Production of Shelf-Stable Ranch Dressing Using High-Pressure Processing


ABSTRACT: High-pressure processing (HPP) can reduce or eliminate microorganisms of concern in food without deteriorating product quality; however, quality benefits must justify the substantial capital investment for the utilization of this technology. HPP is particularly a beneficial preservation technology for products damaged by thermal treatments or when product quality could be improved by reformulation to raise pH or eliminate chemical preservatives. The primary objectives of this study were to determine the efficacy of HPP to protect premium ranch dressing (pH 4.4) from microbial spoilage and to assess changes in physical, chemical, and sensory attributes throughout the product’s shelf life. In inoculated-packages studies, the efficacy of HPP was measured against ranch dressing spoilage organisms: Pediococcus acidilactici, Lactobacillus brevis, and Torulaspora delbrueckii. HPP treatment (600 MPa, 3 min) decreased population of P. acidilactici, the most pressure-resistant spoilage organism tested, by ≥ 6.4 log CFU/g. During a shelf-life study of edible product, treating ranch dressing at 600 MPa for 5 min effectively prevented microbial spoilage throughout the storage period (26 wk at 4 and 26 °C). The pH and emulsion stability of ranch dressing were not adversely influenced by HPP. Extended storage of HPP product for 16 to 26 wk at 26 °C resulted in a decrease in consumer acceptance and significant changes in color and organic acid profile (specifically, increased pyroglutamic acid). These changes were consistent with those expected during extended storage of commercially available products. HPP may be used to produce premium ranch dressing, with defined shelf-life and storage conditions, without significantly changing product attributes.

Keywords: high-pressure processing, organic acid profile, ranch dressing, sensory analysis, spoilage

Introduction

High-pressure processing (HPP) is a novel technology, which has been demonstrated to be effective in nonthermal pasteurization and temperature-assisted sterilization of food. A number of commercial pressure-processed food products are now available in the United States and other countries. Products currently produced using HPP technology are typically processed with pressures ranging between 500 and 600 MPa. The financial commitment of converting existing processing lines to HPP remains a burden for the food industry, and a cost–benefit analysis must show an advantage to justify the conversion. A major advantage of using HPP is the potential for enhancing the quality, functionality, shelf life, and safety of foods that are usually degraded by alternate preservation techniques (for example, heat). Successful utilization of HPP technology for these products will be of great benefit to the food industry and consumers.

In the United States, supermarket sales of pourable salad dressings exceeded $1.4 billion in 2005 (Anonymous 2006). Pourable dressings, with the exception of French dressing, do not have standards of identity and thus vary in composition and physical characteristics (Smittle and Flowers 1982). The shelf life of dressing products should be at least 3 to 6 mo under refrigeration or at room temperature. Product failure modes include microbial spoilage, discoloration, rancidity, presence of off-flavors, and emulsion instability (Mistry and Min 1993). Pourable dressing products are rarely associated with foodborne illness due to their acidic nature (Smittle 1977). Two general approaches may be used to eliminate microbial spoilage of the final product: (1) changing product formulation to prevent microbial growth or (2) including a lethal processing step to inactivate the organisms of concern in the product. Studies reported more than 7 decades ago explored the feasibility of adding lactic and acetic acids to prevent spoilage of mayonnaise-based dressings (Pederson 1930). Iszard (1927) added lactic acid to mayonnaise dressing, determining that total acidities of at least 1.75% (as lactic acid) prevented bacterial spoilage. Further studies suggested that using organic acids (for example, lactic, acetic, or citric) protected these products not only through lowering the pH but also by imparting antimicrobial properties derived from the undissociated forms of the acids (Smittle 1977). Muriana and Kanach (1995) suggested using Nisaplin™ (a commercial nisin) to prevent spoilage of buttermilk ranch dressing by Lactobacillus brevis subsp. lindneri. Nisaplin at 200 ppm was effective against the inoculated bacterium and the product was stable for the 90-d shelf-life study. Other researchers (Castro and others 2002) found EDTA, ascorbic acid, and acetic acid to enhance the stability of sorbate, an antifungal agent, in model salad dressing systems. The researchers suggested that these antimicrobial additives would delay or prevent spoilage of salad dressings by Zygosaccharomyces bailii. Yang and others (2003) explored using fatty acids esters of sucrose and methyglycolose to prevent or delay salad dressing spoilage by Z. bailii and L. fructivorans. Sucrose esters of lauric, myristic, and palmitic fatty acids at 1.0% levels prevented spoilage of salad dressing by Z. bailii; however, none of the esters were effective against L. fructivorans. The food industry relies on a variety of preservatives (for example, sorbate and benzoate) and on high acidity, achieved...
Pressure-processed ranch dressing

primarily by acetic acid, to stabilize shelf-stable dressings (Kurtz-
man and others 1971; Smittle and Flowers 1982; Vargo 1989). Com-
mmercially available shelf-stable ranch dressing products, analyzed in
our laboratories, range in pH between 3.35 and 3.55 (data not
shown). These products have a shelf life that may reach 9 to 12 mo
with room temperature storage (Smittle and Cirigliano 1992).

While acidified products are shelf stable, consumers tend to pre-
fer a less tart and acidic taste in dairy-based dressings such as ranch
(Antaki and Layne 1990). Researchers have attempted substituting a
portion of acetic acid in the formulation with a less tart acidu-
lament. Vargo (1989) investigated the impact of using gluconic acid in
lieu of acetic acid in a model salad dressing with promising results:
formulations containing both acetic and gluconic acids were mi-
icrobiologically stable and were perceived as significantly less tart
by consumers.

Formulation of salad dressing with a low pH (approximately 3.5)
and antimicrobial additives is a common practice to control the
outgrowth of yeast, mold, and lactobacilli. An alternative approach
to achieving microbial stability in salad dressings which addresses
growing consumer interest in “no preservatives” (Anonymous 2008)
and provides a less tart flavor would seem to have commercial
value. Due to its physical characteristics, dressing emulsions are
sensitive to processing and handling conditions leading to product
degradation, specifically emulsion breakdown. Limited studies add-
dressed the efficacy of lethal processing steps to eliminate spoilage
properties of the product. HPP may also decrease the dependence
on additives to stabilize dressing-type products and thus results in
a product with a “cleaner” label.

The primary objective of this study was to determine the feasi-
ibility of using high-pressure processing to produce a shelf-stable
ranch dressing product with qualities associated with improved
consumer acceptance. Therefore, inoculated-pack studies were
conducted as well as long-term shelf-life studies on noninocu-
lated samples, which were then analyzed for sensory, chemical, and
physical attributes.

Materials and Methods

Preparation of experimental ranch dressing

Ranch dressing was prepared according to the following recipe:
2800 g mayonnaise (Kraft Real Mayonnaise, Kraft Foods, Inc.,
Northfield, Ill., U.S.A.), 1258 g buttermilk made with whole milk
(Kroger, The Kroger Co. Cincinnati, Ohio, U.S.A.), 170 g dried
chopped onions (Spice Classics, Por Han-Dee Pak, Inc. Cock-
eysville, Md., U.S.A.), 13 g canned minced garlic (Goya, Goya Foods,
Inc., Secaucus, N.J., U.S.A.), 3.5 g dried parsley flakes (McCormick,
McCormick and Co., Inc., Hunt Valley, Md., U.S.A.), 1.5 g iodized salt
(Morton, Morton Intl., Inc., Chicago, Ill., U.S.A.), 1.2 g dried ground
thyme (McCormick), 1.0 g xanthan gum (Bob’s Red Mill, Newburg,
Oreg., U.S.A.), 0.75 g ground black pepper (McCormick), and 0.75 g
monosodium glutamate (Kroger). All ingredients were dispensed in
a stainless steel mixing bowl and mixed by hand with a whisk to
achieve a uniform consistency. The dressing was covered with plas-
tic wrap and held at room temperature for 30 to 60 min to allow
the pH to stabilize. The pH of the prepared dressing was then mea-
sured using a pH meter (Corning model 430, Corning, Inc., Corn-
ing, N.Y., U.S.A.). After stabilization, product pH was 4 to 4.2; this
was adjusted to 4.4 using 5 M sodium hydroxide. Ranch dressing
was placed in a large stainless steel hotel pan, spread into a thin
layer, less than 6 cm deep, and placed in a vacuum-sealer for 60 s
to degas the product. Dressing prepared according to this pro-
cedure was used in the inoculated-pack and shelf-life studies. The
water activity of the final product was 0.975, measured at the aver-
age HPP-holding temperature for inoculated pack studies (34 °C),
which is sufficiently high (< 0.90) to ensure effectiveness of HPP
on microbial inactivation (Oxen and Knorr 1993; Franceschini and
others 2005).

Inoculated-pack study

Inocula preparation. In a previous study, microorganisms as-
associated with spoilage of ranch dressing were isolated and iden-
tified (Waite and others 2008); these are Pediococcus acidilactici
OSY-JW1, L. brevis OSY-JW1, and Torulaspora delbrueckii OSY-JW1.
Frozen stocks of these microorganisms were prepared by mixing
their cultures in de Man, Rogosa, and Sharpe (MRS) broth with gly-
cool (final concentration of 40%, v/v) and storing the mixture at
−80 °C. Immediately before the experiment, bacterial strains were
transferred from the frozen stock to MRS agar and incubated at
30 °C for 48 h. T. delbrueckii was transferred to ranch dressing agar
(RDA), a newly developed medium for recovery of spoilage yeast
(Waite and others 2008), and plates were incubated at 30 °C for
96 h. Isolated colonies of each strain were transferred to the MRS
broth and incubated overnight at 30 °C. Overnight cultures were
centrifuged at 9000 × g for 10 min (Int'l Equipment Co., IEC Cen-
tra MP4R, Needham Heights, Md., U.S.A.) and resulting pellets were
suspended in phosphate buffered saline (PBS, pH 7.4). The PBS cell
susensions were used in the inoculated pack study.

Pressure treatment. Pressure treatments were performed using
a 2-L capacity hydrostatic food processor (Quintus QFP6,
Flow Pressure Systems, Kent, Wash., U.S.A.) containing 1:1 (v/v)
glycol/water pressure transmitting fluid (Houghton-Safe 620 TY,
Houghton Intl. Inc., Valley Forge, Pa., U.S.A.). Initial temperature of
glycol/water processing fluid was 5 to 10 °C and holding tempera-
ture, measured in the processing fluid at the target pressure, ranged
from 30 to 40 °C. Initial temperature of the dressing was 4 to 6 °C.
Typical come-up times were less than 3 min and decompression
times were less than 10 s.

Comparing pressure resistance of organisms associated with
ranch spoilage. Ranch dressing was prepared as described previ-
ously, then P. acidilactici, L. brevis, and T. delbrueckii were inocu-
lated individually in the product. Inoculation levels were 105 CFU
per gram and 107 CFU per gram for each bacterial
strain. Bags of inoculated dressing (50 g each) were pressure treated
at 200, 400, or 600 MPa with a holding time of 3 min, then survivors
were determined as described subsequently. Controls were bags of
uninoculated dressing and inoculated but not pressure-processed
dressing. Sample bags were stored at 26 °C for 2 wk, observed visu-
ally for gas production every 2 d, and microbiologically analyzed on
days 7 and 14 of storage. For microbial analyses, dressing samples
were initially diluted in a 1:1 (w/v) ratio of ranch dressing and 0.1%
peptone water to obtain a slurry that could be easily pipetted while
maintaining a reasonable detection limit. This initial dilution was
followed by decimal dilutions in 0.1% peptone water, and spread-
plating on appropriate media (MRS agar or RDA). Recovery of inocu-
lated strains was verified by examining growth on agar plates for
colony and cell morphology and the Gram-reaction.

Pressure holding time and inactivation of a processing-
resistant strain. Aliquots of ranch dressing (500 g) were
transferred to sterile polyethylene bags (Fisher Scientific, Pittsburgh, Pa., U.S.A.). Bags were heat-sealed and refrigerated overnight. Packaged dressing was pressure-processed at 600 MPa for 5 min to reduce the natural microbiota in the product and minimize interference with inocula. Treated dressing was aseptically transferred to suitable containers and inoculated with PBS cell-suspension of *P. acidilactici* to achieve approximately 10⁶ CFU/g dressing. Aliquots of inoculated dressing (50 g) were transferred to sterile polyethylene bags (Thompson Equipment and Supply Co., Cincinnati, Ohio, U.S.A.), and bags were vacuum-sealed, refrigerated, and tested as indicated later. Bags of inoculated dressing were pressure-processed at 600 MPa with holding times of 3, 5, and 10 min, then stored at 26 °C for 2 wk. Bags were observed visually for gas production at 2-d intervals and microbial analyses were performed immediately after inoculation and periodically up to 25 d. Microbial analysis was performed as described previously.

**Shell-life study**

**Processing ranch dressing.** Prepared deaerated ranch dressing was transferred to sterile polyethylene bags (Thompson Equipment and Supply Co.) to support microbiological tests, and 8-oz or 20-oz polyethylene terephthalate (PET) bottles for sensory and physical tests. The bottles were drained of their original contents of drinking water prior to use (Dasani®). The Coca-Cola Co., Atlanta, Ga., U.S.A.) Polyethylene bags containing approximately 100 g of dressing were vacuum-sealed and refrigerated. Plastic bottles were filled with dressing to achieve minimal headspace, then capped and the seal wrapped in Parafilm® (Pechiney Plastic Packaging, Chicago, Ill., U.S.A.). The bottles of dressing were then placed in ethylene vinyl alcohol (EVOH) barrier pouches (FoodSaver®, Jarden Corp., Rye, N.Y., U.S.A.) and vacuum sealed to minimize the risk of leakage or ingress to the bottles and to provide a suitable oxygen barrier for subsequent shelf-life studies. Bottles were immediately refrigerated. Bags and bottles were transported overnight in Styrofoam-insulated cardboard boxes with ice packs, from the point of manufacture (The Ohio State Univ, Columbus, Ohio, U.S.A.) to the pressure processing facility (Kraft Foods Global, Inc., Glenview, Ill., U.S.A.).

HPP was conducted in a pilot-scale system with a nominal volume of 6 L (ISO-Lab S-IL-110-625-08-W, Stansted Fluid Power Ltd., Stansted, Essex, U.K.), using a pressure transmitting fluid composed of 1:2 (v/v) food-grade propylene glycol and water. All samples were tempered in an ice water bath to approximately 6 °C immediately prior to pressure processing. The initial temperature of the pressure transmitting fluid was approximately 8 °C, and the jacket of the vessel was maintained at 25 °C. Samples were pressure-processed at 600 MPa for 5 min. Several samples were fit with dressing to achieve minimal headspace, then capped and the seal wrapped in Parafilm® (Pechiney Plastic Packaging, Chicago, Ill., U.S.A.). The bottles of dressing were then placed in ethylene vinyl alcohol (EVOH) barrier pouches (FoodSaver®, Jarden Corp., Rye, N.Y., U.S.A.) and vacuum sealed to minimize the risk of leakage or ingress to the bottles and to provide a suitable oxygen barrier for subsequent shelf-life studies. Bottles were immediately refrigerated. Bags and bottles were transported overnight in Styrofoam-insulated cardboard boxes with ice packs, from the point of manufacture (The Ohio State Univ, Columbus, Ohio, U.S.A.) to the pressure processing facility (Kraft Foods Global, Inc., Glenview, Ill., U.S.A.).

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**Table 1 — Sampling schedule for ranch dressing shelf-life study.**

<table>
<thead>
<tr>
<th>Facility</th>
<th>Product analysis</th>
<th>Time of analysis (wk)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ohio State Univ. (Columbus, Ohio)</td>
<td>Microbial (spoilage)</td>
<td>0, 2, 4, 6, 8, 16</td>
</tr>
<tr>
<td>Silliker Laboratories (Columbus, Ohio)</td>
<td>Microbial (pathogens)</td>
<td>2</td>
</tr>
<tr>
<td>U.S. Army Natick Soldier RDEC® (Natick, Mass.)</td>
<td>Chemical (pH, HPLC organic acids)</td>
<td>0, 4, 8, 16, 26</td>
</tr>
<tr>
<td>Kraft Foods Global, Inc. (Glenview, Ill.)</td>
<td>PBS cell suspensions and PBS</td>
<td>0, 4, 8, 16, 26</td>
</tr>
<tr>
<td></td>
<td>Sensory (quality attribute scales)</td>
<td>0, 4, 8, 16, 26</td>
</tr>
<tr>
<td></td>
<td>Physical (viscosity, emulsion stability)</td>
<td>0, 4, 8, 16, 26</td>
</tr>
</tbody>
</table>

*Time zero samples were analyzed before and after processing. Samples for weeks 2 to 8 were stored at 4, 26, and 37 (± 1 °C). Samples for weeks 8 to 16 were stored at 4 and 26 (± 1 °C).
Pressure-processed ranch dressing... analysis also provides a <2.5% and <97.5% values, indicating oil droplets had a diameter equal to or smaller than these values.

**pH and color analysis.** The pH of salad dressing was measured (Oakton Model 510 pH meter, Oakton Instruments, Vernon Hills, Ill., U.S.A.). Product color (Hunter Lab MiniScan Model MS-S-4500, Hunter Associates Laboratories, Reston, Va., U.S.A.) was also measured; values of L, a, and b were calculated by the instrument's data system (Universal Software V 4.10, Hunter Associates Laboratories).

**HPLC analysis.** High-performance liquid chromatography (HPLC) was used to analyze changes in composition and concentration of organic acids in ranch dressing during storage. Ranch dressing was sampled before and after pressure processing, and after storage at 4, 26, and 37 °C. Samples stored at 37 °C were analyzed at 4 and 8 wk; those stored at 26 and 4 °C were analyzed after 4, 8, 16, and 26 wk. Samples at each time point were prepared in duplicate for HPLC. Samples were homogenized with a Polytron probe homogenizer (PT 10-35, Kineta mica AG, Lucerne, Switzerland) in 0.005 M sulfuric acid (2 g/20 mL) and centrifuged at 12000 × g for 30 min at 4 °C. The supernatant was filtered through a 0.4 μm syringe filter. Samples were analyzed using an HPLC (Waters Associates, Milford, Mass., U.S.A.), equipped with a 50 μL sampling loop and organic ions-exclusion, 300 × 7.8 mm, column (Aminex HPX-87H, Bio-Rad Laboratories, Hercules, Calif., U.S.A.) using 0.005 M sulfuric acid as the mobile phase (flow rate: 0.5 mL/min). The HPLC was equipped with a variable wavelength detector (Waters Associates) set at wavelength of 215 nm. Chromatogram data were collected and analyzed using a commercial software package (SRI, Inc., Torrance, Calif., U.S.A.). Results are reported as mg/100 g (wet weight). Calibration was performed using external standards of pure organic acid salts (Sigma Chemicals, St. Louis, Mo., U.S.A.) by integration of peak areas at wavelength of 215 nm. Pyruvic acid could not be consistently separated from a coeluting unknown peak. Therefore, pyruvic acid concentrations were estimated by adding known quantities of pyruvic acid to ranch dressing samples, and estimating the initial values of pyruvic acid by subtraction.

**Sensory analyses.** Samples of packages stored at 4, 26, and 37 °C for 2 wk were analyzed by a commercial analytical service (Silliker Laboratories, Columbus, Ohio, U.S.A.) for the presence of foodborne pathogens. Freedom of pathogens is required to verify product safety prior to sensory analyses. All samples tested negative for Listeria monocytogenes, Escherichia coli O157:H7, and Salmonella sp.

Sensory analyses were conducted by 12 to 15 member panelists from the U.S. Army Natick Soldier RDEC previously trained in descriptive sensory analysis. Samples were presented randomly to panelists in individual booths, each equipped with a computer for collecting sensory data. The panelists used a 1 to 9 point anchored sliding line scale (Figure 1A) to rate quality attributes of appearance, odor, flavor, texture, and overall quality. The computer system employed allowed panelists to include comments to explain ratings. A pressure-processed ranch dressing sample held at 4 °C was used as a control for each withdrawal period. A sliding scale (0 to 10) (Figure 1B) was used to indicate the overall degree of difference (DOD) of a treated sample from the control dressing. Sensory management system (SIMS 2000, Sensory Computer Systems, Morristown, N.J., U.S.A.) and SAS (SAS Inst. Inc., Cary, N.C., U.S.A.) were used to calculate averages of sensory values and determine standard statistical parameters from the averages and standard errors.

**Results and Discussion**

Comparing pressure resistance of spoilage microorganism in ranch dressing

The population of lactic acid bacteria in ranch dressing was estimated using MRS agar, a medium that favors lactobacilli but may allow the growth of other microorganisms (Difco & BBL Manual 2008). The MRS agar count in freshly prepared dressing was 2.0 × 10^6 CFU/g (Figure 2). Treatment with 200 MPa for 3 min significantly (P < 0.05) decreased MRS agar count in dressing to 1.6 × 10^6 CFU/g. Higher-pressure treatments (400 and 600 MPa) decreased MRS agar count to below detection limit of the enumeration method (<1.2 × 10^1 CFU/g). When ranch dressing was inoculated with P. acidilactici, L. brevis, or T. delbrueckii, and pressure-processed at 200 MPa, the microbial count remained virtually unchanged. Treating the P. acidilactici-inoculated dressing with 400 MPa for 3 min decreased MRS-agar count by 1.8 log, but treatment with 600 MPa decreased the count to below the method’s detection limit (≥ 6.4 log reduction). When ranch dressing was contaminated with L. brevis or T. delbrueckii and pressure-processed at 400 and 600 MPa for 3 min, populations of these spoilage microorganisms were below method’s detection limit.

HPP-treated ranch dressing samples with undetectable levels of contaminants were stored at 26 °C for up to 3 wk to determine if low levels of survivors of these spoilage organisms could recover over time (data not shown). No microorganisms were recovered on MRS agar from uncontaminated and pressure-processed (400 and 600 MPa, 3 min) ranch dressing during 2 wk of storage at 26 °C. Similarly, no yeast colonies were recovered during the same storage period from T. delbrueckii-inoculated dressing that was treated with 400 or 600 MPa for 3 min; however, when ranch dressing was contaminated with L. brevis and treated with 400 MPa for 3 min, survivors were detectable on MRS agar and this population reached 1.6 × 10^6 CFU/g after 1 wk of storage. No colonies were detectable from this contaminated dressing after treatment with 600 MPa for.
Pressure-processed ranch dressing... 

3 min and storage at 26°C for 2 wk. When *P. acidilactici*-inoculated, 600 MPa-treated dressing was held at 26°C, the product contained $7.9 \times 10^3$ and $1.3 \times 10^5$ CFU/g after 1 and 3 wk of storage, respectively. According to these data, *P. acidilactici* was the most pressure-resistant spoilage microorganism tested in this study. Of the remaining strains tested, *L. brevis* was more pressure-resistant than *T. delbrueckii*. HPP of ranch dressing should be designed to eliminate *P. acidilactici*, due to its relative pressure-resistance among the isolated spoilage microorganisms. Nienaber and others (2001) previously reported the efficacy of HPP to inactivate *Z. bailii* and *L. fructivorans* in ranch dressing; however, recovery and growth of spoilage organisms following HPP treatment has not been previously determined.

### Inactivation of *Pediococcus acidilactici* in ranch dressing with various HPP holding times

Ranch dressing with low microbial load was inoculated to contain approximately $10^6$ CFU *P. acidilactici*/g dressing as described previously. Following HPP treatment at 600 MPa, $5.0 \times 10^1$ to $1.6 \times 10^2$ CFU *P. acidilactici* per gram dressing were recovered from the product (Figure 3). Increasing holding time from 3 to 10 min at this pressure did not lead to noticeable increase in lethality. The populations of *P. acidilactici* remained at this level for the first 3 d of storage. During 15 d of storage of pressure-processed product at 26°C, *P. acidilactici* population increased to $>10^7$ CFU/g. An increase in holding time, from 3 to 10 min (600 MPa), had no considerable effect on recovery of *P. acidilactici* in ranch dressing stored at 26°C.

### Shelf-life study

Based on the inoculated-pack results, a shelf-life study was designed to determine if pressure processing of ranch dressing (pH 4.4) at 600 MPa for 5 min could produce a microbiologically stable product. Additionally, physical, chemical, and sensory analyses were completed to determine if pressure processing significantly changed product characteristics.

### Microbial stability of pressure-processed ranch dressing

Ranch dressing, with and without HPP, was analyzed for microbial load immediately after processing and after storage at 4, 26, 3 min and storage at 26°C for 2 wk. When *P. acidilactici*-inoculated, 600 MPa-treated dressing was held at 26°C, the product contained $7.9 \times 10^3$ and $1.3 \times 10^5$ CFU/g after 1 and 3 wk of storage, respectively. According to these data, *P. acidilactici* was the most pressure-resistant spoilage microorganism tested in this study. Of the remaining strains tested, *L. brevis* was more pressure-resistant than *T. delbrueckii*. HPP of ranch dressing should be designed to eliminate *P. acidilactici*, due to its relative pressure-resistance among the isolated spoilage microorganisms. Nienaber and others (2001) previously reported the efficacy of HPP to inactivate *Z. bailii* and *L. fructivorans* in ranch dressing; however, recovery and growth of spoilage organisms following HPP treatment has not been previously determined.

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and 37 °C for up to 16 wk. Recovery of microorganisms (mainly lactic acid bacteria) from dressing stored at 4 and 26 °C is shown in Figure 4A and 4B, respectively. Freshly prepared ranch dressing that was plated on MRS agar contained $2.5 \times 10^4$ CFU/g. HPP treatment at 600 MPa with a holding time of 5 min decreased this population to $2.5 \times 10^2$ CFU/g. Pressure-processed samples, stored at both 4 and 26 °C, maintained a minimal population at approximately $1 \times 10^2$ CFU/g. Isolates from this small population were not likely to be spoilage organisms as these did not grow in the product throughout storage, and no product defects were observed. Morphological examination of isolates suggested that these organisms are likely sporeforming bacteria and members of the natural microbiota of spices used in product formulation. Isolation of *Bacillus* sp. from dressing products has been previously reported (Pederson 1930; Kurtzman and others 1971). No further characterization of these organisms was performed in the current study.

Nonpressure-processed ranch dressing stored at 26 °C supported the growth of contaminants, most likely lactic acid bacteria (Figure 4). During 4 wk storage, the population recovered on MRS agar increased from $2.5 \times 10^4$ to $2.5 \times 10^7$ CFU/g. Untreated ranch dressing held at 4 °C contained a small microbial population throughout the storage period. The population of natural microbiota within the dressing decreased with storage at 4 °C and the product was free from observable defects throughout the storage period. Similarly, few organisms were recovered from ranch dressing stored at 37 °C, with or without HPP treatment (data not shown). There was no microbial growth in the product with extended storage at 37 °C. These results are consistent with a previous study conducted in this laboratory to identify spoilage organisms in ranch dressing (Waite and others 2008). Spoilage could not be induced during incubation at 4 or 37 °C, but was evident upon storage at 26 °C. Additional microbial analyses were performed by plating
dressing samples on PCA and RDA. Counts on PCA were similar to those on MRS agar (data not shown). No microorganisms were recovered on RDA throughout the course of the shelf-life experiment. These results were consistent with the finding that yeasts were not detectable in this freshly prepared dressing. Freshly prepared ranch dressing contains lactic acid bacteria derived mainly from buttermilk. These microorganisms differ in ability to grow and induce spoilage during storage of dressing. According to a recent study (Waite and others 2008), Leuconostoc mesenteroides was isolated initially from ranch dressing but population of the bacterium decreased progressively regardless of storage temperature. These investigators noticed that other microorganisms, including L. brevis, may grow in ranch dressing and induce spoilage with extended storage at 26 °C, but not at 4 °C. Additionally, ranch dressing samples with counts near the detection limit contained microbial populations that were predominantly sporeforming bacteria. It is noteworthy that pressure-processed ranch dressing in the current study was free of L. monocytogenes, Salmonella, and E. coli O157:H7.

Preservation of ranch dressing is commonly achieved by acidification, due to instability of this emulsion upon heat treatment. Investigators have attempted to use novel nonthermal technologies to produce shelf-stable dressing. According to Li and others (2005), pulsed electric field (PEF) treatment (31.8 kV/cm, 45 μs) combined with mild heat (70.4 °C) eliminated L. plantarum ATCC 8014 that was inoculated in a ranch dressing-like product (water, sucrose, whey protein powder, citric acid powder, salt, and modified corn starch). The treated product was microbiologically stable for at least 11 mo at room temperature. Changes in the quality of the product after PEF treatment or with extended storage were not included in the report. While PEF treatment may inactivate potential spoilage organisms, product formulation must be modified to fit the parameters of the equipment with regard to conductivity as well as particle sizes of ingredients (Barbosa-Canovas and others 1999).

Viscosity and emulsion stability. Changes in viscosity of ranch dressing in response to pressure processing and storage are shown in Table 2. Although each sample was only analyzed once, 11 measurements of shear stress/viscosity were made on each sample over a shear rate range of 1 to 100 per second and the results showed a good fit to a power law model with \( R^2 \) values > 0.950. Pressure processing decreased the viscosity of ranch dressing, at a shear rate of 1 per second, from 27.643 to 22.245, but the viscosities of control and pressure-treated dressings were similar at higher shear rates. While the difference in viscosity between the control and pressure-treated dressings is likely significant, it is not large enough to be considered relevant to consumer preference and therefore would not be deemed a flaw in the product. Houska and others (1998) reported that the relative deviation between human perception of the viscosity of non-Newtonian fluids and instrumental measurements is about 18%. Conversely, Nienaber and others (2001) reported that HPP treatments (500 to 800 MPa, 10 min) induced a significant increase in the viscosity of ranch, French, and slaw dressing products. Changes in the rheological properties of emulsions due to HPP are highly dependent on the formulation of a specific product (Dumay and others 1996).

Storage of the product for 4 wk slightly increased product thickness, regardless of storage temperature (Table 2). With extended storage, the viscosity of pressure-processed ranch dressing approached the value for time-zero samples. After 26 wk at 26 °C, there was some evidence of viscosity loss, particularly at low shear rates, but there was no evidence that the emulsion had severely broken down, causing oil separation or becoming unacceptably thin. The loss of viscosity during storage is typical of most emulsions, regardless of processing treatments (Zablocki and others 2000). Other researchers have reported decrease in dressing viscosity with extended storage at 13, 22, and 38 °C (Fetzek 1973). No gel formation was induced in the product as a result of HPP treatment and no gelation occurred throughout the storage period.

Emulsion instability can be manifested in 2 ways: (1) flocculation or creaming (that is, rearrangement of oil droplets resulting in separation of fat or proteins from bulk phase) and (2) coalescence/coagulation leading to formation of larger fat droplets (Fetzek 1973; Mistry and Min 1993). The oil droplet size of ranch dressing was measured before and after pressure processing and with extended storage at various temperatures to determine emulsion stability (Table 3). High-pressuring processing induced limited droplet coalescence as reflected in a very small increase in mean oil droplet diameter \( (d_{32}) \) and oil size distributions. Photomicroscopy of samples before and after HPP treatment showed no obvious differences in droplet size distribution (data not shown).
Pressure-processed ranch dressing...

Similarly, Nienaber and others (2001) reported that HPP treatment did not cause a significant change in particle size distribution in ranch, French, or slaw dressings. Ranch dressing stored at 4 °C appeared to have a relatively stable emulsion with no noticeable changes in oil droplet size throughout the 26-wk storage period (Table 3). Samples stored at 26 °C appeared slightly less stable than those at 4 °C, with evidence of minor coalescence and a skew toward larger droplet size after 16 wk storage. The pressure-processed dressing stored at 37 °C showed evidence of instability with significant coalescence of oil droplets over the first 2 mo of storage; however, in no case was there evidence of complete phase separation (that is, formation of oil layer on dressing surface). Similar changes in oil droplet size are expected for conventionally processed dressings stored under these conditions. Salad dressings, like most emulsions, are thermodynamically unstable and coalescence of oil droplets over time is to be expected due to Oswald ripening. This instability is independent of how the emulsion was prepared, the emulsifiers used, and how the emulsion was processed and stored after preparation, although all of these factors can alter the ultimate shelf life of the dressing. Oil separation will result in a loss of viscosity as described previously, as will breakdown or hydrolysis of starches/gums used as thickeners or emulsifiers (Claesson and others 2003; Ford and others 2003).

There are reports of HPP inducing changes in droplet size of various emulsions. Dumay and others (1996) investigated changes in droplet size in dairy cream and oil-water emulsions due to HPP. Significant changes in droplet size for cream treated with HPP (450 MPa, 15 to 30 min) were more common with HPP holding temperature at 40 °C compared with 25 °C. These findings confirm the need to apply HPP without increasing the heat during processing to maximize emulsion stability in these products.

**pH and color.** The pH of pressure-processed ranch dressing was stable regardless of storage time and temperature (Table 4). Packages stored at 37 °C for 8 wk (an extreme storage condition) showed a significantly lower L value, and there was a downward trend in L values for the product held at 26 °C during the 26 wk of storage. According to results of sensory analysis, as reported in a subsequent section, panelists noticed that stored products became darker with increased storage time at 26 and 37 °C. These observations are consistent with instrumental color analysis.

Color change of salad dressings may result from the oxidation of carotenoids present in the egg yolk used to manufacture the mayonnaise (Weiss 1983). Fetzek (1973) investigated the effects of storage temperatures and time on color of commercially produced mayonnaise, salad dressing, French dressing, and nondefined dressing. The researcher stored the products at 13, 22, and 38 °C with no light exposure. Changes in color were notable in salad dressing and nondefined dressing after storage for 6 mo at room temperature and continued to deteriorate with longer storage.

**Organic acid profile changes.** Concentrations of organic acids present in ranch dressing throughout the storage period are reported in Table 5. Samples held at 37 °C showed considerable deterioration after 8 wk of storage, and were not analyzed for chemical or sensory characteristics after that time. Samples held at 26 and 4 °C showed very little change in organic acid profiles throughout the 26-wk storage period. The most abundant organic acids in ranch dressing were citric, pyruvic, malic, lactic, and acetic. Pyruvic and acetic acid levels increased slightly in product stored at 37 °C, whereas pyruvic acid level increased and then decreased slightly in the product stored at 26 °C. Citric, malic, and lactic acids all showed similar patterns; modest changes in samples held at 26 °C, but a greater increase in concentration in samples held at 37 °C.

Pyroglutamic acid (2-pyrrolidone-5-carboxylic acid) was found in low concentrations in fresh and pressure-processed ranch dressing. The concentration of this organic acid was significantly impacted by storage temperature and time (Figure 5). Increase in pyroglutamic acid concentration was rapid in samples stored at 37 °C with the concentration rising sharply over 8 wk from an initial level of 1.4 mg/100 mg to 10.4 mg/100 mg. With storage at 26 °C, the concentration of pyroglutamic acid rose more gradually over 26 wk to 8.5 mg/100 mg. Pyroglutamic acid concentration increased only slightly (to 2.2 mg/100 mg) with storage at 4 °C for 26 wk. Pyroglutamic acid may be formed by thermal degradation of the free glutamine in a variety of food products (Mahdi and others 1961; Mucchetti and others 2000). Schneider and others (2003) found glutamine could be converted to pyroglutamic acid when subjected to HPP with moderate holding temperature (600 MPa, 50 °C). Pyroglutamic acid is also a product of enzymatic activity (that is, pyrroli done carboxyl peptidase or L-pyroglutamyl-peptide hydrolase) on free glutamine or N-terminal glutamine or glutamic acid present in proteins (Mucchetti and others 2000). In some cases, pyroglutamic acid is associated with off-flavors, but is also associated with the characteristic flavor of some aged cheeses (Abraham and Podell 1981; Mucchetti and others 2000). In the current study, thermal degradation is unlikely considering the low processing temperature (<35 °C) that the dressing experienced. Similarly, the pyroglutamic acid concentration remained low (1.4 mg/100 g sample) following pressure processing and storage at 4 °C, thus pressure-induced formation seems unlikely. Complete destruction of enzyme activity by the HPP conditions utilized in this study seems unlikely (Hendrickx and others 1998). Therefore, enzymes present in ranch dressing ingredients may remain active throughout the storage period leading to increased concentrations of pyroglutamic acid stored at 26 and 37 °C.

**Sensory perception of HPP ranch dressing.** Pressure-processed ranch dressing was analyzed by trained panelists to detect the degree of difference between a reference (4 °C sample) and samples held at a variety of temperatures for up to 26 wk. Panelists also rated the quality attributes of “appearance,” “odor,” “flavor,” “texture,” and “overall.” Overall quality for all samples is shown in Figure 6.

The results indicate that significant sensory changes occur in the dressing with extended storage. For example, identical samples were stored at 4 °C (control reference) and 26 °C (simulated shelf-stable storage) for 26 wk and rated for quality. The 26 °C samples

<table>
<thead>
<tr>
<th>Treatment and storage</th>
<th>pH</th>
<th>L</th>
<th>a</th>
<th>b</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh dressing (HPP)</td>
<td>4.34</td>
<td>87.76</td>
<td>-1.96</td>
<td>11.20</td>
</tr>
<tr>
<td>4 °C storage</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4 wk</td>
<td>4.39</td>
<td>84.11</td>
<td>0.05</td>
<td>10.03</td>
</tr>
<tr>
<td>8 wk</td>
<td>4.42</td>
<td>85.69</td>
<td>0.34</td>
<td>10.30</td>
</tr>
<tr>
<td>16 wk</td>
<td>4.34</td>
<td>83.63</td>
<td>0.27</td>
<td>10.14</td>
</tr>
<tr>
<td>26 wk</td>
<td>4.12</td>
<td>85.18</td>
<td>0.21</td>
<td>10.36</td>
</tr>
<tr>
<td>26 °C storage</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4 wk</td>
<td>4.38</td>
<td>82.51</td>
<td>0.28</td>
<td>10.51</td>
</tr>
<tr>
<td>8 wk</td>
<td>4.41</td>
<td>82.76</td>
<td>0.60</td>
<td>11.74</td>
</tr>
<tr>
<td>16 wk</td>
<td>4.31</td>
<td>81.49</td>
<td>0.50</td>
<td>12.55</td>
</tr>
<tr>
<td>26 wk</td>
<td>4.05</td>
<td>78.73</td>
<td>0.82</td>
<td>12.69</td>
</tr>
<tr>
<td>37 °C storage</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4 wk</td>
<td>4.42</td>
<td>81.43</td>
<td>0.29</td>
<td>11.87</td>
</tr>
<tr>
<td>8 wk</td>
<td>4.31</td>
<td>71.28</td>
<td>2.72</td>
<td>15.34</td>
</tr>
</tbody>
</table>
Pressure-processed ranch dressing . . .

Table 5—Organic acid concentration in ranch dressing before and after high-pressure processing (HPP; 600 MPa, 5 min) and during product extended storage at 4, 26, and 37 °C.

<table>
<thead>
<tr>
<th>Treatment and storage</th>
<th>Lactic</th>
<th>Acetic</th>
<th>Citric</th>
<th>Malic</th>
<th>Pyruvic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh dressing (no HPP)</td>
<td>229.7 ± 18.8</td>
<td>210.2 ± 18.8</td>
<td>120.0 ± 9.7</td>
<td>45.1 ± 2.0</td>
<td>8.7 ± 1.4</td>
</tr>
<tr>
<td>Fresh dressing (HPP) 4 °C storage 4 wk</td>
<td>246.4 ± 13.4</td>
<td>225.9 ± 13.6</td>
<td>122.5 ± 10.0</td>
<td>42.4 ± 2.1</td>
<td>7.8 ± 0.7</td>
</tr>
<tr>
<td>8 wk</td>
<td>257.2 ± 24.4</td>
<td>232.8 ± 8.3</td>
<td>109.3 ± 9.8</td>
<td>49.7 ± 10.7</td>
<td>8.1 ± 0.9</td>
</tr>
<tr>
<td>16 wk</td>
<td>246.0 ± 2.6</td>
<td>188.4 ± 14.6</td>
<td>95.0 ± 14.7</td>
<td>35.8 ± 0.9</td>
<td>8.9 ± 3.5</td>
</tr>
<tr>
<td>26 wk</td>
<td>240.3 ± 4.2</td>
<td>193.7 ± 1.6</td>
<td>104.0 ± 0.4</td>
<td>39.2 ± 0.7</td>
<td>6.5 ± 0.7</td>
</tr>
<tr>
<td>Fresh dressing (HPP) 26 °C storage 4 wk</td>
<td>249.9 ± 0.9</td>
<td>220.5 ± 15.4</td>
<td>122.8 ± 0.2</td>
<td>54.8 ± 2.1</td>
<td>8.2 ± 0.3</td>
</tr>
<tr>
<td>8 wk</td>
<td>267.2 ± 11.6</td>
<td>229.4 ± 13.8</td>
<td>127.3 ± 9.8</td>
<td>59.2 ± 0.6</td>
<td>8.8 ± 0.4</td>
</tr>
<tr>
<td>16 wk</td>
<td>236.3 ± 17.4</td>
<td>170.0 ± 14.6</td>
<td>100.1 ± 7.2</td>
<td>33.8 ± 2.9</td>
<td>5.7 ± 0.2</td>
</tr>
<tr>
<td>26 wk</td>
<td>241.0 ± 8.4</td>
<td>179.9 ± 6.4</td>
<td>101.6 ± 3.5</td>
<td>46.5 ± 1.4</td>
<td>5.9 ± 0.4</td>
</tr>
<tr>
<td>Fresh dressing (HPP) 37 °C storage 4 wk</td>
<td>282.2 ± 1.6</td>
<td>240.6 ± 4.0</td>
<td>138.2 ± 3.1</td>
<td>61.78 ± 2.1</td>
<td>7.4 ± 0.5</td>
</tr>
<tr>
<td>8 wk</td>
<td>303.1 ± 17.5</td>
<td>249.8 ± 7.2</td>
<td>159.5 ± 13.6</td>
<td>77.7 ± 6.4</td>
<td>9.4 ± 0.7</td>
</tr>
</tbody>
</table>

An asterisk (*) indicates a significant difference (P < 0.05) in organic acid concentration between fresh dressing (HPP) time 0 and stored sample.

Figure 5—Pyroglutamic acid concentration in pressure-processed ranch dressing stored at 4, 26, and 37 °C for up to 26 wk. Concentration (in mg/100 g wet weight) was estimated by HPLC. Error bars indicate standard deviation, n = 2.

Figure 6—Overall quality of pressure-treated ranch dressing stored at 4, 26, and 37 °C for up to 26 wk as determined by a trained sensory panel. Values are averaged from panels of 12 to 15 people using a 9-point anchor sliding line scale. Error bars indicate standard error. An asterisk (*) indicates a significant difference (P < 0.01) between indicated sample and 4 °C sample with the same storage time.
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were rated 1 category lower in overall quality than the 4 °C samples. From these data it can be inferred that the product stored at 26 °C for 26 wk would drop in overall quality from “Good” (a score of 7 on the scale) to “Below Good, Above Fair” (a score of 6), which is still an acceptable value on a 9-point quality scale (Figure 6). The differences between these samples were significant (P = 0.0012). In general, the 26 °C product became slightly darker and developed an “eggy/soured” odor and a slight “old/off/oxydized” flavor note when compared with the 4 °C sample stored for the same time period. Samples stored at 37 °C for 8 wk were significantly inferior in odor, flavor, and overall quality compared to 4 °C- and 26 °C-stored samples. Similarly, samples stored at 26 °C for greater than 16 wk were also inferior to the 4 °C samples in all quality attributes. Panelists provided descriptive feedback on the quality attributes of the samples analyzed at 26 wk. An overall summary of these comments is shown in Table 6. Storage at 26 °C resulted in samples with undesirable odors, rancidity, and a change in color. Flavor retention has historically been a major problem for dressing manufacturers. Off-flavors may develop due to lipid oxidation (Fetzek 1973).

Under the tested storage conditions, the changes observed for pressure-processed ranch dressing are not unusual or extreme. Whether consumers would find these levels of difference acceptable or unacceptable was not determined, but differences exist and trained panelists clearly indicated the degree of difference between ranch dressing stored at 4, 26, and 37 °C (Table 6). The darkening tendency during storage at 26 °C but not at 4 °C was also indicated by the downward trend of the Hunter L value over the storage life (Table 4). There are limited published studies on sensory analysis of ranch dressing. Yackinion and others (1999) investigated consumer preference for fat and garlic percentages in ranch salad dressing using a factorial design. Statistical analyses suggested that samples higher in garlic, sourness, and pepper characteristics were favored over samples with low flavor levels. Wendin and Hall (2001) investigated the impacts of fat, thickeners, and emulsifiers on the sensory and rheological properties of salad dressing. The base salad dressing was composed of water, sucrose, beta-carotene, mustard, acetic acid, vinegar, and salt. Fat levels were varied by adding 100 to 300 g/kg rapeseed oil. A mixture of xanthan and guar gums was used as the thickener, and milk protein was used as an emulsifier. Sensory analysis was performed using a panel trained to identify the following parameters: yellow color, presence of bubbles, thickness, fattiness, sourness, sweetness, saltiness, and off-flavor. The panel identified differences in off-flavor, fattiness, thickness, bubbles, and yellow color between the various product formulations.

Conclusions

This study demonstrated that high-pressure processing is an effective method for producing a good quality, relatively high pH ranch dressing that exhibits good microbiological, physical, and chemical stability for 6-mo at refrigerated and for at least 2 mo at room temperature conditions. P. acidilactici was identified as a pressure-resistant spoilage organism that could be targeted when establishing pressure processing parameters.

Acknowledgments

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References

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