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Applications of Ozonation and Membrane Treatment in Poultry Processing

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Applications of Ozonation and Membrane Treatment in Poultry Processing

In Plant Evaluation of

Pre-washing Chickens with Ozonated Water,

Ultrafiltration of Poultry Chiller Overflow for Reuse, and

Ozone as Anti-Microbial Agent in Chiller Bath

Conducted at

Petaluma Poultry Processors, Petaluma, California

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EXECUTIVE SUMMARY

Chlorine is widely used in sanitation of poultry operations. Chlorine generates several by-products that are proven to be harmful from food safety and environmental point of view. The search for alternatives to chlorine in poultry operations, particularly in the chiller is of interest to the poultry industry. Poultry processing plants use large volumes of water and the cost of obtaining and disposal of water is increasing rapidly. Hazard Analysis Critical Control Point (HACCP) quality control procedures introduced recently have increased the water usage and compounded the situation. The cost of electrical energy for cooling chiller water is another major concern. The objectives of this study were to address these issues.

Previous tests conducted at another poultry plant in Northern California indicated that ozonated water is effective in controlling the microbial levels on chickens. Tests conducted at Butterball Turkey in Huntsville, Arkansas, confirmed that the ultrafiltration of chiller water overflow meets the regulatory requirement for reuse in the chiller. The results of these tests were used to prepare the three comprehensive test protocols that were approved by the USDA for processing birds on a pilot scale for marketing. These protocols are

1. Plant evaluation of pre-wash of chickens with ozonated water
2. Plant evaluation of ultrafiltration of poultry chiller water for reuse
3. Eliminate use of chlorine as anti-microbial agent for poultry products and chiller bath water

The mobile membrane test and demonstration unit, a mobile ozone generator, a pilot bird pre-wash unit, and a pilot chiller bath were stationed at the poultry plant for six months. After the initial break-in, approximately 3,000 chickens were processed under each of the three protocols and marketed. The birds and the process water were tested microbiologically and chemically as specified in the protocols.

Plant evaluation of pre-wash of chickens with ozonated water indicated that ozonated water is as effective as chlorinated water in this application. The volume of ozonated water used was 30% less than volume of chlorinated water used. Ozone release into the environment can be kept within regulatory limits. Filtration of commercial chiller overflow water through an ultrafiltration membrane rated at 10,000 mwco met all USDA requirements for maximum use of reconditioned chiller water, including, light transmission and reduction in microorganisms.

Pilot chiller bath water at 38^{o} F maintained at 2 to 4 ppm ozone using 1/4 gallon of makeup water per bird remained clear and microbial counts were equivalent to a commercial 3-stage chlorinated chiller. Potential oxidative degradation measured by TBA and fatty acid profiles did not differ significantly from commercial chickens processed with chlorine. Sensory evaluations also failed to detect any difference between chickens processed with ozone and chlorine.

The plant consumes about 7.0 gallons of water per bird in the processing operation at present. Ultrafiltration of chiller water overflow, ultrafiltration of pre-wash water and coarse filtration of evisceration water are suggested to reduce the water consumption to 4.5 gallons per bird. Recovery of chiller overflow results in substantial energy savings.

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1.0 BACKGROUND

Chlorine and its derivatives are widely used as antimicrobial agents in poultry processing operations. Ozone has several advantages over chlorine in these applications. It does not produce halogenated organic compounds that are considered harmful to health. Ozone is a more powerful oxidizer and is considered more effective than chlorine compounds against several emerging pathogens. It can be generated on site and does not require storage.

Ozone has been used safely and effectively for treatment of drinking water for over nine decades mostly in Europe. It was approved in US for as generally recognized as safe (GRAS) in bottled water (FDA, 1995). More recently it was approved for use as an anti microbial agent in food processing including meat and poultry (FDA, 2001). This allows for its safe and effective use in poultry processing applications such as pre-washing and chilling of birds.

Ultrafiltration of chiller overflow was found to meet the guidelines for use of reconditioned water and to be cost effective under some local conditions. (Mannapperuma, et. al, 1996). Reuse of poultry chiller overflow reduces fresh water use, effluent volume and electrical energy used in chilling. USDA declared it as an accepted technology (Basu, 1996) based on an application made by Koch Membrane Systems.

A preliminary investigation of ozonation and ultrafiltration of poultry chiller overflow water was conducted as Phase I of this project at a poultry processor in Northern California during February through April 1999. Ultrafiltration of poultry chiller overflow water reduced the chemical oxygen demand to about 200 to 300 mg/L from over 1,000 mg/L. It also reduced microbial levels and improved light transmission to levels required for its reuse in the chiller.

Ozone is known to react with organic matter in water. Presence of organic matter in water reduces the antimicrobial effectiveness. Ultrafiltration reduces the COD in poultry chiller water and increases the effectiveness of ozone. Ozonated ultrafiltrate in combination with hydrogen peroxide and Tween 80 was used successfully to sanitize the birds during Phase I of the project.

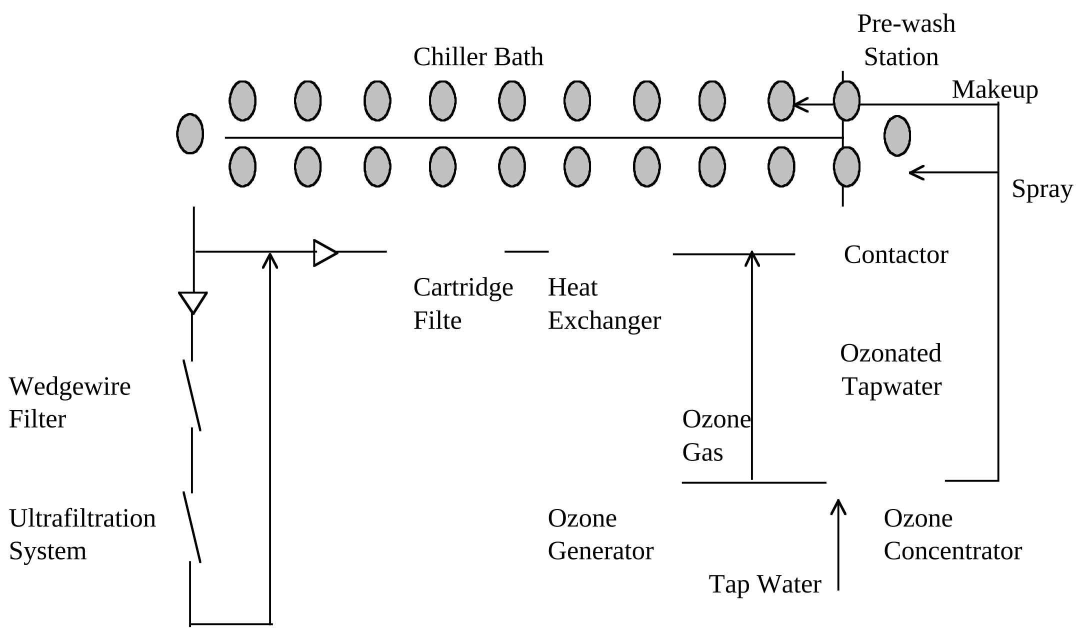
The results of these tests were used to prepare the three comprehensive test protocols for evaluation of application of ozonation and membrane treatment in poultry processing. These protocols were;

1. Plant evaluation of pre-wash of chickens with ozonated water
2. Plant evaluation of ultrafiltration of poultry chiller water for reuse
3. Eliminate use of chlorine for sanitizing poultry products and chiller bath water

The protocols were submitted to Food Safety Inspection Service (FSIS) of USDA in June 1999. They were approved after several queries and revisions by USDA in December 2000. Phase II of the project for in plant evaluation of the protocols began in April 2001.

Petaluma Poultry Processors Plant in Petaluma was the location of the Phase II of the project. The membrane demonstration and testing trailer (MTDU) with the membrane system, a mobile ozone generator, and a pilot chiller bath with a pilot bird pre-wash section were stationed at the poultry plant in April 2001. Figure 1.1 is a block diagram of the pilot chiller system. After the initial break-in, the in plant evaluations were conducted according to the protocols from April through October 2001.

Fig 1.1 Block Diagram of the Pilot Chiller Line



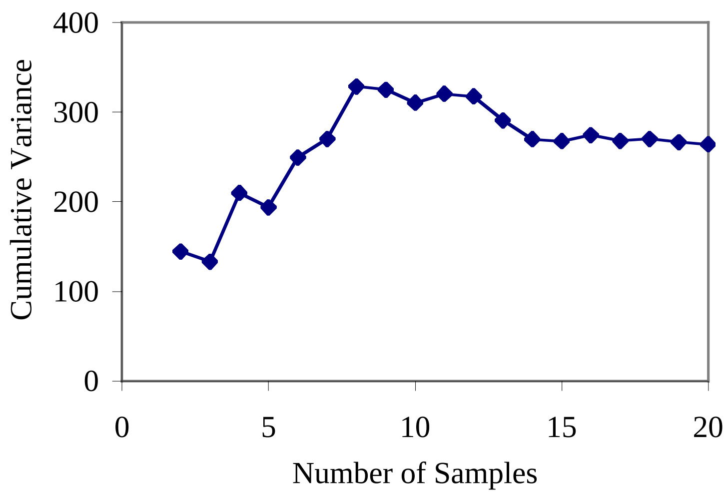
Water management issues of the plant and energy management issues related to water use at the plant were also evaluated in addition to in-plant evaluation of the protocols. This report is a comprehensive presentation of all these studies.

2.0 BASELINE STUDY

Microbial counts vary widely among birds. During Phase I of the project, the initial treatments trials conducted using single bird samples failed to draw any conclusions. Five-bird samples were used in subsequently tests and this improved the results. Larger samples reduce the variance of the mean and improve its confidence level. However, larger samples increase costs of testing. Therefore, it is important to select and optimum sample size before testing of protocols.

The plant collects two bird after chiller for microbial analysis daily. The results of these test over a ten consecutive days was obtained from the plant management. The E. coli counts of these samples were subjected to an analysis of variance. The analysis involved determination of the variance as number of birds was increased from 2 to 20 in steps on one bird. The cumulative variance was plotted against the number of birds. The variance is expected to increase and level of as the optimum number of birds is approached. The results of this analysis are presented in Figure 2.1.

Figure 2.1 Cumulative Variance of E. coli Counts on Rinse Test of Chickens from Commercial Chiller - Two Samples Daily for 10 Days



Two more base line tests was conducted by sampling sixteen birds randomly drawn from the process line over a two hour period after the chiller (Test 1-B) and after the prewash on two different days. Microbial counts were made on individual birds with the standard rinse test using 400 ml of sterile buffer. The E. coli counts of these samples were subjected to similar analysis of variance. The results on the analysis are presented in Figures 2.2 and 2.3 respectively.

Figure 2.2 Cumulative Variance of E. coli Counts on Rinse Test of Chickens after Pre-wash – June 19, 2001 – Test 1B

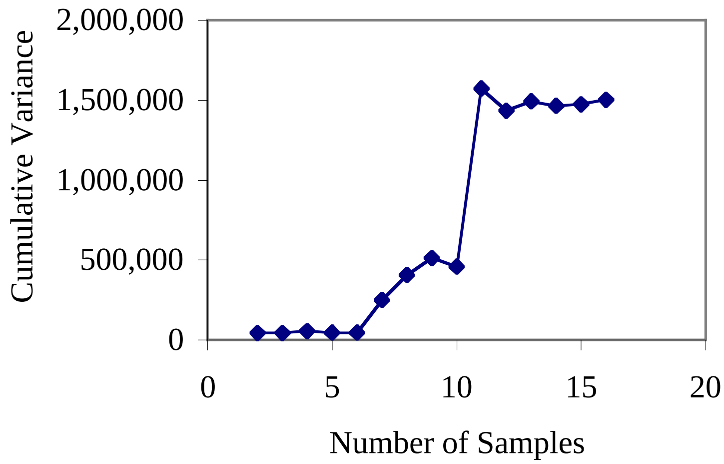
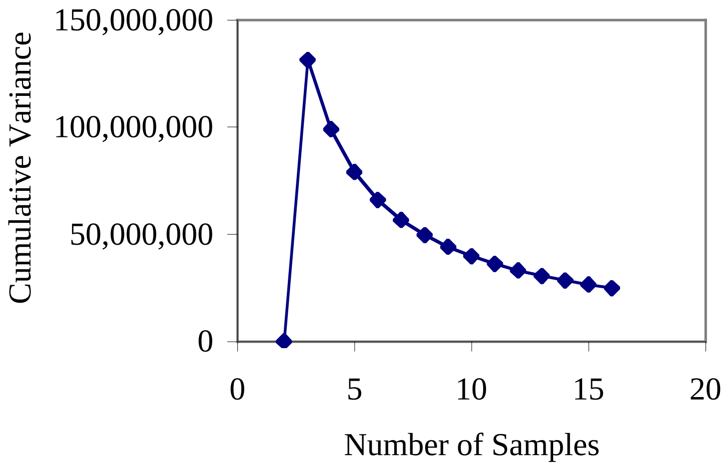


Figure 2.3 Cumulative Variance of E. coli Counts on Rinse Test of Chickens after Chiller – June 29, 2001 – Test 1C



In all three tests the cumulative variance leveled off after about 12 replicates, indicating little gain in statistical confidence with a larger number of samples. Thus we used a standard sample number of 16 samples selected randomly for each test in Protocols 1, 2 and 3.

3.0 PLANT EVALUATION OF PRE-WASHING OF CHICKENS WITH OZONATED WATER – PROTOCOL 1

3.1 Objective

This protocol was designed to demonstrate that washing with ozonated water before entry into the chiller bath could effectively reduce the microbial count of warm poultry carcasses. Birds were processed normally with use of ozonated water in lieu of other sanitizers in the pre-wash step. A total of approximately 2000 birds were processed and marketed under this protocol

3.2 Procedure

The plant processed the birds normally up to the commercial pre-wash station. Control birds (Test 1B and 1H) passed through the commercial pre-wash and were sampled just before entry into the commercial chiller. For treatments, warm birds were removed before the commercial pre-wash and transferred manually to the test pre-wash station. Ozonated water, selected adjuncts, and water were used in lieu of other sanitizers in the test pre-wash step. In this series, 16 birds were selected randomly from 5,400 birds on the process line. The 400-ml sterile buffer jars were pre-cooled in ice. Immediately after rinsing individual birds, the rinse solutions were iced and all samples were transferred to the microbiology laboratory for plating within 3 hours.

3.3 Results

The data obtained during the tests are summarized in Tables 3.1 and 3.2. Results of Tests1B, Test 1D, and Test 3G are presented in Figures 3.1, 3.2 and 3.3. Each test used 16 individual birds drawn randomly from the processing line.

The plant pre-wash consists of a continuous spray with hot water containing 25-ppm gaseous chlorine using 0.3 gallons of water per bird applied on line in a closed chamber, followed by an on-line “in and out” washer using 1 gallon per minute of 74°F water containing 25-ppm gaseous chlorine.

Ozone pre-wash was applied manually at plant water pressure (35 psig) for one minute; birds were dipped in fresh adjunct solution (peroxide, Tween 80, or Water) for 30 seconds before applying ozone spray for 1 minute using 1 gallon of 74° F water per bird.

Table 3.1. Rinse Test Counts on Warm Birds After Pre-Wash

Test No	Coliforms (CFU/ml) Average	Median	Std. Dev	*E. coli* (CFU/ml) Average	Median	Std. Dev	Description
1-A	15,238	4,400	31,439	6,075	2,450	7,360	Not Washed
1-B	2,669	2,300	1,849	1,638	1,250	1,226	Plant Pre-wash
3-H	990	330	1,381	705	250	794	Plant Pre-wash
1-D	1,413	1,250	1,031	775	450	713	Ozone 1.8 ppm
1-G	1,473	1,200	1,361	758	300	1,320	Ozone 2.7 ppm
3-G	189	110	230	139	70	235	Ozone 8 ppm
1-E	658	525	421	369	300	421	Ozone/Peroxide, Ozone 2.2 ppm
1-F	927	1,000	2,289	416	500	176	Ozone/Tween80, Ozone 3.96 ppm
1-H	858	445	980	323	235	271	Ozone/Water, Ozone 2.07 ppm
3-E	658	475	474	403	300	325	Tap water without ozone

Table 3.2. Comparison of No Pre-wash, Plant Pre-wash, Tap water Pre-wash, and Ozone Pre-wash on Warm Chickens Before the Chiller, Rinse Test

Treatment	APC (cfu/mL) Average	Std. Dev.	Coliforms (cfu/mL) Average	Std. Dev.
No Pre-wash	na	na	15,238	31,439
Plant Pre-wash	na	na	2,669	1,849
Tap Water	3433	1,588	436	262
Ozone[c]	3,150	2,013	189	230

[c] Water containing 8ppm Ozone

Figure 3.1 Rinse Test on Commercially Pre-washed Birds
Test 1B – June 19, 2001

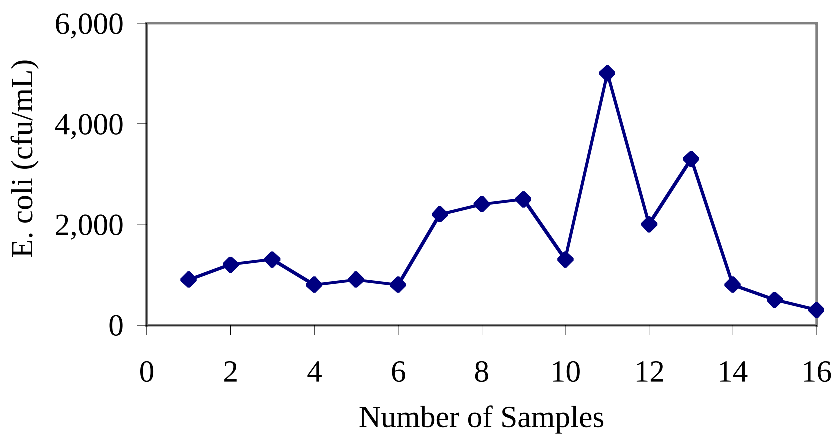


Figure 3.2 Rinse Test on Warm Birds Pre-washed with Ozonated Water
Test 1D – August 8, 2001

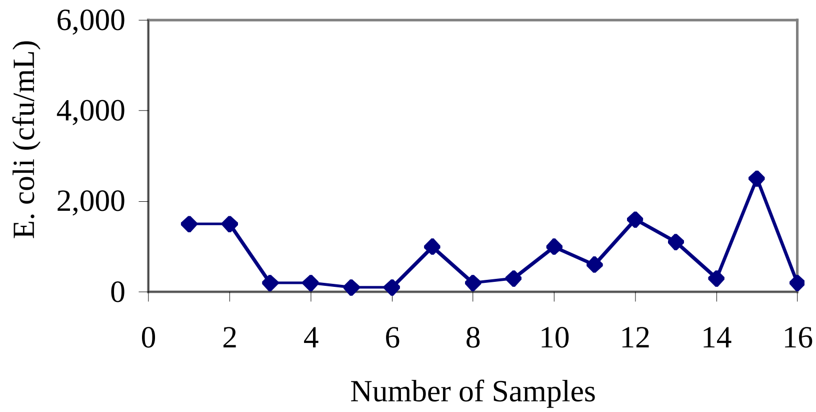
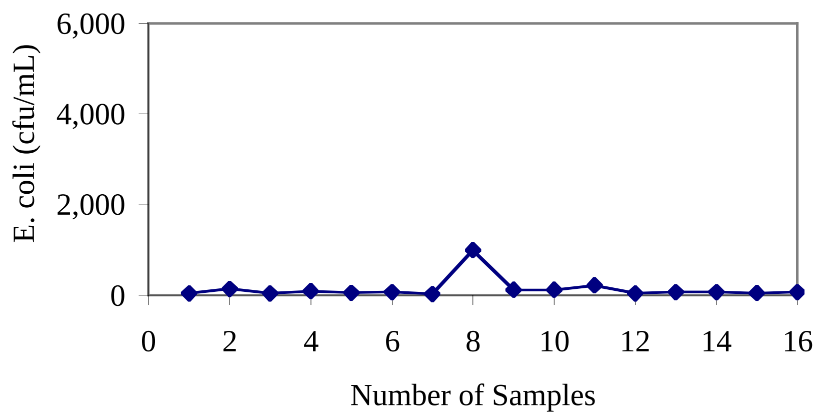


Figure 3.3 Rinse Test on Warm Birds Pre-washed with Ozonated Water
Test 3G – October 3, 2001



3.4 Conclusions

1. Plant continuous on-line pre-wash of 0.3 gallons of hot water containing 25 ppm gaseous chlorine per bird followed by on-line in-and out washer using 1.4 gallons 74°F water containing 25 ppm gaseous chlorine per bird reduced *E. coli* counts by 73 to 88%.

2. Manual pre-wash applied for one minute using 1 gallon of 74°F water containing 4 to 8 ppm ozone per bird reduced *E. coli* counts by 87 to 98%. Thus ozone effectively reduced microbial counts on warm birds before the Chiller. The volume of ozonated water (1 gallon per bird) usage was 30% less than the standard commercial pre-wash (1.7 gallons per bird).

3. Adjuncts (Peroxide, Tween80, and water) plus ozone improved count reductions slightly but not significantly.

4. Fugitive ozone off gassing is an issue. The test pre-wash was applied manually with a hand nozzle. The operator used a facemask and worked inside a shrouded area equipped with an exhaust fan to avoid in-plant emissions. Ozone levels in air immediately above the pilot chiller, as measured with an ENMET Spectrum on-line analyzer, occasionally reached 0.3 ppm. An O3DTKR ozone detector located adjacent to the pilot chiller across a four-foot aisle consistently recorded less than 0.1 ppm ozone in the ambient air.

5. We recommend that an enclosed continuous in line “in- and-out” pre-wash station supplied with ozonated water be used with adequate ventilation on a commercial process line.

4.0 PLANT EVALUATION OF ULTRAFILTRATION OF POULTRY CHILLER WATER FOR REUSE – PROTOCOL 2

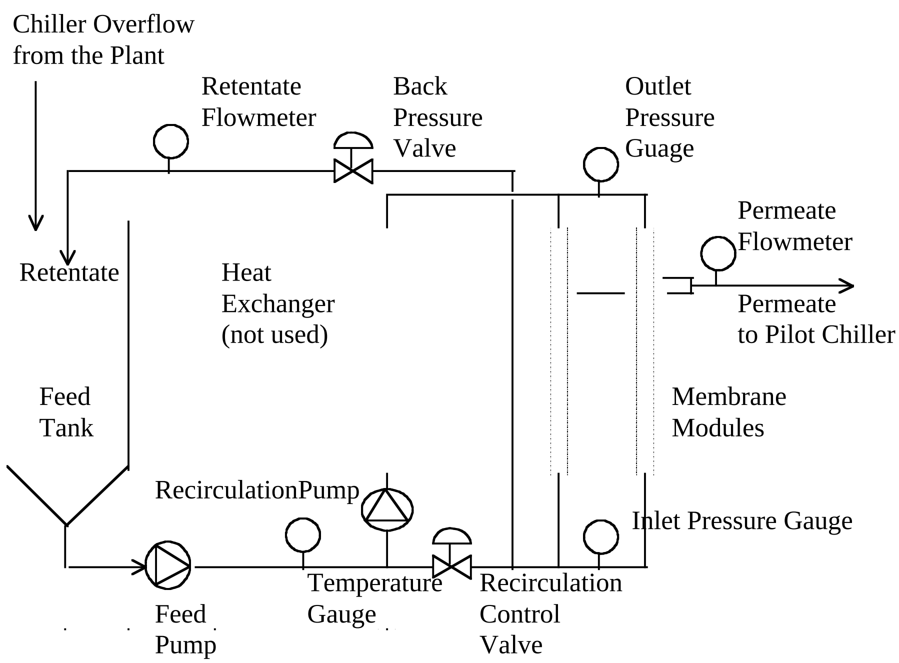
4. 1 Objective

These trials were designed to demonstrate that the poultry chiller overflow water could be reconditioned by ultrafiltration and reused in the chiller to reduce the fresh water intake requirement. This extends and amplifies lab scale tests conducted previously in conjunction with three commercial poultry processing plants. A total of approximately 1000 birds were processed and marketed under this protocol.

4.2 Procedure

The low-pressure system in the membrane test and demonstration unit (MTDU) was modified to accommodate two Romicon PM10 hollow fiber ultrafiltration modules. These module consists of hollow fibers with 1.1 mm internal diameter rated at 10,000 molecular weight cutoff (mwco). The total effective filtration area is 6.6 square meters per module. A schematic diagram of the membrane system with the instrumentation is shown in Figure 4.1.

Figure 4.1 Schematic Diagram of the Ultrafiltration System



The ultrafiltration system was configured to receive overflow water from the plant chiller No. 1. The permeate from the ultrafiltration system was pumped to the test chiller. The membrane system provided the test chiller with the specified make-up rate of 0.5 gallons per bird.

Several extended trials were conducted with the membrane system. During these tests, operating parameters and permeate flow rate were recorded. Samples of feed, permeate and retentate were taken several times for microbiological analysis and other quality determinations. Data collection spanned the entire operating day of the plant, including collecting enough feed stream to continue membrane processing during the plants break and lunch periods.

Microbiological evaluation included Aerobic Plate Count, Coliforms, E. coli, and a pathogen series comprising *Staphylococcus, Clostridium perfringens, Salmonella, Listeria, E. coli* O157:H7, and *Campylobacter.* Water quality tests included light transmission, chemical oxygen demand (COD), and chlorine levels. Pre-filtration ahead of the membranes comprised a coarse wire screen, and a 30-micron wedge wire screen. Temperature of the overflow water after pumping to the membrane system was about 40°F.

4.3 Results

4.3.1 Pathogen Removal with Membranes

Microbiological tests were designed to demonstrate that the membrane system could remove a variety of microorganisms, including pathogens, which may occur in chiller water. Two ultrafiltration trials were conducted on two different days using overflow from plant chiller #1 as the feed. Samples of overflow water and permeate were obtained three times, start, middle and end, of each trial.

All the overflow and permeate samples were tested for APC, E. coli and coliforms. All the permeate samples were tested individually for pathogens. However, three overflow samples were combined, for testing of pathogens to reduce the cost. The samples from Test 2B were quenched with Captor (calcium thiosulfate) to inactivate possible residual chlorine dioxide. This was not done with samples from Test2A. Table 4.1 is a listing of all the microbiological Test results.

Ultrafiltration permeate samples throughout the runs on both days were free of all pathogens. The range of *E. coli*, and Coliforms; APC was from <10 to 30 cfu/mL of permeate returning to the chiller. These results demonstrate the complete removal of pathogens with the ultrafiltration treatment by the membrane system.

Table 4.1. Filtration of Overflow Water from Plant Chiller No. 1 with 10,000 MWCO Ultrafiltration Membrane - Tests 2A September 5 and 2B September 17, 2001

Test			Cfu/mL					25 mL			50 mL
	Sample	time	APC	Coliforms	*E. coli*	Staph.	*Clostridium perfringens*	Salmonella	*Listeria*	E. coli 0157:H7	*Campylo-bacter*
2A	Overflow	Start	2,000	10	<10						
		Middle	20,000	<10	<10						
		End	10,000	30	<10						
		Mix	60,000	<10	<10	40	10	ND	ND	NEG	POS
	Permeate	Start	40	<10	<10	<10	<10	ND	ND	NEG	NEG
		Middle	<10	<10	<10	<10	<10	ND	ND	NEG	NEG
		End	<10	<10	<10	<10	<10	ND	ND	NEG	NEG
2B	Overflow	Start	90	10	<10						
		Middle	10	<10	<10						
		End	9,100	300	10						
		Mix	2,100	70	<10	60	<10	ND	ND	NEG	POS
	Permeate	Start	<10	<10	<10	<10	<10	ND	ND	NEG	NEG
		Middle	30	<10	<10	<10	<10	ND	ND	NEG	NEG
		End	20	<10	<10	<10	<10	ND	ND	NEG	NEG

ND = None Detected; NEG = Negative; POS = Positive

4.3.2 Flux and Rejection Characteristics during Ultrafiltration

Permeate flux, defined as the permeate flow per unit area per unit time is the major operating parameter monitored during membrane trials. Figure 4.2 shows the permeate flux characteristics observed during two ultrafiltration trials. The gradual decline in permeate flux observed during the trial is attributed to fouling of the membranes. This is to be expected in processing water containing foulants. Cleaning test conducted after the trials indicated that most of the fouling was due to proteins. A membrane less prone to protein fouling should be selected for this application. In test conducted under phase I of this project, the process of “bubbled air flotation” before membrane filtration was found to reduce fouling compounds, particularly fats and oils.

Test samples of feed, permeate and retentate were drawn three times during the trials. These samples were analyzed for electrical conductivity, chemical oxygen demand and light transmission. The results of this analysis are presented in Table 4.2. Electrical conductivity, which is a measure of ionized solutes, was not affected by the membrane. The rejection of chemical oxygen demand due to fats, proteins and sugars was around 90%. Ultrafiltration membranes rated at 10,000 mwco reject fats and proteins almost completely and pass nearly all sugars. Removal of turbidity in the chiller overflow was nearly complete as indicated by over

99% light transmission. Photographs of feed and permeate samples shown in Plate 4.1 is a further illustration of the turbidity removal.

Figure 4.2 Permeate Flux Characteristics during Ultrafiltration of Chiller Overflow

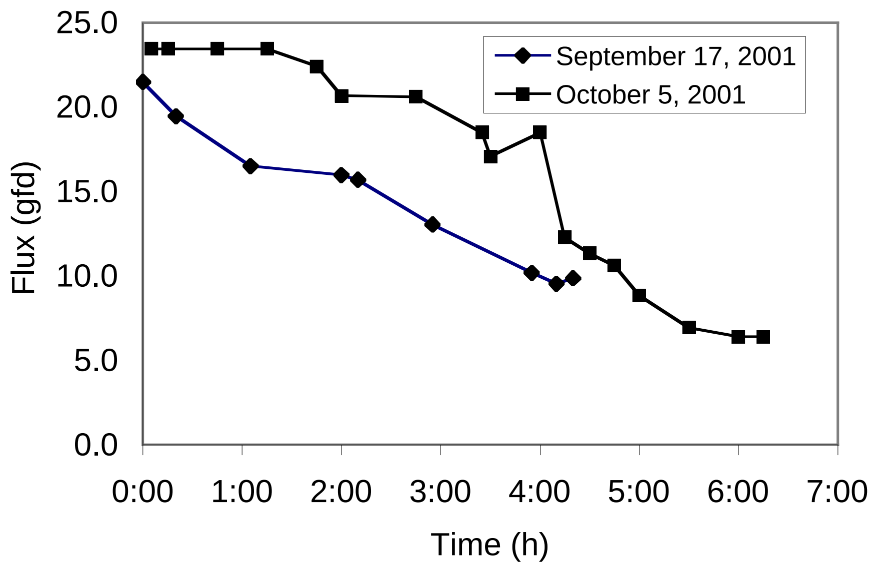
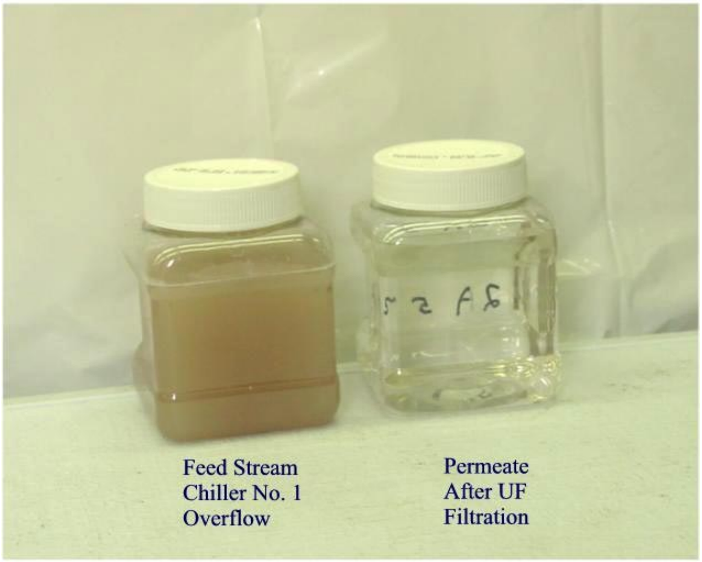


Table 4.2 Rejection Characteristics during Ultrafiltration of Chiller Overflow

Time of Sampling (hr)	Electrical Conductivity (μS/cm)			Chemical Oxygen Demand (mg/L)			Light Transmission (%)		
	Feed	Retentate	Permeate	Feed	Retentate	Permeate	Feed	Retentate	Permeate
Trial on September 17, 2001									
0:20	1,574	1,543	1,500	1,304	2,296	249	26.7	12.1	99.9
2:10	1,594	1,610	1,591	1,642	3,040	277	26.3	3.4	99.7
4:10	1,666	1,902	1,847	1,782	4,790	397	7.4	1.2	99.2
Trial on October 1, 2001									
0:15	1,149	1,179	1,092	658	698	42	36.7	28.9	99.8
4:15	1,455	1,456	1,444	1,430	1,574	283	29.3	19.4	99.9
6:15	1,558	1,543	1,471	2,018	2,734	370	17.7	7.5	99.9

Plate 4.1 Chiller water overflow form Chiller No. 1 before (left) and after (right) Ultrafiltration through a 10,000 mwco Ultrafiltration Membrane



4.4 Conclusions

1. Filtration of commercial chiller overflow water through an ultrafiltration membrane rated at 10,000 mwco met all USDA requirements for maximum use of reconditioned chiller water, including , light transmission and reduction in microorganisms.

2. Removal of microbial pathogens including *Campylobacter*, *Clostridium perfringens*, and *Staphylococcus* was demonstrated. APC was reduced by 4 to 5 logs.

3. Ultrafiltration using spiral membrane modules rated at 10,000 mwco can be economically attractive and meet all the regulatory requirements.

5.0 ELIMINATE USE OF CHLORINE FOR SANITIZING POULTRY PRODUCTS AND CHILLER BATH WATER - PROTOCOL 3

5.1 Objective

This protocol was designed to demonstrate that ozone could replace chlorine for sanitizing poultry products and Chiller water in poultry processing. A total of approximately 2000 birds were processed and marketed under this protocol.

Present USDA Regulations permit the use of up to 50-ppm chlorine in Chiller bath water to reduce the risk of pathogenic microorganisms on chilled poultry products. Chlorine is generally effective for this purpose, but is hazardous to store and handle in the processing plant. Concern exists because chlorine reacts with organics and may yield potentially carcinogenic trihalomethane (THM) compounds. These create problems for receiving body for wastewater effluent. Also USDA is conducting studies to determine whether THM compounds adsorbed by the carcasses during chilling present a food-safety risk. Recovery and reuse of spent chiller water can save energy and reduce total water usage, but may tend to concentrate such undesirable residues.

Ozone is an oxidizer and also can react with organics, but it is not known to yield persistent, toxic residues because of its short half-life and will not concentrate over time. Thus ozonation may be preferred if chiller water is to be reconditioned. Methods to measure the extent of oxidative reactions with chicken tissue were used to gauge the potential degradation. This included sensory evaluation of roasted birds, determination of TBA values, and fatty acid profiles on uncooked and roasted birds.

TBA (ThioBarbituric Acid) Value is recognized as a general measure of oxidation. Almost all living tissues contain oxidizable components, such as sulfhydryl groups in enzymes and aldyhydic structures in fatty acids, which may be altered by oxidation. Thus TBA value is used to estimate the extent of oxidative change, which has occurred in a substance. FDA requested determination of TBA values in ozone-processed chickens to estimate the extent of oxidative change caused by washing chicken carcasses with ozonated water.

The amount of fatty acids based on the number of carbon atoms and degree of unsaturation (double bonds) is determined as the fatty acid profile. The naming convention for fatty acids (e.g. 18:3) is two numbers representing number of carbon atoms (18) and degrees of unsaturation (3). Oxidation of fats during a process is expected to affect the profile by increasing the content of more saturated fatty acids at the expense of less saturated fatty acids. Fatty acid profiles were also included in the protocol to determine the fat oxidation effects of ozone.

The oxidized products of fatty acids contribute to stale or tallowy flavors, and usually are identifiable by taste more readily than by TBA analysis. For this reason, taste panels also compared chickens with and without washing in ozonated water.

5.2 Procedure

The pilot chiller line constructed for this study is a complete, freestanding system installed adjacent to plant chiller # 1. It operates independently at one bird per minute parallel to the plant chillers, with a separate pre-wash, water supply, cooling system, conveyor, filtration, and ozonation systems. Freshly slaughtered birds were removed from the commercial process line just before the plant pre-wash. Birds were transferred manually to the pilot chiller line adjacent to the plant chiller.

Birds were pre-washed using 1 gallon per bird of tap water with 4 to 8 ppm ozone at approximately 74°F. Water was applied manually for one minute with a spray nozzle to the interior cavity and outer surfaces of the birds. The birds were then cooled through the pilot chiller with water maintained at 38°F and 2 to 4 ppm ozone. Reconditioned overflow water from plant chiller No. 1 was ultrafiltered and returned to the test chiller at the rate of _ gallon per bird.

The pilot chiller test was repeated several times. A final rinse with ozonated water on birds emerging from the test chiller was evaluated in one test (Test 3-D). Sixteen birds were drawn from each of these tests for microbiological evaluations. Four birds were drawn from two tests (C and D) for chemical evaluations. Six birds were drawn form Test 3-D for sensory evaluations.

Four chickens were collected from the pilot chiller line after Test 3-D and four chicken were collected on the same day from plant chiller line chemical tests. Chickens from the plant chiller received the normal chlorine treatment. Chickens from the pilot chiller line were pre-washed and chilled in ozonated water.

5.3 Results

5.3.1 Microbial tests

The sample birds drawn form the pilot chiller (Tests C and D) and a set of control samples drawn from the plant chiller were used in the microbiological tests. USDA standard carcass rinse was done with the birds and the 400 mL rinse water was used in the tests. Microbiological tests conducted included total aerobic plate counts (APC), Coliforms, and *E. coli* counts. Microbial counts on birds emerging from the test system in full operation on two days are shown together with counts on birds from the plant chiller on one day in Table 5.1.

Table 5.1. Microbial Counts (cfu/mL of buffer) on Birds emerging from the Pilot Chiller and the Plant Chiller in Continuous Operation

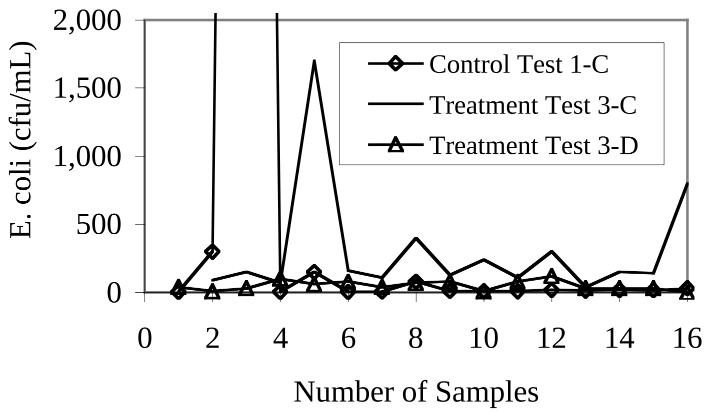
Bird Number	Birds from Pilot Chiller Test 3-C September 10, 2001			Birds from Pilot Chiller Test 3-D September 19, 2001			Birds from Plant Chiller Test 1-C June 29, 2001	
	APC	Coliforms	*E. coli*	APC	Coliforms	*E. coli*	Coliforms	*E. coli*
1	13,500	400		1,830	40	40	6	5
2	2,500	210	90	1,640	30	10	1,000	300
3	4,000	310	150	2,070	40	30	26,000	20,000
4	1,200	90	70	7,700	240	100	13	5
5	8,000	2,000	1,700	5,400	670	60	160	150
6	1,500	210	160	5,600	300	80	11	4
7	6,200	260	110	1,520	80	40	10	8
8	4,300	600	400	1,530	70	70	150	80
9	2,400	430	130	2,210	140	80	23	11
10	1,300	410	240	1,460	30	10	18	9
11	2,700	240	110	1,230	140	80	50	10
12	3,500	800	300	2,110	190	120	29	18
13	2,100	230	40	1,410	60	30	25	13
14	3,000	240	150	1,290	50	30	22	19
15	1,800	180	140	2,010	90	30	22	17
16	6,000	2,600	800	1,040	30	0	90	30
Average	4,000	576	306	2,503	138	51	1,727	1,292
Median	2,850	285	150	1,735	75	40	24	15
Std. Dev.	3,191	704	429	1,937	163	35	6,477	4,989
Ave. Dev.	2,250	462	264	1,399	107	29	3,034	2,338

The pre-wash in test 3-C and 3-D were done with water with 4 to 8 ppm Ozone. Test chiller was maintained at 2-4 ppm of ozone at birds were immersed for 22 minutes in the pilot chiller. A final rinse of one-minute duration with water ozonated to 8 ppm was done in Test 3-D.

Birds are chilled in the plant in a three-stage chiller with 25 ppm chlorine first and second stages and 15 ppm chlorine dioxide in third stage. The combined residence time in all three stages was 60 minutes. Sixteen birds were drawn randomly after the plant chiller and used in the Test 1-C.

The E. coli counts for tests 3-C and 3-D with birds treated in the pilot chiller and test 1-C with birds from the plant chiller are also presented in Figure 5.1. The E. coli counts of treatment (Test 3-D are comparable to the control (Test 1-C).

Table 5.1. E. coli Counts (cfu/mL of buffer) on Birds Emerging from the Test Chiller and the Plant Chiller in Continuous Operation



5.3.2 Chemical Tests

FDA requested thiobarbituric (TBA) analyses and fatty acid profiles as measures of potential oxidation of the tissue of ozonated birds. Both TBA values and fatty acid profiles were determined on tissue of fresh and roasted birds treated in the pilot chiller and compared with birds from the plant chiller. These results will strengthen the existing database on food contacted with ozone.

These analyses were conducted on birds from Test 3-D. Whole birds were collected from the chiller exit near the end of the test, along with similar birds emerging from the plant chiller. Birds were iced immediately, then frozen and held frozen until analyses were made. The birds were thawed in the refrigerator before testing.

Both control and treatment birds were roasted in the same oven, uncovered on pans, lightly salted, with no added spices. Birds were roasted at 350 F for one hour. Roasting was continued for another 15 to 30 minutes based on visual observation. Roasted birds were cooled and refrigerated overnight before testing.

Tissue samples were removed and analyzed for TBA value in a commercial analytical laboratory. TBA values in the fresh chicken samples were insignificant. The cooked samples had higher TBA values and were analyzed in duplicate. Results of the TBA analyses are listed in Table 5.2.

Table 5.2. Thiobarbituric Acid (TBA) Values of Commercially Processed (Control) and Ozone Treated (Treatment) Fresh and Roasted Chickens

Sample		Fat %	TBA	Ratio TBA/Fat
Fresh Chicken	Treatment	6.7	0.011	0.002
	Control	8.8	0.14	0.002
Roasted Chicken	Treatment	10.4	0.391 / 0.386 *	0.038 / 0.037 *
	Control	10.5	0.398 / 0.421 *	0.038 / 0.039 *

* Duplicate samples were tested

Since TBA value is directly proportional to the amount of fat present in a sample the ratio of TBA to Fat content was considered a better statistic for comparison. This ration almost doubled due to roasting. However, the ratio did not differ significantly between treatment and control for both fresh and roasted samples.

Table 5.3 Fatty Acid Profiles on Commercially Processed (Control) and Ozone Treated (Treatment) Chickens and Statistical Analysis

			Fatty Acid (%)			Statistical Analysis			
		Fresh	Fresh	Fresh	Roasted	Average	SD	F value	P value
14:0	Control	0.7	0.5	0.6	0.6	0.60	0.08	0.27	0.62
	Treatment	0.6	0.6	0.6	0.7	0.63	0.05		
16:0	Control	27.1	26.0	22.7	22.3	24.5	2.39	2.43	0.17
	Treatment	26.9	25.9	25.8	27.3	26.5	0.74		
16:1	Control	6.4	5.5	3.7	2.8	4.60	1.64	3.08	0.13
	Treatment	5.9	6.7	5.6	6.2	6.10	0.47		
17:0	Control	0.3	0.3		0.3	0.30	0.00		
	Treatment								
18:0	Control	6.1	6.8	9.5	8.9	7.83	1.63	0.03	0.87
	Treatment	6.1	5.7	8.9	9.7	7.60	2.00		
18:1	Control	35.4	35	33.1	33.8	34.3	1.06	0.65	0.45
	Treatment	32.7	36.6	39.2	33.8	35.6	2.92		
18:2	Control	22.5	24.3	25.4	29.4	25.4	2.92	3.39	0.12
	Treatment	25.4	22.4	17.7	19.8	21.3	3.33		
18:3	Control	1.6	1.6	3.6	4.2	2.75	1.35	3.00	0.13
	Treatment	1.8	1.7	1.1	1.6	1.55	0.31		
20:1	Control	0.3	0.2	0.4	0.3	0.30	0.08		
	Treatment	0.3	0.3	0.3	0.4	0.33	0.05		

Tissue samples from fresh and roasted chicken were also tested for fatty acid profiles in a commercial analytical laboratory. Three fresh samples and one roasted sample were used for both control and treatment. Fatty acid profiles of commercially processed chicken (control) were compared with those of chicken treated with ozonated water (treatment) conducting an analysis of variance. F values for all the comparisons were well lower than 5.99, which is $F_{0.05}$ for two-group eight-sample test. Therefore, no significant difference is detected between control and treatment in any of the fatty acids. The fatty acid profiles and statistical analysis are summarized in Table 5.3.

5.3.3 Visual and Sensory Evaluations

Skin color and odor of test birds was compared with commercial birds processed concurrently. Taste panels in a commercial laboratory with experienced judges compared the taste of birds processed in the plant chiller and the pilot chiller. They were also compared in home-cooked meal in three different homes.

Appearance of birds from the pilot chiller (treatment) was compared with control birds from the plant chiller (control) during each test. The birds were very similar with no clearly identifiable differences noted when viewed by experienced operating personnel and by the research team. A panel of 10 experienced tasters, divided into two groups, in a commercial laboratory evaluated flavor of roasted birds.

The birds were roasted on pans in the same oven, uncovered, lightly salted, with no spices added. Roasting temperature was 350° F for one hour plus 15 to 30 minutes based on visual observation. Roasted birds were cooled and refrigerated overnight before tasting. After removing portions of roasted birds for chemical analyses, a triangle difference/preference panel compared treated and untreated birds. The panel comprised 10 experienced panelists. Each panelist received a set of three coded samples for tasting, identifying and recording the preferred sample. Each set comprised two identical and one different portion. The panelists found no taste preference between treated and untreated samples. Order of presentation of paired and unpaired samples was randomized. Details of the taste test are in Table 7.

Table 5.4 Sensory Evaluation of Commercially Processed (Control) and Ozone Treated (Treatment) Roasted Chickens
Preferred Sample Identified with + sign

Set	Group 1			Group 2		
Number	Control		Treatment	Control	Treatment	
1	+			+		
2		+				+
3			+			+
4			+		+	
5		+		+		

Sensory tests on roasted birds comprised triangle preference tests in a commercial laboratory using experienced panelists. Additionally, three birds were refrigerated until roasted and evaluated separately by in-home tests.

In group 1 two panelists preferred treatment and three preferred control while in group 2 three panelists preferred treatment and two preferred control. The total vote was 5 preferred treatment and 5 preferred control, indicating no difference between treated and control samples.

Three pairs of treated and control coded samples were prepared for meal service in three homes, with a total of nine family members. No consistent taste preferences were reported. Two panelists noted slightly darker color in the treatment samples. This was noted also when samples were prepared for the taste panel. We believe the slightly darker color in ozone treated samples was due to less vigorous squeezing of the carcasses in the test chiller, which pulled the birds through the bath whereas the commercial chiller used a screw conveyor to push carcasses through the bath resulting in more "massaging" of the birds in the commercial chiller

5.4 Conclusions

1. Pilot chiller bath water at 38^{o} F maintained at 2 to 4 ppm ozone using 1/4 gallon of makeup water per bird remained clear and microbial counts were equivalent to a commercial 3-stage chlorinated chiller.

2. A final rinse of birds emerging from the chiller using tap water containing 6 to 8 ppm ozone further reduced microbial counts.

3. Potential oxidative degradation measured by TBA and fatty acid profiles did not differ significantly from commercial chickens processed with chlorine.

4. Sensory evaluation by expert panels and at-home food service judged ozonated chickens equal to commercial chickens from the same line processed with chlorine.

6.0 WATER AND ENERGY MANAGEMENT

Petaluma Poultry Processors Plant processes free range chicken and organic chicken. The plant has one process line that has a capacity of about 90 birds per minute and operates a single shift for five days per week. It operates longer shifts and holidays occasionally to meet the demand.

On a typical day it processes 32,700 birds in an 8-hour shift. The average production is about 4,000 birds per hour. The weights of birds vary from 5 lbs for juniors and Buddhist exempt pullets and 7.5 lbs for range chicken. Average weight of birds processed is 6 lbs. The plant obtains fresh water from wells in the promises and also from the City of Petaluma and the plant effluent is disposed to the treatment plant operated by the city of Petaluma.

6.1 Fresh Water Supply

The total water consumption by the plant is about 230,000 gallons per day. The plant has four wells within its premises. These wells are approximately 200 feet deep. The water supply system consists of pumps at each of the wells, four booster pumps and six bladder tanks. The capacity and installed power for the pumps are listed in Table 6.1

Table 6.1 Schedule of Pumps

Pump Description	Capacity (gpm)	Installed Power (kW)
Well #1	48	5.6
Well #2	120	5.6
Well #3	28	5.6
Well #4	117	7.5
Main Booster	200	5.6
Secondary Booster	300	11.2
Bird Wash Boosters (2 nos)	n.a.	7.5

The wells are not capable of supplying the total water requirement of the plant. The deficit is met with by obtaining water from the City. The volume of water obtained from the city varies from 7,500 to 30,000 gallons per day. The city charges $2.60 per ccf or $ 3.47 per thousand gallons (kgal) of water.

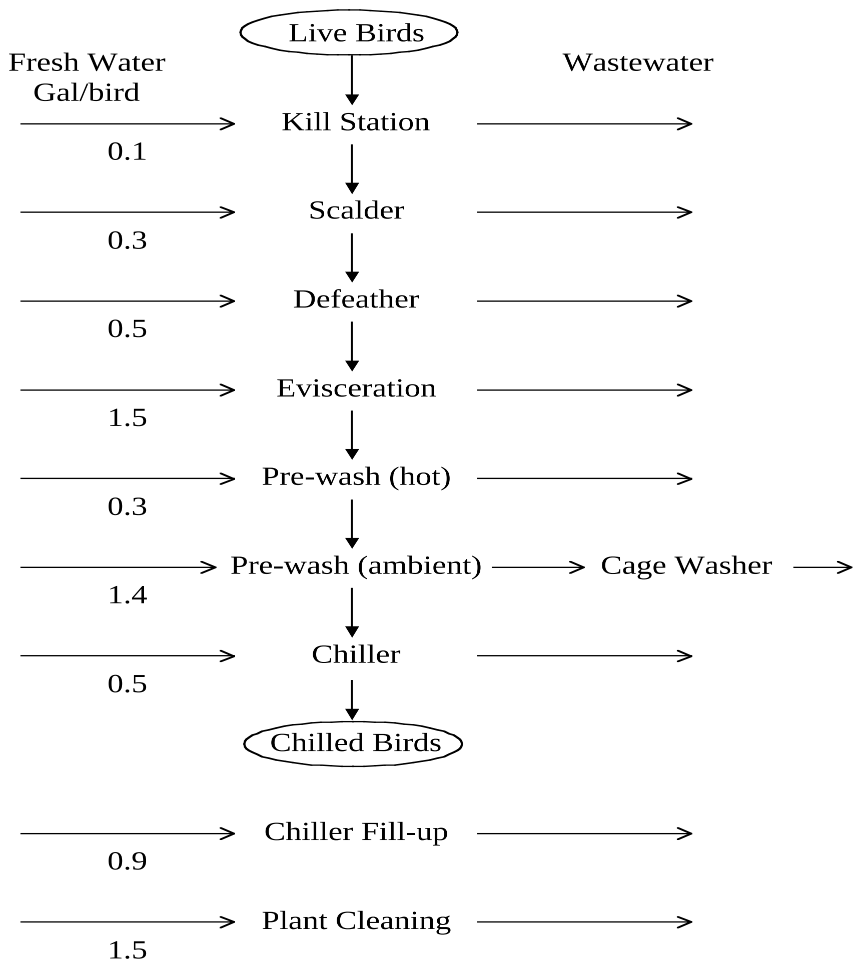
The average water consumption by the plant is about 20,300 gph (gallons per hour), 18,500 gph from the wells and 1,800 gph from the City. The specific water consumption is estimated at 7.0 gallons of water per bird processed.

The total installed power in the well water supply system is 48.6 kW. Assuming 80% utilization of pumps and 18,500 gallons per hour of well water consumption, the specific energy consumption for well water supply is 2.10 kWh per kgal.

6.2 Water Use in the Plant

The plant uses water in several unit operations during processing and for cleaning of the plant and machinery at the end of the shift. The total water use amounts to about 7.0 gallons per bird. Figure 6.1 is a flow diagram illustrating the water use pattern.

Figure 6.1 Flow Diagram of In-Plant Water Use



Scalder and hot pre-wash use hot water while chiller uses low temperature water. All other operations use ambient water. The estimated recoverable energy content in hot and cold process water streams is summarized in Table 6.2. Incoming water was assumed to be at the average ambient temperature of 70 F. Overall conversion efficiency of 85% was assumed for heating and a COP of 3.0 was assumed for cooling.

Table 6.2. Recoverable Energy Contents in Hot and Cold Process Water Streams

Unit Operation	Temperature (F)	Flow (gal/bird)	Thermal Energy (Btu)		Electrical Energy (kWh)	
			Per bird	Per hour	Per bird	Per hour
Scalder	140	0.25	145	667,000		
Hot Pre-wash	180	0.25	228	1,048,000		
Chiller	35	0.50	-145	-667,000	0.014	78.4

6.3 Wastewater Disposal

Process water from the plant is screened at several points to remove feathers and offal. Screened effluent is treated by a dissolved air flotation (DAF) system with polymer injection. Solid waste including feathers, offal and DAF skimmings are disposed separately.

All the process water from the plant is discharged to the wastewater treatment facility operated by the City of Petaluma. The city charges $3.47 per kgal of wastewater. There are surcharges of $0.4670 per lb of BOD above a threshold of 250 ml/L. and $0.4242 per lb of TSS above a threshold of 250 ml/L. In addition fines are imposed above 900 mg/L of BOD or 700 mg/L TSS. Total sewer charge is about $20,000 per month. This is expected to increase in the near future.

Petaluma wastewater treatment plant treats about 2 billion gallons per year. The plant consists of primary solids separation, activated sludge digestion followed by polishing in trickling filters. The sludge dewatered in a press and trucked to Redwood Landfill. Treated water is disinfected by chlorination and then used in irrigation of a golf course and a fodder field during summer or discharged to Petaluma River rest of the year.

Energy consumption for water treatment has been studied in 1996. The electrical energy for primary and secondary treatment amounted to 3,142 MWh during this year. The energy for the aeration, chlorination and dechlorination treatment was 1,800 MWh and for the irrigation distribution was 1,286 MWh.

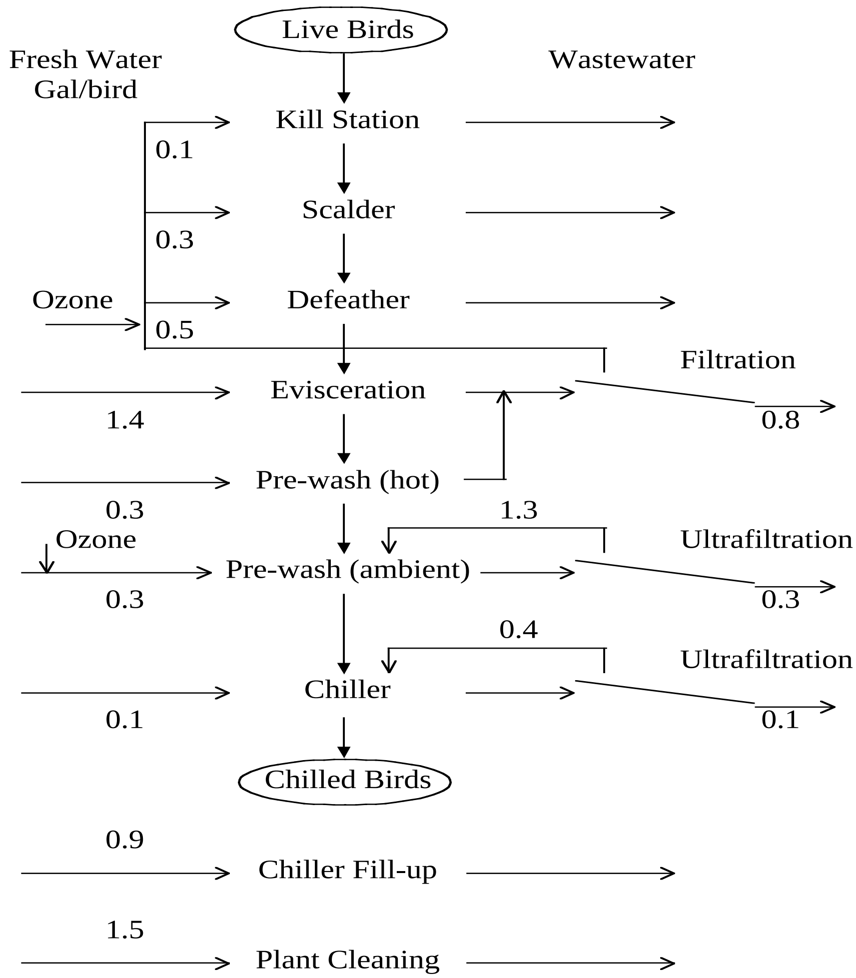
The total electrical energy consumption was 6,228 MWh for treatment of 2 billion gallons. This amount to 3.1 kWh/kgal of wastewater treated. The cost of electrical energy was $300,000 during the year, which amounted to $0.15/kgal.

Disposal of the sludge involves transport that also consumes energy. The trucks made 400 trips per year to the landfill ten miles away and used 5,400 gal of gasoline/year. At 126,000 Btu/gal of gasoline this amounts to 340 Btu/kgal of wastewater treated. This translated to 0.03 kWh electrical equivalent per kgal of wastewater treated.

6.4 Water and Energy Conservation

Petaluma Poultry Processors plant processes about 90 birds per minute and operates one eight-hour shift per day. It has plans to increase production by two-shift operation in near future. The plant uses about 200,000 gallons of fresh water per day in its operations. Adequate fresh water supply is a critical requirement to increase production.

Figure 6.2 Flow Diagram with Proposed Recovery Systems



The cost of fresh water supplied by the city is $3.48 per kgal and the cost of disposal is also $3.48 per kgal. It is possible to recover process water using advance treatments like membrane filtration and yet be cost effective compared to these cots. Three water recovery schemes were identified to reduce the total water use from present 7.0 gallons per bird to 4.5 gallons per bird. Figure 6.2 is a flow diagram illustrating these possibilities.

6.4.1 Chiller Overflow Recovery by Ultrafiltration

The plant uses a water bath chillers to cool birds before storage. The chiller system has a continuous make-up overflow system that uses 0.5 gallons of fresh water per bird or about 45 gallons of water per minute. This water is discharged at a temperature of about 42 F. It is possible to reuse about 90% of this water by reconditioning it through ultrafiltration. This will reduce fresh water use, effluent disposal volume and a substantial part of electrical energy used in chilling.

A full size ultrafiltration membrane system is proposed to treat 45 gpm of chiller overflow and produce 40 gpm of reconditioned water for reuse in the chiller. This will be used as make-up to the chiller partially replacing fresh water make-up. The membrane system comprises of 4,800 sft of spiral ultrafiltration membrane modules rated at 10,000 molecular weight cut-off. A 100 micron prefilter and a check filter are included in the system to prevent suspended solids from entering the system. The system contains a feed pump and two recirculation pumps. The power consumption is estimated at 2.1 kW for feed pump and 6.9 kW for each of the recirculation pumps.

Table 6.3. Economic Assessment of Membrane System for Chiller Overflow Recovery

System Parameters	
Ultrafiltration capacity (gpm)	40
Capital investment ($)	200,000
Electric power (kW)	16
Hours of production (hours/day)	15
Hours of cleaning (hours/day)	2
Days of operation (days/year)	250
Water recovery (kgal/year)	9,000
Cleaning water use (kgal/year)	500
Net water saving (kgal/year)	8,500
Expenses ($/year)	
Energy cost (68,000 kWh @ $0.10)	6,800
Membrane cost ($19,200 every two years)	9,600
Cleaning (lump sum)	2,000
Labor (2 hours/day @ $20)	10,000
Total operating costs	**28,400**
Potential Benefits ($/year)	
Electrical energy saving (9,000kgal @ $2.31)	20,790
Fresh water saving (8,500kgal @ $3.48)	29,580
Disposal saving (8,500kgal @ $3.48)	29,580
Total Savings	**79,950**

The benefits of the system includes savings in fresh water at the rate of $3.48 per kgal, savings in disposal cost at the rate of $3.48 per kgal, and the energy saving due to avoided cost of chilling. Latter is estimated based on a set of assumptions. Annual average incoming water temperature is about 70 F. The chiller water overflow is at 42 F. The water recovered by filtration would be about 45 F due to heat dissipation at pumps. Therefore, the net saving is a temperature difference of about 25 F. The electrical energy saving due use of reconditioned water is estimated as 23.1 kWh per kgal based on a coefficient of performance of 3.0 for the refrigeration system and electric motor efficiency of 0.85. This is valued at $2.31 per kgal at an electricity cost of $0.10 per kWh.

The economic assessment of the membrane application based on these assumptions for double shift plant operation is presented in Table 6.3. The capital cost listed is only a budgetary estimate. More accurate estimate should be obtained after conducting pilot tests with a membrane system manufacturer.

The cost of recovered water is $3.34 per kgal or $2.50 per ccf based on this preliminary economic assessment. This is very attractive compared to the present costs of fresh water, sewer and chilling. The return on investment is 29%. This may be low compared to industry standard. However, substantial financial incentives are available due to possible savings in electrical energy consumption. These will reduce the capital investment and improve the cost effectiveness of the project.

6.4.2 Pre-wash Water Recovery by Ultrafiltration

The birds after evisceration are washed with ambient water in an enclosure. This operation uses 1.4 gallons of fresh water per bird or about 126 gallons of water per minute. It is possible to treat this water by ultrafiltration followed by anti-microbial treatment and reuse for the pre-wash. This will reduce fresh water use, effluent disposal volume chilling.

An ultrafiltration membrane system similar but larger than the chiller water recovery system is proposed to treat 125 gpm pre-wash and recover 110 gpm for reuse. The membrane system comprises of 15,000 sft of spiral ultrafiltration membrane modules rated at 10,000 molecular weight cut-off. The system contains a feed pump and two recirculation pumps. The power consumption is estimated at 6 kW for feed pump and 22 kW for each of the recirculation pumps.

The economic assessment of the membrane application based on these assumptions for two shift plant operation is presented in Table 6.4. The capital cost listed is only a budgetary estimate. More accurate estimate should be obtained after conducting pilot tests with a membrane system manufacturer. The costs for anti-microbial treatment should also be included. The cost of recovered water is $2.60 per kgal based on this preliminary economic assessment. This is also attractive compared to the present costs.

Table 6.4 Economic Assessment of Membrane System for Pre-wash Water Recovery

System Parameters	
Ultrafiltration capacity (gpm)	110
Capital investment ($)	400,000
Electric power (kW)	50
Water recovery (kgal/year)	25,000
Cleaning water use (kgal/year)	1,500
Net water saving (kgal/year)	23,500
Expenses ($/year)	
Energy cost (160,000 kWh @ $0.10)	16,000
Membrane cost ($60,000 every two years)	30,000
Cleaning (lump sum)	5,000
Labor (2 hours/day @ $20)	10,000
Total operating costs	**61,000**
Potential Benefits ($/year)	
Fresh water saving (23,500 @ $3.48)	81,780
Disposal saving (23,500 @ $3.48)	81,780
Total Savings	**163,560**

6.4.3 Reuse of Evisceration Wash Water by Coarse Filtration

Water discharged from the evisceration section can be treated by coarse filtration, followed by anti-microbial treatment and used in the killing, scalding and defeathering sections. A broad choice of technologies is available for this process.

Technology and economic assessment was not performed for reusing evisceration water because this application was not pilot-tested as part of this project. However, such reuse is already being done at other poultry plants. This reuse possibility is included in arriving at the 4.5 gal/bird net water-use figure that can be achieved with implementation of the process changes recommended as a result of this project.

7.0 CONCLUSIONS AND RECOMMENDATIONS

This study evaluated three protocols approved by USDA for processing chicken on a pilot scale for marketing. Water and energy management practices of the plant were also studied and conservation strategies were proposed.

Plant evaluation of pre-wash of chickens with ozonated water indicated that ozonated water is as effective as chlorinated water in this application. The current practice of using water containing 25 ppm chlorine per bird reduced *E. coli* counts by 73 to 88%. Pre-wash using water containing 4 to 8 ppm ozone reduced *E. coli* counts by 87 to 98%. The volume of ozonated water used was 30% less than volume of chlorinated water used. Ozone release into the working environment was a concern but can be kept within regulatory limits with proper precautions. A well enclosed continuous in line "in- and-out" pre-wash station supplied with ozonated water used with adequate ventilation on a commercial process line is recommended.

Filtration of commercial chiller overflow water through an ultrafiltration membrane rated at 10,000 mwco met all USDA requirements for maximum use of reconditioned chiller water, including, light transmission and reduction in microorganisms. Removal of microbial pathogens including *Campylobacter, Clostridium perfringens,* and *Staphylococcus* was demonstrated. APC was reduced by 4 to 5 logs. Ultrafiltration using spiral membrane modules rated at 10,000 mwco can be economically attractive and meet all the regulatory requirements.

Pilot chiller bath water at 38^{o} F maintained at 2 to 4 ppm ozone using 1/4 gallon of makeup water per bird remained clear and microbial counts were equivalent to a commercial 3-stage chlorinated chiller. A final rinse of birds emerging from the chiller using tap water containing 6 to 8 ppm ozone further reduced microbial counts. Potential oxidative degradation measured by TBA and fatty acid profiles did not differ significantly from commercial chickens processed with chlorine. Sensory evaluation by expert panels and at-home food service judged ozonated chickens equal to commercial chickens from the same line processed with chlorine.

At present the plant consumes about 7 gallons of water per bird in the processing. Ultrafiltration of chiller water overflow, ultrafiltration of pre-was water and coarse filtration of evisceration water are suggested to reduce the water consumption to 4.5 gallons per bird. Recovery of chiller overflow results in substantial energy savings.

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