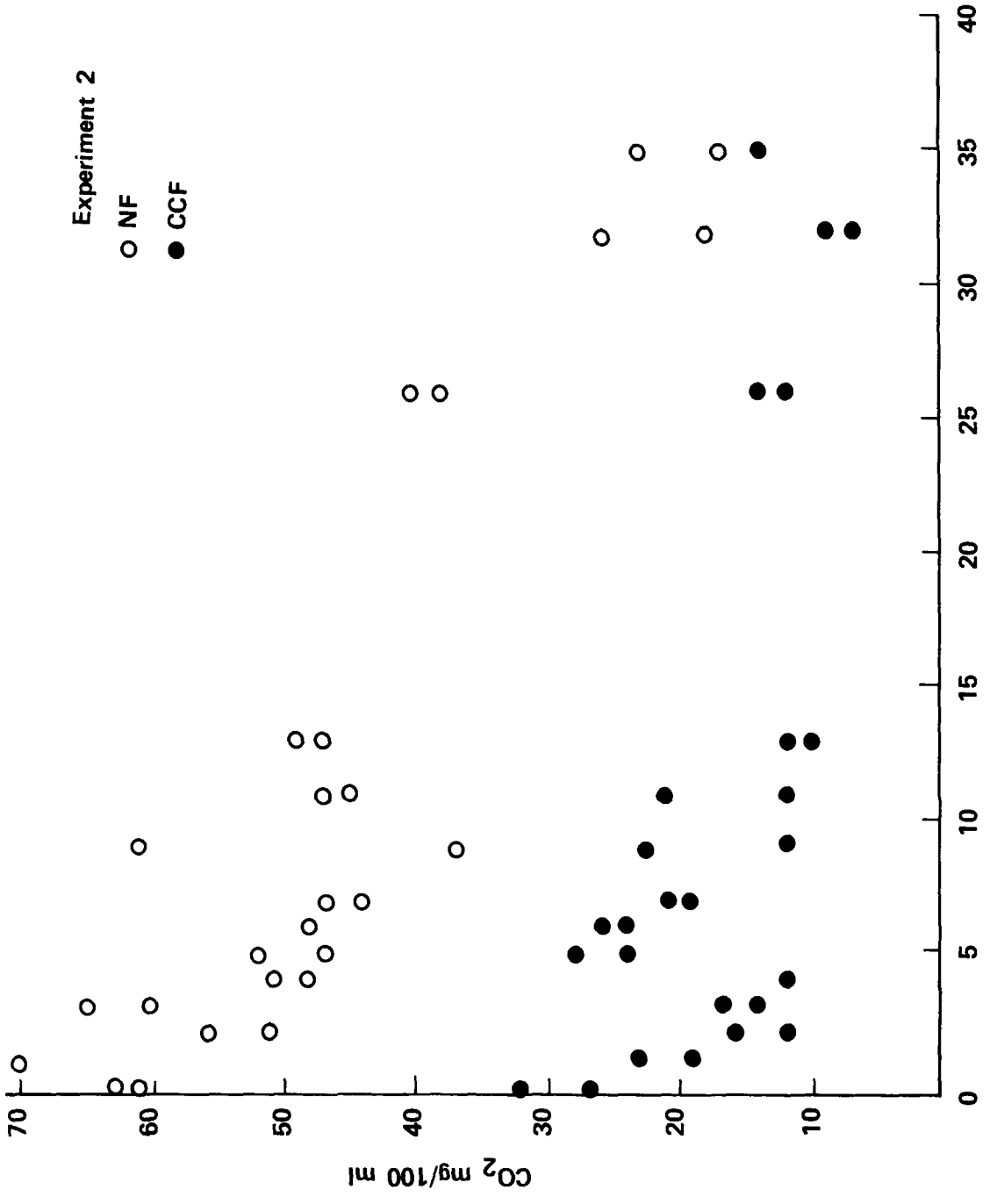


Figure 7. Carbon dioxide accumulation in experimental tanks, experiment 2.

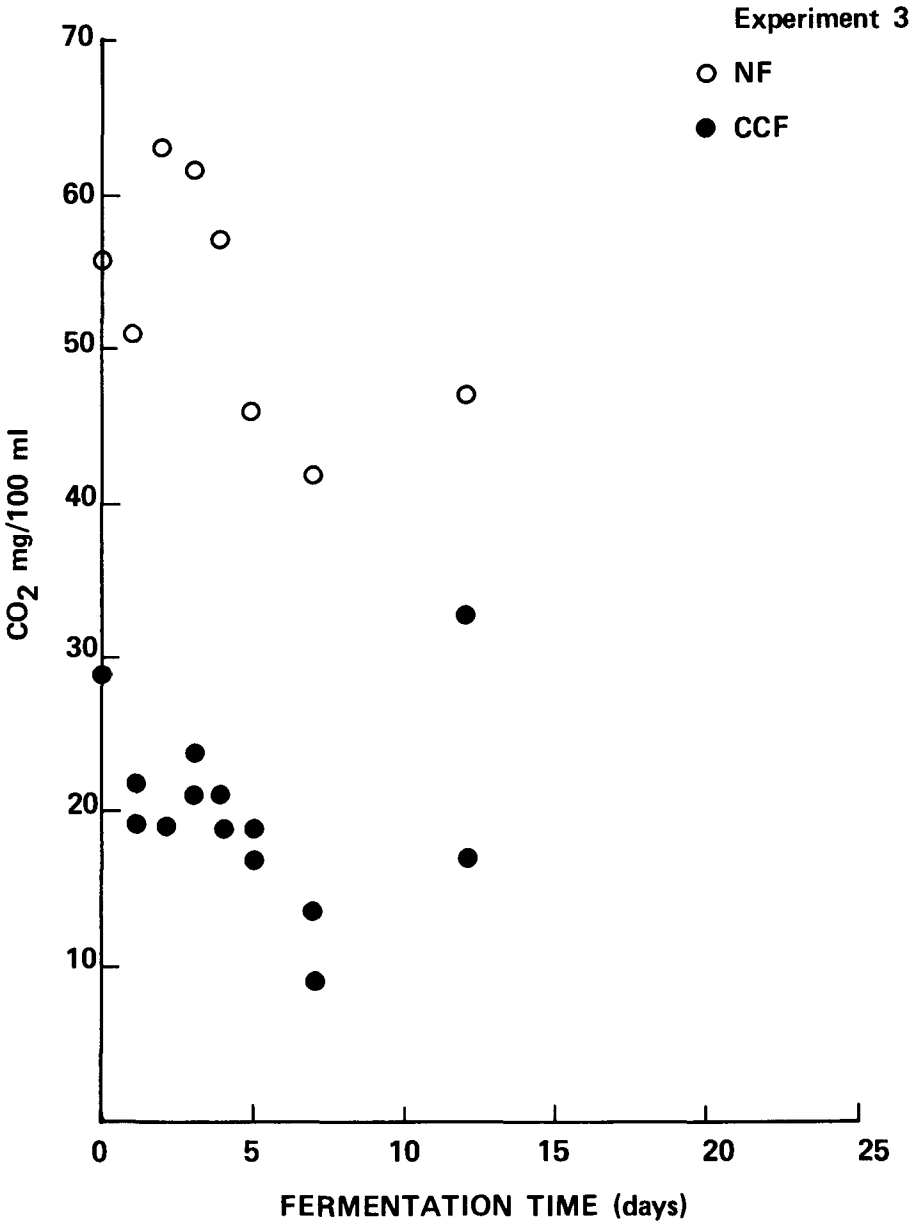


Figure 8. Carbon dioxide accumulation in experimental tanks, experiment 3.

TABLE 11. CO_2 ACCUMULATION IN LARGE COMMERCIAL BRININGS

Fermentation time (days)	CO_2 (mg/100 ml)			
	1	2	3	4
0	77	72	70	68
1	81	78	76	76
2	79	68	70	79
3	82	69	78	66
6	98	88	88	84

BRINE RECYCLING STUDIES

Laboratory studies of brine recycling were initially directed toward finding a coagulant or coagulant aid which, at the low pH typical of spent brine, would effectively reduce enzyme activity. In addition, an attempt was made to correlate turbidity removal with reduction of enzyme activity. Preliminary experiments, however, indicated that substantial removals of turbidity could occur with little or no effect on enzyme activity. Since in each of these cases enough turbidity remained to be visible to the naked eye, further work was directed to provide a "completely clarified brine," defined as a treated brine which in a test tube does not contain visible colloids and has a turbidity of less than 7 JTU.

Low pH Coagulation with Sodium Aluminate

Sodium aluminate ($NaAlO_2$), a coagulant not previously investigated with spent brines, was evaluated at low pH. It was selected primarily for its alkaline properties. Characteristic spent brine pH of about 3.2 is below the range of the isoelectric point for enzymes (Tenney and Stumm, 1965) and it was postulated that addition of sodium aluminate might raise the pH to a point at which coagulation of the enzyme and other colloids would occur by adsorption and charge neutralization. It was also hypothesized that raising the pH might bring about formation of a "sweep floc" of gelatinous aluminum hydroxide which would adsorb the enzyme.

Results of the first jar test (Table 12) indicated that increasing doses of sodium aluminate reduced turbidity of the brine, but even at large doses residual brine turbidity was still substantial. Although the reduction in turbidity did not result in a reduction in enzyme activity, these results were considered inconclusive since the enzyme activities were so high that accurate determinations were not possible.

Several additional jar tests were performed with sodium aluminate in an effort to find a reasonable dosage that would provide complete clarification. Visual inspection, in each case, was sufficient to indicate that even massive doses (up to 576 mg/l as Al^{+3}) failed to completely clarify the brine.

However, substantial floc formation was observed with sodium aluminate. Immediately after the aluminate addition, large amounts of floc could be seen, but the amount appeared to decrease during flocculation and settling. This behavior could be due to initial formation of aluminum hydroxide on contact between brine and the coagulant, followed by dissolution of the hydroxide in the acid environment in the brine; if so, use of sodium aluminate alone for coagulation would require tremendouse doses to achieve a pH high enough to maintain the precipitate.

Since these experiments with sodium aluminate did indicate the ability of aluminum to form a floc in the brine, use of alum addition and pH adjustment was investigated. Selection of alum appeared more favorable from the standpoint of coagulant requirements and in addition settling properties of alum precipitates are frequently superior to those produced by sodium aluminate.

TABLE 12. SODIUM ALUMINATE JAR TEST

Sodium aluminate dosage (mg/l)	Initial pH	Final pH	Turbidity (JTU)	Enzyme activity units
0 (raw brine)	3.2	3.2	---	800
250	3.2	3.4	240	800
500	3.2	3.5	190	800
750	3.2	3.6	100	800
1,000	3.2	3.7	75	800
1,500	3.2	3.9	56	800

Low pH Coagulation with Alum, $Al_2(SO_4)_3 \cdot 18\ H_2O$

In initial jar tests with alum, the brine was raised with sodium hydroxide to pH 5, a level at which precipitation of aluminum hydroxide would be expected to occur. The results from the first experiment are shown in Table 13. Although floc formation was observed at all alum dosages, greater amounts were present at the lower dosages. Some turbidity reduction was realized, but it was insufficient to actually clarify the brine or to noticably affect pectinase activity. The turbidity data actually indicated that the lowest dose (100 mg/l) was more effective than higher doses. In a second jar test (Table 14) these results were confirmed. In this test pH was monitored more closely. Addition of alum depressed the pH. Gelatinous floc formed at pH 5.45, which indicated that the brine already contained precipitable metal species. Turbidity reduction at pH 5.45 was better than at pH 5, but the brine still had a cloudy appearance.

Increased turbidity removal with incrased pH was further obtained in a third experiment in which initial pH of the brine was set at pH 7. Results are shown in Figure 9 and Table 15. Again, significant turbidity removal was accomplished by simply raising the brine pH, flocculating and settling, indicating possible presence of aluminum in the brine. To confirm this hypothesis, metals analyses were conducted, as discussed later below.

Effect of alum dosage on turbidity is apparent in Figure 9 . Increasing alum dosage increased turbidity removal to a point at which further alum addition resulted in increased turbidity. Addition of alum at 150 mg/l at pH 7 reduced brine turbidity from 90 to 10 JTU. The treated brine, though visibly better than that obtained in previous experiments, still contained faintly visible cloudiness. At the same time, enzyme activity remained virtually unaffected. Although the enzyme activities were again so high as to prevent accurate measurement, the data indicate that

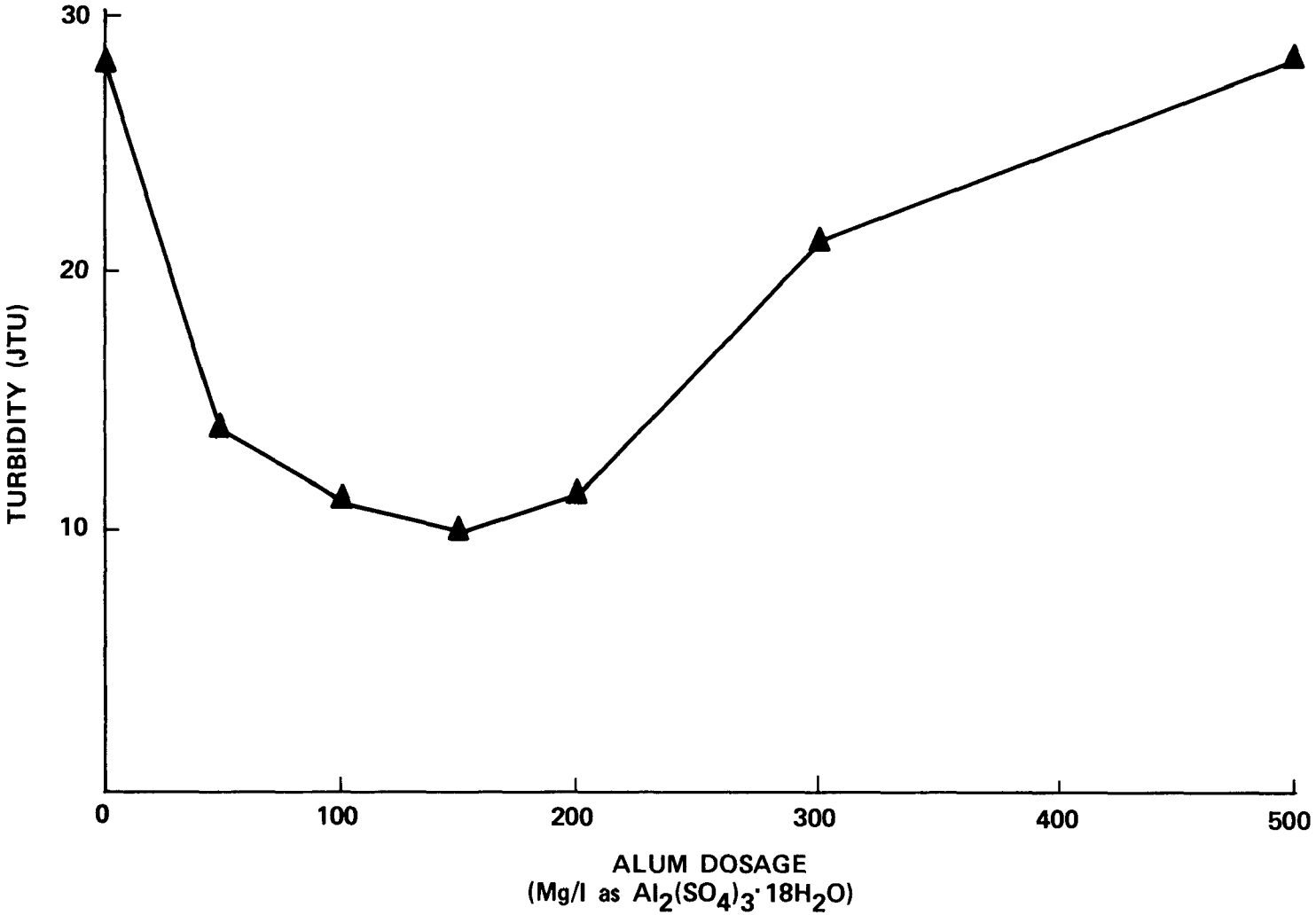


Figure 9. Brine turbidity as a function of alum dosage, initial pH = 7.

TABLE 13. TURBIDITY AND ENZYME ACTIVITY AS A FUNCTION OF ALUM DOSAGE, INITIAL pH = 5

Alum dosage mg/l*	Turbidity (JTU)	Enzyme activity units
0	89	385
100	44	378
200	48	380
300	48	400
500	55	389
700	54	387

*As $[Al_2(SO_4)_3 \cdot 18H_2O]$

removal of most of the turbidity-causing colloids had little if any effect on reducing the enzyme activity. Since it was still uncertain, however, if removal of the remaining visible colloids would result in physical removal of the enzyme, further experiments considered use of coagulant aids to enhance precipitation.

Low pH Coagulation with Alum and Clay

The first coagulant aid investigated was a clay solution, Nalco 8151, a slightly anionic bentonite. Use the "optimum" alum dosage previously determined, 150 mg/l, two tests were performed with two different brine samples, each spiked with pectinase. Results are shown in Tables 16 and 17. Turbidity was not measured in these experiments since in both cases visible colloids remained after settling and because the major objective was to reduce enzyme activity. It is apparent from the results that addition of Nalco 8151 at the selected alum dosage did not reduce enzyme activity. The enzyme activities in each experiment merely indicate expected variations based on the sensitivity of the enzyme analysis. The results in Table 16 also illustrated the failure of even large clay dosages to affect enzyme removal. No further investigations appeared warranted.

Kaolinite was also briefly examined as a possible coagulant aid. Brine at pH 5.5 with a spiked enzyme activity of about 170 units was flocculated with 1000 mg/l of kaolinite, then settled. Enzyme activity in the supernatant was 161, not a significant reduction.

TABLE 14 . TURBIDITY AS A FUNCTION OF ALUM DOSAGE, INITIAL pH = 5.45

Alum dosage mg/l*	Initial pH	Final pH	Turbidity (JTU)
0 (raw brine)	3.2	3.2	90
0	5.45	5.45	23
20	5.45	5.4	24
50	5.45	5.35	25
100	5.45	5.3	26
200	5.45	5.2	29
400	5.45	4.95	34

*As $[Al_2(SO_4)_3 \cdot 18H_2O]$

TABLE 15 . TURBIDITY AND ENZYME ACTIVITY AS A FUNCTION OF ALUM DOSAGE @ INITIAL pH = 7

Alum dosage mg/l*	Initial pH	Turbidity (JTU)	Enzyme activity units
0	3.2	90	373
0	7	27	350
50	7	14	577
100	7	12	522
150	7	10	580
200	7	12	570
300	7	21	605
500	7	27	543

*As $[Al_2(SO_4)_3 \cdot 18H_2O]$

TABLE 16. EFFECTS OF NALCO 8151 AND OPTIMUM ALUM DOSAGE, INITIAL pH = 7

Alum dosage mg/l*	Nalco 8151 mg/l	Enzyme activity units
0	0	185
150	50	168
150	75	175
150	100	180
150	125	182
150	150	178
150	200	140

*As $[Al_2(SO_4)_3 \cdot 18H_2O]$

TABLE 17. EFFECTS OF NALCO 8151 AND OPTIMUM ALUM DOSAGE, INITIAL pH = 7

Alum dosage mg/l*	Nalco 8151 mg/l	Enzyme activity units
0	0	16
150	5	21
150	10	34
150	20	28
150	30	35
150	40	24
150	50	24

*As $[Al_2(SO_4)_3 \cdot 18H_2O]$

TABLE 18. METALS CONTENT IN A NO. 2 SPENT BRINE USED FOR JAR TESTS, COMPARED WITH THOSE REPORTED BY HENNE AND GEISMAN (1973)

Metal	Concentration (mg/l)	
	Brine #2	Geisman brine
Aluminum	41	13
Magnesium	58	300
Manganese	1.5	11
Zinc	1.4	12
Calcium	400	1,000
Iron	10	40

Metal Analyses of Spent Brines

During earlier jar tests with alum, it was observed that turbidity of the brine was substantially reduced if the pH was raised to between 5 and 6 and then the brine was allowed to flocculate (30 min) and settle. Appearance of a white precipitate at these pH levels suggested that aluminum (or other metal) might be present in the untreated brine. Subsequent metal analyses revealed that not only was there a substantial concentration of aluminum, but of other metals as well. The results of the metal analyses are provided in Table 18 along with those reported by Henne and Geisman (1973), for comparison. The observed differences would be expected, since metals content of the brines should vary with differences in soils in which the cucumbers were grown, in the water used for brine preparation, and in the rock salt used for the brine. Apparently, some plants also add alum directly to the brines on occasion for help in controlling unwanted algal growth.

Presence of metals in the brines has several implications. An aluminum concentration of 40 mg/l is equivalent to an alum dosage of 450 mg/l as $Al_2(SO_3)_4 \cdot H_2O$, a substantial dose in terms of wastewater treatment, yet at pH 5 little precipitation was observed. Presence of metals which resist precipitation during recycling could possibly lead to buildup, which appears to be the case in studies by McFeeters et al. (1978) though the significance of the amount they observed may be small.

TABLE 19. EFFECTS OF HIGH MOLECULAR WEIGHT, ANIONIC POLYMER (NALCO 7744A) ON TURBIDITY AND ENZYME REMOVAL AT pH = 5.6

Nalco 7744A (mg/1)	Initial pH	Final pH	Turbidity (JTU)	Enzyme activity Units
0	5.6	5.6	24	135
0.1	5.6	5.6	21	123
1	5.6	5.6	19	121
3	5.6	5.6	20	134
10	5.6	5.6	21	120
30	5.6	5.6	23	116

Low pH Coagulation, Polymer Addition

Addition of polymers was evaluated in an effort to find some means to generate the expected precipitation and formation of aluminum hydroxide floc at pH 5-6. Since the aluminum hydroxide, as well as the enzyme, is expected to have a slight positive charge at this pH, a high molecular weight anionic polymer, Nalco 7744A, was first evaluated. It was hoped that the polymer might bridge the positively charged particles to form large settleable floc. Results (Table 19) indicated that a slight improvement in turbidity was obtained with increased polymer dosage to about 1 mg/1. However, all tested dosages failed to produce heavy floc and brine clarity. There also appeared to be no significant reduction in enzyme activity. The evaluation of Nalco 7744A was discontinued since larger dosages gave no indication of improvement and polymer doses above 30 mg/1 were considered to be economically prohibitive.

Other polymers were evaluated. Dow A-21, a high molecular weight anionic polymer, and Dow C-31, a cationic polymer, were tested at pH 5.5 at 0.3, 1, 3, 10, and 30 mg/1, but neither provided complete clarification or sufficient reduction of enzyme activity.

Low pH Coagulation with Ludox and Celite Addition

Since the pectinase enzyme should be positively charged at low pH, it was hypothesized that the addition of a negatively charged adsorbent to the untreated brine (pH=3.2) might effect enzyme removal by adsorption. Two such negative adsorbents were evaluated. Ludox, a negatively charged silica, was considered a potentially useful adsorbent as its high surface area: weight ratio provides a large number of adsorption sites per unit dose. Ludox solution was evaluated in jar tests at doses of 359-3590 mg/1 (Figure 10). As indicated, there was no significant reduction of enzyme activity. Further tests were conducted with Celite, another negatively charged silica. A brine

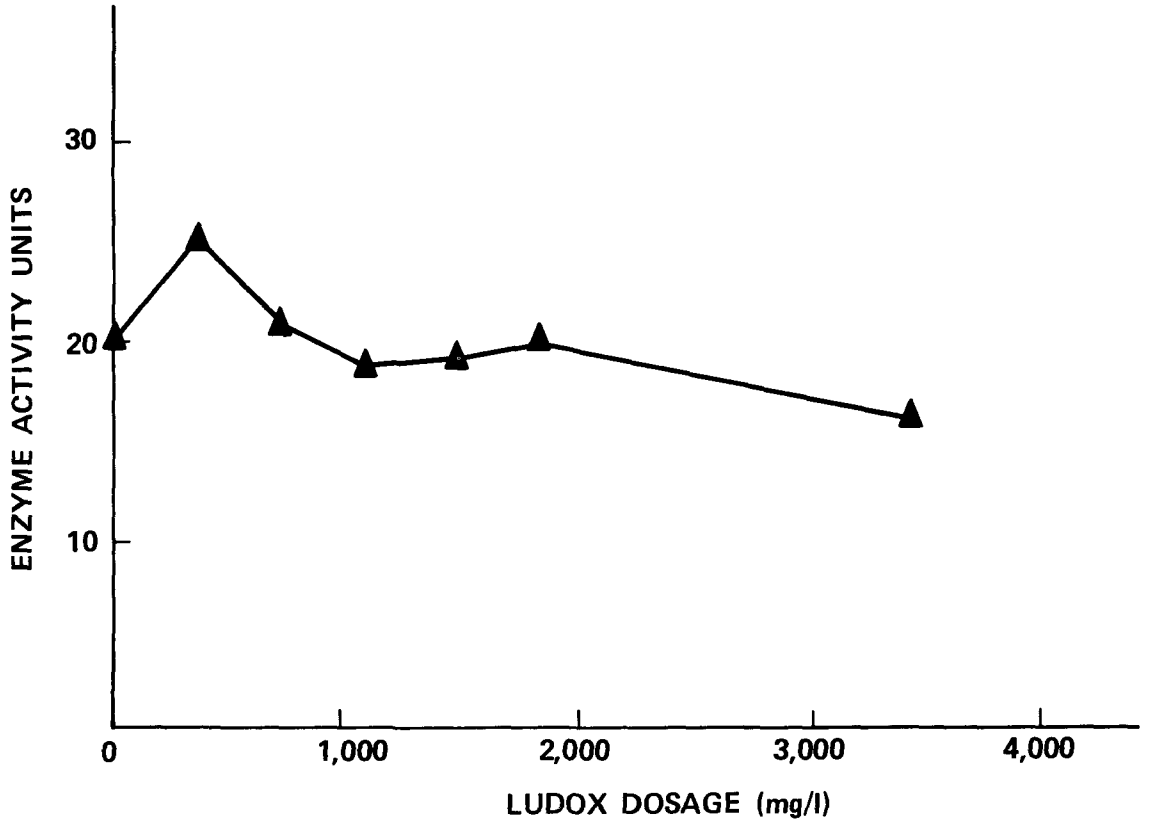


Figure 10. Enzyme activity as a function of Ludox dosage.

"spiked" with a larger enzyme dosage was used in these tests, but again no effects on enzymes activity were seen even at doses as high as 1 g/l (**Fig. 11**). Most probable cause for failure of these materials to sorb the enzyme is the nature of the brine, which contains high salt concentration and large amounts of organics which could interfere with sorption.

pH Effects on Coagulation and Enzyme Activity

A study of the pH effects on coagulation and enzyme activity was initiated for two reasons: (1) all attempts to reduce pectinase activity or generate a completely clarified brine by coagulation and sedimentation at pH less than 7 were unsuccessful, and (2) the metals analyses indicated that the brines contained substantial amounts of aluminum, magnesium, and calcium and it was anticipated that these metals might precipitate at higher pH.

To assess effects of pH, a "spiked" spent brine (designated #2) was treated at a series of pH values ranging from 3.2 (raw) to 10.1. pH of the samples was adjusted with NaOH. Following pH adjustment, each sample was flocculated and settled and the supernatant was withdrawn for enzyme analysis. At pH 6.7 after flocculation and settling, the brine supernatant was completely clarified (Table 20). Complete clarification was also achieved at every pH above 6.7. Of especial significance are the corresponding enzyme activity measurements. Even though the brine was completely clarified at pH 6.7, no significant reduction in enzyme activity was observed below pH 10.1, thus indicating that no correlation exists between enzyme activity and turbidity removal.

To establish this conclusion, an additional series of samples was examined, this time adjusting pH with a lime slurry. Effect of pH on enzyme activity is shown in Figure 12 . Again the brine was completely clarified at pH 7, but there was no substantial effect on enzyme activity below pH 9, with the most drastic effects occurring between pH 10.1 and 10.4. The fact that significant enzyme reductions did not occur below pH 10.1 even though massive precipitation and complete clarification occurred at pH as low as 6.7 appeared to indicate that reduction of enzyme activity was not the result of physical removal by adsorption on the precipitate but was, instead, the result of denaturation at high pH. This denaturation appeared to be irreversible since readjustment of the sample to pH 5 for enzyme analysis failed to restore enzyme activity to the original value.

Several experiments were performed to confirm these findings. Tests of spent brine spiked with pectinase enzyme and subjected to high pH by various procedures indicated that high pH destroyed enzyme activity regardless of the method of pH adjustment (Figure 13).

Since there was also heavy precipitation and flocculation accompanying the high pH levels, and since the brines were completely clarified, there still remained some question as to whether the enzyme was removed physically by sorption or whether denaturation destroyed enzyme activity. Several analyses which were performed appeared to favor the denaturation theory.

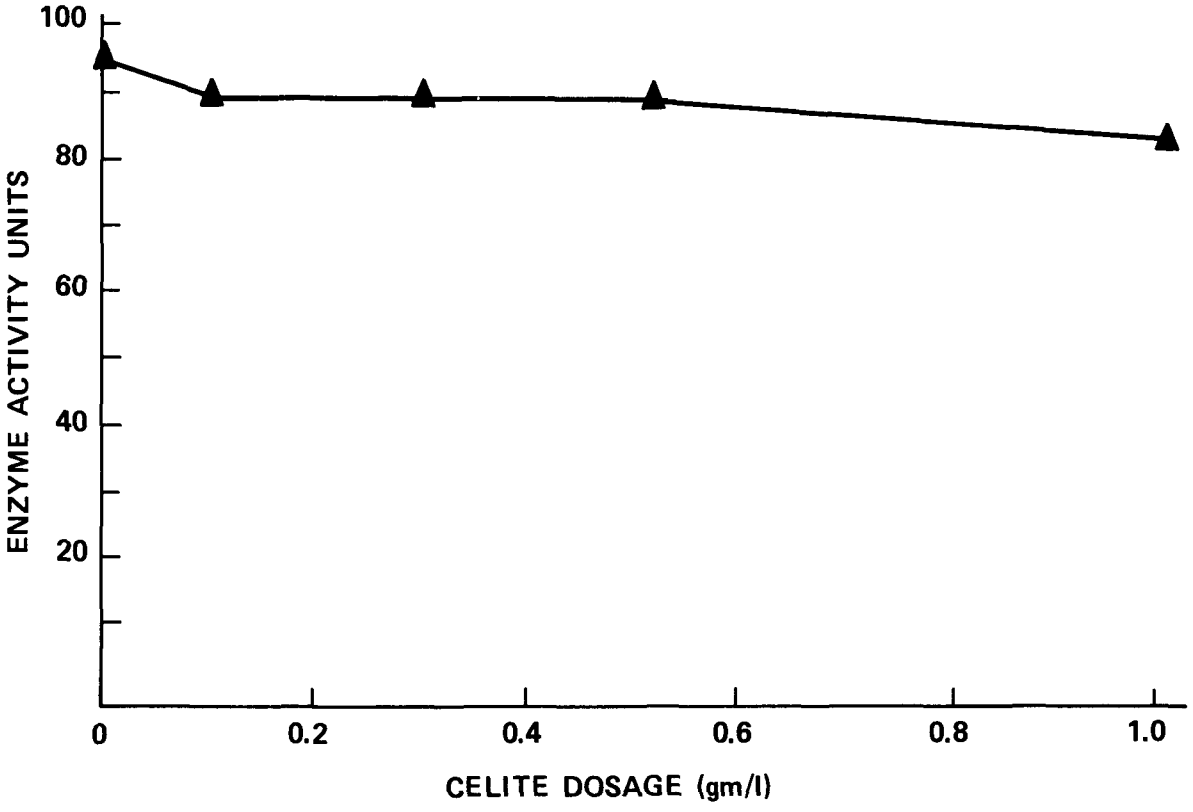


Figure 11. Enzyme activity as a function of Celite dosage.

TABLE 20. THE EFFECTS OF pH ON TURBIDITY AND ENZYME ACTIVITY

pH	Turbidity (JTU)		Enzyme activity units
3.2 (raw)	90	Little or no precipitation	93
4	72		--
4.5	39		--
5.2	27	Some small precipitation, but brine remains cloudy	--
5.5	21		--
6.1	16	Larger amount of precipitation, but visible colloids remain	90
6.7	7	Massive precipitation initiated during flocculation	100
7.1	7		96
8.1	7	Massive precipitation in evidence before flocculation	90
9.1	7		86
10.1	7		74

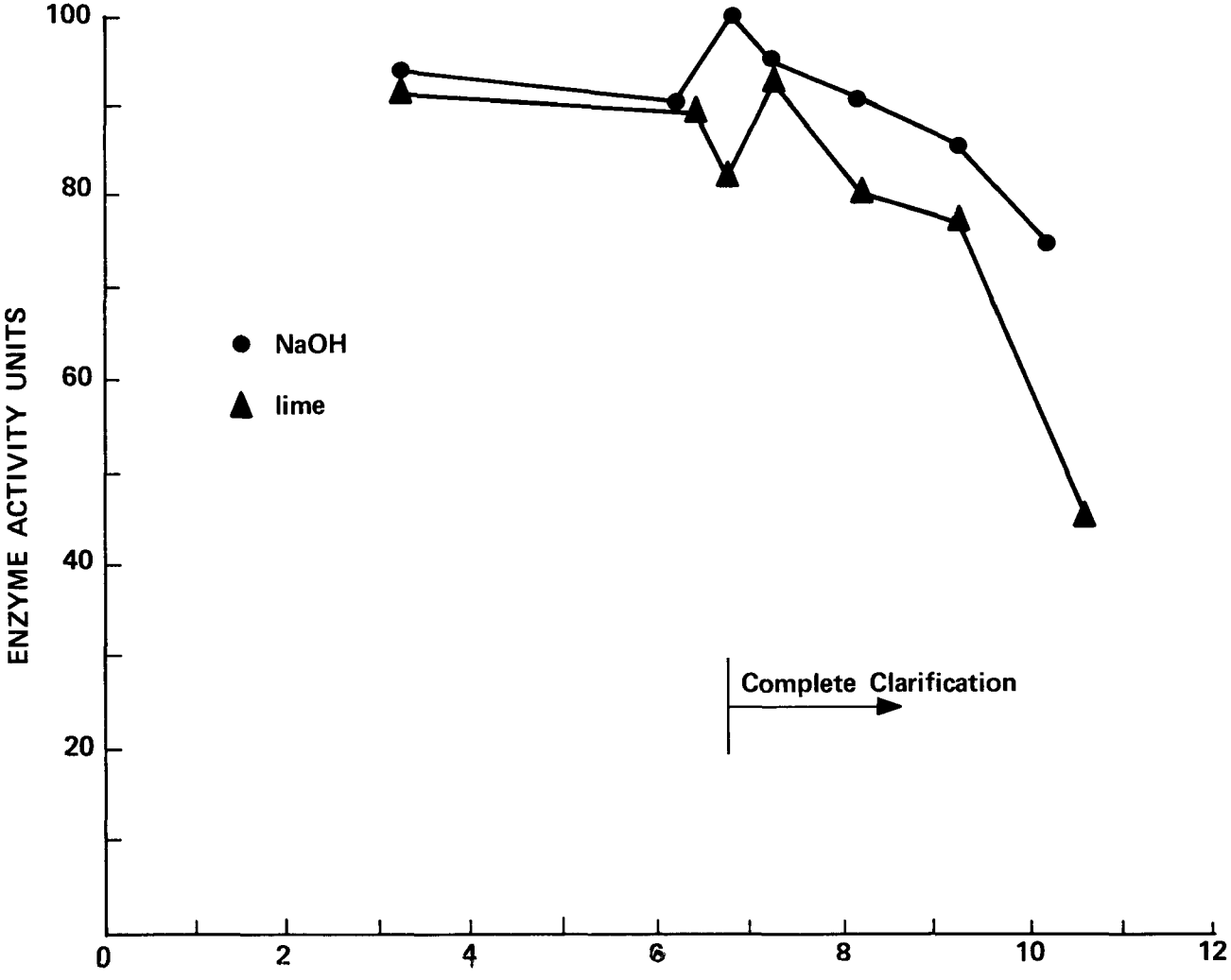


Figure 12. The effect of pH on the enzyme activity of a no. 2 spent brine.

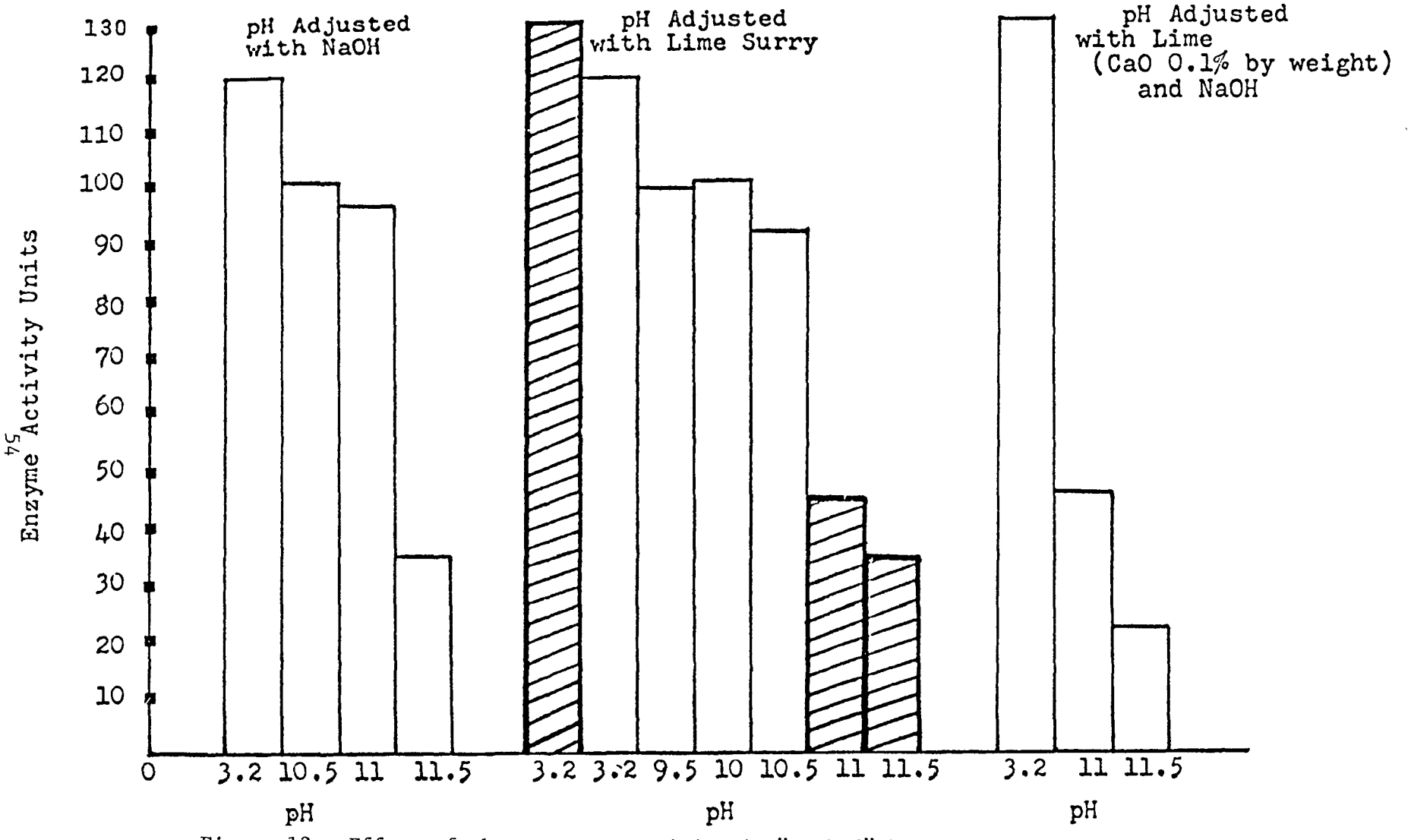


Figure 13. Effect of ph on enzyme activity in "spiked" brine samples.

A sludge sample was removed from a brine at pH 11.5, dissolved, and the resulting solution adjusted to pH 5. An enzyme analysis indicated an enzyme activity of about 21 units, about the same as that of the supernatant.

To determine the enzyme activity as a function of pH without interference from precipitation and floc formation, "synthetic" brines, free of metals, were produced in the laboratory, one of 50°S acidified to pH 3.2 with acetic acid; the other, a 35°S 0.1 M lactic acid solution. After "spiking" the synthetic brines with enzyme, several aliquots were taken from each for pH adjustment and subsequent enzyme analysis. Enzyme activities of these brines as a function of pH are shown in Figures 14 and 15. In both brines pH decreased sharply above pH 10. However, in the higher salinity brine difficulties were encountered in maintaining the high pH levels. After allowing the samples to stand for 30 min, pH was again recorded, indicating that the pH 11.5 brine had dropped to pH 8-9 and possibly accounting for the lesser reduction expected. In either case, dependence of enzyme activity on pH is clearly demonstrated. In these brines there was no possibility of physical removal of the enzyme since there was no precipitation in the solution; the enzyme activity losses in these brines corresponds to those observed in the actual brines.

Further confirmation is provided in Figure 16 in which enzyme activity of a pectinase-containing spent brine from an actual fermentation is plotted as a function of pH. As in the cases with "spiked" spent brines and synthetic brines, the enzyme activity decreased rapidly between pH 9 and 11.

Discussion of Failure of Coagulation and Precipitation to Remove Pectinase

In the course of the studies described above, it was observed that in spite of massive precipitation of aluminum hydroxide and subsequent brine clarification, enzyme activity remained unaffected, indicating that aluminum floc had failed to adsorb the pectinase. Several factors described in the literature may account, at least in part, for this phenomenon. These include interference by high salt concentration and by competing metal species, as well as interference by organics (Dixon and Webb, 1964). It is not known which of these factors, or combination thereof, were responsible for the observed failure of aluminum precipitation. However, a further experiment did demonstrate that in itself aluminum hydroxide precipitate is capable of sorbing the enzyme. A solution of distilled water "spiked" with pectinase was treated with alum doses of 300 and 500 mg/l. After buffering at pH 5.8 with sodium bicarbonate, the sample was flocculated and settled. Analysis of the supernatant (Figure 17) indicated that enzyme activity was reduced from 220 units to 24 units by the 300 mg/l dose and further reduced to 15 units by the 500 mg/l dose. Since the pH of the samples never was extreme enough to inhibit enzyme activity these results show that in the absence of interferences alumina floc is an effective sorbent of the pectinase enzyme.

In summary, removal of pectinase activity by high pH coagulation-precipitation is due to denaturation of the enzyme at the high pH rather than to physical removal by precipitation when real or synthetic brines are employed. Failure of widely used coagulants to remove pectinase activity from spent brine is probably due, at least in part, to interferences such as salt.

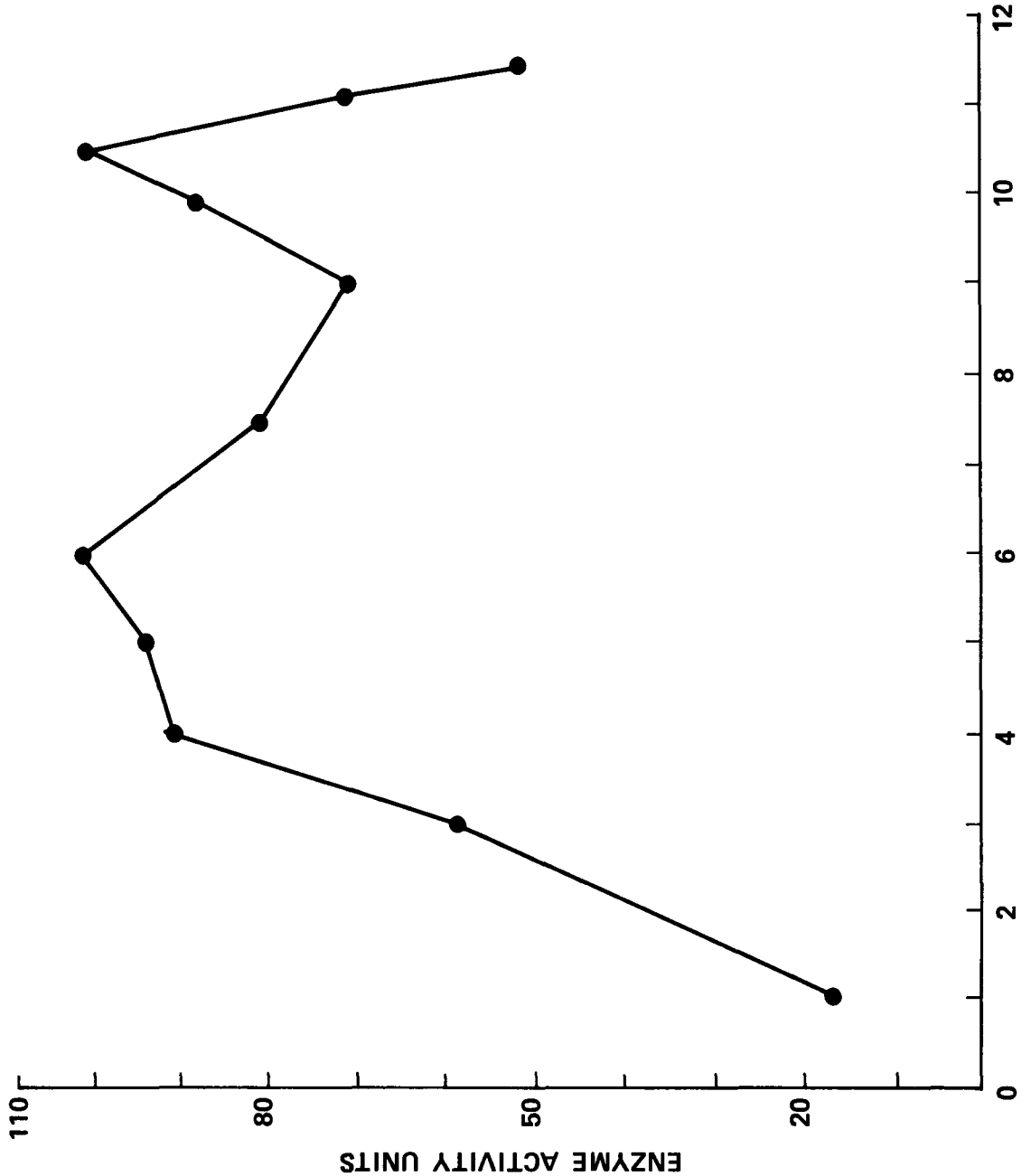


Figure 14. Enzyme activity as a function of pH for 35°S, 0.1 lactic acid "synthetic" brine.

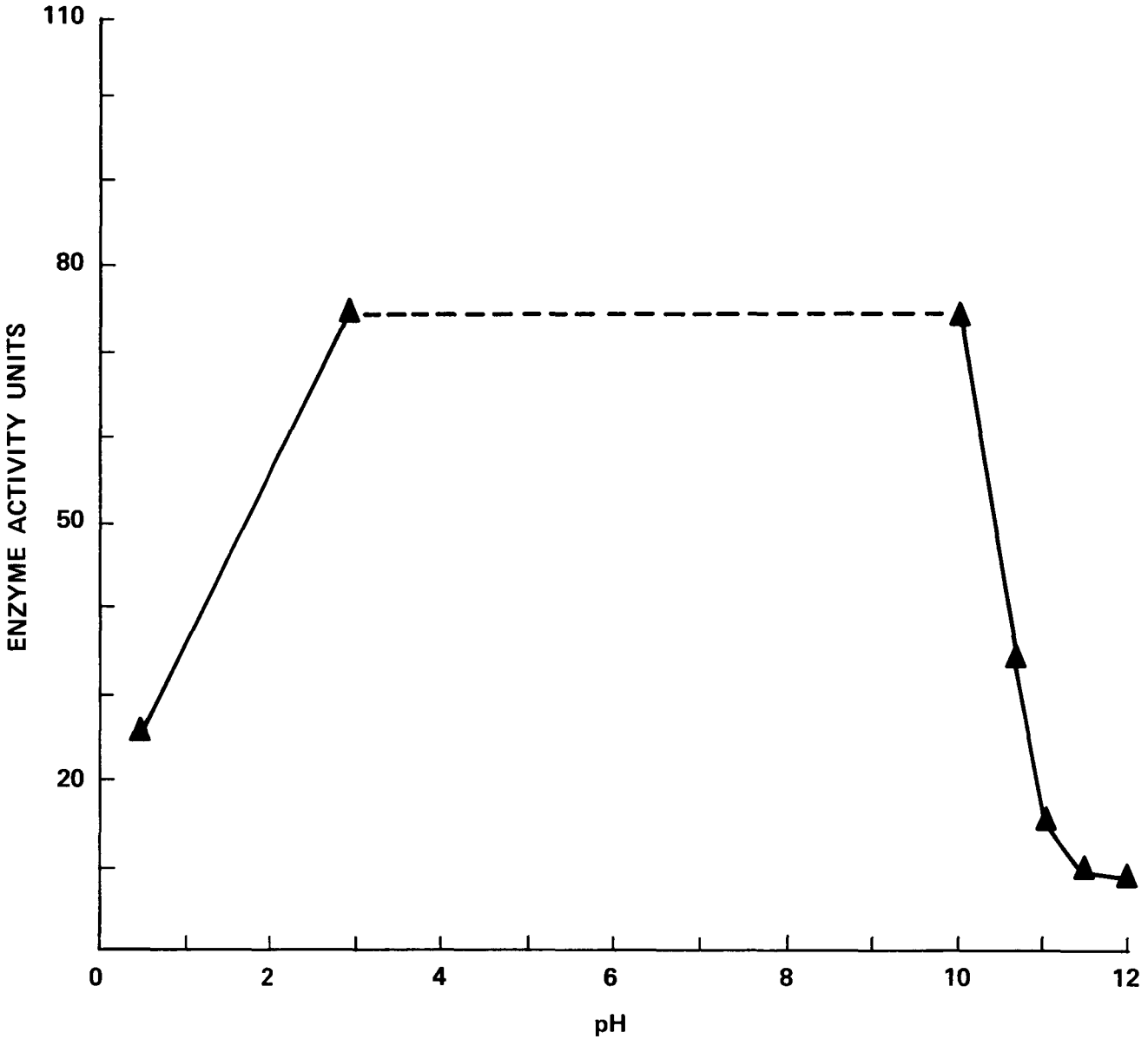


Figure 15. Enzyme activity as a function of pH synthetic brine, $35^{o}S$, acetic acid.

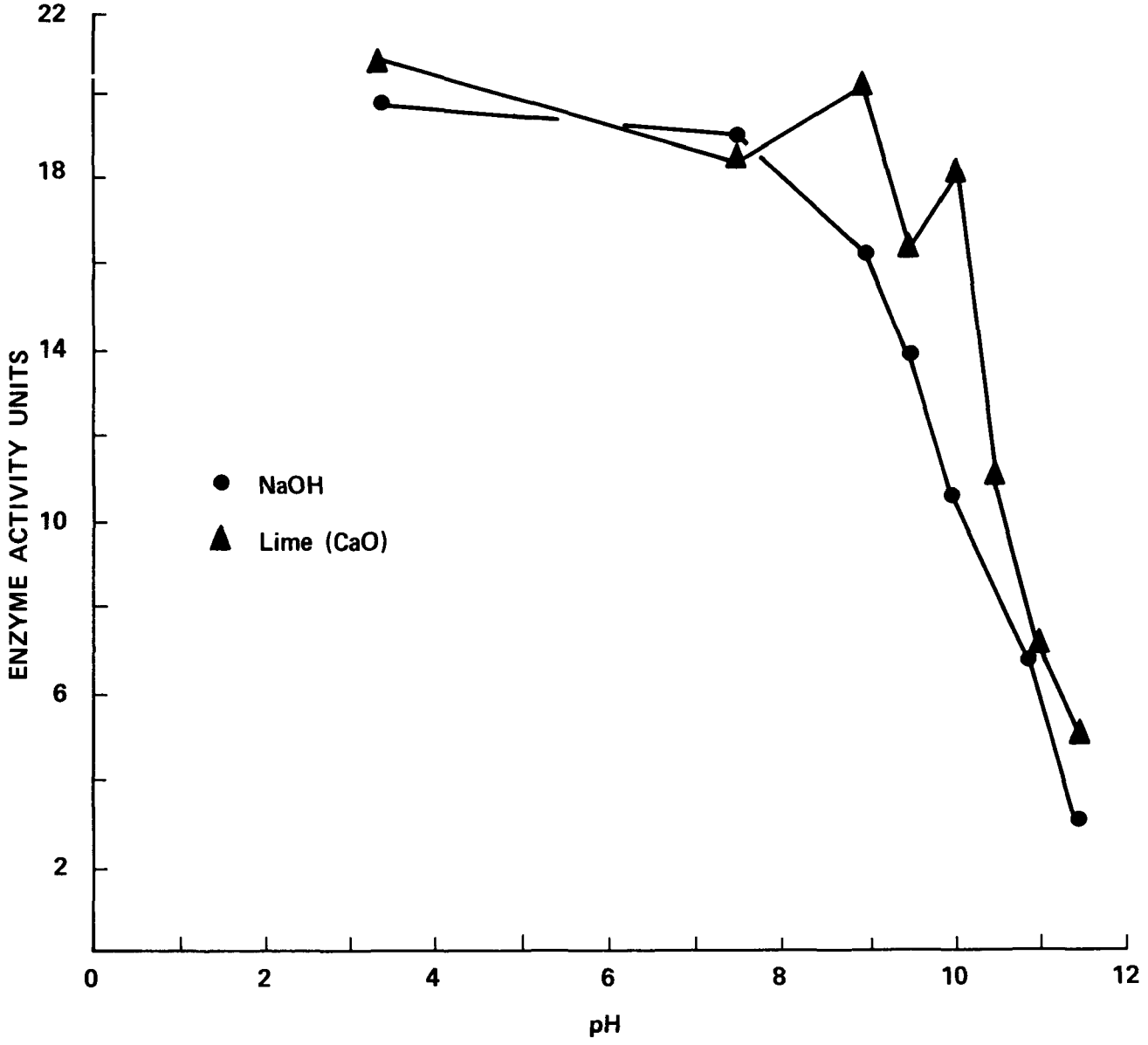


Figure 16. Enzyme activity of a spent brine as a function of pH.

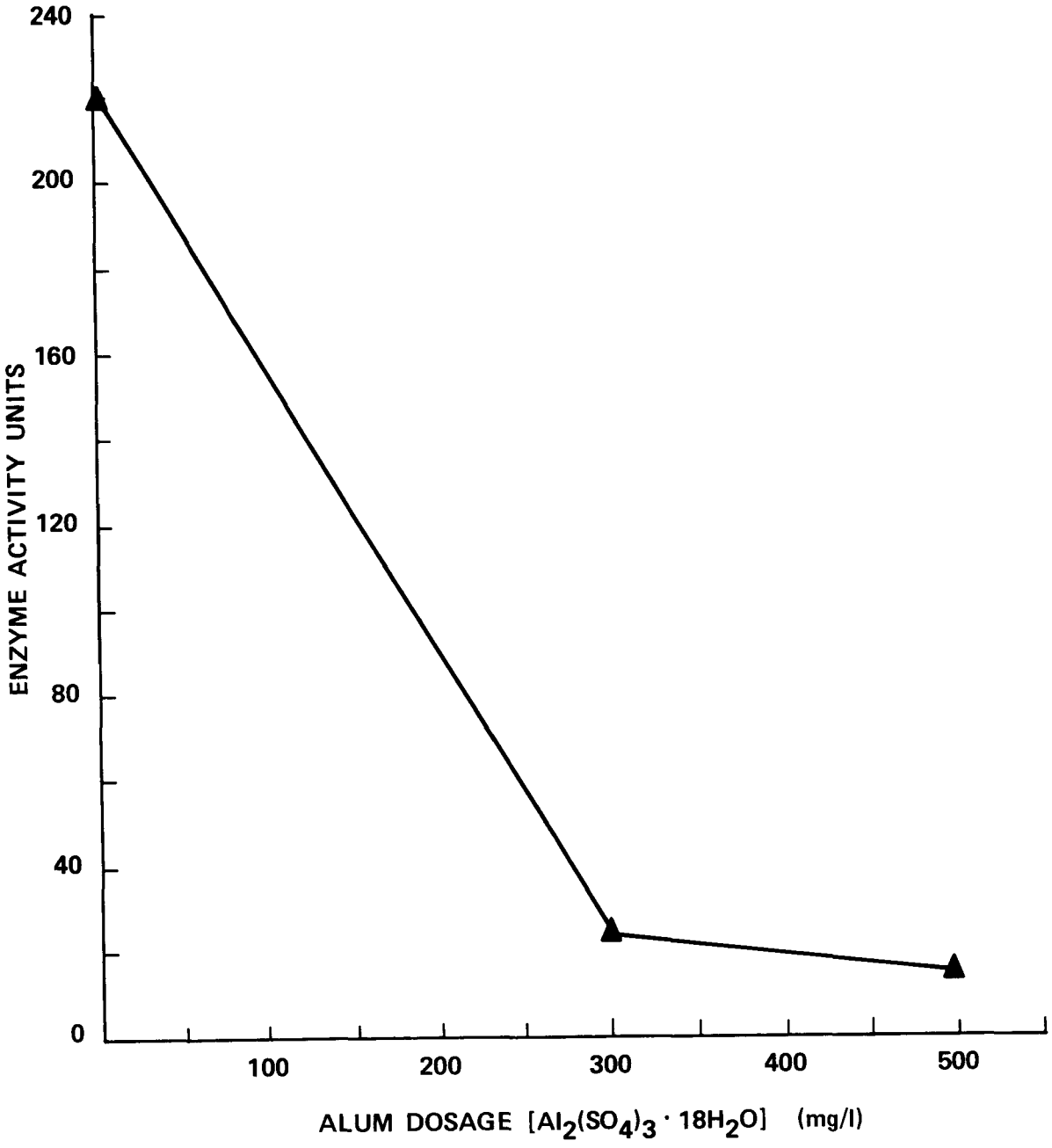


Figure 17. Pectinase activity as a function of alum dosage in a distilled water sample.

Due to constraints of space, time, and money, only limited studies were conducted on recycling spent brines. These laboratory studies were conducted as described above (p. 18), using large (no. 3) cucumbers and the conventional NF procedure. Two control pails received fresh 30°S brine according to plant specifications. Two pails were brined with recycled NF brine from the tankyard studies, diluted to 30°S. One of these received brine which had received the high pH (lime, sodium hydroxide) treatment; the other, untreated brine from the same tank. The NF brine came from Tank 6, which had a high residual pectinase activity. The final two pails received undiluted, untreated CCF brine from Tank 5. Salometer data for the studies is shown in the Appendix (Table A-11). Results of the studies are shown in Tables 21 and 22. Following active fermentation and a three month curing period, the brinestock was examined for its quality. While these studies were done on a very limited scale, some observations seem warranted. Acidity in the pails receiving recycled CCF brine was high to begin with and increased rapidly in the early part of the fermentation (Table 22). Acidity developed the slowest in the untreated recycled NF brine and reached the lowest final value(equal, in fact, to the starting value of the CCF brines). Treated recycled NF brine started with low acidity, but reached a level nearly as high as that achieved in recycled CCF brine.

The most intriguing results were noted in evaluating brinestock quality (Table 21). In terms of bloaters, brinestock quality was poorest in the case of the directly recycled CCF brines. It was conjectured that this was due to the high initial acidity of the brines and possible inhibition of the desired fermenting organisms. However, since the pails were relatively shallow they were not purged. Therefore, an alternate possibility was that in the CCF brine there was such a high inoculum of the desired organisms that the fermentation was intense and accompanied by unusually extensive carbon dioxide production, concommitant with bloating. In terms of bloaters, the high quality of the NF brinestock is worth noting. On the whole, differences among the tests in terms of firmness (pressure test) is unremarkable. It is remarkable that untreated recycled NF brine produced firm brinestock, despite the pectinase content. From a subjective standpoint, those evaluating the brinestock felt that the quality of brinestock from CCF recycling was substantially poorer from that from other treatments.

Degree of cure in different treatments deserves comment, as this study corroborates studies with tankyard recycling of spent NF brines. That is, degree of cure achieved was more variable and tended to be lower in brinestock brined in high pH treated NF brine.

ECONOMIC EVALUATION

A realistic evaluation of the differences in cost of the NF and CCF procedures could not be made. It became apparent early in the study that on the typical commercial tankyard the monitoring and care of NF tanks was considerably less extensive than that outlined in the recommended procedure and therefore less extensive than that provided for our experimental NF tanks. It was our distinct impression that the overall high quality of the NF brinestock produced in our studies was due in large part to the care given the tanks during the early fermentation period. It should be noted, however,

TABLE 21. EFFECTS OF TREATMENT ON BRINESTOCK QUALITY, RECYCLING STUDIES

Pail	Treatment	Bloaters found (%)[1] No.	Balloon	Lens	Honeycomb	Firmness[2] No.	Lb.PT	Cure %
1	Control	19	10.5 (M)	26.0(S-M)	15.5 (S)	10	17.5	80(30-100)
2	"	18	0	5.5(M)	44.0 (S)	10	16.9	80(40-100)
	Recycle,NF							
3	-Treated	18	16.5 (M)	27.5(S-M)	16.5 (S)	10	18.4	70(10-100)
4	-Untreated	19	10.5 (M)	31.5(S-A)	10.5 (S-A)	10	18.9	100
5	Recycled CCF	22	22.5(M-A)	27.0(M-A)	41.0 (S-M)	7	17.6	99
6	" "	20	20.0(A-M)	25.0(S-M)	25.0 (S)	10	17.8	98

[1] No. refers to number examined. Values in parentheses indicate severity of bloating: S=slight; M=moderate; A=advanced. Where two letters are given, the first indicates the severity in the majority of the cucumbers. When a cucumber had more than one type, the balloon-type was given priority. (Fleming et al., 1973a).

[2] U.S. Pressure test in pounds; No. indicates number tested.

TABLE 22. PROGRESS OF FERMENTATION IN RECYCLING STUDIES

Fermentation Time,days	Temp. C	Acid, % as Lactic (Sugar, %)[1] Controls 1	Controls 2	NF Recycle Treated	NF Recycle Untreated	CCF Recycle 1	CCF Recycle 2
0	22						
3	21	0.05 (T)	0.15 (¼)	0.14 (¼)	0.32 (T)	0.55 (T)	0.55 (T)
5	19	0.14 (¼)	0.21 (¼)	0.23 (¼)	0.48 (0)	0.59 (.1)	0.71 (0)
7	19	0.24 (¼)	0.32 (½)	0.26 (¼)	0.41 (0)	0.68 (0)	0.72 (0)
11	19	0.38	0.47	0.47	0.53	0.75	0.79
12	19	0.43 (0)	0.53 (0)	0.59 (0)	0.51 (0)	0.77 (0)	0.73 (0)
18	19	0.50 (0)	0.63 (0)	0.72 (0)	0.55 (0)	0.84 (0)	0.83 (0)
25	19	0.62	0.73	0.90	0.55	0.99	0.92
35	19	0.74	0.76	0.92	0.54	1.09	0.95

[1] T=trace

that we did achieve better consistency in the CCF than in the NF fermentations, despite our care, leading us to believe that while the NF stock can be markedly improved by closer attention to recommended procedures, it is unlikely to reach the levels attained in CCF.

In essence, the only extra labor required for CCF is as follows: 1) initial washing of the cucumbers, 2) sanitizing the brine, 3) acidifying and adjusting the brine, 4) adding the culture, 5) monitoring the purging. If purging is widely adopted for NF brinings, as appears likely, item 5 will no longer constitute a difference. CCF tanks consistently showed less scum and film growth than did NF tanks, and consequently required less maintenance after fermentation was underway.

The only additional capital costs associated with CCF brining are those for the purging apparatus and for storage facilities for the starter cultures. As noted above, the cost differential due to purging will be eliminated if purging is generally accepted for all brinings. Proper storage of currently available commercial cultures turned out to be a major nuisance in this project, since the frozen cultures had to be shipped in dry ice by air freight, picked up immediately, transported 50 miles to the plant site, and stored at approximately -70 C until use. Maintaining such low temperatures requires a source of dry ice and preferably an ultracold freezer. These amenities are rarely available at commercial tankyards or even in nearby towns. A local dairy was able to assist us during the project. However, on a tankyard scale, procuring and storing starter cultures would be a major operation. The manufacturer of one of the starter cultures (Chr. Hansen) indicates to us the possibility that there will soon be available commercial freeze-dried cultures which can be stored in an ordinary deep-freezer.

Water use is initially greater with CCF, since the cucumbers must be washed before brining. However, less water is required for desalting CCF brinestock, providing that plant personnel are educated to gauge volume of desalting water to the amount of salt to be removed from the brinestock.

Relative costs of recycling CCF and NF brines must await further studies. As indicated above, it appears at least possible, if not likely, that direct recycling of CCF brines without treatment may not be an option. Obviously, additional assessment of CCF brine recycle must be made. The aspects briefly noted above (purging vs no purging; neutralized vs untreated; diluted vs direct recycle) could be readily studied in the laboratory to determine the conditions necessary for recycling. Such questions will need resolution before large scale studies should be attempted with recycled CCF brines, since the concept is evidently not quite as simple and straightforward as initially expected.

The major advantage of CCF appeared to be the reliable production of a consistently high quality brinestock and final product. Less variation was noted in quality and in course of active fermentation in the case of the CCF tanks. Taste of the CCF products was quite similar from one brining to another, in contrast to marked taste differences among NF products. However, it must be noted that some people do not favor the pronounced lactic acid flavor of CCF products. With rising costs of raw products (1979 NC costs per hundredweight: 1's, $12; 2's, $6; 3's, $3.75) it appears likely that

cost due to CCF will be minimal when balanced against the possibility of losing a tank of brinestock.

In addition, even if recycling is not practiced, the CCF procedure greatly reduces the salt content of the wastewaters, an important factor in producing a satisfactory plant effluent.

In reference to costs of recycling NF brines, Wendle (1977) performed a desktop evaluation of costs of the three alternate treatment methods usually suggested in the literature: ultrafiltration, coagulation/sedimentation, and heat treatment. For the purpose of this cost analysis, all assumptions in regard to the volume of brine to be recycled were based on operations of the Perfect Packed Products Co. in Henderson. The size of this plant was assumed to be typical of the industry. At this facility, during the first six weeks of green season, there are generated approximately 320,000 gal of 36-hr brines from brining the smaller cucumbers likely to be associated with high pectinase activity. This volume would be generated from brining about 60,000 bushels of no. 1 cucumbers. In addition, about 270,000 bushels of larger sizes would be brined during the season, producing bout 1,450,00 gal of spent brine. For purposes of this comparison, it is assumed that enzyme levels would be checked prior to treatment, and only those with significant pectinase activity would be treated. This is estimated to be about 393,000 gal (all from the no. 1 cucumbers and 5 % from the larger cucumbers).

It was assumed that coagulation/sedimentation would be accomplished by lime addition to a pH of 11, followed by mixing with compressed air, then by sedimentation and clarification under quiescent conditions. It was assumed that ultrafiltration would be accomplished by passage through a commercial hollow-fiber type unit, at a rate of 10 gpm or 15 gpm, at pore sizes suitable for pectinase removal (Little et al., 1976). It was assumed that heat treatment would be accomplished by flash pasteurizagion, by raising the brine temperature to 165°F for 15 sec, followed by cooling to 50°F. Costs of three fuels were estimated, i.e., natural gas, no.2 fuel oil, and propane. Complete details and calculations are given in Wendle (1977). In brief, for heat treatment fuel costs alone would be, per year, $1400 for natural gas, $2100 for no. 2 fuel oil, and $3000 for propane. Current prices would be somewhat higher, as costs of fuels are generally rising. For coagulation/sedimentation, followed by recycle after neutralization with acetic acid, total annual costs were estimated to be $1900; without the neutralization, $1200. Costs of ultrafiltration would be largely associated with the capital cost and the life of the unit would be critical. At 15 gpm capacity with an estimated 10 yr economic life, cost would be per year $2450; with a 20 yr life, $1950. At 10 gpm capacity, at 10 yr cost would be $2050; 20 yr life, $1650. Labor costs are not considered for any of these alternatives. Wendle pointed out that ultrafiltration might provide an attractive treatment alternative since in addition to economic feasibility there would be the following advantages: 1) the chemical addition needed for coagulation/precipitation would not be a factor, 2) the brine would be basically unchanged in soluble chemical content, but removal of enzymes and bacteria would leave a clear sterile brine, 3) wastes from the process would be minimal, 4) the process would be relatively independent of rising fuel and chemical costs. The major disadvantage would be that estimating the life of the equipment under the low pH-high salt conditions imposed by the contact with the brines is presently impossible. It

would be possible to get realistic information on performance only by large scale studies over a period of time.

Wendle (1977) also made the interesting observation that each of the three alternate methods would provide a net savings to the industry just in terms of reduction of salt use. He noted that under the assumptions he employed, approximately 120 tons of salt would be wasted by discarding only the 36-hr brines. If only 80% of this could be recovered for reuse, at a cost of $32 per ton for rock salt an annual savings of around $3100 could be realized, more than enough to pay for high pH treatment or ultrafiltration.

REFERENCES

APHA, AWWA, WPCF. 1976. Standard Methods for the Examination of Water and Wastewater, 14th ed. American Public Health Association, Washington, DC.

Bell, T.A., J.L. Etchells, and I.D. Jones. 1955. A Method for Testing Cucumber Salt-Stock Brine for Softening Activity. Agricultural Research Service ARS-72-5, U.S. Dept. of Agriculture.

Dixon, M., and E.C. Webb. 1964. Enzymes. Academic Press, Inc., N.Y.

EPA. 1974. Methods for Chemical Analysis of Water and Wastes. US-EPA, Methods Development and Quality Assurance Research Laboratory, Cincinnati, OH.

Etchells, J.L., T.A. Bell, H.P. Fleming, R.E. Kelling, and R.L. Thompson. 1973. Suggested Procedure for the Controlled Fermentation of Commercially Brined Pickling Cucumbers--The Use of Starter Cultures and Reduction of Carbon Dioxide Accumulation. Pickle Pak Sci. 3: 4-14.

Etchells, J.L., T.A. Bell, H.P. Fleming, R.E. Kelling, and R.L. Thompson. 1974. Bloater Chart. Published and distributed by Pickle Packers International, Inc., St. Charles, IL.

Fleming, H.P., R.L. Thompson, J.L. Etchells, R.E. Kelling, and T.A. Bell. 1973a. Bloater Formation in Brined Cucumbers Fermented by Lactobacillus plantarum. Jour. Food Sci. 38: 499-503.

Fleming, H.P., R.L. Thompson, J.L. Etchells, R.E. Kelling, and T.A. Bell. 1973b. Carbon Dioxide Formation in the Fermentation of Brined Cucumbers. Jour. Food Sci. 38: 504-506.

Fleming, H.P., R.L. Thompson, and T.A. Bell. 1974. Quick Method for Estimating CO_2 in Cucumber Brines. Advisory Statement from the U.S. Food Fermentation Laboratory, Agricultural Research Service, USDA, Raleigh, NC.

Fleming, H.P., R.L. Thompson, T.A. Bell, and R.J. Monroe. 1977. Effect of Brine Depth on Physical Properties of Brine-Stock Cucumbers. Jour. Food Sci. 42: 1464-1470.

Geisman, J.R., and R.E. Henne. 1973. Recycling Brine from Pickling. Ohio Report 58(4): 76-77.

Henne, R.E., and J.R. Geisman. 1973. Recycling Spent Cucumber Brines. Presented at 1973 Food Processing Waste Management Conf., Syracuse, NY, Mar. 26-28, 1973.

Little, L.W., J.C. Lamb III, and L.F. Horney. 1976. Characterization and Treatment of Brine Wastewaters from the Cucumber Pickle Industry. Report No. 99, Univ. of NC Water Resources Research Institute, Raleigh, NC.

McFeeters, R.F., W. Coon, M.P. Palnitkar, M. Velting, and N. Fehringer. 1978. Reuse of Fermentation Brines in the Cucumber Pickling Industry. EPA-600/2-78-207, US Environmental Protection Agency, Cincinnati, OH.

Palnitkar, M.P., and R.F. McFeeters. 1974. Use of Recycled Brines for Cucumber Fermentations. Paper presented at annual meeting of Pickle Packers International, Inc., Chicago, IL, December, 1974.

Popper, K., W.M. Camirand, G.G. Walters, R.J. Bouthilet, and F.P. Boyle. 1967. Recycles Process Brine, Prevents Pollution. Food Engng. 39: 78-80.

Tenney, M.W., and W. Stumm. 1965. Chemical Flocculation of Microorganisms in Biological Waste Treatment. Jour. Water Poll. Control Fed. 37: 1370-1388.

Wendle, J.G. 1977. Treatment Methods for Removal of Pectinase Activity in Spent Cucumber Pickling Brines. Master's Report for M.S. in Environmental Engineering in the Dept. of Environmental Sciences and Engineering, University of North Carolina at Chapel Hill.

SECTION 7

LIST OF PUBLICATIONS

To date, publications resulting from this project are as follows:

Little, L. W., R. Harrison, J. Davis, J. Harris, and S. J. Dunn. 1977. Reduction of Wastes from Cucumber Pickle Processing by Use of the Controlled Culture Fermentation Process, pp. 322-332, in Proc. Eighth National Symp. on Food Processing Wastes, EPA - 600/ 2-17-184.

Wendle, J. G. 1977. Treatment Methods for Removal of Pectinase Activity in Spent Cucumber Pickling Brines. Master's report for M.S. in Environmental Engineering in the Department of Environmental Sciences & Engineering, University of North Carolina at Chapel Hill. 121 pp.

Appendix

TABLE A-1. BRINE CHARACTERISTICS DURING NATURAL FERMENTATION PROCESS
TANK NO. 1, EXPERIMENT NO. 1

Time		Degrees Salom		% Acid as Lactic	pH	Temp °F
Date	Days	Top	Bottom			
June 23	0	--	--	--	--	--
June 24	1	17	18	0.05	5.3	82
June 25	2	20	21	0.21	5.2	87
June 26	3	20	22	0.23	4.6	85
June 27	4	24	26	0.28	3.9	84
June 29	6	24	25	0.38	3.7	87
July 1	8	24	25	0.50	3.4	83
July 5	12	15	26	0.65	3.3	86
July 9	16	24	27	0.68	3.2	82
July 13	20	30	34	0.53	3.2	84
July 19	26	33	36	0.41	3.2	84
July 23	30	32	35	0.48	3.0	85
Aug. 5	43	23	40	0.47	3.2	78
Aug. 11	49	39	41	0.50	3.2	80
Aug. 16	54	34	43	0.45	3.5	85

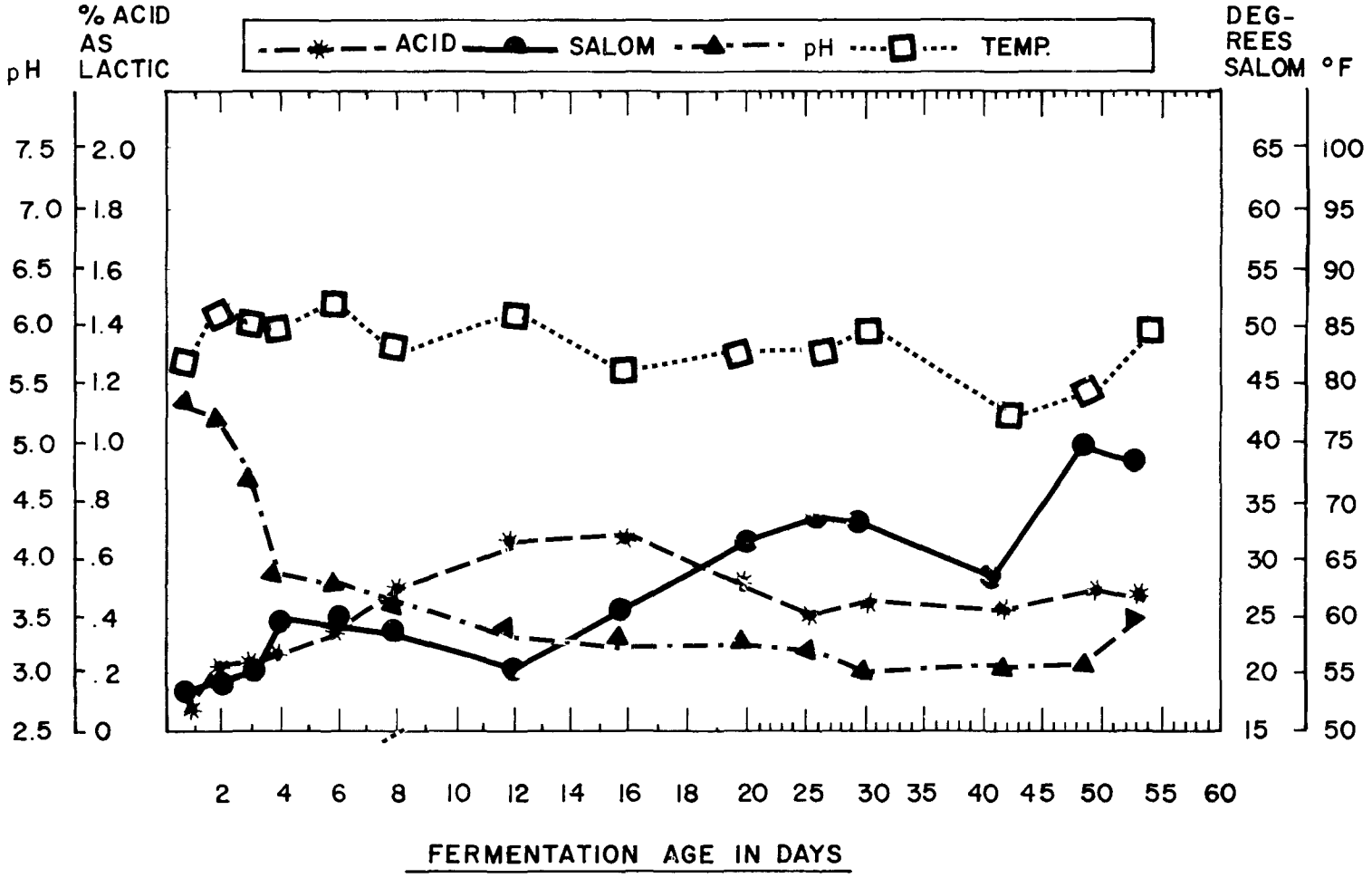


Figure A-1. Fermentation curves for natural fermentation process, tank no. 1, experiment no. 1.

TABLE A-2. BRINE CHARACTERISTICS DURING CONTROLLED CULTURE FERMENTATION PROCESS, TANK NO. 3, EXPERIMENT NO. 1

Time		Degrees Salom		% Acid as Lactic	pH	Temp °F
Date	Days	Top	Bottom			
June 23	0	--	--	--	--	--
June 24	1	26	27	0.21	3.6	80
June 25	2	22	21	0.27	4.7	86
June 26	3	21	21	0.48	4.0	86
June 27	4	23	23	0.68	3.9	88
June 29	6	22	23	0.86	3.8	87
July 1	8	25	26	0.91	3.6	83
July 5	12	25	26	0.94	3.6	84
July 9	16	25	26	0.97	3.4	82
July 13	20	25	27	0.98	3.3	80
July 19	26	27	28	1.04	3.3	84
July 23	30	27	28	0.96	3.3	85
Aug. 5	43	27	27	1.03	3.5	77
Aug. 11	49	27	29	1.04	3.5	83
Aug. 16	54	25	32	0.84	3.7	83

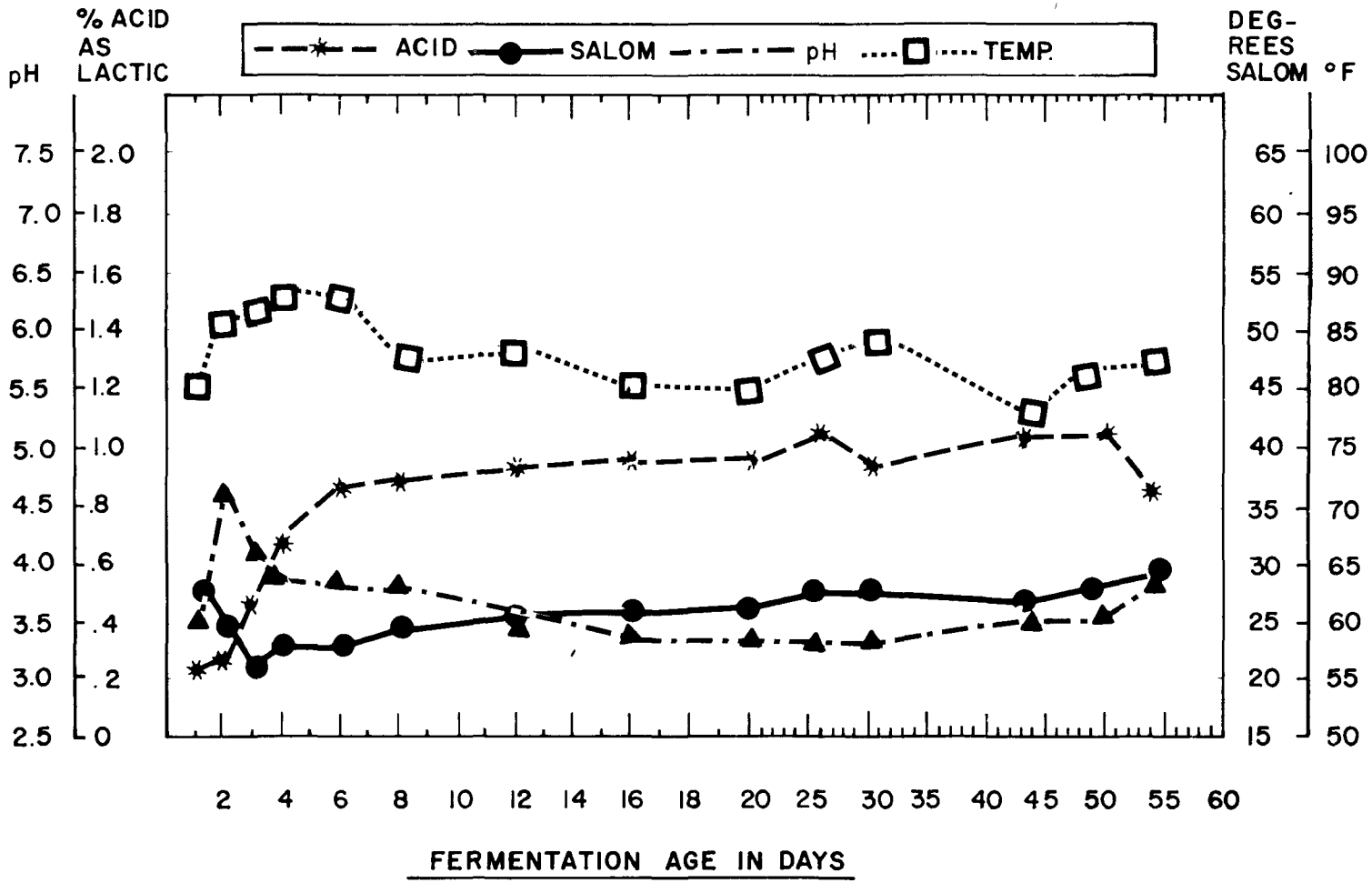


Figure A-2. Fermentation curves for controlled culture fermentation process, tank no. 3, experiment no. 1.

TABLE A-3. BRINE CHARACTERISTICS DURING NATURAL FERMENTATION PROCESS, TANK NO. 7, EXPERIMENT NO. 2

Time		Degrees Salom		% Acid As Lactic	pH	Temp °F
Date	Days	Top	Bottom			
July 8	0	--	--	--	--	--
July 9	1	--	--	--	--	--
July 10	2	19	22	0.06	4.9	81
July 11	3	24	28	0.08	4.0	86
July 12	4	23	28	0.24	3.4	86
July 14	6	24	28	0.51	3.0	84
July 16	8	24	27	0.63	3.0	85
July 19	11	25	31	0.41	3.2	85
July 21	13	26	30	0.72	3.2	86
July 23	15	25	28	0.77	3.1	85
Aug. 5	28	20	30	0.81	3.2	78
Aug. 11	34	32	33	0.83	3.3	82
Aug. 16	39	35	39	0.71	3,4	80
Aug. 25	48	38	40	0.70	3.2	82

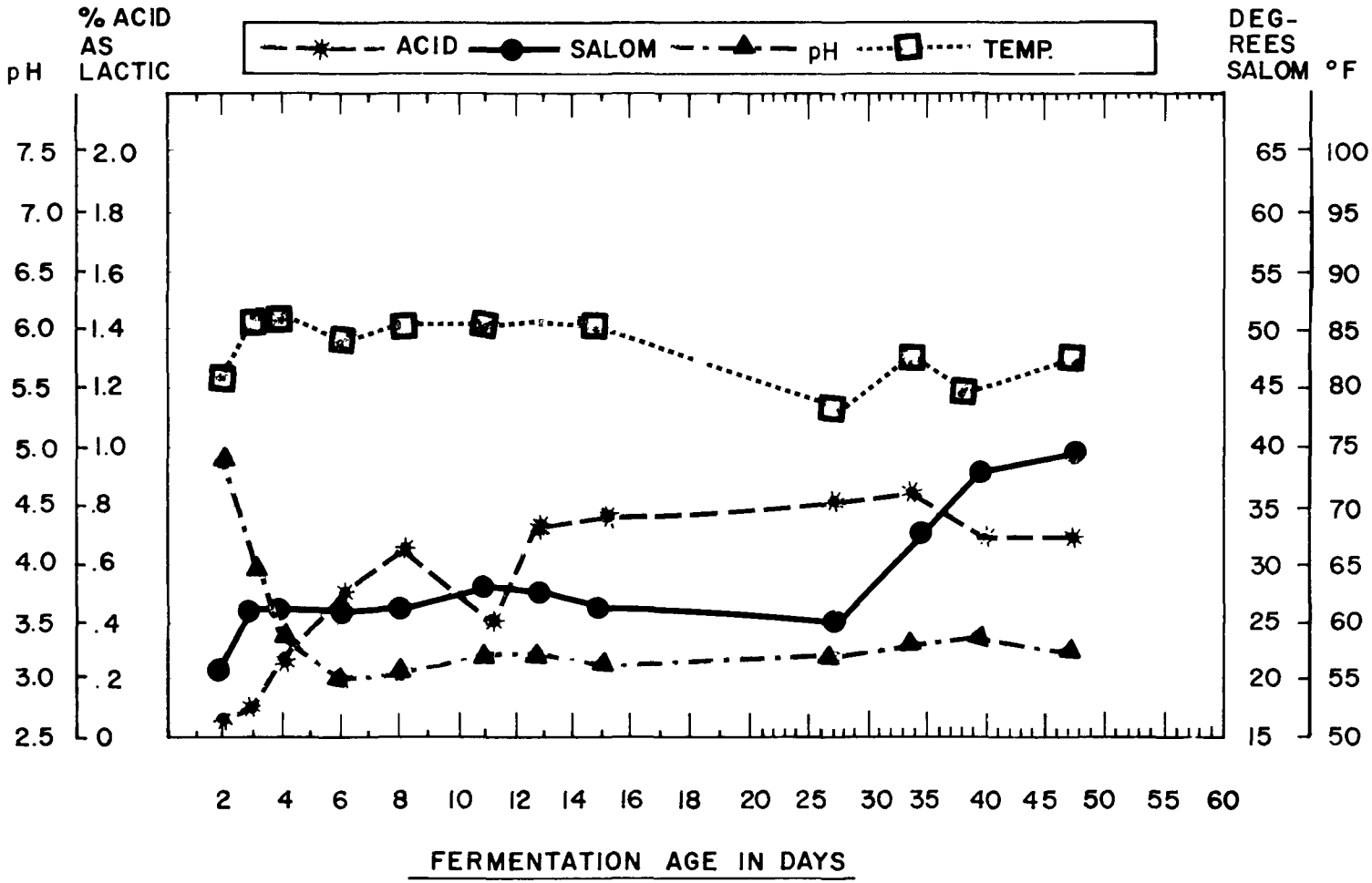


Figure A-3. Fermentation curves for natural fermentation process, tank no. 7, experiment no. 2.

TABLE A-4. BRINE CHARACTERISTICS DURING CONTROLLED CULTURE FERMENTATION PROCESS, TANK NO. 8, EXPERIMENT NO. 2

Time		Degrees Salom		% Acid As Lactic	pH	Temp °F
Date	Days	Top	Bottom			
July 9	0	--	--	--	--	--
July 10	1	--	--	--	--	--
July 11	2	25	26	0.25	4.6	83
July 12	3	23	24	0.35	4.1	82
July 13	4	24	25	0.47	3.8	82
July 15	6	22	25	0.72	3.4	86
July 17	8	23	25	0.90	3.5	86
July 19	10	24	26	0.96	3.4	86
July 21	12	24	26	0.96	3.6	86
July 23	14	24	25	1.02	3.2	86
Aug. 5	27	25	25	1.12	3.6	74
Aug. 11	33	--	--	1.13	--	--
Aug. 16	38	21	31	0.92	3.6	84
Aug. 25	47	27	28	1.03	3.3	82

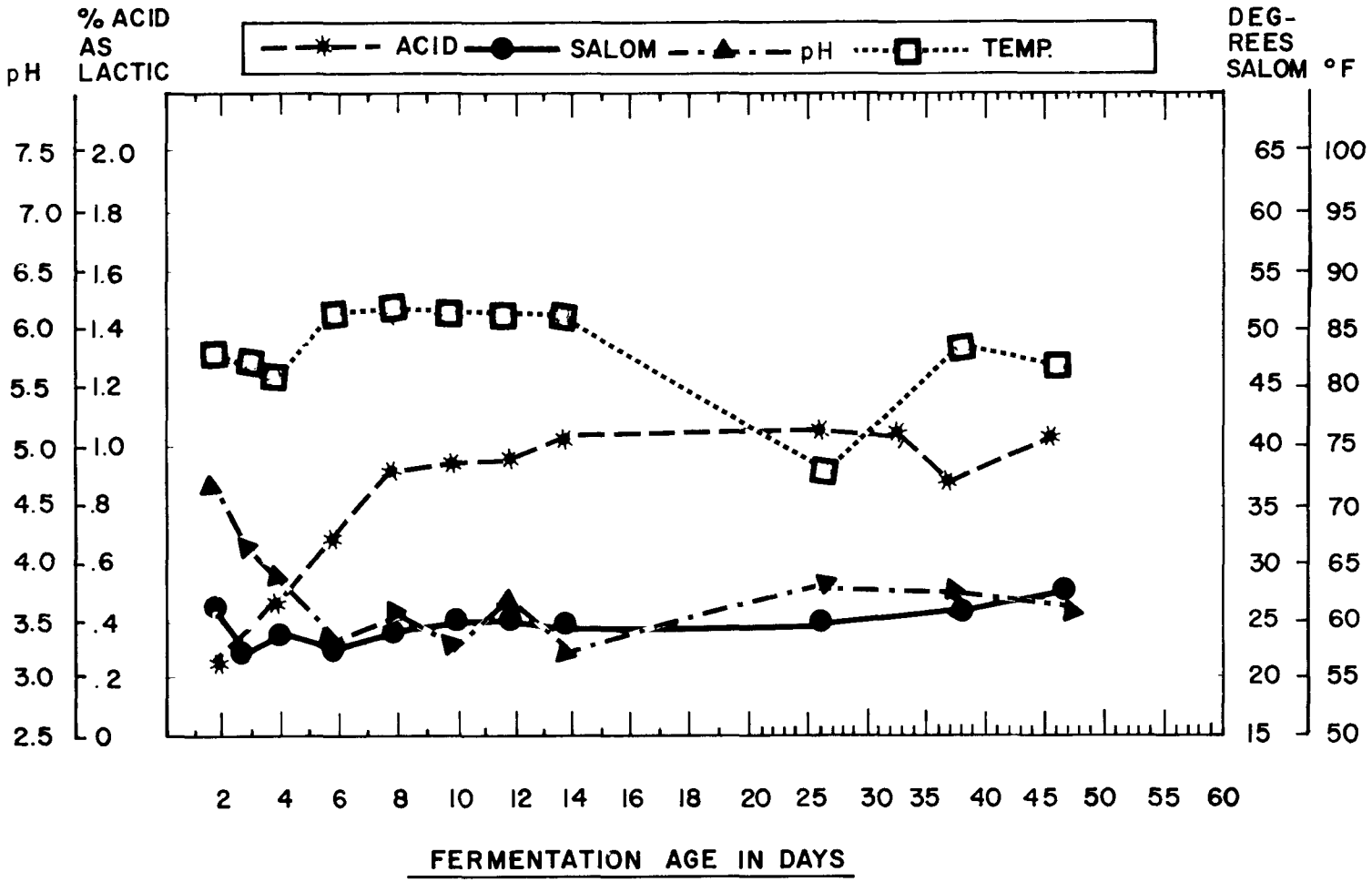


Figure A-4. Fermentation curves for controlled culture fermentation process, tank no. 8, experiment no. 2.

TABLE A-5. BRINE CHARACTERISTICS DURING NATURAL FERMENTATION PROCESS, TANK NO. 9, EXPERIMENT NO. 2

Time		Degrees Salom		% Acid As Lactic	pH	Temp °F
Date	Days	Top	Bottom			
July 8	0	--	--	--	--	--
July 9	1	--	--	--	--	--
July 10	2	18	20	.06	4.9	80
July 11	3	24	27	.14	3.8	84
July 12	4	23	26	.25	3.5	84
July 14	6	23	25	.47	3.2	83
July 16	8	23	26	.54	3.2	83
July 19	11	25	30	.47	3.2	84
July 21	13	24	29	.68	3.0	85
Aug. 5	28	20	32	.58	3.0	77
Aug. 11	34	32	32	.70	3.3	82
Aug. 16	39	36	39	.63	3.6	82
Aug. 25	48	36	38	.67	3.3	82

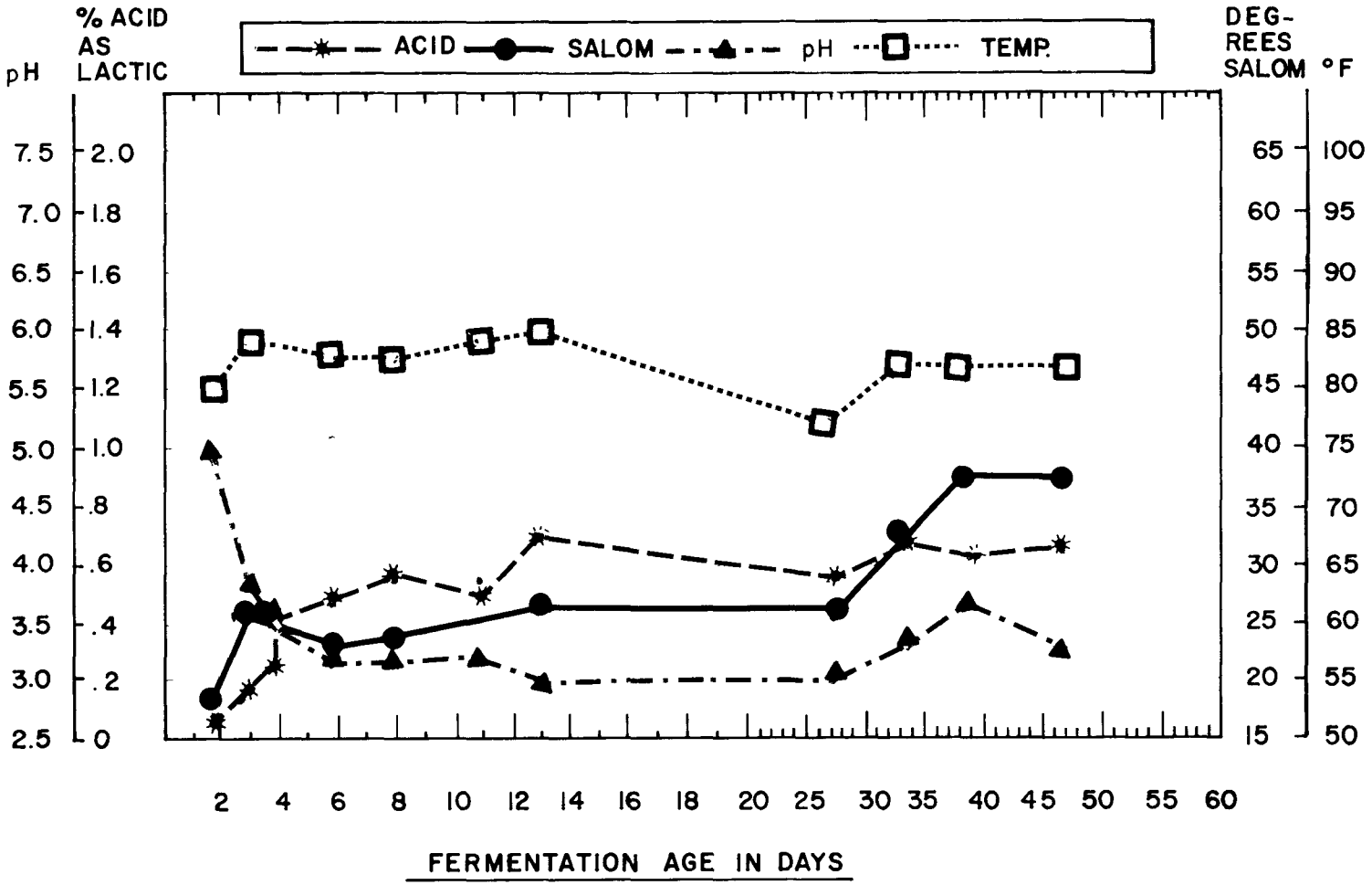


Figure A-5. Fermentation curves for controlled culture fermentation process, tank no. 9, experiment no. 2.

TABLE A-6. BRINE CHARACTERISTICS DURING CONTROLLED CULTURE FERMENTATION PROCESS, TANK NO. 10, EXPERIMENT NO. 2

Time		Degrees Salom		% Acid As Lactic	pH	Temp °F
Date	Days	Top	Bottom			
July 9	0	--	--	--	3.6	--
July 10	1	--	--	--	4.8	--
July 11	2	26	28	0.23	4.6	82
July 12	3	24	25	0.32	4.2	83
July 13	4	24	26	0.48	3.9	83
July 15	6	23	24	0.70	3.6	84
July 17	8	24	26	0.82	3.4	87
July 19	10	24	27	0.91	3.3	85
July 21	12	24	26	0.97	3.5	87
July 23	14	24	26	1.03	3.3	87
Aug. 5	27	26	27	1.12	3.4	75
Aug. 11	33	--	--	1.07	--	--
Aug. 16	38	22	26	0.95	3.7	85
Aug. 25	47	26	27	0.95	3.4	83

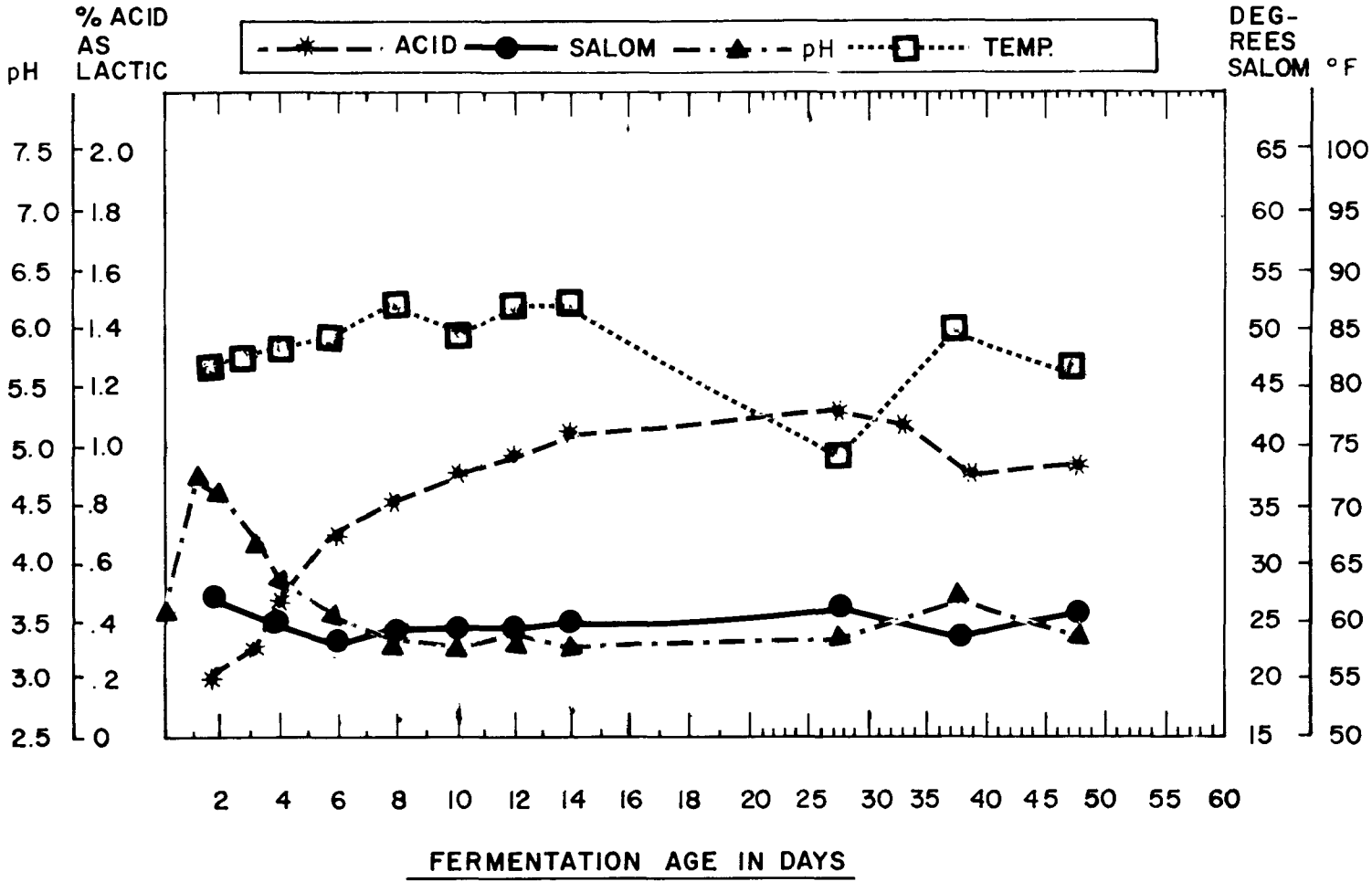


Figure A-6. Fermentation curves for controlled culture fermentation process, tank no. 10, experiment no. 2.

TABLE A-7. BRINE CHARACTERISTICS DURING CONTROLLED CULTURE FERMENTATION PROCESS, TANK NO. 4, EXPERIMENT NO. 3

Time		Degrees Salom		% Acid As Lactic	pH	Temp °F
Date	Days	Top	Bottom			
Aug. 3	0	--	--	--	--	--
Aug. 4	1	27	29	0.27	4.7	81
Aug. 5	2	21	22	0.24	--	81
Aug. 6	3	26	27	0.33	4.6	80
Aug. 7	4	25	28	0.68	3.9	80
Aug. 8	5	24	26	0.77	3.8	77
Aug. 11	8	23	24	1.05	3.6	83
Aug. 16	13	27	31	1.05	3.6	81
Aug. 19	16	28	30	1.13	3.4	78
Aug. 25	22	28	30	1.15	3.2	82
Aug. 30	27	25	30	1.40	--	76
Sept. 2	30	25	35	1.40	3.2	69
Sept. 8	36	35	36	1.30	--	69
Sept. 23	51	35	35	1.30	3.6	72

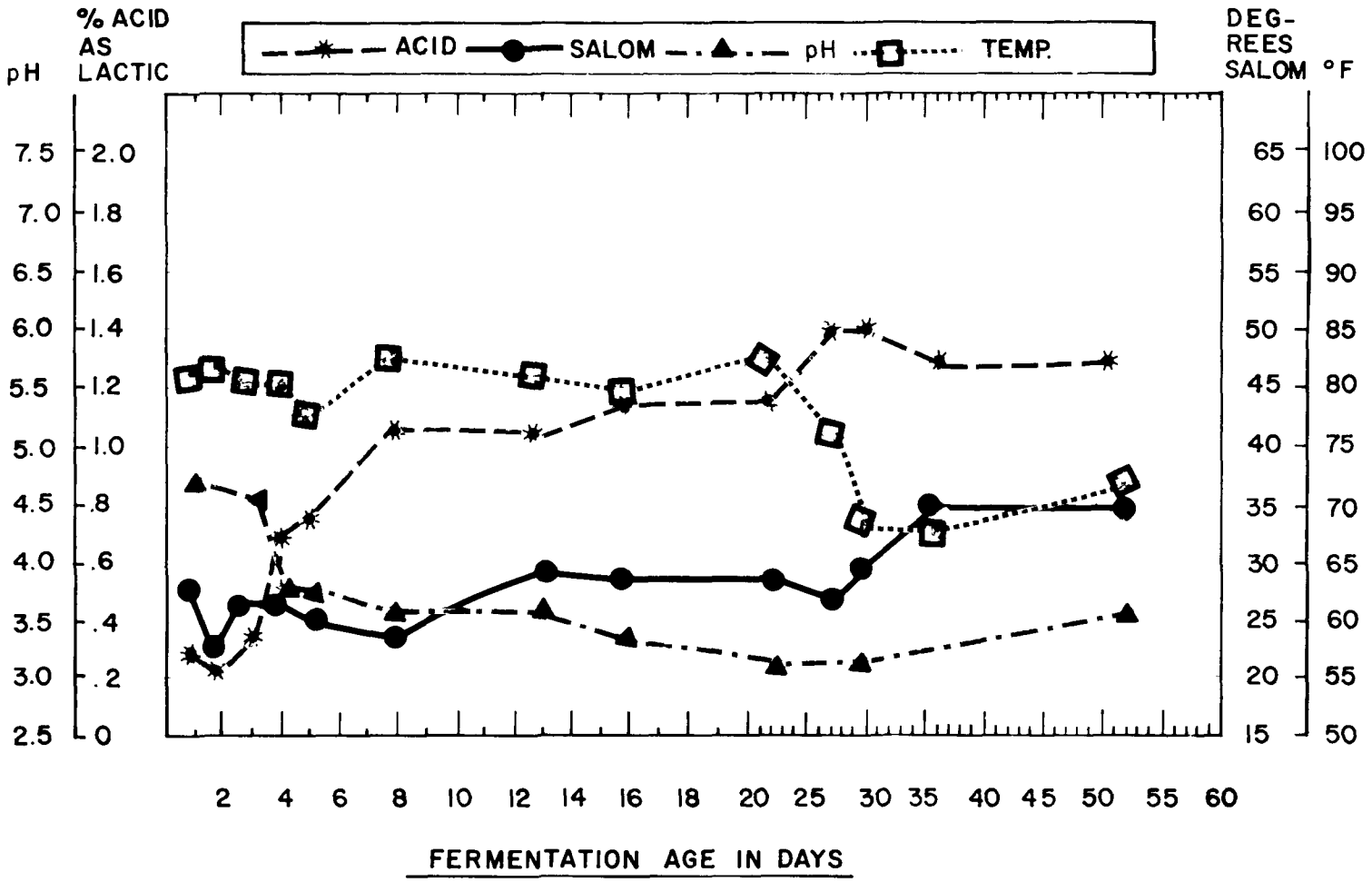


Figure A-7. Fermentation curves for controlled culture fermentation process, tank no. 4, experiment no. 3.

TABLE A-8. BRINE CHARACTERISTICS DURING CONTROLLED CULTURE FERMENTATION PROCESS, TANK NO. 5, EXPERIMENT NO. 3

Time		Degrees Salom		% Acid As Lactic	pH	Temp °F
Date	Days	Top	Bottom			
Aug. 3	0	--	--	--	--	--
Aug. 4	1	27	27	0.26	4.6	81
Aug. 5	2	21	23	0.24	--	81
Aug. 6	3	27	28	0.36	4.4	80
Aug. 7	4	26	33	0.56	4.0	80
Aug. 8	5	25	26	0.71	3.8	77
Aug. 11	8	25	26	0.96	3.6	83
Aug. 16	13	22	26	1.03	3.5	86
Aug. 19	16	26	27	1.10	3.6	81
Aug. 25	22	26	27	1.07	3.4	83
Aug. 30	27	25	28	1.31	--	76
Sept. 2	30	25	29	1.31	3.3	69
Sept. 8	36	26	27	1.21	--	70
Sept. 23	51	25	26	1.17	3.7	72

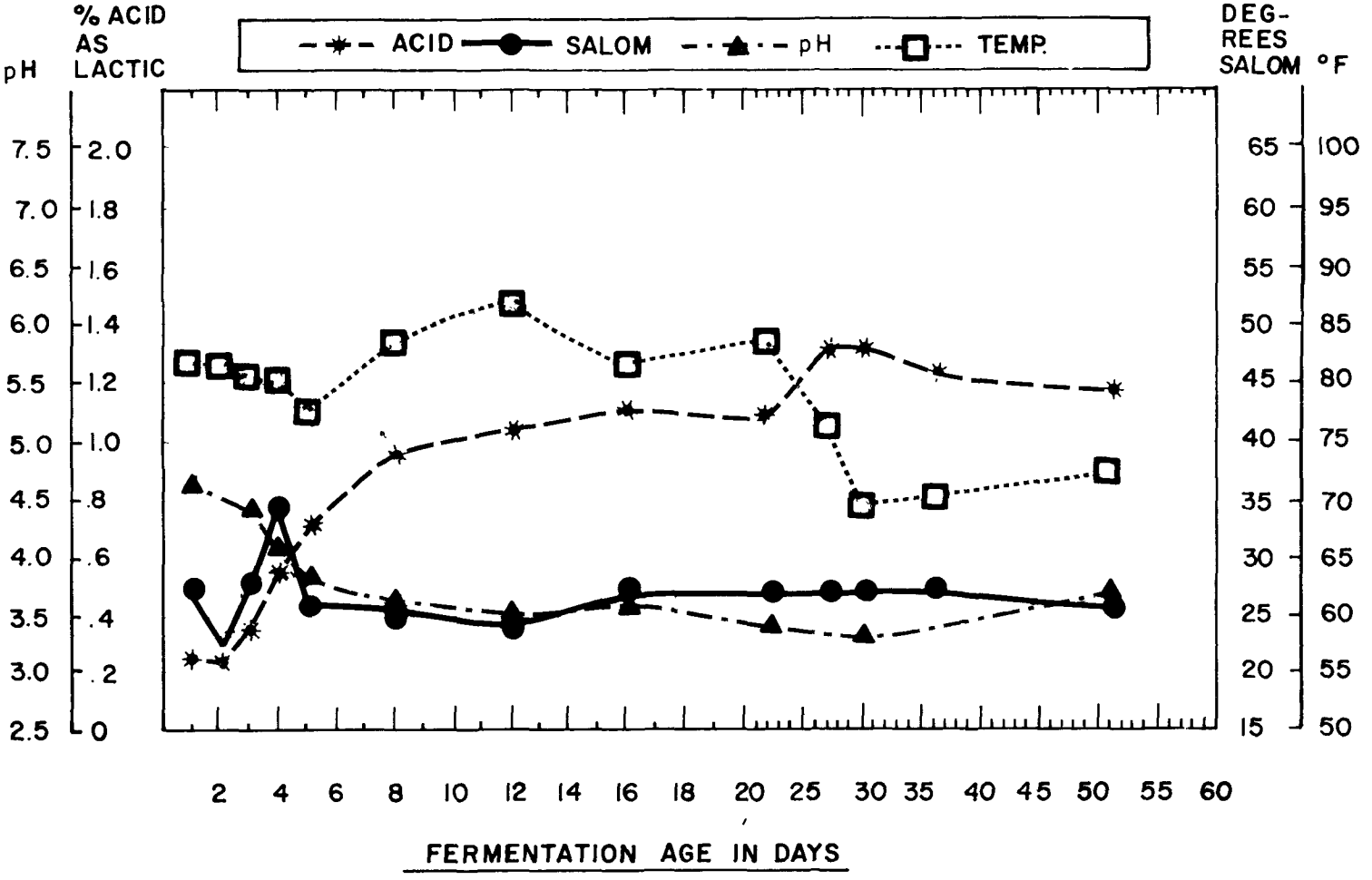


Figure A-8. Fermentation curves for controlled culture fermentation process, Tank no. 5, experiment no. 3.

TABLE A-9. BRINE CHARACTERISTICS DURING NATURAL FERMENTATION PROCESS, TANK NO. 6, EXPERIMENT NO. 3

Time		Degrees Salom		% Acid As Lactic	pH	Temp °F
Date	Days	Top	Bottom			
Aug. 3	0	--	--	--	--	--
Aug. 4	1	27	29	0.04	4.8	81
Aug. 5	2	20	23	0.05	--	83
Aug. 6	3	27	29	0.10	4.0	78
Aug. 7	4	25	26	0.18	3.9	78
Aug. 8	5	26	27	0.24	3.8	77
Aug. 11	8	26	26	0.55	3.6	82
Aug. 16	13	16	26	0.61	3.7	84
Aug. 19	16	28	30	0.76	3.4	82
Aug. 25	22	29	30	0.78	3.2	80
Aug. 30	27	30	30	0.91	--	76
Sept. 2	30	27	30	0.89	3.2	70
Sept. 8	36	34	35	0.86	--	70
Sept. 21	49	36	--	--	--	75
Sept. 23	51	39	39	0.87	3.5	72

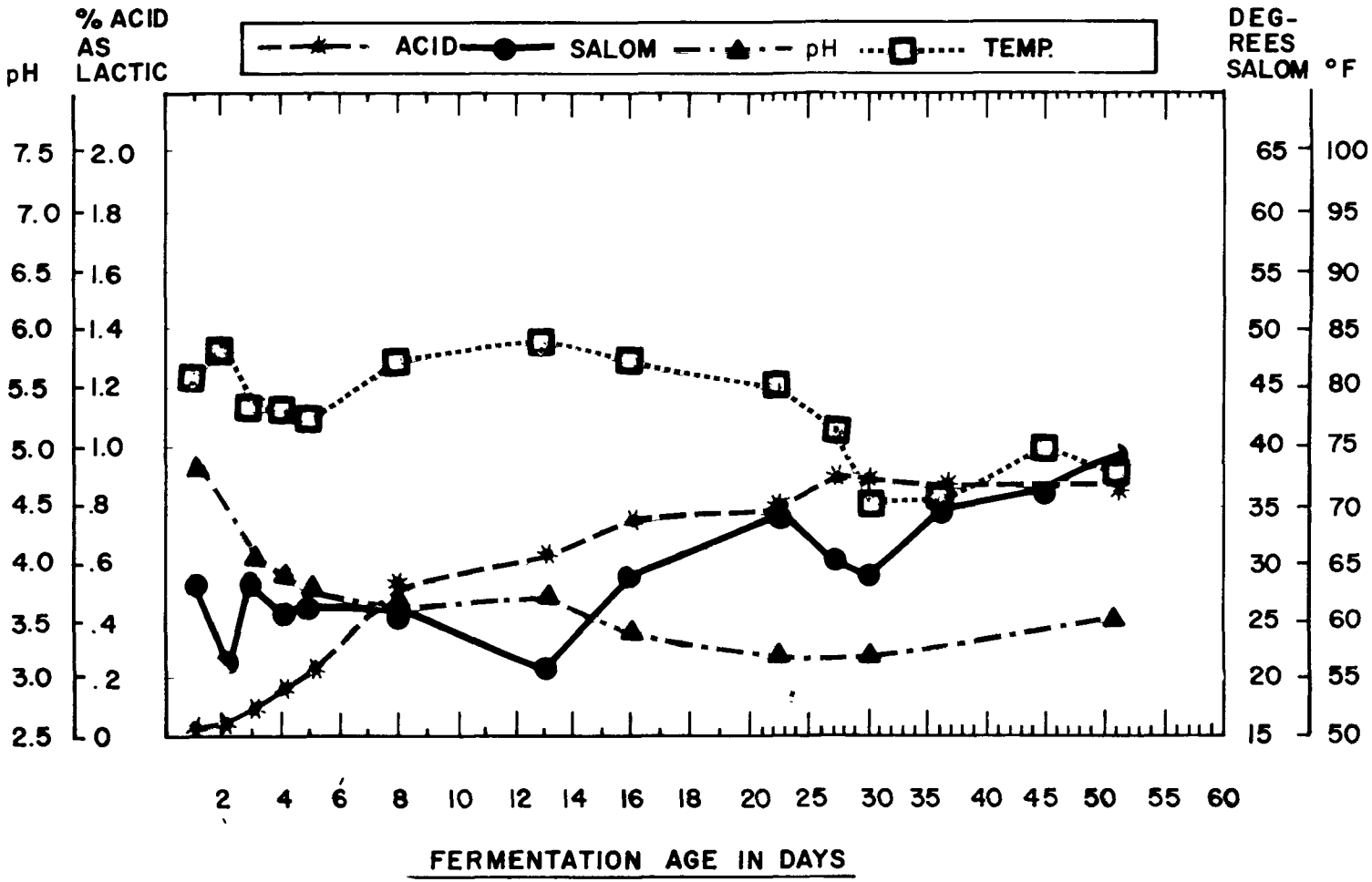


Figure A-9. Fermentation curves for natural fermentation process, tank no. 6, experiment no. 3.

TABLE A-10. STATISTICAL ANALYSIS OF ENZYME ANALYSIS DATA

	Brine #1 (weak)	Brine #1 (moderately active)	Brine #3 (extremely strong)
	6.3	19	500
	4	17	350
	6	20	280
	6	19	280
	5	18	300
	4	16	146
	3	19	100
	4	20	250
	4		
	4		
Sample Size	10	8	8
Mean	4.6	18.5	276
Variance (s^2)	1.26	2	15,130
Std. Dev. (s)	1.12	1.4	123
95% confidence	2.4-6.8	15.7-21.3	30-522

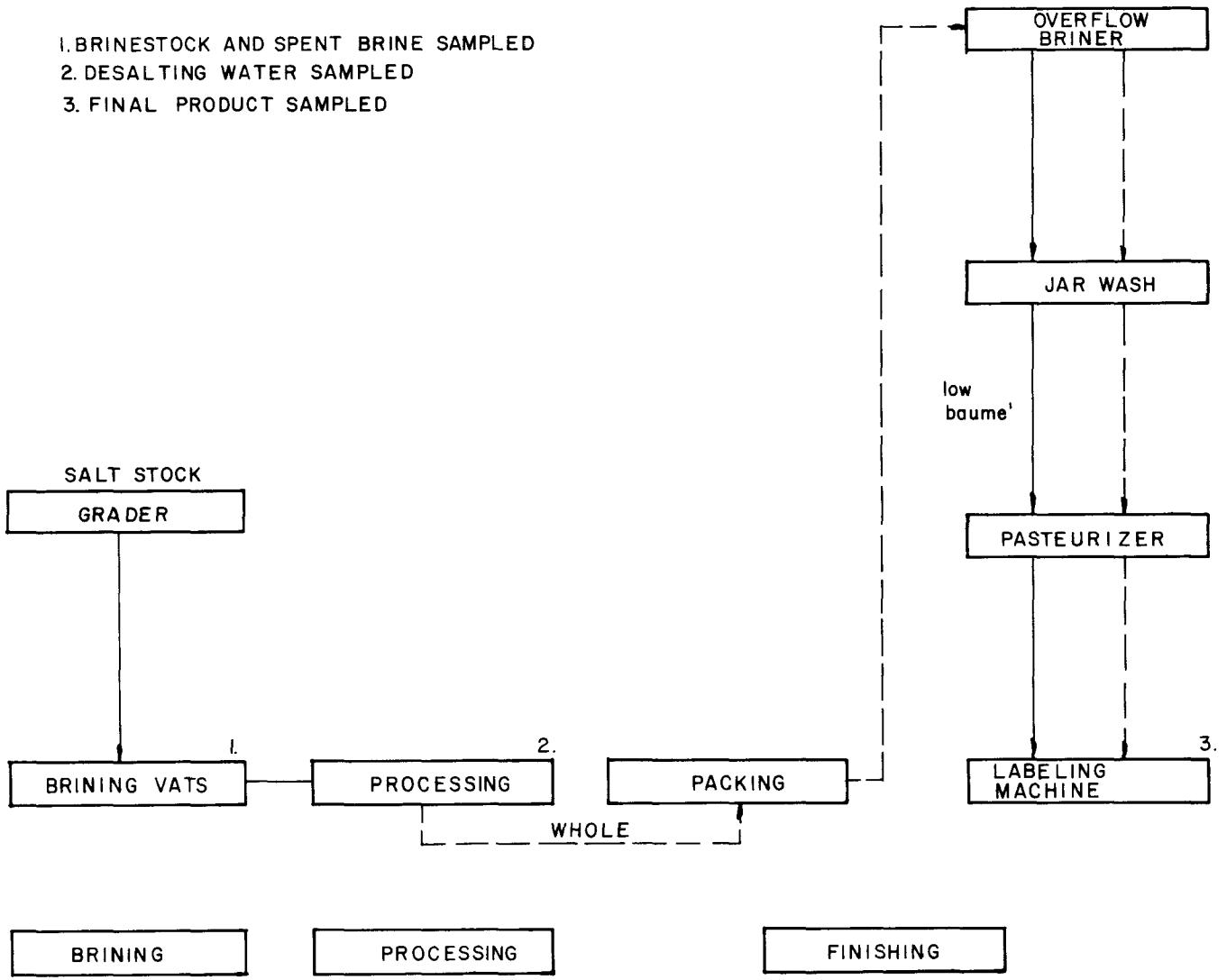


Figure A-10. Flow chart of typical unit processes in the production of processed cucumber pickles.

TABLE A-11. SALOMETER DATA FOR RECYCLING STUDIES

Fermentation time, days	°S					
	Controls		NF, trt.	NF, untrt.	CCF	
0	30	30	30	30	25	25
3	21	20	20	20	17	17
5	21	18	22	24	19	17
7	24	25	24	26	22	22
11	22	25	25	27	25	30
12	24	24	25	30	25	31
18	25	26	26	30	28	31
25	25	25	28	31	30	32
35	31	31	31	31	32	38

TECHNICAL REPORT DATA
(Please read Instructions on the reverse before completing)

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15. SUPPLEMENTARY NOTES

*North Carolina A&T State University, Greensboro, North Carolina

16. ABSTRACT On a demonstration scale, the controlled culture fermentation process (CCF) developed by the U.S. Food Fermentation Laboratory was compared with the conventional natural fermentation process (NF) in regard to product quality and yield and volume and concentration of wastewaters. Weight of cucumbers, volume of water, and amounts of additives were recorded. pH, acidity, salinity, and temperature were closely monitored. After brining, brinestock quality was evaluated by a panel of experts from the US Food Fermentation Laboratory and the Heinz Company. The brinestock was then processed; spent brines and processing waters were collected. Volume and wastewater characteristics (salinity, BOD, N and P forms, residues) were determined for the waters and weight of brinestock was determined. The cucumbers were then packed using a conventional finishing procedure for whole dill pickles and hamburger dill chips. Yield of final product was determined. Acceptability of the finished products was evaluated by a panel.

Analysis of data indicates that the CCF produces a product of quality equal to or exceeding that of NF; that a reduction of the total dissolved solids load in the wastewaters was achieved; and that fermentation occurs more rapidly and predictably.

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Food Processing Fermentation Quality Control wastewater	Cucumber Pickling Process Modification	

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