# Effects of Sodium Chloride Concentration on Firmness Retention of Cucumbers Fermented and Stored with Calcium Chloride

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## ABSTRACT

The firmness of cucumbers brined at 0.2% $CaCl_2$ was retained during fermentation (1 month) and storage (12 months) when 2.6, 4.2 or 5.8% NaCl also was present. Firmness retention was not improved ($P \geq 0.05$) during storage by increase in NaCl concentration to 11.9% after fermentation. In the absence of added NaCl, cucumbers were firm after fermentation, but firmness was reduced during storage to 69% of initial for whole cucumbers, 64% for mesocarp tissue, and 32% for endocarp tissue. Addition of uncharacterized softening enzymes, extracted from debris collected at a cucumber grading operation, resulted in softening of cucumbers brined at 1.8% NaCl in the absence of added $CaCl_2$. Addition of $CaCl_2$ reduced but did not eliminate softening by ths extract. Results indicated that the firmness of brined cucumbers could be retained at appreciably lower NaCl concentrations than those traditionally used but that the lower limit of NaCl required to prevent softening by possible contaminating enzymes is yet to be established.

## INTRODUCTION

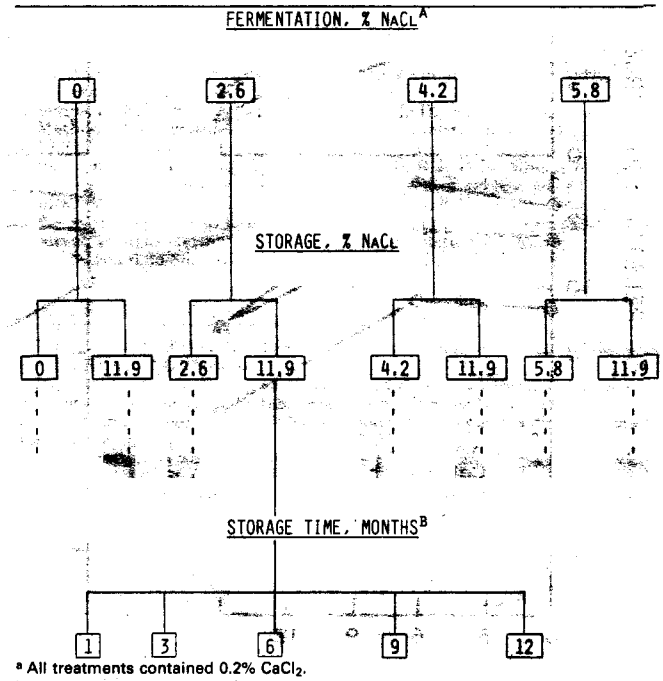
APPROXIMATELY 40% of the United States pickling cucumber crop is temporarily preserved by brine fermentation and storage. During the harvest season the cucumbers are placed in 2,000- to 20,000-gallon (7,600- to 76,000L) tanks along with a brine solution in which the cucumbers ferment. Dry salt is added to the tops of headboards that are positioned to restrain cucumbers below the brine surface. The tanks, which have open tops, are kept out of doors to permit exposure of the brine surface to sunlight. Sunlight prevents growth of film yeasts and other oxidative microorganisms on the brine surface. The salt concentration is held relatively low (5–8%) during fermentation to permit rapid growth of lactic acid bacteria which convert fermentable sugars to lactic acid and other end products. Traditionally, dry salt has been added after fermentation to gradually increase the concentration to 12–16% for storage (Etchells et al., 1951). This added salt serves as insurance against spoilage, particularly softening, during storage. When the brine stock cucumbers are removed from the tanks for further processing during the year, as needed, they must be desalted to an extent that the final product will contain the desired salt concentration, which may range from less than 1% for sweet pickles to 2 to 4% for certain types of dill pickles. Thus, the amount of salt that must be leached from the salt stock cucumbers depends mainly upon the concentration used for storage. The salt leached from the brine stock normally is discarded, which may create a disposal problem. Leaching also removes desirable flavor and nutrient components. It would be preferable, therefore, to brine and store cucumbers at a sufficiently low concentration of salt to reduce or eliminate the necessity for desalting.

A series of studies has implicated pectinolytic enzymes of filamentous fungi as the cause of softening of brined cucumbers under commercial conditions (Bell et al., 1950, 1955; Etchells et al., 1955, 1958). Cucumber flowers retained on the fruit (flower retention was more prominent with smaller fruit) were found to harbor most of the pectinolytic activity, apparently from fungi that had grown in the flowers (Etchells et al., 1958). Bell and Etchells (1961) demonstrated that increasing concentrations of NaCl prevented softening of brined cucumbers to which fungal pectinolytic enzymes had been added, thus justifying the use of higher concentrations of NaCl as insurance against softening.

Thompson et al. (1979) found that in the absence of detectable polygalacturonase activity, storage temperature had the greatest effect upon firmness retention in brined cucumbers; pH, NaCl concentration, and storage time had lesser effects. They developed a prediction equation for rate of firmness loss which incorporated these four variables. They concluded that firmness of brined cucumbers can be retained at concentrations of NaCl lower than traditionally used, provided the cucumbers are washed to remove softening enzymes and the storage temperature is 15.5°C or lower. These conclusions were reached for cucumbers that were brined without the addition of calcium compounds.

Recent studies have indicated that addition of calcium to initial cover brines can improve firmness retention in cucumbers brined at lower concentrations of salt than those traditionally used. Fleming et al. (1978) found that firmness retention in small cucumbers and in sliced large cucumbers was greatly enhanced by addition of calcium acetate. Tang and McFeeters

*Table 1—Experimental design for the fermentation and storage of size no. 2 cucumbers at various concentrations of NaCl.*




[a] All treatments contained 0.2% $CaCl_2$.
[b] Storage at 11.9% NaCl relates only to 3, 6, 9, and 12 months.

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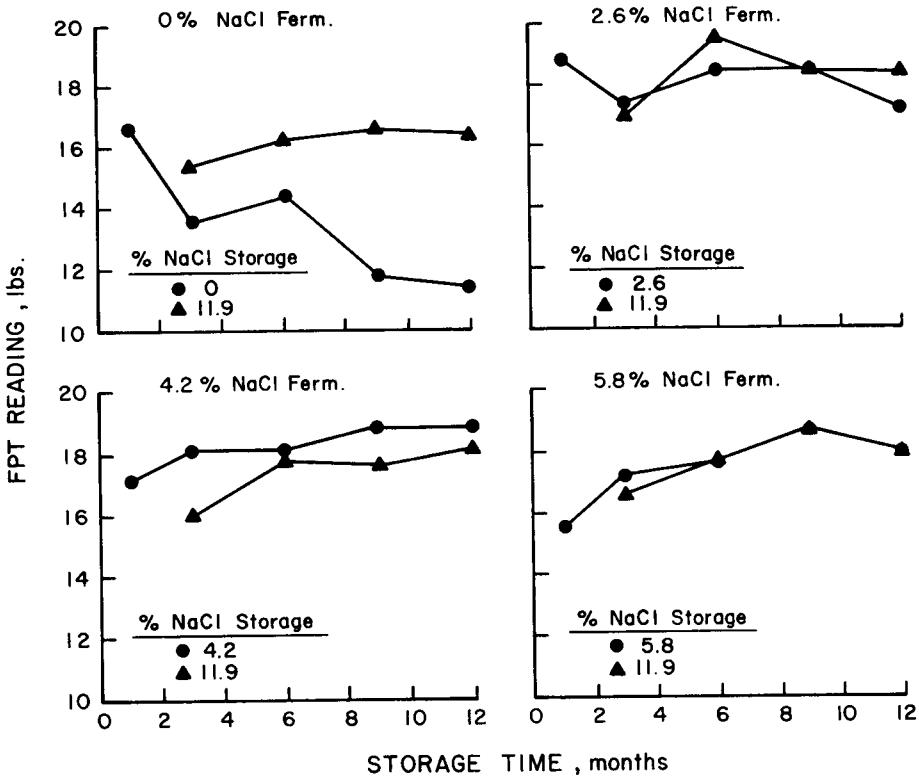


*Fig. 1—Firmness changes in size no. 2 cucumbers fermented and stored at various concentrations of Nacl. All brines contained 0.2% $CaCl_2$. Firmness was determined on whole cucumbers by a Magness-Taylor Fruit Pressure Tester.*

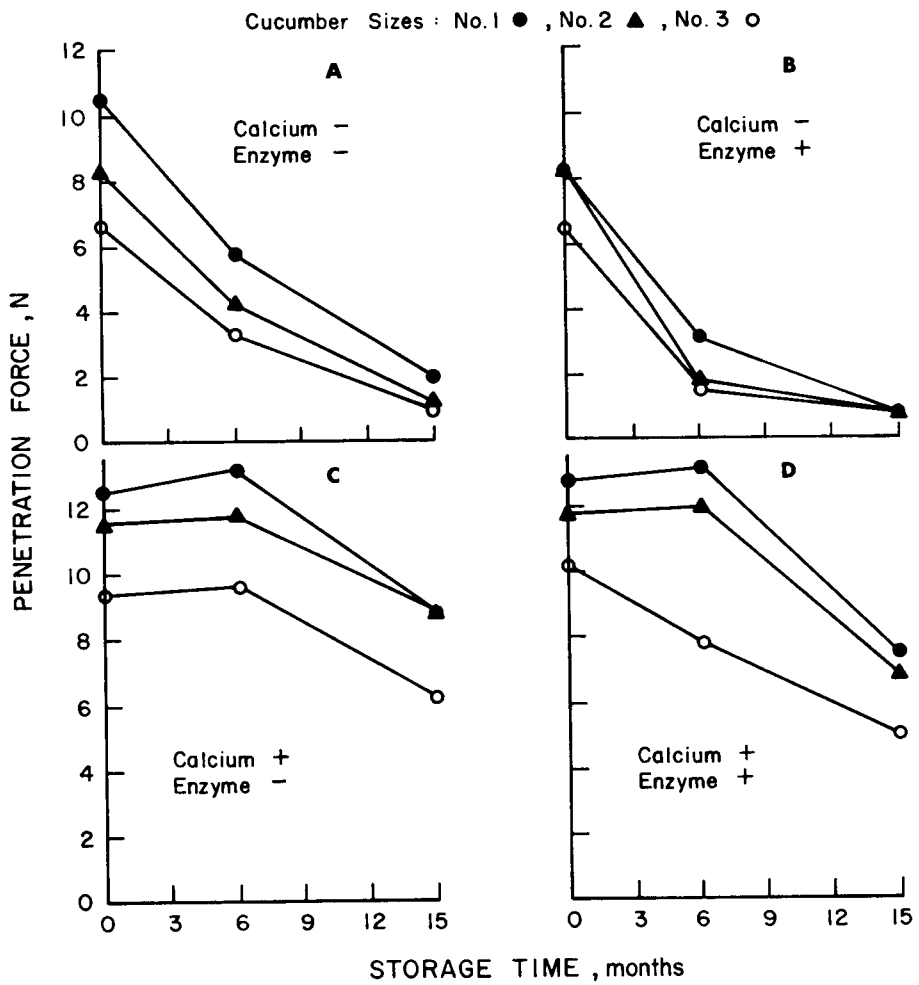


*Fig. 2—Firmness changes in three sizes of cucumbers as influenced by the addition of $CaCl_2$ and a softening enzyme preparation. All brines contained 1.8% NaCl. Firmness was determined on mesocarp tissue by the Instron UTM.*

*Table 2—Effect of NaCl concentration on firmness stability of fermented cucumbers*

| NaCl, % | | FPT, lb | | Instron UTM, N | | | |
|---|---|---|---|---|---|---|---|
| | | Whole[b] | | Mesocarp[b] | | Endocarp[b] | |
| Fermentation | Storage | Initial[a] | 12 Months | Initial[a] | 12 Months | Initial[a] | 12 Months |
| **NaCl not increased for storage** | | | | | | | |
| 0.0 | 0.0 | 16.6cd | 11.5e | 11.4ab | 7.3d | 1.9ab | 0.6c |
| 2.6 | 2.6 | 18.9c | 17.2bc | 12.1a | 9.5c | 1.9ab | 1.8ab |
| 4.2 | 4.2 | 17.2bc | 18.8a | 11.4ab | 9.4c | 2.0ab | 1.7b |
| 5.8 | 5.8 | 15.6d | 18.0abc | 12.1a | 10.6bc | 2.0ab | 2.0ab |
| **NaCl increased for storage** | | | | | | | |
| 0.0 | 11.9 | — | 16.6cd | — | 9.4c | — | 1.8ab |
| 2.6 | 11.9 | — | 18.4ab | — | 10.5bc | — | 2.1a |
| 4.2 | 11.9 | — | 18.2ab | — | 10.1bc | — | 2.0ab |
| 5.8 | 11.9 | — | 18.0abc | — | 10.6bc | — | 1.8ab |

[a] Initial firmness measurements were taken 1 month after brining (0 month storage) at which time the fermentation was complete.
[b] Means under the same category (i.e., whole, mesocarp, endocarp) followed by a common letter are not significantly different ($P \geq 0.05$) by Duncan's New Multiple Range Test.

*Table 3—Analysis of Variance for firmness of cucumber mesocarp as influenced by calcium, softening enzymes, fruit size and storage time*

| Source of variation | Degrees of freedom | Mean square | F-ratio[a] |
|---|---|---|---|
| Calcium | 1 | 10,802 | 394** |
| Enzyme | 1 | 214 | 7.8* |
| C X E | 1 | 38 | 1.4 |
| Error A | 4 | 27 | |
| Size | 2 | 705 | 57** |
| C X S | 2 | 95 | 7.6** |
| E X S | 2 | 8 | 0.7 |
| C X E X S | 2 | 23 | 1.9 |
| Time | 2 | 2,898 | 233** |
| C X T | 2 | 918 | 74** |
| E X T | 2 | 66 | 5.3 |
| T X S | 4 | 21 | 1.7 |
| C X E X T | 2 | 39 | 3.2 |
| C X S X T | 4 | 26 | 2.1 |
| E X S X T | 4 | 13 | 1.1 |
| C X E X S X T | 4 | 13 | 1.0 |
| Error B | 32 | 12 | |

[a] Significant at the 0.05 (*) and 0.01 (**) probability levels.

*Table 4—Effect of calcium chloride and softening enzyme extract on firmness of brined, whole no. 3 size cucumbers*

| | | Firmness by FPT, lb | |
|---|---|---|---|
| $CaCl_2$[a] | Enzyme extract[b] | After fermentation[c] | After 6 months' storage |
| | | **1.8% NaCl** | |
| – | – | 14.5de | 5.8f |
| – | + | 13.0e | 3.4f |
| + | – | 21.5ab | 19.8b |
| + | + | 21.4ab | 16.7cd |
| | | **5.8% NaCl** | |
| + | + | 23.4a | 18.8bc |

[a] $CaCl_2$ added to equilibrate at 0.2%.
[b] Softening enzyme extract from cucumber grading waste added at 40 units/mL of cover brine.
[c] Means followed by a common letter are not significantly different ($P \geq 0.05$) by Duncan's New Multiple Range Test.

(1983) reported similar results using $CaCl_2$. Buescher et al. (1979, 1981) reported that addition of $CaCl_2$ to the cover brine prevented softening of whole cucumbers, even when softening enzymes of fungal origin were intentionally added to the brines. Recent research has been directed toward the development of a better understanding of the role of calcium in firmness retention (Buescher and Hudson, 1986; Hudson and Buescher, 1985, 1986; McFeeters et al., 1985).

The use of controlled fermentation procedures such as that suggested by Etchells et al. (1973) and the anaerobic tanks suggested by Fleming et al. (1983), along with addition of calcium-containing compounds offer the potential for reducing the concentration of salt used for fermentation and storage of cucumbers. In fact, there has been a trend by the pickle industry during the past few years to reduce the concentration of salt used for storage of cucumbers. However, the concentration used for fermentation has not changed appreciably. Some companies may not increase the salt concentration after fermentation if the brine stock is to be used within a few weeks. Some increase the concentrations to 8 to 12% for storage, while others retain the higher salt concentrations as traditionally used. In cold climates, companies are more likely to use higher concentrations for storage to reduce problems associated with freezing.

Lower limits of salt required for firmness retention in brined cucumbers have not been established. The purpose of this study was to determine firmness retention in cucumbers brined to ferment at 0–5.8% NaCl with 0.2% $CaCl_2$, with or without increase in salt concentration to 11.9% for the storage period. The effects of uncharacterized softening enzyme(s), extracted from debris collected at a cucumber grading operation, on firmness retention in the presence and absence of added $CaCl_2$ were also determined.

## MATERIALS & METHODS

### Cucumber brining

Pickling cucumber size nos. 1, 2, and 3 (1.9–2.7 cm, 2.7–3.8 cm, and 3.8–5.1 cm diameter, respectively) were obtained from local commercial growers. Only disease-free cucumbers without obvious physical injury were used. The cucumbers were brined in duplicate, 5-gal (19-L) plastic pails at a ratio of 60/40, cucumbers/brine (w/v) by modifications of the Etchells et al. (1973) controlled fermentation procedure. Details of this procedure that were followed in the present experiments included: washing of the cucumbers, addition of 30 mL glacial acetic acid per 19L of brined cucumbers at the time of brining. addition of 95g sodium acetate trihydrate 24 hr after brining followed by immediate inoculation of the brines with *Lactobacillus plantarum* WSO at $10^6$ cells/mL of brined cucumber volume, and continuous purging of the brines with $N_2$ (25 mL/min/pail). Modifications of the original procedure included omission of the chlorination treatment, addition of $CaCl_2$ (38g per 19-L pail, when added) and addition of varying NaCl concentrations. The above additions to the pails of cucumbers were intended to achieve equilibrium concentrations within the pails of 0.16% acetic acid, 0.5% sodium acetate and 0.2% $CaCl_2$, w/v. The cucumbers were fermented at 26°C.

### Analyses

Titratable acidity (expressed as lactic acid), pH, reducing sugars, and softening enzyme activity were measured as previuosly described (Fleming et al., 1984).

Firmness of whole cucumbers was determined with a Magness-Taylor Fruit Pressure Tester (FPT) with a 5/16 in (0.8 cm) tip, as described by Jones et al. (1954), with adjective ratings as described by Bell et al. (1955). Firmness of mesocarp and endocarp tissues was determined with an Instron Universal Testing Machine (UTM) according to Thompson et al. (1982). The FPT provided an overall firmness response which included the force needed to penetrate the skin and underlying flesh. The force unit for this instrument is indicated in pounds and is expressed likewise herein for ready comprehension by those familiar with this commonly used instrument. The Instron UTM was used to assess firmness of component tissues since the FPT is not considered to be sufficiently sensitive for this purpose.

Instron UTM force readings are expressed as Newtons. Since the purpose of the present work was to compare treatment effects, no attempt was made to interrelate force units by the two methods.

### Softening enzyme extract

An extract was made of debris obtained from a commercial cucumber grading operation. The debris contained leaves, flowers, soil and other foreign materials that were shaken from the fruit during grading. The debris was extracted with 4.5% (w/v) NaCl and was considered to be a good source of softening enzymes for use in determining the influence of brining treatments on softening of the cucumbers. This extract, when analyzed by the softening enzyme assay method of Bell et al. (1955), was found to reduce the viscosity of the sodium polypectate substrate solution. The extract thus prepared was added to specified brines to provide an initial activity in the cover brine of 40 units per mL brine. No attempt was made to determine the nature of the enzymes that catalyzed viscosity loss in the pectate assay solution. Thus, we refer to the activity herein as pectate depolymerase (PD) activity, referring only to its apparent action on the pectate substrate.

### Experimental design

The study consisted of two major experiments, one of which is outlined in Table 1. In this experiment, size no. 2 cucumbers were brined and fermented in quadruplicate 19-L pails to give equilibrated NaCl concentrations of 0, 2.6, 4.2, and 5.8%. Food-grade salt was used for brine preparation. All pails contained 0.2% $CaCl_2$, equilibrated. After fermentation for 1 month at 26°C, two pails of each treatment were supplemented with dry NaCl to increase the equilibrated NaCl concentration to 11.9%. The cucumbers were then packed into 1-gal (3.8-L) jars (4 jars per pail) in the brine from the pails from which they came. The jars were stored at room temperature for various periods, as indicated in Table 1.

In a second experiment, a mixture of size nos. 1, 2, and 3 cucumbers was packed into 19-L pails to equilibrate at 1.8% NaCl. Each pail contained 40 no. 1, 35 no. 2, and about 60 no. 3 size cucumbers. The experimental design consisted of the following four treatments, with duplicate pails of each: (1) no $CaCl_2$, no PD extract; (2) no $CaCl_2$, plus PD extract; (3) $CaCl_2$, no PD extract; and (4) $CaCl_2$, plus PD extract. The brines were inoculated with *L. plantarum* WSO and the pails incubated at 28°C for 22 days, after which the cucumbers were removed and packed into 3.8-L jars containing 10 each of the 3 sizes of cucumbers along with their fermentation brine. The jars were stored at room temperature until opened for assay at 0, 6, and 15 months of storage.

For statistical evaluation, both experiments were split plot designs. The data were evaluated by Analysis of Variance.

## RESULTS

### Fermentation and storage at various salt concentrations

Size no. 2 cucumbers fermented normally in 0, 2.6, 4.2, and 5.8% NaCl, as indicated by acid production, pH, and sugar utilization, with a slightly lower fermetnation rate at higher concentrations of NaCl. All fermentations were essentially complete after 22 days, as indicated by little or no reducing sugars in the brine (0.1% or less), pH 3.5 ± 0.1 and brine acidity of 1.4% ± 0.1% (calculated as lactic acid).

Cucumbers fermented and stored at 2.6 to 5.8% NaCl (0.2% $CaCl_2$ also present) remained firm over a 12-month period, as determined by FPT of the whole fruit (Fig. 1). Addition of NaCl to 11.9% after fermentation did not improve firmness retention during storage. Cucumbers fermented at 0% NaCl, however, steadily lost firmness during the 12-month storage period. Addition of NaCl to 11.9% after fermentation prevented firmness loss in these cucumbers (Fig. 1).

A comparison of firmness retention in whole cucumbers, as determined by the FPT, and in the internal flesh (mesocarp and endocarp tissues), as determined by the Instron UTM, is summarized in Table 2. Immediately after fermentation (initial), cucumbers fermented at all salt concentrations were firm as determined by the FPT measurement of the whole fruit, including those fermented at 0% NaCl (Table 2). However, there were differences in firmness, with cucumbers fermented at 2.6% NaCl being most firm ($P \leq 0.05$). No significant differences were observed in initial firmness of mesocarp and endocarp tissues among the four salt concentrations as determiend by the Instron UTM.

It is not clear why initial firmness of whole cucumbers by FPT measurement varied as it did, or even if the effect was important. After 12 months' storage, cucumbers fermented and stored at 0% NaCl were significantly less firm than those fermented at 2.6 to 5.8% NaCl (Table 2). The firmness of cucumbers fermented and stored at 0% NaCl was rated "soft" according to the firmness scale of Bell et al. (1955) [and adjustment for size no. 2 cucumbers as suggested by Jones et al. (1954)]. When 11.9% NaCl was added after fermentation, cucumbers fermented at 0% NaCl retained the initial firmness during 12 months' storage. No important differences in firmness of whole cucumbers by FPT measurement were observed in cucumbers fermented at 2.6–5.8% NaCl, regardless if the NaCl concentration was increased to 11.9% for storage.

The firmness of mesocarp tissue was reduced during storage at all NaCl concentrations, and most of the reductions among treatment means were significant ($P \leq 0.05$). The greatest firmness reduction occurred in the mesocarp of cucumbers fermented and stored in the absence of NaCl (Table 2). The only significant reduction in firmness of endocarp tissue occurred in cucumbers fermented and stored at 0% NaCl.

Relative responses of whole cucumbers and mesocarp and endocarp tissues to the NaCl treatments varied. Retention of firmness, as % of initial, in the absence of NaCl during fermentation and storage was lower for the internal flesh (endocarp, 32%; mesocarp, 64%) than for whole cucumbers (69%). When fermented at 2.6–5.8% NaCl, however, whole cucumbers approximately maintained or increased in firmness (91–115% of initial), mesocarp itssue became slightly less firm 78–89% of initial) and endocarp tissue approximately maintained firmness (86–111% of initial) during the 12-month storage period (Table 2). The slight loss in mesocarp firmness during storage at 2.6–5.8% NaCl, although statistically significant, may not be of great practical importance. The 9.4–10.6N force readings after storage (Table 2) corresponded to about 7 on the sensory scale (1 to 10, with 10 firmest) of Thompson et al. (1982).

### Effects of $CaCl_2$ and PD activity on firmness

Three sizes of cucumbers were brined together (1.8% NaCl) in the presence and absence of added $CaCl_2$ and PD activity. Results of ANOVA for this experiment are summarized in Table 3. Addition of $CaCl_2$ and PD activity, cucumber size and storage time all significantly influenced firmness of mesocarp and endocarp tissues, as determined by the Instron UTM. Addition of $CaCl_2$ resulted in greater initial firmness and greater retention of firmness for all three sizes of cucumbers over the 15-month storage period (Fig. 2). Firmness of all sizes decreased rapidly during storage in the absence of added $CaCl_2$. and firmness loss was greater when PD activity was added (Fig. 2A and 2B). Addition of $CaCl_2$ resulted in greater firmness retention in both the presence and absence of added PD activity (Fig. 2C and 2D). However, mesocarp firmness retention of size no. 3 cucumbers was reduced ($P \leq 0.05$) after 6 months when PD activity was added, even in the presence of $CaCl_2$. Firmness loss in sizes 1 and 2 due to PD activity in the presence of $CaCl_2$ was not apparent after 6 months of storage, but firmness was reduced ($P \leq 0.05$) for size 2 after 15 months when compared to cucumbers to which PD activity was not added (Fig. 2C and 2D).

Treatment effects on the firmness of whole, size no. 3 cucumbers, as determined by the FPT, are summarized in Table 4. The firmness of these cucumbers was determined immediately after fermentation and storage for 6 months. Again, omission of $CaCl_2$ resulted in a significant loss in firmness after

fermentation and a 6-month storage period, and these cucumbers were judged to be unacceptable for commercial use. Addition of $CaCl_2$ resulted in greater firmness retention ($P \leq 0.05$). In the absence of added PD activity, the slight decrease in firmness over the 6-month storage period was not significant ($P \geq 0.05$). When cucumbers were brined at 1.8% NaCl in the presence of PD activity, however, firmness retention was significantly reduced ($P \leq 0.05$) over the 6-month period. Increase in NaCl concentration to 5.8% resulted in greater firmness in the presence of PD activity compared to the 1.8% NaCl concentration, but the difference was not statistically significant ($P \geq 0.05$).

## DISCUSSION

THESE RESULTS indicated that the firmness of cucumbers can be retained during brine storage at much lower NaCl concentrations than those traditionally used. Addition of calcium enhances the potential for fermentation and storage at relatively low concentrations of NaCl based on previous studies (Fleming et al., 1978; Buescher et al., 1979, 1981; Hudson and Buescher, 1986) and the present work. However, considerable experience may be required to establish the minimum concentration of NaCl that can be safely used. Sodium chloride is reputed to serve two major functions in the preservation of vegetables by fermentation; namely, to help retain firmness of the vegetable and to direct the course of the microbial activities (Pederson, 1979).

Addition of calcium may result in a lessened risk of loss in cucumber firmness at relatively low concentrations of NaCl but may not be a panacea for protection. Softening enzyme activity (40 units/mL) obtained from a cucumber grading operation caused slight softening of large, but not small, cucumbers, even in the presence of added calcium. Perhaps a higher equilibrated concentration of $CaCl_2$ than the 0.2% used herein would have improved firmness retention in the large cucumbers exposed to softening enzymes. Buescher et al. (1981) reported that 0.2% $CaCl_2$ greatly enhanced firmness retention in the presence of PG (338 units/mL), and that a small increase in firmness retention was achieved by higher concentrations of $CaCl_2$. The 0.2% concentration was selected because it did not create a "chalky" off flavor problem that was noted at 0.3–0.4% $CaCl_2$ (unpublished observation). It should be noted that Buescher et al. (1981) used PG obtained from a commercial source (from *Aspergillus niger*), while our source was associated with the cucumber field operation. Also, they added a much higher PG activity than we did. McFeeters et al. (1984) reported that calcium caused a moderate decrease in the rate of tissue softening by fungal enzymes, but even high calcium concentrations cannot prevent tissue softening. Protection by calcium against softening was influenced by the fungal source of the softening enzymes. Walter et al. (1985) reported stem end softening of cucumbers fermented at 2.6% NaCl in the presence of added calcium. They speculated that the softening probably was due to fungal infection of the cucumbers before brining. Thus, questions may be raised regarding the magnitude and universality of the protective effect of calcium against softening of brined cucumbers. These include variables associated with the cucumber (size, cultivar, condition) and the source of softening enzyme.

In reducing salt concentration, effects on the microbial fermentation should be considered. Recently, an aberrant, secondary fermentation of small cucumbers that were brined at 2.3% NaCl was observed (Fleming et al., 1986). The spoilage that resulted was attributed to the use of relatively low salt concentration and small cucumbers to which soil readily adhered.

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