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A survey of Iowa apple cider: microbial loads and producers' production practices

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A Survey of Iowa Apple Cider: Microbial Loads and Producers' Production Practices

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INTRODUCTION

Escherichia coli O157:H7 was first recognized as a human pathogen in 1982 and is now known as a recurring causative agent in food-associated illnesses. The vehicle of infection is usually undercooked food, especially ground beef products such as beef burgers (Kay et al., 1994). However, in the past decade a wider variety of foods have been identified in *E.coli* O157:H7 outbreaks, including apple cider, apple juice, mayonnaise, salad dressing, lettuce, and water. *E.coli* O157:H7 is now known as an important cause of hemorrhagic colitis and hemolytic uremic syndrome (HUS) (Griffin et al., 1991).

E.coli O157:H7 was first associated with apple cider in a 1980 outbreak of HUS in Canada. The patients had bloody diarrhea and cramps before developing HUS (Steele et al., 1982). In the United States, an outbreak of hemorrhagic colitis was first associated with apple cider in 1991 in Massachusetts (Besser et al., 1992). Again, *E.coli* O157:H7 was believed to be the causative agent. Since the 1991 incident, several *E.coli* O157:H7 outbreaks have been linked with apple cider including one with 45 cases associated with drinking Odwalla brand unpasteurized apple juice (MMWR,1996).

Previously, high-acid foods such as apple cider were not regarded as potentially hazardous or conducive to the survival and growth of pathogens. Apple cider with a typical pH of 3.4 to 4.0 is considered a high-acid food. Demonstration of the survival of *E.coli* O157:H7 in apple cider, mayonnaise, and salad dressing suggests that *E.coli* O157:H7 possesses unusual tolerance to low pH (Semanchek et al., 1996).

In 1998 the Food and Drug Administration (FDA) mandated the requirement of a warning label on fruit juices that were not processed in a manner to produce at least a 5-log (100,000-fold) reduction in the pertinent target microorganism. Exceptions to the rule included juices sold in restaurants, juice bars, and other retail establishments that sell ready-to-consume products. The warning label reads as follows: "WARNING: This product has not been pasteurized and, therefore, may contain harmful bacteria which can cause serious illness in children, the elderly, and persons with weakened immune systems" (21 CFR Part 101 [Docket No. 97N-0524] RIN 0910-AA43). The 5-log reduction standard was determined by groups at the National Advisory Committee on Microbiological Criteria for Foods (NACMCF) who considered what levels of *E.coli* might typically occur in juice and then added a standard 100-fold safety margin. The 5-log reduction performance standard has this built-in safety factor that provides additional consumer protection (FDA, 2001)

Thermal pasteurization of apple juice/cider can achieve this reduction readily; however, there are no definitions of time and temperature requirements for pasteurization in the cider industry. Concerns about quality changes imparted by pasteurization and about the costs of installing a pasteurization unit affect the decisions made by cider producers about whether or not they can afford to produce pasteurized cider. Other means of *E.coli* O157:H7 reduction in cider currently under investigation include ultraviolet light treatment, irradiation, high pressure, pulsed electric fields, and the use of ozone (Buchanan et al., 1998; Linton et al., 1998; Wright et al., 1999; Iu, et. al, 2001; Garcia-Graells et al., 1998). Any combinations of treatments that lower the pathogen load by 5 logs in the apple cider can be

implemented. Once the 5-log reduction is reached, the warning label is no longer necessary.

The FDA passed the final Hazard Analysis Critical Control Point (HACCP) regulation on January 19, 2001. This ruling requires all apple cider producers to have a HACCP program in place by January 20, 2004 (FDA, CFR 21.2001). The FDA intends that this final rule cover both "interstate juice" and "intrastate juice". The agency proposed in Sec. 120.9 that failure of a processor to have and to implement a HACCP system that complies with Secs. 120.6, 120.7, 120.8, or otherwise to operate in accordance with these requirements, renders the juice products of that processor adulterated.

The purpose of this study was to look in detail at cider production in Iowa to determine the microbial load of cider and the production practices affecting these microbial loads. With knowledge gained through the analysis of cider production, HACCP plans were created and were given to the producers involved in the study. Cider was followed during storage to trace the microbial load, determine shelf life, and evaluate the survival of *E.coli* and two other common contaminants under various conditions. All of the information gathered was shared with the cider processors to assist them in producing a safer product.

LITERATURE REVIEW

Theodor Escherich first characterized *Escherichia coli* in 1885. The organism was first termed *Bacterium coli* commune and was found to be commonly isolated from feces (Janda et al., 1998). *E.coli* belongs to the family *Enterobacteriaceae*, which encompasses at least 37 different species of microorganisms. Several features identify isolates as members of the family *Enterobacteriaceae*: small Gram-negative rods, aerobic/facultative anaerobic metabolism, oxidase-negative, catalase-positive, 38-60% G-C content, the ability to ferment D-glucose and to convert nitrates to nitrites (Janda et al., 1998). *E.coli* is also a coliform, i.e. those members of *Enterobacteriaceae* that are able to decompose lactose with acid and gas production. However, slow lactose-fermenting or lactose-negative *Enterobacteriaceae* do exist and should be taken into account when enumerating coliforms (Kay et al., 1997).

The intestinal tract of warm-blooded animals (including humans) serves as the natural reservoir for *E.coli*. Birds may also harbor *E.coli* as part of their normal bowel flora (Janda et al., 1998). *E.coli* is commonly found in external environments (soil and water) that have been affected by human and animal activity (Sussman,1997). Although cattle appear to be the main environmental reservoir, with transmission from animal to animal or from animal to human a major means of infection, the distribution of *E.coli* O157:H7 in the environment as a whole is relatively unknown. Other transmission modes need to be determined (Phillips, 1999). *E.coli* can find its way into the food system through at least two mechanisms: during the processing of slaughtered animals at abattoirs, meats destined for retail

consumption can become fecally contaminated with *E.coli*; alternatively excretion of biologic wastes by both domesticated and wild animals onto lands used for agricultural purposes can adulterate the food supply (Sussman, 1997).

Because most strains of *E.coli* are not pathogens, and because different strains cause various types of disease, it is important to be able to differentiate strains or groups of strains that may be responsible for a particular outbreak (Salyers et al., 1996). Those *E.coli* associated with foodborne illness are usually grouped into four categories based on virulence properties, clinical syndrome, epidemiology and O:H serogroups. The O:H serological classification system is based on the O antigen of lipopolysaccharides and the H antigen of flagella. The O antigen identifies the serogroup of a strain, and the H antigen identifies its serotype (Salyers et al.,1994). Currently, there are five virotypes: enteropathogenic (EPEC), enteroinvasive (EIEC), enterotoxigenic (ETEC), enteroaggregative (EaggEC), and enterohaemorrhagic (EHEC). *E.coli* O157:H7 is an EHEC, which can cause haemorrhagic colitis in humans and usually presents as bloody diarrhea. (Kay et al., 1997).

Apple Cider Production

The term "cider" varies in meaning around the world, but in the United States, the term commonly refers to the freshly pressed juice of apples (Semanchek et al., 1996). Although there is no legal definition of cider, apple cider is typically distinguished from apple juice on the basis of a darker color, less clarity, and the presence of suspended solids (Downing, 1989).

Apple cider production begins with the harvesting of apples. The maturation level of the apples is important to the overall taste and quality of the cider. Because apple cultivars ripen at different rates (Childers, 1983), the use of a particular cultivar in cider production to create the desired blend of flavors will vary throughout the cider production season. Early in the production season (i.e., late August through mid-October) the harvested apples are used very quickly (stored less than 7 days after harvest) because of the demand for apple cider. As the season progresses, apples that have been stored at 4°C for one month or longer are predominantly used.

Once the apples are picked, they may go into refrigerated storage (7 to 12°C) or directly into production. Depending on the processor, apples may be washed and/or brushed prior to use. Washing may be accomplished through a dip treatment in which the apples are dumped into a tank of chlorinated water and allowed to soak for a period of time. A fresh water rinse may also be used in which the apples are placed on a moving belt that undergoes a series of brushing and rinsing applications (personal observation). Once the apples are washed and brushed, they are ground into a pulp. The pulp and pomace (cores, stems, etc) are sent through a crushing system of belts or press that extracts the juice from the pulp. The leftover pulp and pomace are disposed of by hand or through an auger system (personal observation).

The extracted juice is pumped through a filter into a holding tank. Once in the holding tank, preservatives can be added. Potassium sorbate and/or sodium benzoate at concentrations of 0.1% have been approved for use in cider (Sofos et

al., 1980). Usually the cider is held in a tank overnight to allow for settling of solids. Once settling has taken place, the cider can be bottled or further treated through pasteurization. Various times and temperatures of pasteurization are being used; times range from 1 sec. to 11 sec. and temperatures range from 163°F to 183°F (73°C to 84°C).

Spoilage Organisms Associated with Apples and Cider

A variety of yeasts can usually be found on fruits, and these organisms often bring about the spoilage of fruit products, especially in the field. Many types of yeast are capable of attacking the sugars found in fruits and bringing about fermentation with the production of alcohol and carbon dioxide (Jay, 2000).

Surveys of the microflora of apples have focused on three distinct yet interrelated areas: in the orchard, during storage, and within processing facilities (Deak et al., 1996). Weakly fermentative yeasts rather than molds or bacteria typify the predominant primary flora of sound apples. Yeast populations range from 10^2 to 10^6 cells per apple and show a seasonal variation, reaching a peak in fall. The main resident yeast species on apples include *Hanseniaspora uvarum, Metschnikowia pulcherrima, Debaryomyces hansenii, Sporidiobolus roseus and Cryptococcus albidus*. *Aureobasidium pullulans* and *Cladosporium herbarum* (black yeasts) are also permanent members of the microflora (Deak et al., 1996).

A finished fresh apple cider product contains small amounts of ethanol in addition to acetaldehyde. The holding of nonpasteurized or unpreserved cider at

suitable temperatures invariably leads to the development of cider vinegar, which indicates the presence of acetic acid bacteria in these products (Jay, 2000).

In their study of the ecology of acetic acid bacteria in cider manufacture, Passmore and Carr (1975) found six species of *Acetobacter.* Those that display a preference for sugars tend to be found early in the cider process, whereas those that are more acid-tolerant and capable of oxidizing alcohols appear after the yeasts have converted most of the sugars to ethanol. *Zymomonas spp.*, gram-negative bacteria that ferment glucose to ethanol, have been isolated from cider, but they are presumed to be present in low numbers (Jay, 2000).

Molds

Many molds are capable of utilizing alcohols as sources of energy; when these and other simple compounds have been depleted, these organisms proceed to destroy the remaining parts of fruit, such as the structural polysaccharides and rinds (Jay, 2000). Molds also play a role in the spoilage of cider, mostly in unpasteurized cider without preservatives.

Some molds present on apples and in apple cider produce the mycotoxin patulin; these include a large number of penicillia, including *P. claviforme, P. expansum,* and *P. patulum*, as well as some aspergilli and *Byssochlamys nivea* and *B. fulva* (Jay, 2000). The presence of mold, however, does not guarantee the presence of mycotoxins such as patulin, which is produced only under certain conditions (Klaassen, 1996). Production of patulin can occur over a wide range of temperatures (5 to –20°C) with only small amounts produced at 30°C.

Patulin is recognized as a carcinogen and has been found to cause liver damage (Klaassen, 1996). In apple juice, levels of patulin may be as high as 440 ppb and in cider up to 45 ppm. The FDA has recently issued guidelines specifying 50 ppb as the maximum allowable level for patulin (FDA, 2000).

Frequently patulin presence is associated with mold contamination on apples with surface damage. The FDA believes that control by processors of patulin levels can be achieved principally by removing spoiled and visibly damaged apples from the product stream used for the production of apple cider and juice (FDA, 2000). Patulin is reported to be destroyed by fermentation and thus is not found in either alcoholic fruit beverages or vinegars produced from fruit juices (FDA, 2000). However, thermal processing appears to cause only moderate reductions in patulin levels, thus patulin present in apple juice will survive the pasteurization process (WHO IARC, 1990).

Other Pathogens in Cider

Outbreaks of cryptosporidiosis and salmonellosis have been associated with the consumption of unpasteurized apple cider (MMWR, 1997). These documented outbreaks have sparked further research into the survival and destruction of *Salmonella, Cryptosporidium,* and other microorganisms in apple cider.

Salmonella typhimurium has been found to grow in some apple juices, depending on the variety of apple used and the pH of the cider (Goverd et al., 1979). Salmonellae can survive in apple juice for as long as 30 days and can also grow well as low as pH 3.68. However, temperatures below 4°C were not conducive to *S.*

typhimurium growth in the apple juice and actually caused a decline in numbers (Goverd et al., 1979). Uljas and Ingham (1999) found that *S. typhimurium* DT104 decreased by 5 log units in pH 3.3 apple cider that had been frozen and thawed only once. *Salmonella* sp. appears to be more heat-sensitive than *E.coli* O157:H7 and *Listeria monocytogenes* (Mazzota, 2000). Acid adaptation increased the heat resistance of *Salmonella typhimurium* and *Salmonella enteritidis*, but the D-values were consistently lower than the other two microorganisms tested.

Pasteurization of apple cider at 70°C for 10 sec. or longer was demonstrated to cause at least a 4:9-log reduction of *Cryptosporidium* oocysts (Deng et al., 2001). These findings can provide a base when designing a flash pasteurization process sufficient to kill 5 logs of *Cryptosporidium*. Heat treatment would be the most effective way to destroy *Cryptosporidium* as the oocyts are resistant to most common disinfectants such as bleach, iodine, and sodium hydroxide.

Although there has not been a documented outbreak of illness caused by *L. monocytogenes* in apple cider, its prevalence in the environment increases the likelihood of its appearance. Mazzotta (2000) found that *E.coli* O157:H7 had greater heat resistance than *L. monocytogenes*. However, when the data were extrapolated, the kill curve for *E.coli* O157:H7 crossed that of *L. monocytogenes* at 65°C; this may indicate that *L. monocytogenes* is more heat-resistant than *E.coli* O157:H7 at typical juice-processing temperatures. *L. monocytogenes* has been suggested as a target microorganism in apple cider; therefore, more research is needed on its behavior in cider under various conditions.

Fermentation Effects on Cider

Previously, cider was identified as an alcoholic beverage produced by natural fermentation (Martin, 1976). Today, alcoholic ciders are classified into several types, depending on the character and ethanol content of the product (Downing, 1989). Upon complete alcoholic fermentation of fresh apple cider, a noneffervescent fermented product containing 6 to 7% ethanol is obtained. This fermented end product commonly is referred to as hard or dry cider (Semanchek et al., 1996).

Yeasts of significance in the fermentation of cider include weakly fermenting *Hanseniaspora uvarum* and strongly fermenting *Saccharomyces cerevisiae*. In pressed apple juice and in the early stages of fermentation, *Metschnikowia pulcherrima, Saccharomycodes ludwigii*, and *Dekkera sp*. may be present (Deak et al., 1996). Carr (1984) pointed out that *H. uvarum* imparts an off-flavor and *S. ludwigii* causes spoilage of bottled cider. However, under good manufacturing practices, most spoilage organisms can be avoided.

The fermentation of cider greatly changes the flavor, aroma, shelf life, and microflora of the product. The production of alcohol in the cider has deleterious effects on a wide array of microorganisms. The microflora that once flourished in the cider may not adapt well to the ethanol in the hard cider product. Fermentation decreases the ability of salmonellae to survive, partly because of the presence of ethanol and partly because of the changes in the nutritional or physiological conditions brought about by the fermenting yeasts (Goverd et al., 1979).

Semanchek et al. (1996) found that populations of *E.coli* O157:H7 in fermenting cider decreased from an initial load of 6.4 log CFU/ml to undetectable

levels after 2 to 3 days. The cider pH did not change significantly in fermenting cider samples during a 10-day test period; this suggested that factors other than pH (ethanol or other fermentation by-products) were responsible for the total inactivation of *E.coli* O157:H7 in fermenting cider. The authors suggested that alcoholic fermentation (6 to 7% ethanol) of fresh cider is an effective means of destroying *E.coli* O157:H7.

Acid Tolerance of *E.coli* O157:H7

The survival of *E.coli* O157:H7 has been monitored in various products under many conditions. Survival under acidic conditions is of particular interest because of the low pH of apple cider. According to Miller et al. (1993), *E.coli* O157:H7 strains survived better in apple cider than the control *E.coli* strain, which decreased in numbers more rapidly and survived for a shorter period of time.

There are two ways to introduce an organism to an acid: through acid shock or acid adaptation. Generally, induction of acid shock involves the sudden shifting of cells from a neutral or alkaline condition to an acid condition ($pH \leq 5.8$) (Ryu et al., 1998). Cells exposed to fermentation conditions may undergo acid adaptation rather than acid shock, since the pH gradually decreases as fermentation progresses (Ryu et al., 1998). The term acid resistance is generally used to refer to the extended exposure of a microorganism to moderately acid conditions. Conversely, acid tolerance refers to the enhanced survival of a microorganism exposed to pH values between 2.0 and 4.0 after a brief exposure to moderately acidic conditions (Buchanan et al., 1992).

When evaluating a growth study dependent on pH, one needs to look at the acidulant being used and the concentration of undissociated acid at the pH being evaluated. At neutral pH, there is very little undissociated acid (HA), but HA increases logarithmically as the pH declines. Higher concentrations of the undissociated acid generally will have an increased antibacterial effect on the microorganism being studied. However, that may not always be the case; Buchanan et al. (1992) observed that acetic acid was less inhibitory than lactic acid, even though at pH 3.0, the calculated concentration of undissociated acetic acid was almost twice that of lactic acid. The relative activity of different organic acids is also dependent on the presence of acid-specific membrane transport systems (Buchanan et al., 1992).

Conner et al. (1994) screened several *E.coli* O157:H7 strains for growth in various types of acids at various pH levels (4.0, 4.5, 5.0, 5.5, and 7.0) and at different temperatures. Acetic and lactic acids were most inhibitory, while tartaric acid was least inhibitory. Malic acid was also less inhibitory than lactic acid (Buchanan et al., 1999). The growth of *E.coli* O157:H7 was also affected by the temperature, storage time, and the inoculum size (Conner et al., 1994). Buchanan et al. (1992) found that hydrochloric acid was consistently the gentlest acid tested and lactic acid was consistently the most deleterious organic acid when tested at pH 3.0.

Changes in heat tolerance have been observed in *E.coli* O157:H7 strains that have been exposed to acidic environments. At various heating temperatures (52, 54, 56°C), the D-values of acid-adapted cells were significantly higher than D-values

of acid-shocked or control cells (Ryu et al., 1998). Survival of heat-shocked *E.coli* O157:H7 cells in minimal glucose medium at pH 2.5 was about 10-100 times greater than that of untreated cells, depending on the strain (Wang et al., 1998). This study used hydrochloric acid as the acidulant. Other studies employed organic acids, with different results. The thermotolerance of *E.coli* O157:H7 decreased upon short-term storage in apple cider and apple juice at pH 3.4. Changes occurred more quickly at 2°C than at 4°C, suggesting that sublethal injury caused by exposure to acid pH is temperature-dependent (Uljas et al., 1999). These results indicate that the type of acidulant and the procedure used to expose *E.coli* O157:H7 to reduced pH can markedly influence the level of tolerance to subsequent stress conditions.

Three acid resistance systems, i.e. an acid-induced oxidative system, a glutamate-dependent system, and an arginine-dependent system, have been identified in *E.coli* O157:H7 (Lin et al., 1996). Wang et al. (1998) suggested the oxidative system in heat-shocked or acid-adapted *E.coli* O157:H7 cells plays an important role in enhancing acid tolerance. Furthermore, the relative importance of the different acid resistance systems appeared to vary with acidulant (Buchanan et al, 1992). For example, it was observed that the glutamate decarboxylase system was the primary mechanism for survival at pH 4.0 in the presence of benzoic acid, whereas arginine decarboxylase and glutamate decarboxylic systems were both important for resistance to a mixture of fatty acids designed to mimic the environment of the small intestine (Buchanan et al., 1992).

The oxidative or glucose-repressed system is active when cells are growing aerobically or anaerobically in the absence of glucose. The glutamate-dependent

and arginine-dependent systems are active during fermentation. All three systems are active during stationary-phase growth, which suggests the involvement of an alternate, stationary-phase sigma factor S, encoded by rpoS (Price et al., 2000). RpoS is involved in regulating the expression of a variety of stress response genes (Jyhshiun et al., 1996). Price et al. (2000) found that rpoS inactivation selectively prevents the induction of the oxidative system, which indicates that the oxidative or glucose-repressed system is rpoS-dependent. The same effect was not observed on the arginine-and glutamate-dependent systems. Further findings by Price et al. (2000) indicated that rpoS plays a role in *E.coli* O157:H7 shedding in calves, possibly by inducing resistance to gastrointestinal stress, including acid stress offset by the glucose-repressed rpoS-dependent system. Once induced, the acid resistance systems (rpoS-dependent, glutamate-dependent, and arginine-dependent) will persist for at least one month at 4°C (Price et al., 2000).

Temperature Effects on Growth and Survival of *E.coli* O157:H7

Temperature has a significant effect on the growth kinetics of *E.coli* O157:H7. A lower growth temperature (15°C) significantly lengthens the lag phase and slows the growth rate from that observed at 37°C (Duffy et al., 1999).

Refrigeration has been shown to enhance the survival of *E.coli* O157:H7 in acidic foods. *E.coli* O157:H7, when initially present at 10^5 CFU/ml, survived for up to 31 days in one lot of apple cider (pH 3.7) held at 8°C (Zhao et al., 1993). In the same study with cider held at 25°C, survivors were detected at 2 to 3 days but not at 6 days post inoculation. Miller et al. (1993) reported that *E.coli* O157:H7 inoculated

into apple cider with 0.1% sodium benzoate was detectable for 21 days at 4°C, (Buchanan et al., 1998). Dingman (1999) reported that refrigeration (1 to 4°C) and freezing (-20°C) of cider preserved *E.coli* O157:H7 gfp-72ec during a 50-day period. By 64 days, refrigerated cider had exhibited a 1-log decline in CFU, whereas frozen cider exhibited only an approximate 30% drop. These results indicate that *E.coli* O157:H7 can indeed survive in refrigerated cider for the greater portion of its shelf life.

Preservatives and Apple Cider

Preservatives are often used in apple cider to prolong the shelf life of the product. A maximum concentration of 0.1% is mandated for potassium sorbate and sodium benzoate, whose antimicrobial effects are mainly due to the undissociated form of the molecule (Sofos et al., 1980). Benzoic acid is especially suitable for inhibiting yeasts and molds, but is less effective against bacteria. Sorbic acid is a potent inhibitor of the growth of a wide variety of yeasts, molds, and bacteria, with less effect on lactic acid bacteria (Turantas et al., 1999).

The inhibition of bacteria by sorbate appears to be more complex than simple inhibition of "growth" or metabolite production. Sorbate can inhibit spore germination, outgrowth, and vegetative cell division (Sofos et al., 1985).

Inhibition of bacterial growth by weak acid preservatives has been proposed to be due to a number of actions, including membrane disruption, and more recently, inhibition of essential metabolic reactions, stress on intracellular pH homeostasis and the accumulation of toxic anions. In yeasts, it has been proposed that the actual

inhibitory action could be due to the induction of an energetically expensive stress response that attempts to restore homeostasis and results in the reduction of available energy pools for growth and other essential metabolic functions (Brul et al., 1999).

Zhao et al. (1993) found that potassium sorbate did not significantly affect the survival of *E.coli* O157:H7 at 8°C; organisms were detectable in four of six lots of cider at 20 days. *E.coli* O157:H7 survived longer in the presence of 0.1% potassium sorbate than in control samples. In contrast, 0.1% sodium benzoate was inhibitory to *E.coli* O157:H7 at 8°C, reducing the number of organisms to undetectable populations within 7 days in five of six lots of cider.

Dock et al. (2000) showed that the addition of sorbate, benzoate, and malic acid, solely and in combination, significantly decreased the D-values of *E.coli* O157:H7 in apple cider. The largest effect was due to malic acid and benzoate, while sorbate had a lesser but still significant effect. The addition of 0.1% benzoate increased the z-value in cider, which meant that the D-values decreased more slowly as temperature increased. Therefore, increased processing time would be required for a 5D process in cider with benzoate compared to cider without benzoate (Dock et al., 2000).

These various results suggest that one should not confidently rely on preservatives such as potassium sorbate and sodium benzoate to reduce populations of *E.coli* O157:H7. These preservatives are much better suited to increase the shelf life of the product by lowering populations of yeasts and molds.

E.coli and Apples

The quality of the apples used to make cider and their bacterial load play a large role in the quality of the final cider product. Fruits and vegetables can become contaminated with pathogenic microorganisms while growing in fields or orchards, or during harvesting, post-harvest handling, processing, and distribution. *E.coli* O157:H7 in apple cider has been thought to be linked to the use of "drop apples", that is, apples that have fallen from the tree and have contacted animal feces. However, no direct evidence linking the use of dropped apples to fecal contamination of cider has been presented. Cider manufactured using only tree-picked (i.e., obtained directly from the tree) fruit has been found to contain *E. coli* (Dingman, 1999). Potential sources of *E.coli* O157:H7 may be bird droppings and feces of domestic or feral animals (Janiesievicz et al., 1998). Other possible sources of contamination may be the storage environment of the apples (outside or warehouse) and pest control measures.

Maule (cited in Kay et al., 1997) found that *E.coli* O157:H7 could survive for long periods both in cattle feces and in soil. With an *E.coli* O157:H7 inoculum level of 10^7, the organism was reduced by only 2 logs in cattle feces after 54 days and by only 1 log in soil after 63 days. One could assume from these results that once the organism is present in the apple orchard, it may remain viable for several months. The environmental survival of *E.coli* O157:H7 is particularly important because of its extremely low infective dose (100 cells) required to cause disease (Salyers et al., 1994).

E.coli survival and penetration into apples has been evaluated. Wounded or punctured apples are often used in apple cider. Wounded apple tissue has been found to be an excellent substrate for the growth of *E.coli*. *E.coli* O157:H7 inoculated onto wounded apple tissue at a level of 10^4 CFU/ml increased by more than 3 logs within 48 hours (Janisievicz et al., 1998). Similarly, Dingman (1999) found that all tested cultivars (McIntosh, Red Delicious, Macoun, Melrose, and Golden Delicious) promoted growth of *E. coli* O157:H7 in damaged apple tissue independent of the apple source (i.e., harvested as tree-picked or dropped fruit). Bruised apple tissue had a significantly higher pH and significantly lower Brix value than undamaged apple tissue, which may have played a role in the growth of the bacteria. Also, the tissue pH of Red Delicious apples was significantly higher than those of the other cultivars, independent of the apple source or whether the tissue was undamaged or bruised (Dingman, 1999).

Janisievicz et al. (1998) analyzed the ability of fruit flies to spread *E.coli* O157:H7. The high frequency of external and internal contamination of fruit flies during relatively short periods (24-48 h) of exposure to an *E.coli* source, and the high incidence of contamination of apple wounds with *E.coli* by these flies, demonstrated the potential for fruit flies to transmit *E.coli* to apples (Janisievicz et al., 1998). This information is significant, given the abundant levels of fruit flies present during apple cider processing. Houseflies have also been found to harbor 100 different pathogens and have been shown to transmit 65 of these pathogens (Kettle, 1982).

The uptake of *E.coli* O157:H7 into different regions of the apple (skin, outer core, inner core, and pulp) varies. The greatest contamination on a per gram basis

was consistently in the outer core region (Buchanan et al., 1998). Concentration of pathogens in this region of the apple has serious implications for sanitization treatments. It is very difficult to displace any fluid trapped in the core cavity, therefore making it hard to rinse thoroughly with water (Buchanan et al., 1998).

Temperature differentials between apples and external washes affect uptake by the apples, as demonstrated with a model system using a marker dye (Buchanan et al., 1998). When cold apples (4°C) were immersed in a warm (21°C) dye solution, no uptake of the dye into the inner core region was observed. However, when warm apples (22°C) were submerged in a cold water (9°C) dye solution, approximately 6% of the apples had substantial accumulation of the dye in the inner core region. During cider making, apples are sometimes submerged in a dump tank of chlorinated water. If this tank is not properly maintained, it can be a major source of contaminants. Results of the Buchanan study suggest that the apples coming into the dump tank should be colder than the water, to prevent infiltration of microorganisms into the apple.

Chemical Treatments of Apples

Fruits commonly contain populations of 10^4 to 10^6 microorganisms/g when they arrive at the packing house or processing plant (Brackett, 1994). Only a 1-log reduction has been observed when washing fruits in water alone (Beuchat, 1992).

The use of chlorine in wash water may reduce microbial populations by an additional log (Annous et al., 2001). Only about a 1.4-log reduction was seen on tomato surfaces dipped in 320 ug/ml chlorine solution (Beuchat et al., 1995).

Apples washed on a flatbed washer, instead of in a water dip, did not show a decrease in bacterial populations, even when antimicrobial agents were used (Annous et al., 2001). The authors attributed this to the short exposure time and ineffective brushing of the washer. Although chlorine cannot be relied upon as a disinfectant for fresh produce, chlorination of wash water reduces the likelihood of contaminated produce reaching the consumer. Depending on the fruit or vegetable, 200 to 300 ug/ml of chlorine is recommended as a sanitizer in wash water.

Other means of reduction of *E.coli* O157:H7 on fruits and vegetables through chemical treatments have been studied to provide alternatives to pasteurization. Wisniewsky et al. (2000) found that peroxyacetic acid, chlorine dioxide, and a chlorine/phosphate buffer solution were not very effective in achieving a 5-log reduction of *E.coli* O157:H7 inoculated at 10^8 and 10^6 cells per apple. These sanitizers would need to be used at high concentrations for at least 15 minutes to achieve the 5-log reduction; this would be impractical for use in production (Wisniewsky et al., 2000).

Similar results were obtained in a study of the removal of *Salmonella chester* from apples. Hydrogen peroxide treatment reduced the number of *Salmonella chester* on the apple skin by 3 to 4 logs/fruit, and on the stem and calyx by 1 to 2 logs/fruit; this gave an overall reduction of only 2 logs/fruit (Liao et al., 2000). The authors suggested that failure of sanitizers to completely inactivate *Salmonella chester* on apples was likely due to the firm attachment of bacteria on stem and calyx, where they were either resistant to or protected from the sanitizer treatment.

E.coli-inoculated Golden Delicious apple halves washed with 0.5% hydrogen peroxide experienced population reductions generally in the range of 3 - 4 logs; however, a residual H_2O_2 level of 1000 ppm was found on the apples immediately after washing. This could be lowered to <20 ppm if apples were immediately rinsed with water (Sapers et al., 1999). In the same study, 200 ppm chlorine solution was found to reduce *E.coli* numbers by 2 logs on apple halves. The use of apple halves represents the "worst case" scenario because of absorption of the microorganism into the tissue. Trials performed on whole apples showed lower adherence of *E.coli* to the apples, with less than a 1-log reduction upon exposure to various sanitizers (Sapers et al., 1999).

Similar results for chemical sanitizers have been found in other food products. Various sanitizers (NaOCl, H_2O_2, Tsunami, Vortexx, Vegi-Clean, ethanol, and chlorine) all proved to be ineffective at eliminating *E.coli* O157:H7 from alfalfa seeds initially containing 2.0 to 3.2 $\log_{10}$ CFU/g of the pathogen (Taormina et al., 1998). The inability of the test chemicals to eliminate *E.coli* O157:H7 from alfalfa seeds was attributed more to the nature of the seeds than to the efficacy of the chemicals tested or the durability of the pathogen. Cells of *E.coli* O157:H7 were thought to be protected in cracks and crevices in the seeds (Taormina et al., 1998).

Heat may also be used to reduce microorganisms on the surface of fruits and vegetables. However, the type of fruit plays a great role in the microbial reduction efficacy of hot water immersion. Pao et al. (2000) found that fresh juice extracted from oranges that were immersed in hot water at 80°C for 1 or 2 min. had >5.4 log

reduction of the inoculated *E.coli.* In contrast, the *E.coli* level detected in the juice of homogenized fruit without hot water immersion was over 5 $\log_{10}$ CFU/ml.

Fleischman et al. (2001) used two inoculation methods to determine the efficacy of hot water immersion in removing organisms from apples. Surface-inoculated apples immersed at 80 or 95°C experienced a significant reduction of *E.coli* contamination, with a 6-to 7-log drop occurring in the first 30 seconds. However, apples that had been inoculated by submersion experienced a much lower reduction of *E.coli*: an approximate 2-log decrease at 95°C. These results suggest that the organism was protected from surface temperature through internalization into the apple (Fleischman et al., 2001). Hot water immersion could be useful in reducing large numbers of microorganisms on the surface of apples, but would not be effective for internalized organisms.

HACCP

The Pillsbury Company developed the Hazard Analysis Critical Control Point (HACCP) system as a management system focused on prevention of problems to assure the production of food products that are safe to consume. The HACCP concept covers biological, chemical, and physical hazards that can naturally occur in the food, contributed by the environment or generated by a mistake in the manufacturing process. Several prerequisite programs such as good manufacturing practices (GMPs) and standard operating procedures (SOPs) are the foundation to the successful implementation of a HACCP program (Stevenson et al., 1999).

Because the FDA will require apple cider producers to incorporate a HACCP plan, the use of HACCP within the apple cider industry is currently under investigation. To study microbiological control, an indicator organism is usually chosen to serve as a useful verification tool for proper sanitation and for the HACCP plan itself. Lang et al. (1999) chose *E.coli* as the most useful indicator organism over coliforms and enterococci in apple cider. That conclusion was based on the exclusively fecal origin of *E.coli*, the generally good survival of this species in refrigerated apple cider compared to other coliforms and enterococci, the association of *E.coli* with drop apples, and the greater selectivity of available *E.coli* testing methods compared to those for coliforms. The FDA has concluded that target pathogens must be chosen on the basis of historical association with a product and the way in which the product is processed. For example, there have been apple juice outbreaks associated with *E.coli* O157:H7, *Salmonella spp.*, and *Cryptosporidium parvum*. The NACMCF recommended the use of *E.coli* O157:H7 or *L. monocytogenes* as the target organism, as appropriate. This recommendation is based on the number of known outbreaks of *E.coli* O157:H7 in juice and the ubiquitous nature of *L. monocytogenes*. The FDA plans to provide additional information in its juice HACCP hazards and controls guidance to assist producers in identifying the pertinent microorganism for measuring the 5-log standard (FDA, 21 CFR Part 120, 2001).

Differences in apple cider processing methods mean that production control steps may also differ. A HACCP plan makes the processor understand the process and any hazards that could be introduced in that process. Possible production

control steps are: pasteurization, exclusion of drops apples, chlorine soak or spray for apples, and temperature control of cider (Senkel et al., 1999).

A microbiological and production practice survey of cider producers in Maryland was conducted to determine the effect of HACCP on bacterial levels in the cider (Senkel et al., 1999). Results from routine microbiological analyses completed between 1993 and 1996 were compared with samples collected during the 1997-1998 season, which was the season that HACCP was incorporated. There was a highly significant difference in the number of bottled cider samples that were positive for generic *E.coli* between the two seasons studied, with a trend toward reduced bacterial levels during the 1997-1998 season. These results suggest improvement in the microbiological quality of fresh cider when HACCP was incorporated. Thus, improvements in sanitation, apple treatments, and the procedures required by the model HACCP plans appeared to reduce the bacterial levels and the likelihood of fecal contamination (Senkel et al., 1999).

The FDA conducted a survey of 237 apple cider-processing facilities in 32 states during the 1997 season (FDA, 1999). The survey looked at harvesting and processing practices as well as the microbiological quality of the product. The FDA found that most processors (59%) did not use drop apples to make cider. Sixty percent of the producers were found to wash apples upon receipt and 84% washed apples immediately before pressing. An additional 21% of the processors sanitized the apples with chlorine before pressing. Seventeen percent of the firms used untested well water as their source of water for processing (FDA, 1999).

A study of *E.coli* incidence in cider was useful in identifying possible factors that might have contributed to the 1996 *E.coli* outbreak associated with cider in Connecticut. A strong point of focus was the use of drop apples. Dingman (1999) found that 64% of the mills used drop apples in addition to tree-picked apples. Of the 314 cider samples tested, *E.coli* was found to be present in 4% of the samples tested in the 1999 season. However, during the peak level of drop apple usage, no *E.coli* were found in the cider. *E.coli* was also isolated from cider samples in which only tree-picked fruit was used. These observations suggest that factors apart from the use of drop apples may contribute to *E.coli* contamination of cider. Dingman (1999) suggested that apple storage conditions, the length of storage, and the quality of the fruit being used contribute to the contamination of cider.

Preliminary research at the FDA Apple Cider pilot plant in Placerville, CA, has confirmed the importance of sanitation as one component of an overall safety strategy in the production of quality apple juices and ciders (Keller et al., 1999). Juice was produced with little or no clean-up or sanitation of the equipment or facilities, and the apples used were not all of good quality with no culling performed. Typical juices made under poor conditions with poor quality fruit resulted in aerobic microbial populations of over 5 logs per ml, despite incoming apples having only 3 logs per gram (Keller et al., 1999).

Senkel et al. (2000) found generic, nonpathogenic *E.coli* in 13% of in-line apple samples and 18% of cider samples, but not on the exterior of incoming apples. This suggests that *E.coli* was introduced during in-plant processing and highlights the importance of appropriate sanitation practices in juice production.

Pasteurization alone does not assure the safety of juice products. Proper handling of the product after pasteurization is required to prevent post-process contamination (FDA, 2001). Many producers bottle cider by hand or add flavorings after pasteurization of cider. These could be potential contamination points of pasteurized cider if care is not taken to ensure the bottling procedure is performed so that post-process contamination does not occur. This again emphasizes the importance of sanitation during the entire process.

MATERIALS AND METHODS

Iowa Cider Survey

Twenty-four yes/no or short-answer questions pertaining to apple cider production practices were compiled into a questionnaire that was mailed to all Iowa certified apple cider producers (those who had attended a series of seminars on cider production and food safety). The cider producers were asked to return the questionnaires within 60 days in a provided pre-addressed envelope. The questionnaires were completely anonymous; individual producers could not be identified. The questionnaire can be found in the appendix.

Cider Facility Survey

Four cider producers, two that pasteurized cider and two that did not, participated in an in-depth study over the course of two cider seasons (1999-2000 and 2000-2001). In the second year of the study, one producer of pasteurized cider dropped out of the study, and was replaced by another pasteurizer. In initial visits to the participating facilities, numerous questions were asked about production practices while a tour of the facility was made. Samples were taken of apples, cider, and the processing environment during the initial visit and three additional visits made over the course of the cider season.

Apples were taken from containers recently brought from the orchard and stored inside or outside, from refrigerated storage before washing, from refrigerated storage after washing, and from the conveyor belt on the processing equipment.

Apples were placed in sterile stomacher bags (Fisher Scientific Co., Itasca, IL) and kept at 7°C until testing (2-24 h).

Cider samples were taken from various points during the process: before preservatives were added, after preservatives were added, and after pasteurization. Samples (1000 ml) were collected in sterile glass containers and held at 7°C until testing (2-24 h). Occasionally producers were asked to save cider samples for pick-up the following day. These samples were in clean ½ - or 1 – gallon containers used for retail sales and were held at refrigerated temperatures.

Environmental samples were taken using sterile cotton swabs that were wetted with sterile 0.1% peptone (Difco Laboratories, Detroit, MI) diluent. Either 10- or 100–cm^2 areas were sampled on the equipment, including conveyor belts, press cloth, stirring paddle, cider filters, holding tanks, and inlet and outlet lines. Samples were taken either after cleaning or prior to start-up. Water samples were taken from the faucet and/or hoses in the processing room and collected in sterile test tubes.

During sampling, cider temperature before and after pasteurization was measured with a thermometer (Comark, range -40 to 300°F) that was dipped in 95% ethanol prior to coming in contact with the cider. Temperature readings were also recorded from the pasteurizing unit, the thermometers on the cider holding tanks, and any refrigerated storage areas.

Media and Sample Preparation

All dilutions were made in sterile 0.1% peptone. To recover microorganisms from the apple surface, 100 ml diluent was added to each bag containing an apple.

The bag was shaken vigorously for 2 min., the apple was removed, and the diluent was serially diluted and plated in duplicate on appropriate media. Environmental swabs were placed in 10 ml diluent in test tubes and shaken vigorously before dilution and plating. Cider was added directly to the diluent.

An aerobic spread plate count was performed on Tryptic Soy Agar (TSA, Difco). Colonies were counted after both a 24-and 48-h incubation at 32°C. The 48-h incubation period was incorporated because of the harsh environment of apple cider and the possibility of resuscitating microorganisms over a longer growth period. Counts increased by about 5% between 24 and 48 h. All counts reported here are from 48 h.

Yeasts and molds were enumerated on Potato Dextrose Agar (PDA, Difco), acidified to pH 3.5 with tartaric acid. Plates were incubated at 25°C for 5-7 days. Presumed yeast colonies were randomly selected for microscopic examination in a gram stain to distinguish them from bacteria.

Coliforms were counted on Violet Red Bile Agar (VRB, Difco) after 24-h incubation at 35°C for samples taken in 1999-2000. The following season (2000-2001), E.coli/Coliform Petrifilm (3M, St. Paul, MN) was used instead of VRB for the enumeration of coliforms and *E.coli*.

AOAC methods were followed to count coliforms and *E.coli* on Petrifilm. Red colonies with an adjacent bubble of gas after 24 h at 35°C were counted as coliforms; blue colonies with an adjacent gas bubble after 48 h at 35°C were counted as *E.coli*. Colonies that did not show gas formation were also enumerated but not

included in the overall coliform count. The Petrifilm was stored at 7°C until use; any opened Petrifilm packages were sealed and held at room temperature as directed by 3M.

Counting procedures followed standard methods (Gerhardt *et al.*, 1994). Representative colonies that were commonly observed on the different media were purified by streaking onto TSA, gram stained, and frozen at –80°C in Tryptic Soy Broth (TSB, BBL, Cockeysville, MD) with 20% glycerol for further study.

Storage Study

Cider samples were stored at 7°C and sampled at 2, 4, 6 and 8 weeks of storage. Microbial growth was detected by observation of gas or off-odor production, and by plate counts as described above.

Identification of Microorganisms

Biochemical tests were performed to identify isolated organisms. The BBL DrySlide indole and oxidase slides were utilized according to the manufacturer's instructions. Voges-Proskauer reagent droppers (BBL) were used according to the standard method. Catalase, citrate (Difco), and methyl red reactions were performed according to standard methods (Gerhardt *et al.*, 1994).

The BBL crystal kit for enteric non-fermenters (E/NF) was used to identify some isolates. The 30-reaction panel was read by means of a color reaction chart; a number value was obtained for each isolate and entered into the BBL Crystal ID

System Electronic Codebook, which assigned a probable identification. All procedures were followed according to the Becton Dickinson instruction manual.

Cider Inoculation Study

Microorganisms

Two strains of *E. coli* O157:H7 obtained from Dr. Pina Fratamico, Agricultural Research Service, Wyndmoor, PA, were used in this study: strain B6-914 90ec that contains a green fluorescent protein gene (*gfp*) and does not produce Shiga-like toxins I and II (Stx1 and Stx 2), and pathogenic strain SEA 13B88 *gfp* 73ec, which was isolated from a cider outbreak and also contains the *gfp* gene.

Representative organisms isolated from samples in the current study were also used: *Klebsiella spp.* and *Enterobacter spp.*, isolated as blue colonies on *E.coli*/coliform Petrifilm from a raw cider sample and from an apple, respectively. Two yeasts strains were also used; these were isolated from two different ciders that had been stored approximately 7 weeks.

To check whether the bacteria could be easily distinguished from the yeasts by colony morphology, all strains were plated on TSA and incubated for 24-48 h. Yeast colonies were easily differentiated from the bacteria by their small size and color. Fluorescent *E.coli* strains were observed under an ultraviolet lamp (UVS-12, Ultra-violet Products, Inc, San Gabriel, CA) to check for purity. Gram stains were also performed throughout the study to ensure that colonies were of the same morphology as the original colonies.

Permanent cultures were stored at -80ºC in 5 ml TSB containing 20% glycerol. Working bacterial cultures were streaked weekly onto TSA, incubated at 35ºC for 24 h, and stored at 4ºC. Working yeast cultures were streaked weekly onto PDA and stored at room temperature.

Inoculation

Individual bacterial cultures were grown at 35°C for 24 h in 10 ml of TSB without agitation, and harvested by centrifugation for 10 min at 8000 x *g*. The cell pellet was resuspended twice in 10 ml of sterile diluent. Cells were inoculated at about 5.6×10^6 or 3.4×10^3 per ml of sterile (autoclaved at 121°C for 15 min) cider. Cider sample sizes were 200 ml. A single lot of cider containing 0.1% potassium sorbate was used for the entire study. The pH of the cider was taken before inoculation and at regular intervals during the study.

Four conditions were used: incubation at room temperature or at 4°C, and with or without concurrent inoculation with yeasts at about 4.0×10^3/ml. The yeasts were pooled together to create a mixture of yeasts with each strain present at about the same concentration (2.0×10^3/ml). Characteristics of individual bacterial strains were of interest; therefore, cultures were not mixed. A plate count on TSA (48 h incubation at 35°C) was performed immediately after inoculation to determine the initial bacterial count. Duplicate or triplicate counts were performed for each set of conditions.

Plate counts were performed every day for the first 5 days and every other day for the next 5 days. If bacteria were still present, samples were plated every 2-3

days thereafter until no colonies were detected. After bacterial levels were below detection, three plate counts were taken the following consecutive 3 days to ensure that all resuscitated cells were recorded.

Statistical Analysis

Tukey's multiple comparison procedure was performed using the SAS statistical analysis system (SAS Institute, Inc., Cary, N.C.). The nonparametric Wilcoxon rank sum test was performed according to methods described in An Introduction to Statistical Methods and Data Analysis (Ott, 1993).

RESULTS AND DISCUSSION

Cider Survey

According to the Farm Fresh 2000 booklet (2000), there were 21 certified apple cider producers in the state of Iowa in 2000. The five producers participating in the in-depth study were interviewed directly. Questionnaires were mailed to the other 16 producers; eight surveys were returned. Two of these producers noted that they had quit processing cider in the past year. The results summarized below thus come from 11 producers.

The questionnaire had two sections, one referring to harvesting and the other to processing practices. Responses are summarized in tables 1 and 2.

Table 1. Harvesting Practices of Iowa Apple Cider Producers

Harvest Practice	Number of Yes Responses	%	Number of No Responses	%
Manure fertilizer used in orchard	0	0	11	100
Deer fence around orchard	1	9	10	91
Drop apples used to make cider	2	18	9	82
Apples from another supplier used to make Cider	5	45	6	55

The first three practices related to the possibility of apple and cider contamination by fecal microorganisms of animal origin. While manure is not used as a fertilizer, deer and other animals do have access to most orchards. Drop apples should not be used for cider, although this was a traditional use for such

apples. The number of producers in Iowa using drop apples is slightly less than the 37% found to be using drops in a 1997 FDA inspection of cider producers around the country (FDA, 1999). However, many producers obtain their apples from another source; harvesting practices of these other apple suppliers are unknown.

Table 2. Processing Practices in Iowa Apple Cider Producers

Processing Method	Number of Yes Responses	%	Number of No Responses	%
Apples are stored in cooler	10	91	1	9
Processing water is chlorinated	5	45	6	55
Apples are washed and brushed	10	91	1	9
Apples are sanitized (chlorine dip, spray, etc)	6	55	5	45
Cider is pasteurized	8	73	3	27
Cider is filtered	9	82	2	18
Bottling is performed by hand	10	91	1	9

Given the rural location of most processors, many used well water to process cider; this was not tested regularly for microorganisms or for chlorine content. Since 91% of the producers washed apples, the water quality becomes a key factor in the microbiological quality of the process. Over half of the producers disposed of the pomace by hand, mostly by wheelbarrow or shovel. This pomace was dumped back onto the orchard fields or fed to animals. Apples were almost always stored inside a cooler at temperatures ranging from 36 to 46°F. As noted, most of the processors

do pasteurize, however temperatures and times of pasteurization ranged from 160 to 179°F for 2 to 20 seconds. Before bottling, cider was allowed to settle for various amounts of time from 3 hours to more than a week. During this settling, cider was stored in a large bulk tank at temperature ranges of 32 to 68°F. Each producer used a slightly different storage temperature that fluctuated very little throughout the seasons studied.

Cider Facility Survey

Results of the in-depth study of five facilities are given separately for the two years of the study. Microbial counts on apples, in the processing environment, and in cider are reported for each sampling time. Producers A, B, and E pasteurized their cider while producers C and D did not. All added 0.1% potassium sorbate as a preservative. In addition to potassium sorbate, Producer B also used 0.1% sodium benzoate.

Sampling visit results are reported in the order in which they were taken (early season to late season). The number of visits made varied with each producer and depended on communication from the producers about when they were making cider. Data for three producers were obtained for both years of the study. Producer B was in the study only in the first year. Producer E was picked as a replacement in the second year. Therefore, data from only one year are available for these two producers.

Initially an *Enterobacteriacae* count was planned for the study using Violet Red Bile Agar with glucose (VRBGA). Because a large number of microorganisms

(not all of which were *Enterobacteriaceae*) grew on this agar, the more selective VRB agar (without glucose) was used for coliform detection in the first year of the study. In the second year, *E.coli*/coliform Petrifilm was used instead of VRB agar because it was easy to use, gave quicker results than standard plating procedures, and could also be used by the processors as a method of verifying the safety of their product. Silk et al (1997) found there was no significant ($P>0.05$) difference when comparing mean coliform CFU obtained on Petrifilm and VRB agar.

The limit of detection for aerobic bacteria and yeasts and molds was 100 CFU/apple and 10 CFU/ml of cider. The limit of detection for coliforms on apples and in cider was 100 CFU/apple and 1 CFU/ml on VRB, respectively. The low pH of cider affected the indicators on the Petrifilm; cider was diluted 1:10 before plating so that correct color reactions for coliform and *E.coli* colonies could be seen. This dilution raised the coliforms and *E.coli* detection limit with Petrifilm to 10 CFU/ml. Data in Figures 1, 2, 7, and 8 reflect these detection limits.

Microbial Contamination of Apples

Four cider outbreaks caused by *E.coli* O157:H7 and cider outbreaks caused by *Cryptosporidium* and *Salmonella* have all been attributed to the fruit; therefore, it was necessary to evaluate the microbial loads of incoming apples (MMWR, 1975, 1996, and 1997, Besser, et al., 1993). Results of the microbial tests performed on apples can be found in Figures 1 and 2. The apple samples were taken as they were going into the grinder and in most cases were already washed, brushed, and/or waxed.

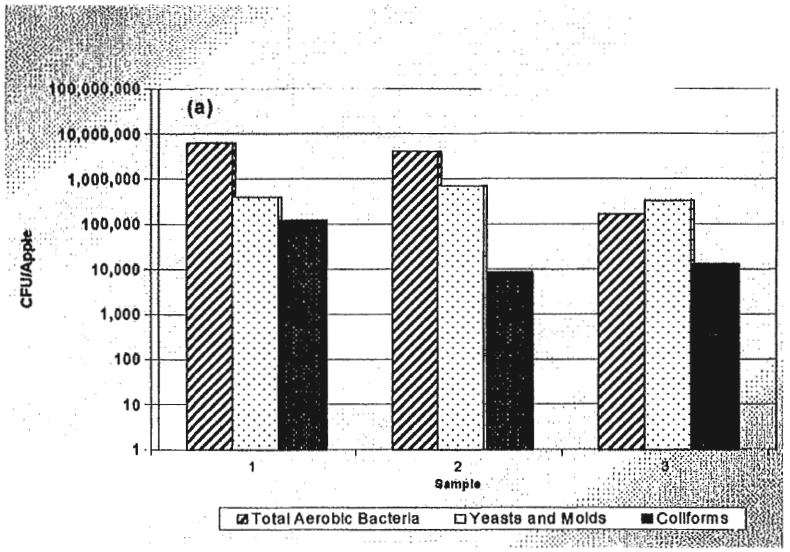
FIGURE 1a. Producer A - Microbial counts on apples during the 1999-2000 season.

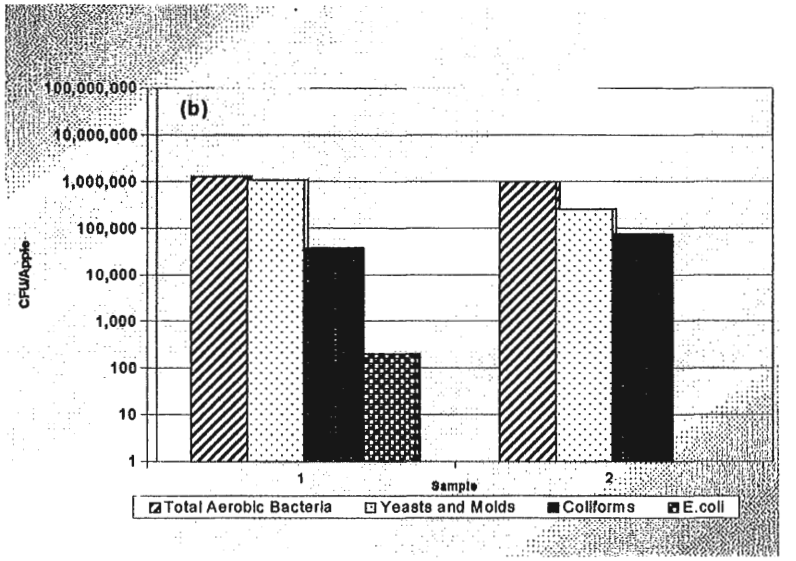
FIGURE 1b. Producer B - Microbial counts on apples during the 1999-2000 season.

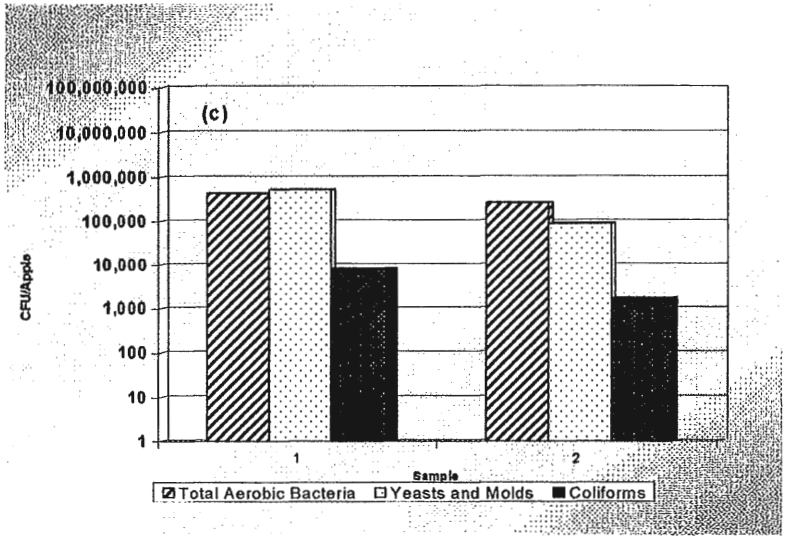
FIGURE 1c. Producer C - Microbial counts on apples during the 1999-2000 season.

FIGURE 1d. Producer D - Microbial counts on apples during the 1999-2000 season.

The limit of detection for all microbial tests was 100 CFU/apple. *E.coli* is noted only if it was found in the sample.







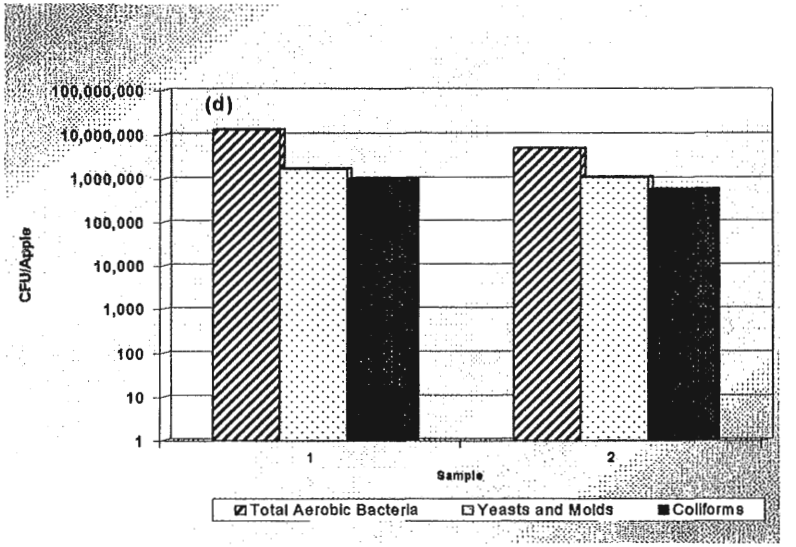


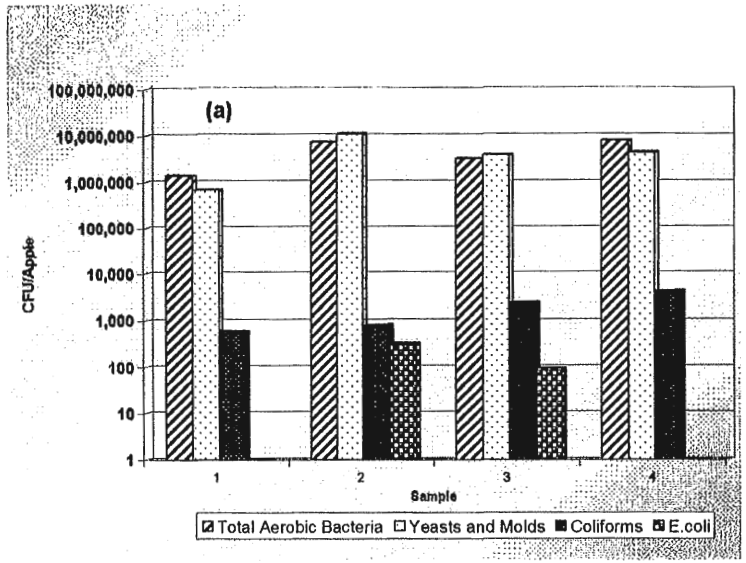
FIGURE 2a. Producer A - Microbial counts on apples during the 2000-2001 season.

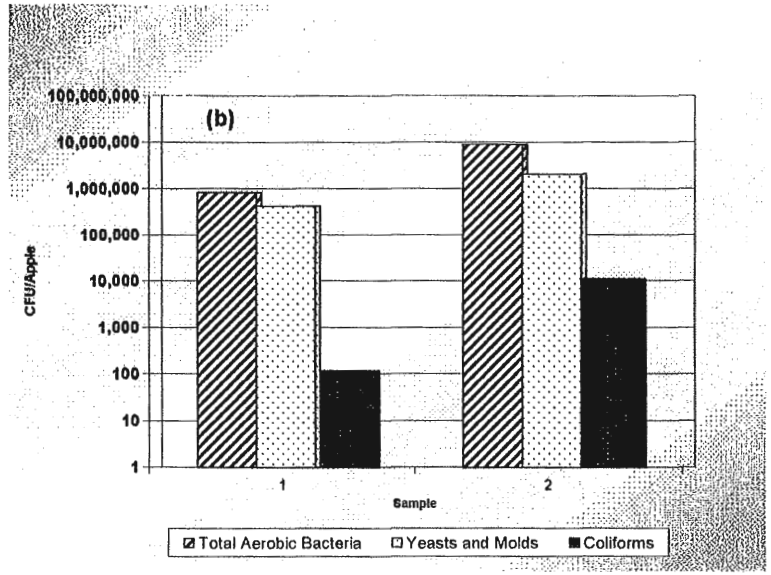
FIGURE 2b. Producer C - Microbial counts on apples during the 2000-2001 season.

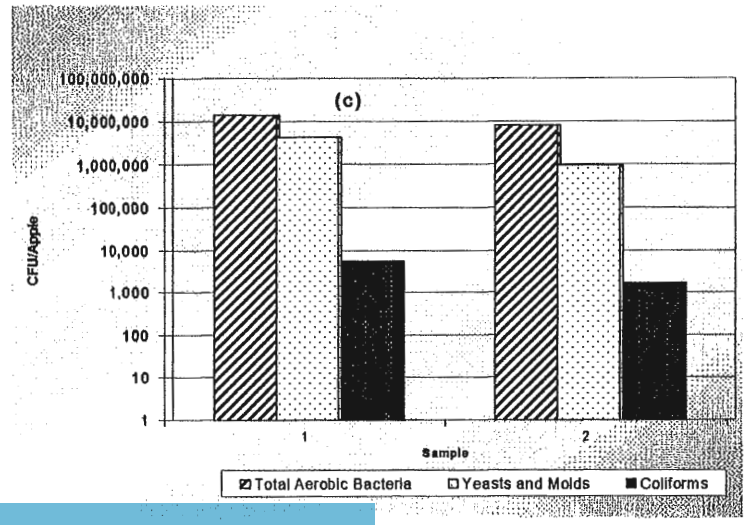
FIGURE 2c. Producer D - Microbial counts on apples during the 2000-2001 season.

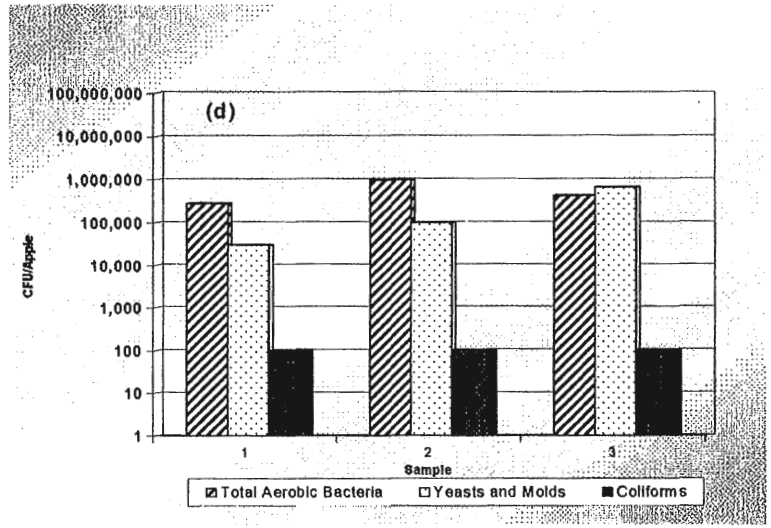
FIGURE 2d. Producer E - Microbial counts on apples during the 2000-2001 season.

The limit of detection for all microbial tests was 100 CFU/apple. *E.coli* is noted only if it was found in the sample.









Average counts of yeasts and molds were similar for the two years of the study, with levels slightly higher in the second year. Average yeast and mold counts were usually between 3.0×10^4 and 1.0×10^6 CFU/apple; the highest average was 1.1×10^7 CFU/apple. Yeasts dominated (85%) over molds on a per-apple basis. The high yeasts counts observed in the current study are similar to the 10^2 to 10^6 CFU/apple ranges Deak et al. (1996) found. Since yeast populations tend to peak in the fall, high levels were expected.

Aerobic bacterial counts on apples were also similar between the two seasons. Aerobic bacteria loads on apples were generally between 2.6×10^5 and 8.0×10^6 CFU/apple; the average high was 1.4×10^7 CFU/apple. The apples tested in the present study were comparable to the average aerobic bacterial count of 4.6×10^4 CFU/g of apple that Senkel et al. (1999) found in their study of Maryland cider producers.

Coliform counts on apples varied with producers and were found to be lower in the second year of the study. Ranges of coliforms on apples were from <100 to 1.0×10^6 CFU/apple. While producer E had no detectable coliforms on apples, producer D had loads of up to 10^6 CFU/apple. Producer E waxed the apples, which may have played a role in the lower levels of microorganisms found. Typical *E.coli* were detected in 8.6% of the apple samples tested. Upon further testing these were determined not to be *E.coli* O157:H7.

The large variability in microbial counts on apples might be attributed to differences in the weather, source of apples, processing methods, and storage conditions. Since the apple harvest generally occurred from late August through

October, apples sampled early in the cider season were freshly picked while those sampled late in the season would have been stored for several weeks.

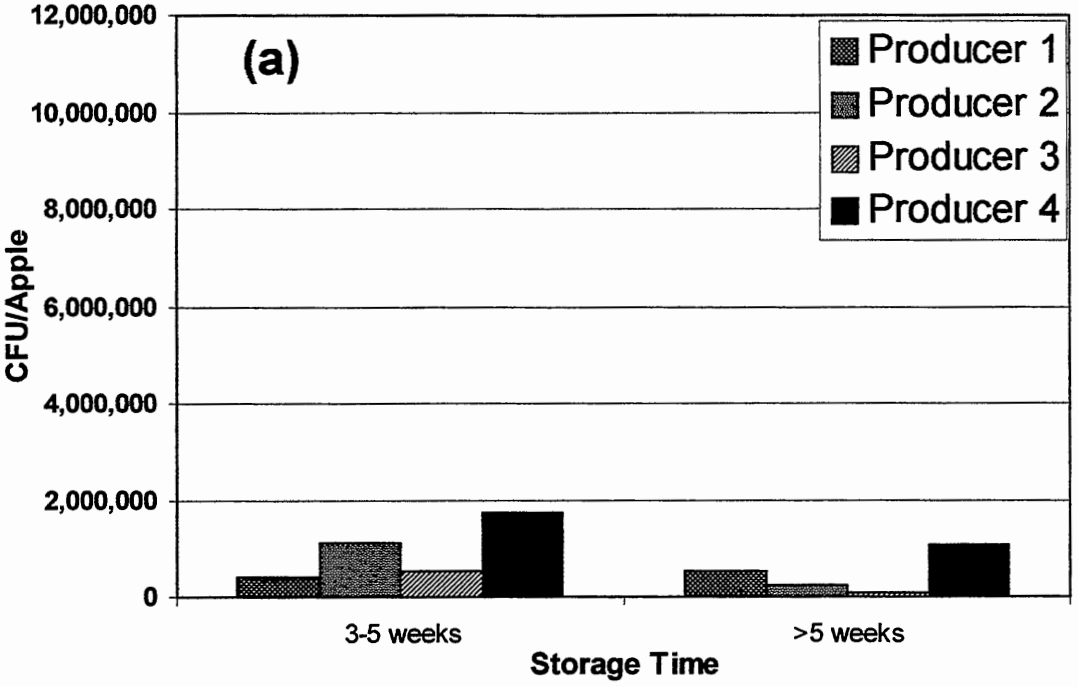
Figures 3, 4, and 5 show the microbial variations in apples during storage. Apples were compared at different storage times to evaluate any microbial changes that might occur on the apples. In most cases, counts were highest on apples stored for 3-4 wks and declined thereafter. However, when counts on stored apples were analyzed by Tukey's method using the SAS statistical analysis system (SAS Institute, Inc., Cary, N.C.), storage time did not significantly affect the level of microorganisms found on the apples ($P > 0.05$). Producers were routinely removing rotten and damaged apples during storage; this systematic culling probably was responsible for counts remaining fairly consistent. Fisher and Golden (1998) found that *E.coli* O157:H7 survived on all tested apple cultivars for 18 days at refrigeration temperatures before visible mold spoilage occurred. Since *E.coli* O157:H7 can survive on apples during a significant portion of their storage, it is essential to incorporate an effective method of sorting apples to prevent further contamination of apples and growth of already present microorganisms.

Various processing methods can play a role in contributing to or lowering microorganisms on apples. Most processors washed and/or brushed their apples immediately upon harvesting, so unwashed apples were not generally available for sampling. However, producer A washed/brushed apples immediately prior to processing; hence the efficacy of this processing step could be evaluated.

FIGURE 3A. Changes in yeasts and molds during storage of apples for the 1999-2000 season.

FIGURE 3B. Changes in yeasts and molds during storage of apples for the 2000-2001 season.

Apples were chosen at different points during storage and were tested for yeasts and molds. When apples were not obtained during a specific storage period, no data are presented for those points.


(a)
Producer 1
Producer 2
Producer 3
Producer 4
12,000,000
10,000,000
8,000,000
6,000,000
4,000,000
2,000,000
0
CFU/Apple
3-5 weeks
>5 weeks
Storage Time

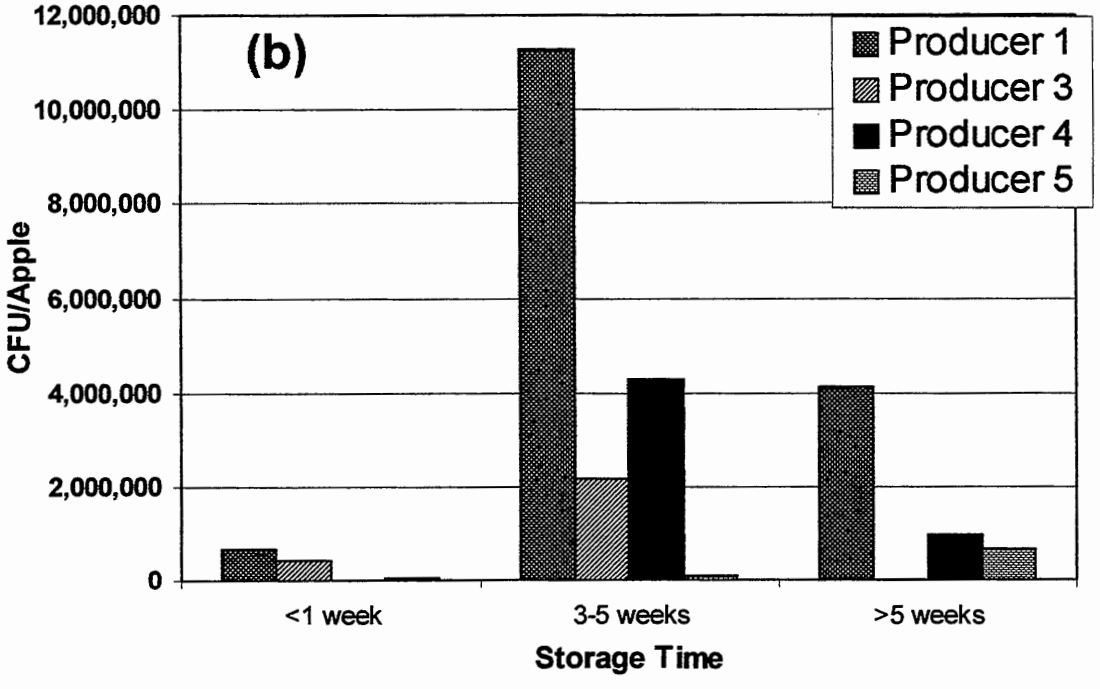
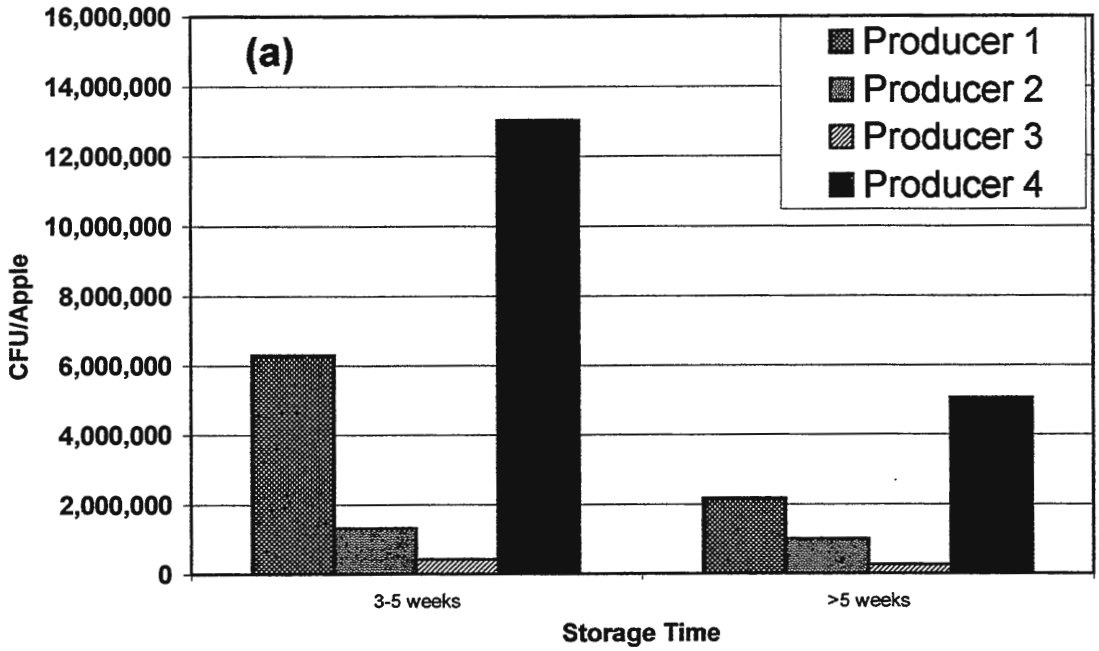

(b)
Producer 1
Producer 3
Producer 4
Producer 5
12,000,000
10,000,000
8,000,000
6,000,000
4,000,000
2,000,000
0
CFU/Apple
<1 week
3-5 weeks
>5 weeks
Storage Time

FIGURE 4A. Changes in aerobic bacteria during storage of apples for the 1999-2000 season.

FIGURE 4B. Changes in aerobic bacteria during storage of apples for the 2000-2001 season.

Apples were chosen at different points during storage and were tested for total aerobic bacteria. When apples were not obtained during a specific storage period, no data are presented for those points.



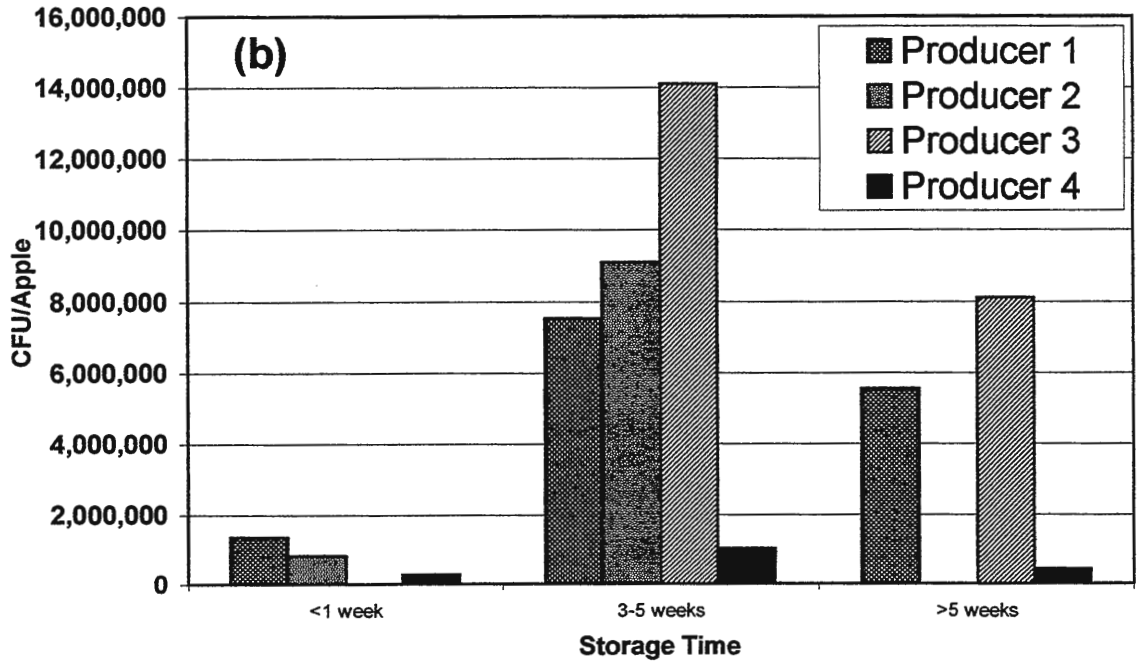
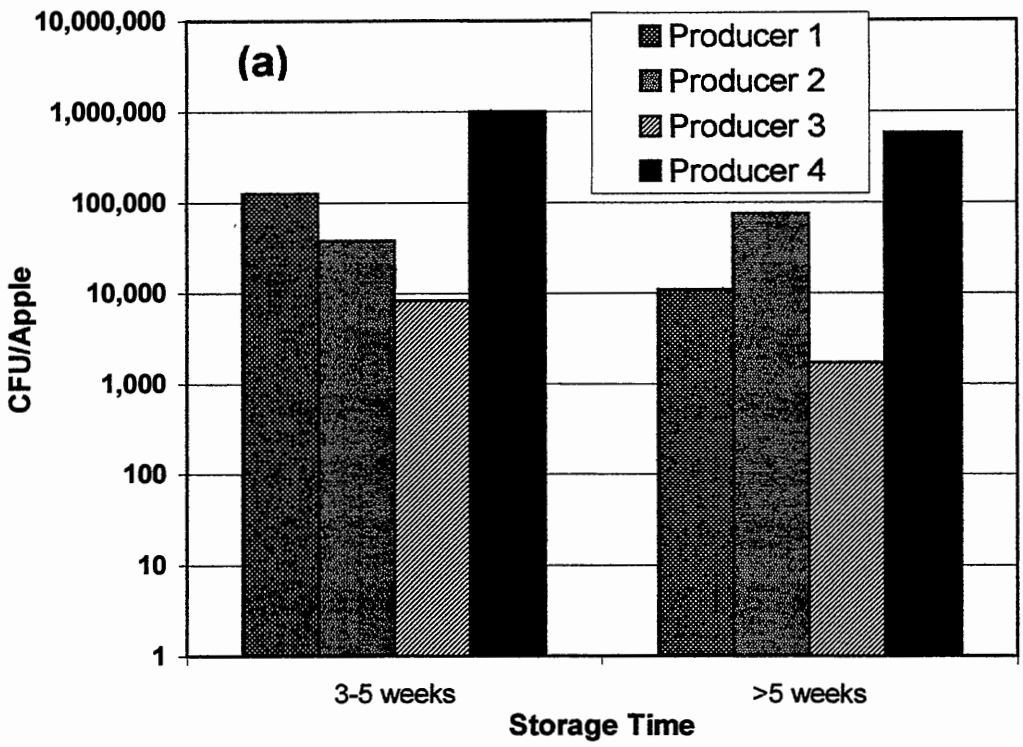
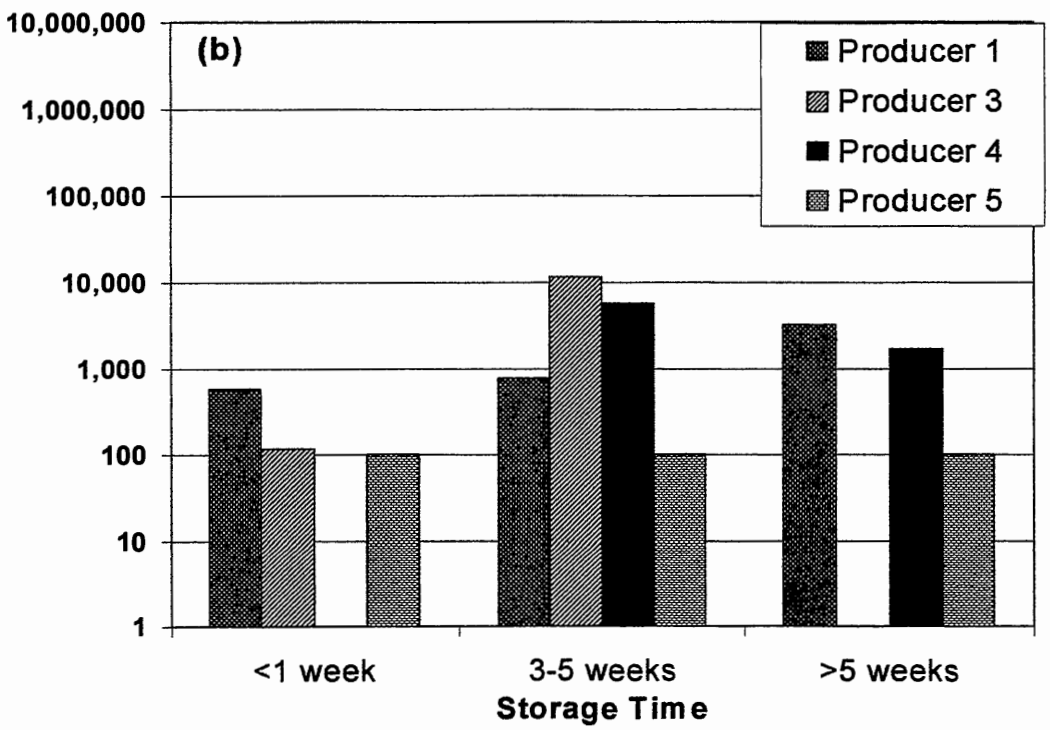


FIGURE 5A. Changes in coliforms during storage of apples for the 1999-2000 season.

FIGURE 5B. Changes in coliforms during storage of apples for the 2000-2001 season.

Apples were chosen at different points during storage and were tested for coliforms. When apples were not obtained during a specific storage period, no data are presented for those points.


(a)
Producer 1
Producer 2
Producer 3
Producer 4
10,000,000
1,000,000
100,000
10,000
1,000
100
10
1
CFU/Apple
3-5 weeks
>5 weeks
Storage Time


(b)
Producer 1
Producer 3
Producer 4
Producer 5
10,000,000
1,000,000
100,000
10,000
1,000
100
10
1
<1 week
3-5 weeks
>5 weeks
Storage Time

Generally, washing/brushing of apples is expected to remove contaminants, but in the case of producer A, the opposite was observed. Figures 6 and 7 show the effect of washing/brushing on the aerobic bacteria and coliforms on the apples during the 2000-2001 season. In using the nonparametric Wilcoxon Rank Test (Ott, 1993) it was found that aerobic bacteria and coliforms increased significantly ($P < 0.001$) after washing/brushing. On average, aerobic bacteria increased by 240% and coliforms increased by over 4000% after the apples were washed. Yeasts and molds were not similarly affected; washing reduced counts by 2.3%. Upon further investigation, aerobic bacteria were found in all six samples of water taken from the processing room. The source of processing water was a well that had not been chlorinated that year, nor had the well water been tested in the previous six months. It is likely that the well water was the source of contamination on the washed apples.

Lowering the microbial load on the apples is an important means of controlling the microbial loads in cider. If microorganisms on incoming apples are low, further treatments (chemical preservatives and pasteurization) could be more effective in keeping final counts in cider very low. It is essential to ensure the quality of the water used in processing to prevent further contamination of equipment and the product itself.

Microbial Contamination of Cider

Microbial counts in apple ciders produced in the 1999 and 2000 seasons are presented in Figures 8 and 9. The data are representative of the producers' final

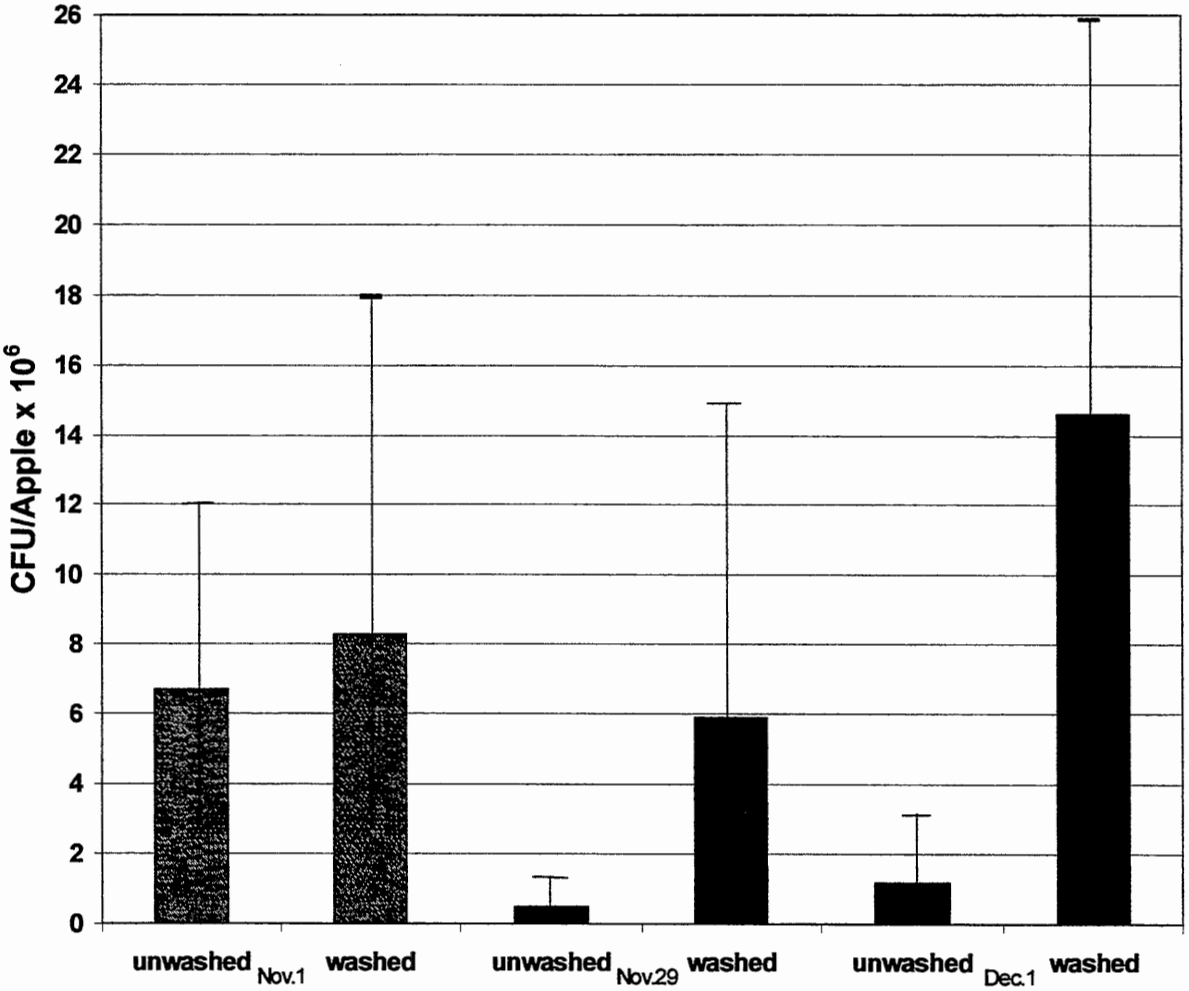


Figure 6. Comparison of average aerobic bacteria counts on apples from Producer A before and after washing during the 2000-2001 season. Three sampling dates are shown. Error bars show the ranges of counts obtained.

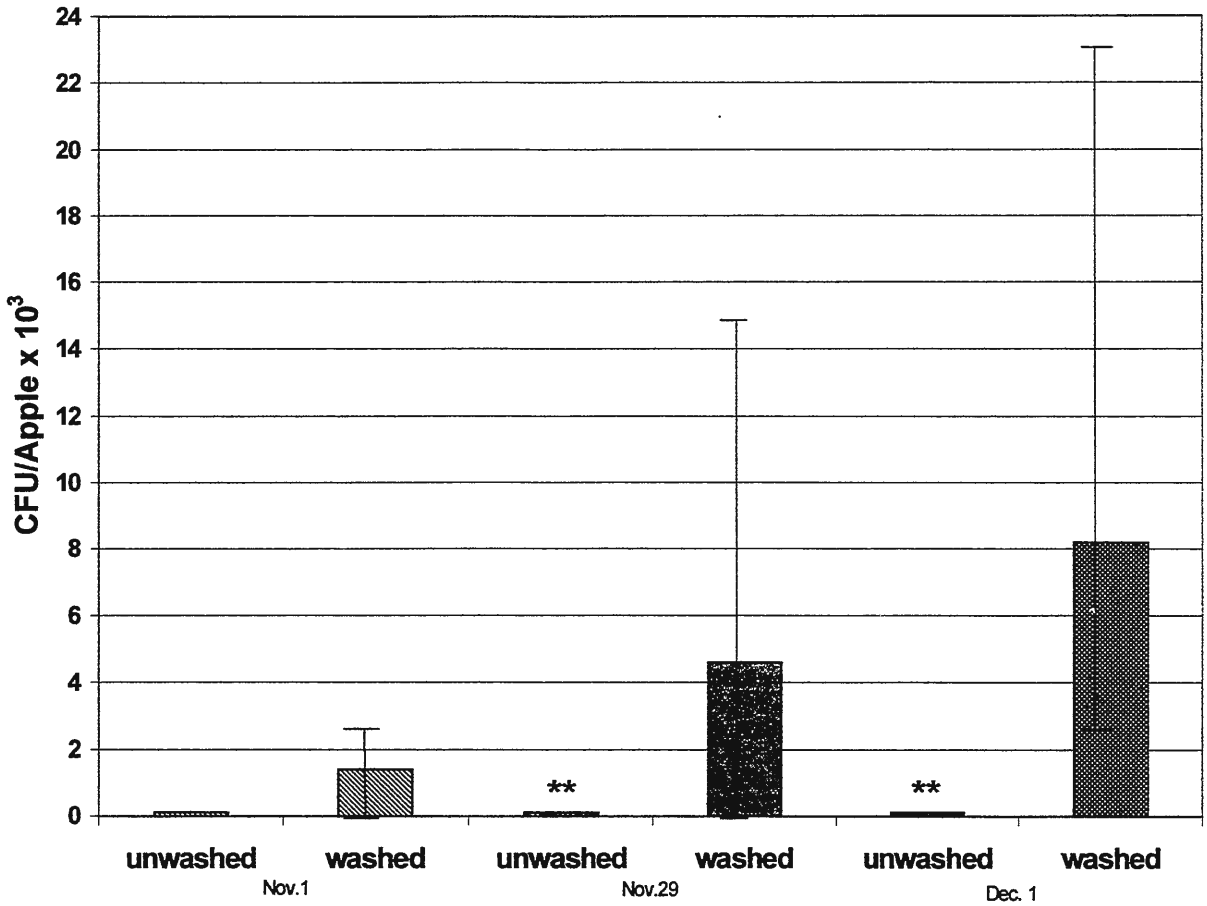


Figure 7. Comparison of average coliform counts on apples from Producer A before and after washing during the 2000-2001 season. Three sampling dates are shown. Those samples that were below the level of detection (100 CFU/apple) are depicted with **. Error bars show the ranges of counts obtained.

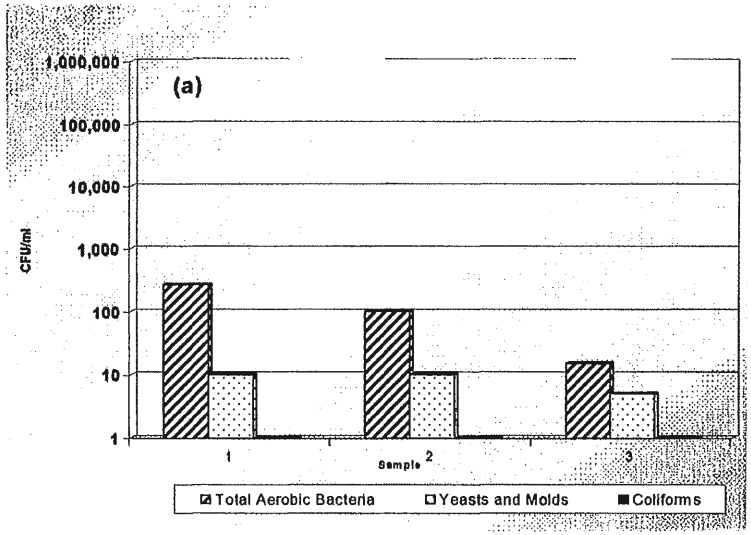
FIGURE 8a. Producer A - Microbial counts in cider during the 1999-2000 season.

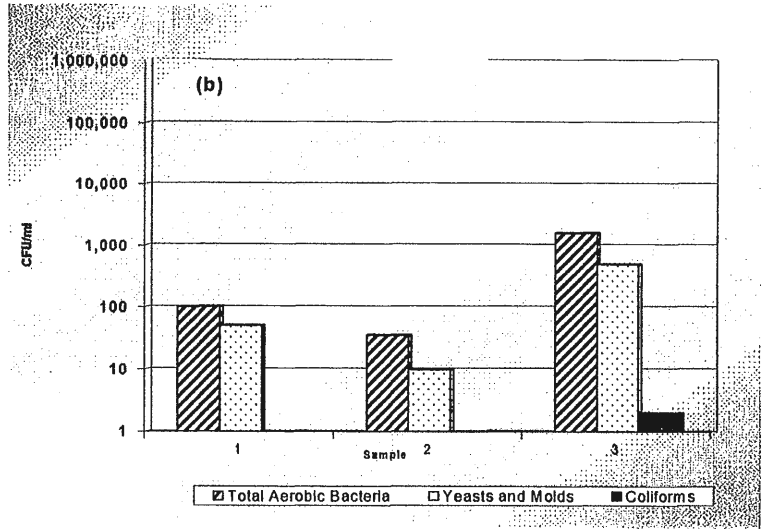
FIGURE 8b. Producer B - Microbial counts in cider during the 1999-2000 season.

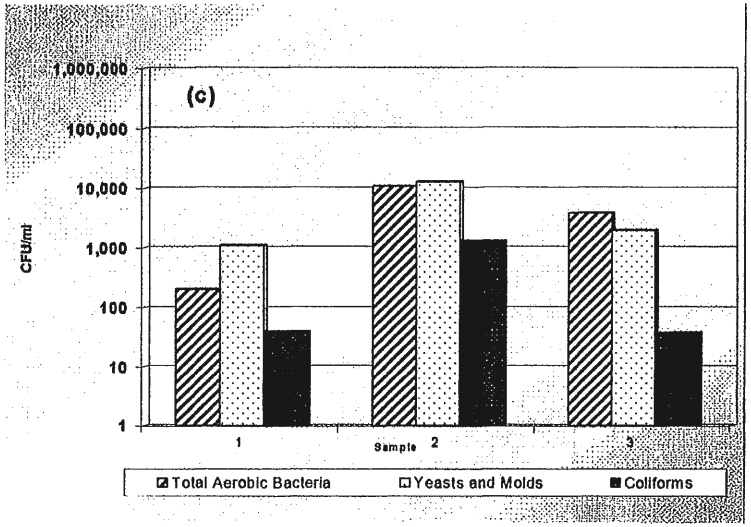
FIGURE 8c. Producer C - Microbial counts in cider during the 1999-2000 season.

FIGURE 8d. Producer D - Microbial counts in cider during the 1999-2000 season.

All cider samples are representative of the producers' final cider. Producers A and B pasteurized cider while producers C and D did not pasteurize their cider. *E.coli* was not detected in any of the cider samples. (Limits of detection for microbial tests were <10 CFU/ml for yeasts and molds and aerobic bacteria and <1 CFU/ml of cider for coliforms).







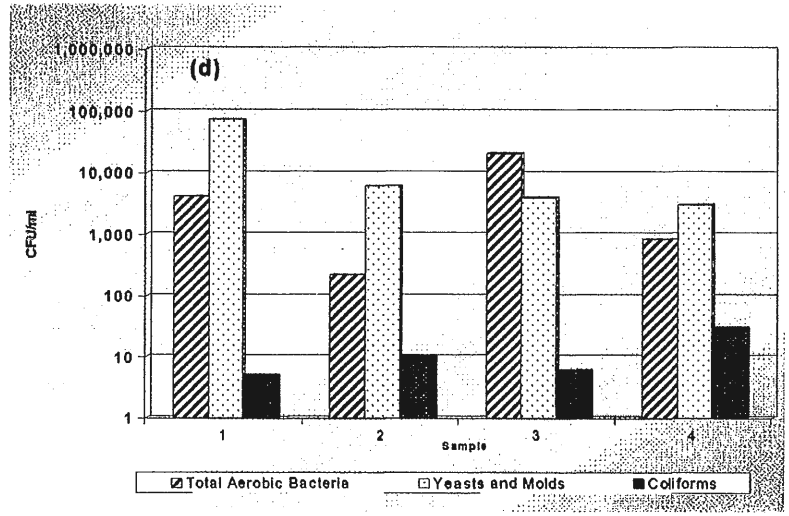


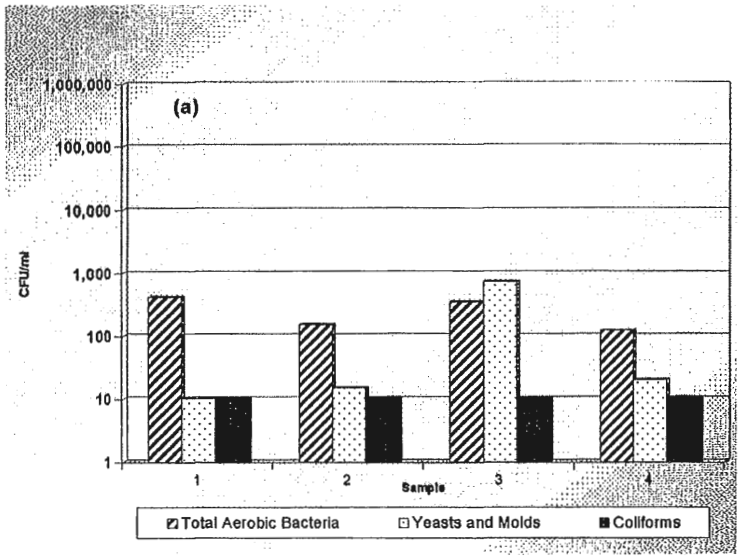
FIGURE 9a. Producer A - Microbial counts in cider during the 2000-2001 season.

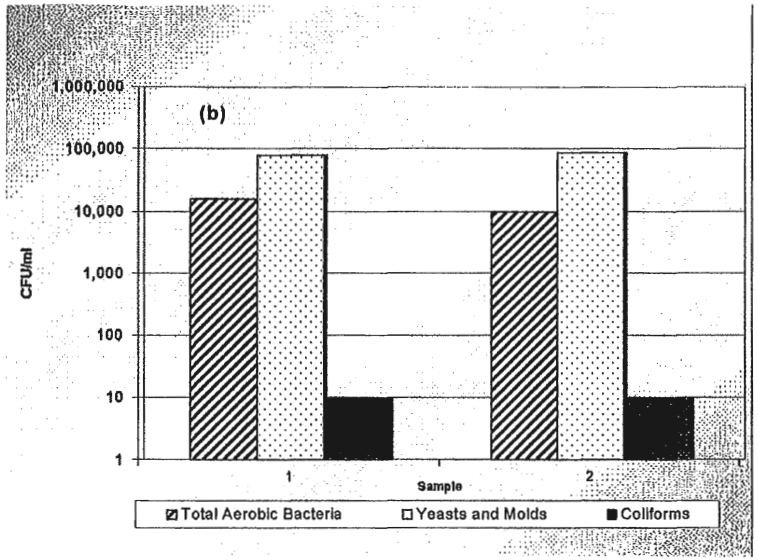
FIGURE 9b. Producer C - Microbial counts in cider during the 2000-2001 season.

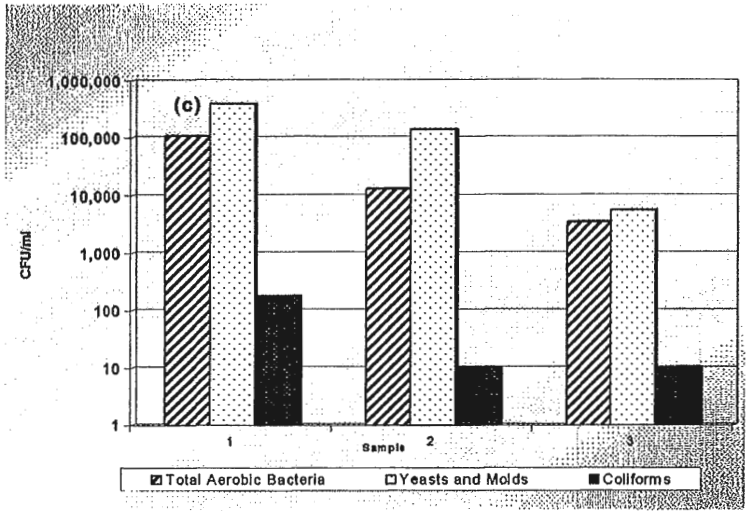
FIGURE 9c. Producer D - Microbial counts in cider during the 2000-2001 season.

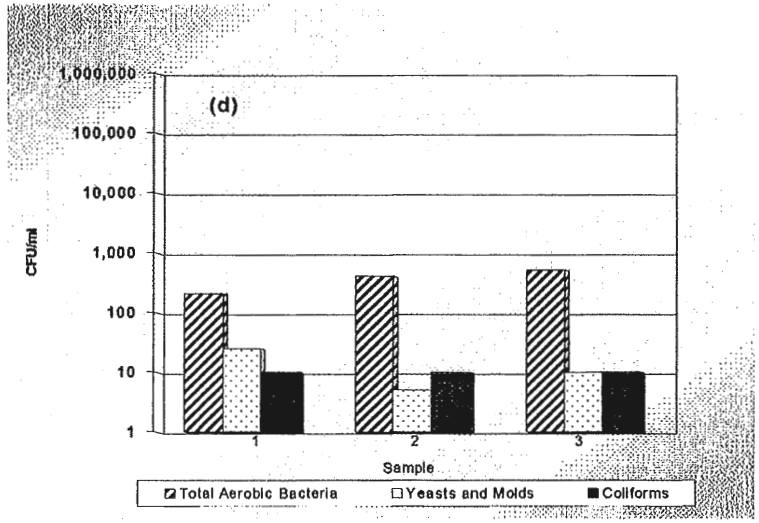
FIGURE 9d. Producer E - Microbial counts in cider during the 2000-2001 season.

All cider samples are representative of the producers' final cider. Producers A and E pasteurized cider while producers C and D did not pasteurize their cider. *E.coli* was not detected in any of the cider samples. (Limit of detection for all microbial tests was <10 CFU/ml of cider)









cider product: pasteurized with preservative for producers A, B, and E and unpasteurized with preservative for producers C and D.

In comparing both years' data, there was a slight increase in yeast and mold counts in the second year of the study. Yeasts and molds in pasteurized cider were all below 1.0×10^3 CFU/ml with most counts in the range of 10-20 CFU/ml. In unpasteurized cider, yeasts and molds were higher with more variability among producers. The highest level of yeasts and molds observed for 1999 was 7.3×10^4 CFU/ml; in 2000 counts reached 3.9×10^5 CFU/ml. Those highest levels were observed in the unpasteurized cider.

Total aerobic bacteria also increased slightly during the second year of the study, with counts of up to 2.0×10^4 CFU/ml in 1999 and as high as 1.1×10^5 CFU/ml in 2000. As expected, counts in pasteurized cider were lower than those in unpasteurized cider, with ranges of 1.5×10^1 to 1.6×10^3 CFU/ml and 2.1×10^2 to 1.1×10^5 CFU/ml, respectively. These results agree with those of a Maryland study that found aerobic bacteria counts in pasteurized cider in the 10^3 to 10^4 CFU/ml range and in unpasteurized cider in the 10^4 to 10^5 CFU/ml range (Senkel et al., 1999).

Coliform levels in cider were generally low and fluctuated very little between the two seasons. Counts below 10 CFU/ml were observed for all pasteurized ciders. The unpasteurized samples were all below 1.3×10^3 CFU/ml with most values in the range of 10 to 200 CFU/ml. These values were much lower than those found in the Senkel et al. (1999) study. The authors reported finding coliforms in unpasteurized cider at an average level of 1.3×10^5 CFU/ml and 3.2×10^3 CFU/ml in pasteurized

samples (Senkel et al., 1999). *E.coli* was not detected in any final cider samples in the present study.

Raw cider (without preservative and unpasteurized) counts showed less variation than the final cider counts. Raw cider generally had higher microbial counts than the final cider or cider with preservatives. However, the addition of preservatives was found to raise the yeast and mold counts for producers A and B, and increased the aerobic bacteria and coliforms for producer A. It is hypothesized that contamination of the cider occurred when the preservatives were added to the cider. The introduction of contamination may have been through the equipment used to stir the preservative or contaminated water in which the preservative was dissolved.

Effects of Pasteurization

Tables 3, 4, 5, summarize the average percent kill achieved by use of preservative and pasteurization for the five cider producers. The values in the table were obtained by comparing fresh cider to cider with preservative and to pasteurized cider (for the processors that pasteurized). Therefore, the data illustrate each producer's process effectiveness in reducing microorganisms during the two years of the study.

According to the nonparametric Wilcoxon rank test (Ott, 1993), pasteurization in 1999 was found to significantly ($P<0.001$) reduce yeasts and molds, aerobic bacteria, and coliforms. Similar results were found in 2000; however, effects on

Table 3. Average Percent Kill of Yeasts and Molds in Apple Cider by Pasteurization and Addition of Preservative during the 1999 and 2000 Seasons

	1999			2000		
Producer	**Preservative Alone**	**Pasteurization Alone**	**Combination of 2 Steps**	**Preservative Alone**	**Pasteurization Alone**	**Combination of 2 Steps**
A	0% Y	99.98%	99.98%	0% Y	99.93%	99.91%
B	0% Y	99.86%	99.85%	N	N	N
C	88%	S	S	32%	S	S
D	80%	S	S	74%	S	S
E	N	N	N	S	S	99.98%

Y Samples showing an increase in yeasts and molds after addition of preservatives
N Producer was not part of the study in this year
S Samples were not available at these points

Table 4. Average Percent Kill of Aerobic Bacteria in Apple Cider by Pasteurization and Addition of Preservative during the 1999 and 2000 Seasons

	1999			2000		
Producer	**Preservative Alone**	**Pasteurization Alone**	**Combination of 2 Steps**	**Preservative Alone**	**Pasteurization Alone**	**Combination of 2 Steps**
A	61%	95.10%	98.09%	0% Y	96.69%	96.11%
B	2%	99.09%	99.11%	N	N	N
C	83%	S	S	65%	S	S
D	99.02%	S	S	53%	S	S
E	N	N	N	S	S	99.98%

Y Samples showing an increase in yeasts and molds after addition of preservatives
N Producer was not part of the study in this year
S Samples were not available at these points

Table 5. Average Percent Kill of Coliforms in Apple Cider by Pasteurization and Addition of Preservative during the 1999 and 2000 Seasons

	1999			2000		
Producer	**Preservative Alone**	**Pasteurization Alone**	**Combination of 2 Steps**	**Preservative Alone**	**Pasteurization Alone**	**Combination of 2 Steps**
A	0% Y	93.70%	96.98%	71%	79%	94%
B	67%	97.49%	99.17%	N	N	N
C	58%	S	S	90%	S	S
D	99.98%	S	S	40%	S	S
E	N	N	N	S	S	99.34%

Y Samples showing an increase in yeasts and molds after addition of preservatives
N Producer was not part of the study in this year
S Samples were not available at these points

coliforms were not found to be significant because the levels of coliforms detected were so low. Percent reduction calculations can greatly depend on the initial level of microorganisms on the incoming apples or in the raw cider; only high levels will allow a high log reduction to be demonstrated.

Yeasts and molds were found to be the most affected by pasteurization, with all pasteurizing processors reaching at least 99.9% destruction (reduction by 3 logs). Similar results were observed for aerobic bacteria, with pasteurization eradicating at least 95%. These results were similar to the findings of Senkel et al. (1999) that pasteurization of cider reduced aerobic bacteria by 2.63 logs. At least 79% of coliforms were killed through pasteurization alone with an average kill among all producers of 90%. Because the number of coliforms per ml was low (less than 10^3) and the detection limit was 10/ml, it was possible only to detect up to 99% kill. At least 10^6 organisms/ml would need to be present to observe a 5-log kill. Higher percent kill values were observed for yeasts and molds and aerobic bacteria because there were higher incoming levels on the apples and in the raw cider.

A best linear unbiased predictor model was utilized to predict the destructive power that each producer would continue to achieve through pasteurization. There was no significant difference found among the producers in terms of coliforms and aerobic bacteria. Therefore, the predicted percent kill for all pasteurization processes of coliforms and aerobic bacteria was 86% and 96.7%, respectively. Although all producers were predicted to have at least 99.6% kill for yeasts and molds, there was a small amount (0.3%) of producer variability observed.

Effect of Preservatives

Tables 3, 4, 5, also summarize the average percent kill achieved by addition of preservative for each individual producer. Potassium sorbate showed a range of destructive capability, reducing viable counts of yeasts and molds by 32% to 88%, aerobic bacteria by 2% to 99%, and coliforms by 40% to 99.98%. Sources of variation may have been application methods used, cleanliness of equipment utilized in the transfer, and the actual concentration of preservative used. Some producers did not weigh the amount of potassium sorbate for each use but rather relied on a volume estimate. Therefore, concentrations of preservative would likely vary for each batch of cider.

Based on the results in tables 3, 4, and 5, it can be concluded that only pasteurization is capable of destroying microorganisms and that potassium sorbate should not be relied upon to kill microorganisms. The cider storage study, discussed later, illustrates the effect of preservative in lengthening shelf life.

Environmental Swab Results

Environmental swabs taken after cleaning equipment are useful tools in examining the efficacy of cleaning and sanitation procedures. The bottler, cider press, and holding tanks were common pieces of equipment that were evaluated for each processor. Water samples were taken from the processing room; unused lids were also collected. The setup of the processing facility sometimes made it impossible to obtain samples from all pieces of equipment; therefore, those points

are reported as not available. Tables 6, 7, and 8 summarize the results; raw data can be found in the appendix.

The following specifications have been set to differentiate among "clean", "marginal", and "failed" equipment: approx. 16 cfu/cm^2 separates clean from marginal and approx. 75 cfu/cm^2 separates marginal from failed (Dickson, 1998). Most of the equipment fell in the "clean" to "marginal" categories. Coliforms were not found in any of the locations tested; either cleaning regimes were sufficient to remove those microorganisms, or they were rarely present at detectable levels. However, on occasion, trouble spots were noted for high counts of aerobic bacteria and yeasts and molds. Samples that fell within the "failed" category are shown in Table 9.

Table 9: Environmental Trouble Spots*

Producer	Location
A	Water used in processing
B	None
C	Water used in processing
D	Bottler Tip
E	Cider Press

* Defined as those areas that had levels of microorganisms greater than 75 CFU/cm^2.

Most facilities utilized a clean-in-place (CIP) regime immediately after production but rarely, if ever, before start-up. The effectiveness of CIP depends on

Table 6. Contamination of the Processing Environment With Yeasts and Molds (Averages)

Location	Producer A	Producer B	Producer C	Producer D	Producer E
Water from Processing room (count per ml)	<10 est	N/a	< 10 est	N/a	< 10 est
Cider Press (count per cm2)	< 1.0 est	N/a	3.0 est	1 est	>6500 est
Cider Holding Tank (count per cm2)	N/a	N/a	< 0.5 est	36 est	0.18 est
Bottler Tip (count per cm2)	< 1.0 est	52 est	5.0 est	190 est	< 10 est
Unused Lid (count per lid)	<10 est	<10 est	7.5 est	<10 est	53 est

N/a = swabs could not be taken from these areas either because equipment was inaccessible or the facilities were operating during sampling
Est = counts fell above (>) or below (<) the countable range

Table 7. Contamination of the Processing Environment With Aerobic Bacteria (Averages)

Location	Producer A	Producer B	Producer C	Producer D	Producer E
Water from Processing room (count per ml)	630	N/a	640	N/a	10 est
Cider Press (count per cm2)	< 0.5 est	N/a	4.0 est	0.5 est	>6500 est
Cider Holding Tank (count per cm2)	N/a	N/a	< 0.5 est	19	0.18 est
Bottler Tip (count per cm2)	< 1.0 est	18 est	10 est	25 est	<10 est
Unused Lid (count per lid)	<10 est	< 10 est	10 est	<10 est	55 est

N/a = swabs could not be taken from these areas either because equipment was inaccessible or the facilities were operating during sampling
Est = counts fell above (>) or below (<) the countable range

Table 8. Contamination of the Processing Environment With Coliforms (Averages)

Location	Producer A	Producer B	Producer C	Producer D	Producer E
Water from Processing room (count per ml)	1.2 est	N/a	< 1.0 est	N/a	< 1.0 est
Cider Press (count per cm2)	< 1.0 est	N/a	< 0.1 est	< 0.1 est	< 0.3 est
Cider Holding Tank (count per cm2)	N/a	N/a	< 0.05 est	0.11 est	< 0.2 est
Bottler Tip (count per cm2)	< 1.0 est	< 1.0 est	< 1.0 est	< 1.0 est	< 1.0 est
Unused Lid (count per lid)	< 1.0 est	< 1.0 est	< 1.0 est	< 1.0 est	< 1.0 est

N/a = swabs could not be taken from these areas either because equipment was inaccessible or the facilities were operating during sampling
Est = counts fell above (>) or below (<) the countable range

time, temperature, concentration, force, and good equipment design (Marriot, 1997). The processors in this study did not record the temperature of the water, the concentration of the cleaning compound and/or sanitizer, and the time the cleaning compounds were in direct contact with the surface. Concentrations of cleaning and sanitizing compounds need to be monitored as residues could contaminate the cider and/or corrode equipment. A rinsing step before start-up could further remove any cleaning compound residues or residual microorganisms remaining after the last production time. Because of the design of various pieces of equipment and the difficulty of removing hardened sugar residues, brushing is recommended as a supplemental step in the cleaning process.

The 1997 FDA inspection revealed that 41% of the firms surveyed had open passageways and entries into the processing room, which provided easy access for large numbers of bees and fruit flies (FDA, 1997). By comparison, 55% of the processors in the current survey left doors and windows open during processing. The occurrence of open passageways during processing is categorized as an environmental sanitation deficiency that could easily be prevented.

The FDA also reported deficiencies in employee hygiene for 25% of the firms surveyed in 1997 (FDA, 1997). The most common deficiencies observed in the current survey that fell into the FDA's categories were: failure to wash and sanitize hands when handling product; inadequate toilet facilities; employees eating or smoking in the processing area; and lack of proper protective clothing and hair restraints. In addition, 55% of the processors in Iowa had not developed standard operating procedures (SOPs) and sanitation standard operating procedures

(SSOPs). Compliance with SOPs and SSOPs can help ensure that all employees follow proper cleaning procedures and maintain appropriate hygiene.

Storage Study Results

Ciders at various stages of processing (unpreserved, with preservative before pasteurization, and pasteurized with preservative) were held at refrigeration temperatures, and microbial counts were determined throughout storage. By comparing the counts obtained, the effect of preservatives and pasteurization on shelf life could be evaluated. Figures 10, 11, 12, 13, and 14 present changes over time for yeasts and molds and aerobic bacteria in cider from each of the five producers in the study. Coliforms decreased to undetectable levels after two weeks of storage in all cider samples tested, and their counts are not shown here.

As storage time progressed, it was noted that more than one type of organism grew on the media used. Thus, care had to be taken when making counts. Because bacterial colonies appeared on PDA plates, gram stains were performed to differentiate bacteria from yeasts. Also, TSA plates were kept an additional 4 days at room temperature to distinguish molds from bacterial colonies.

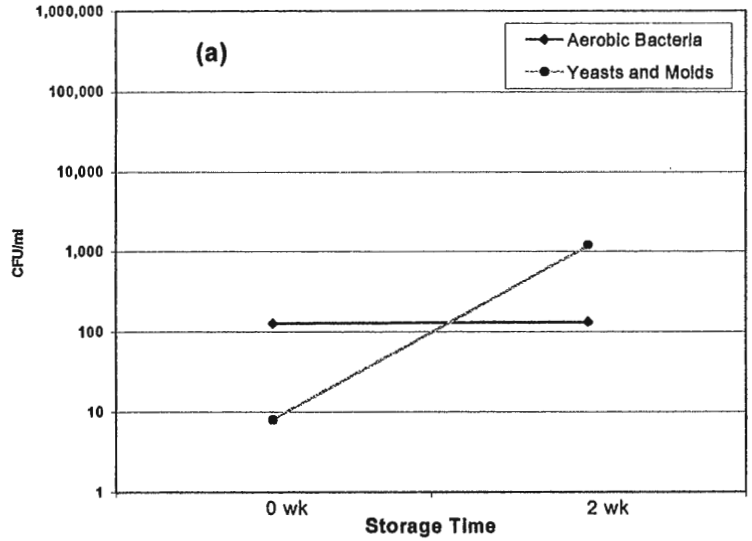
Two general trends were observed: either aerobic bacteria increased over time, with a simultaneous decrease in yeast and mold counts, or yeasts and molds increased while aerobic bacteria decreased. These were likely competing populations whose initial numbers and/or physiological state would determine which would predominate.

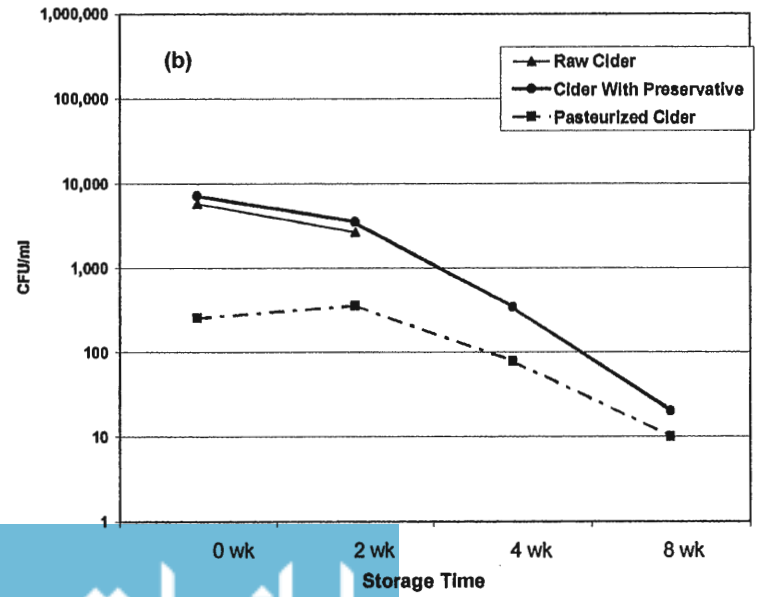
FIGURE 10a. Producer A - Changes in aerobic bacteria and yeasts and molds during storage of pasteurized cider, 1999-2000 season.

FIGURE 10b. Producer A - Changes in aerobic bacteria during cider storage, 2000-2001 season.

FIGURE 10c. Producer A - Changes in yeasts and molds during cider storage, 2000-2001 season.

Cider was held at 7 C. Raw cider is unpasteurized without preservative. Ciders noted as "with preservative" were not pasteurized. Ciders noted as "pasteurized" contained 0.1% potassium sorbate. Storage studies were performed only on pasteurized cider in the 1999-2000 seasons. Storage studies were not performed on raw cider samples because of gas production at 2 wks; therefore, data in figure 10a are for pasteurized cider only.





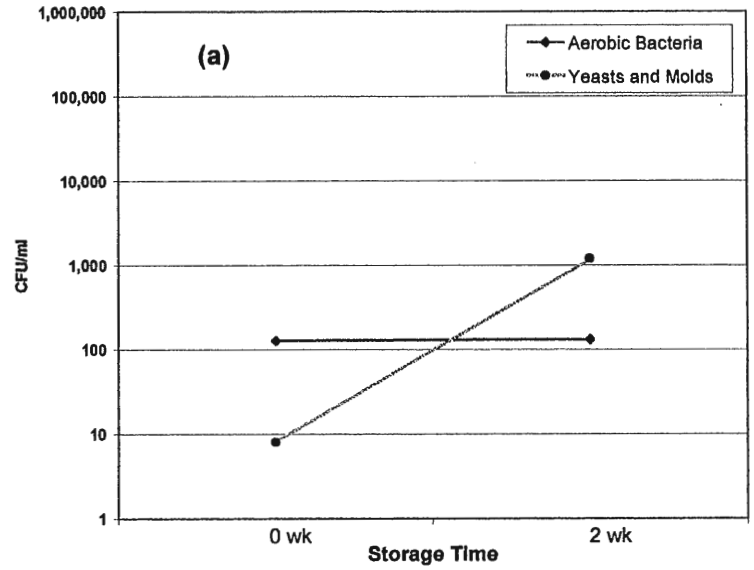
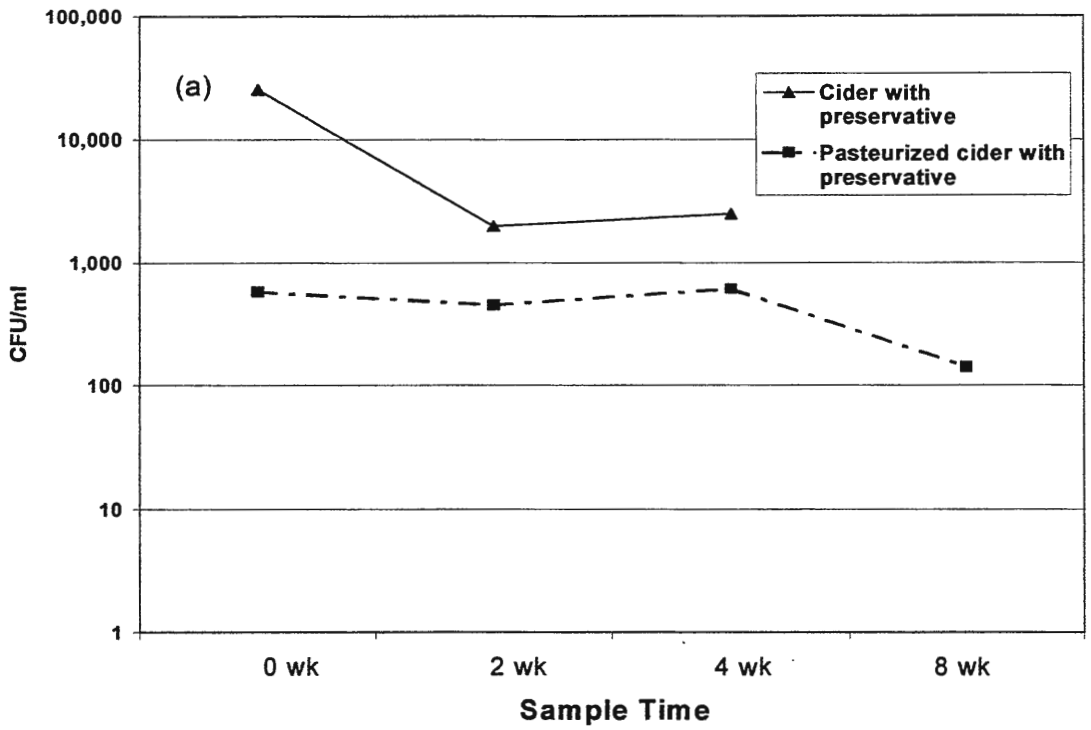


FIGURE 11A. Producer B - Changes in aerobic bacteria during cider storage, 1999-2000 season.

FIGURE 11B. Producer B - Changes in yeasts and molds during cider storage, 1999-2000 season.

Cider was held at 7 C. Ciders noted as "with preservative" were not pasteurized. Ciders noted as "pasteurized" contained 0.1% potassium sorbate and 0.1% sodium benzoate. Storage studies were not performed on raw cider samples because of gas production at 2 wks.



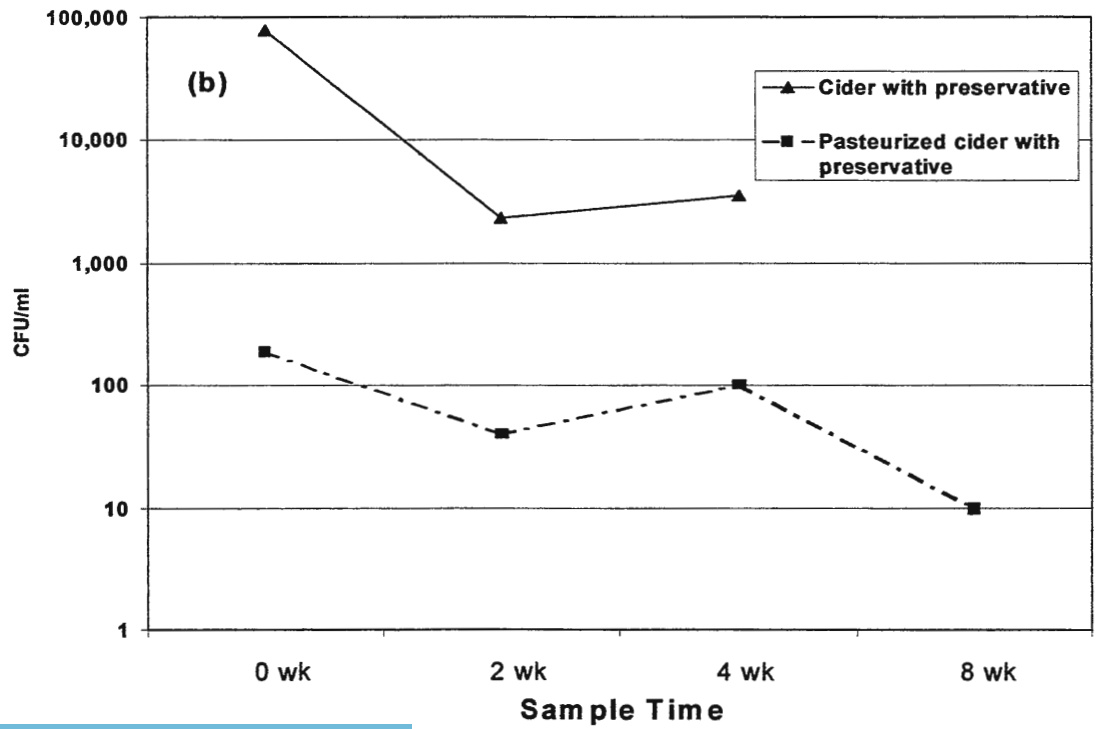
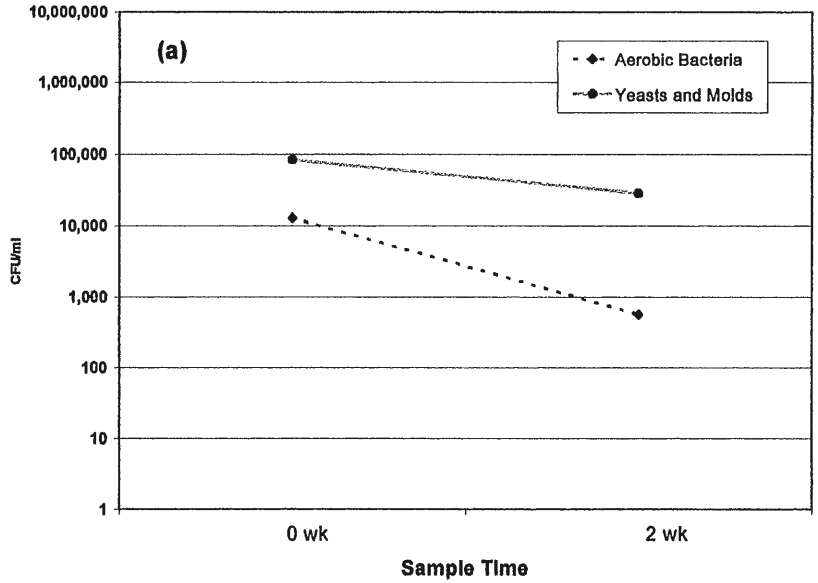


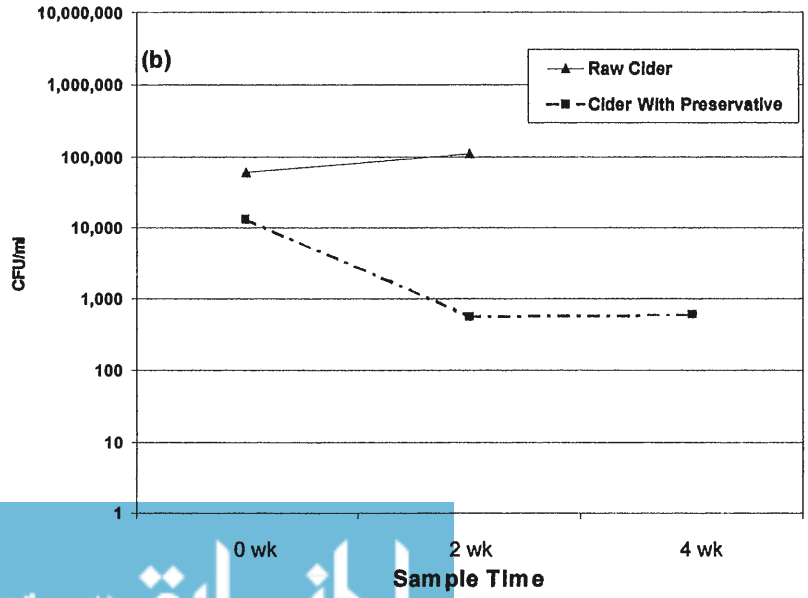
FIGURE 12a. Producer C - Changes in aerobic bacteria and yeasts and mold during storage of cider with preservative, 1999-2000 season.

FIGURE 12b. Producer C - Changes in aerobic bacteria during cider storage, 2000-2001 season.

FIGURE 12c. Producer C - Changes in yeasts and molds during cider storage, 2000-2001 season.

Cider was held at 7 C. Ciders noted as "raw" were unpasteurized without preservative. Ciders noted as "with preservative" were not pasteurized. Storage studies were not performed on raw cider samples beyond two weeks of storage because of gas production.





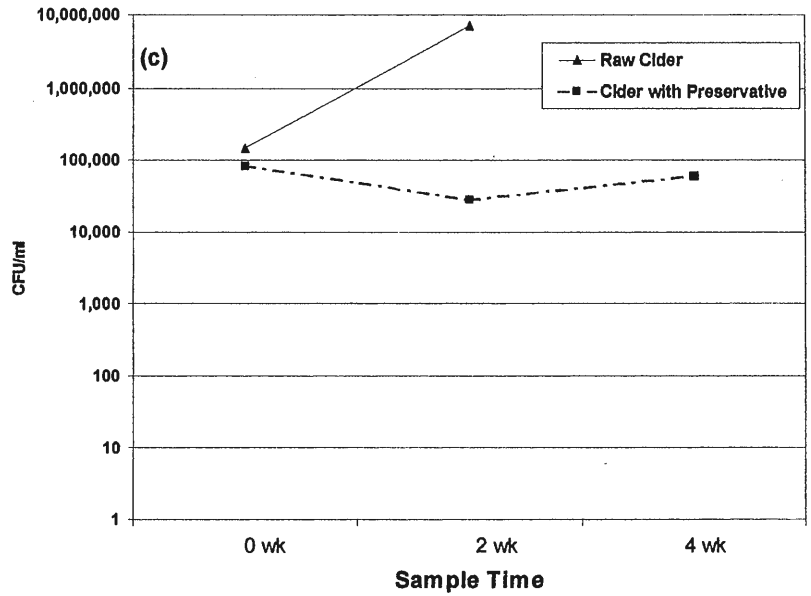


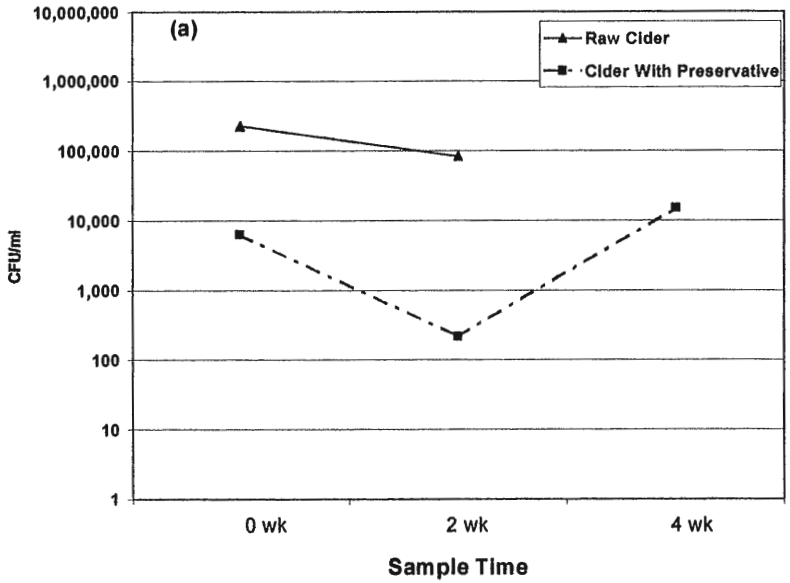
FIGURE 13a. Producer D - Changes in aerobic bacteria during cider storage, 1999-2000 season.

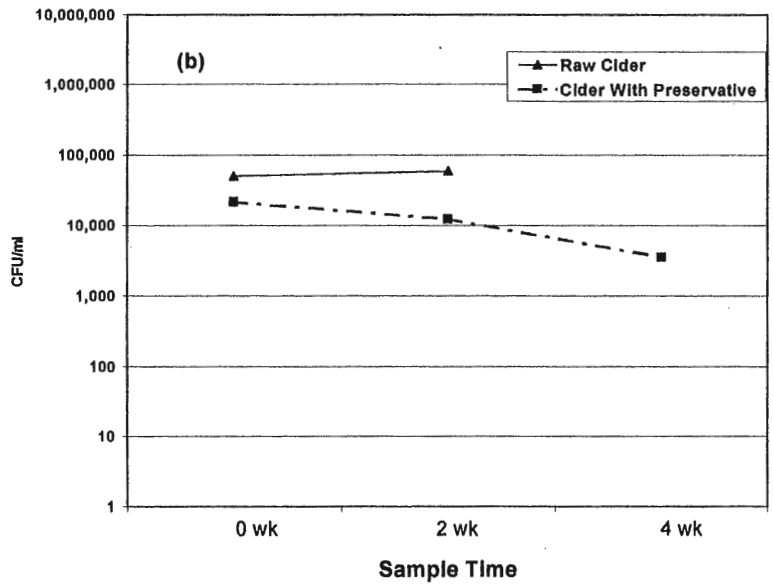
FIGURE 13b. Producer D - Changes in yeasts and molds during cider storage, 1999-2000 season.

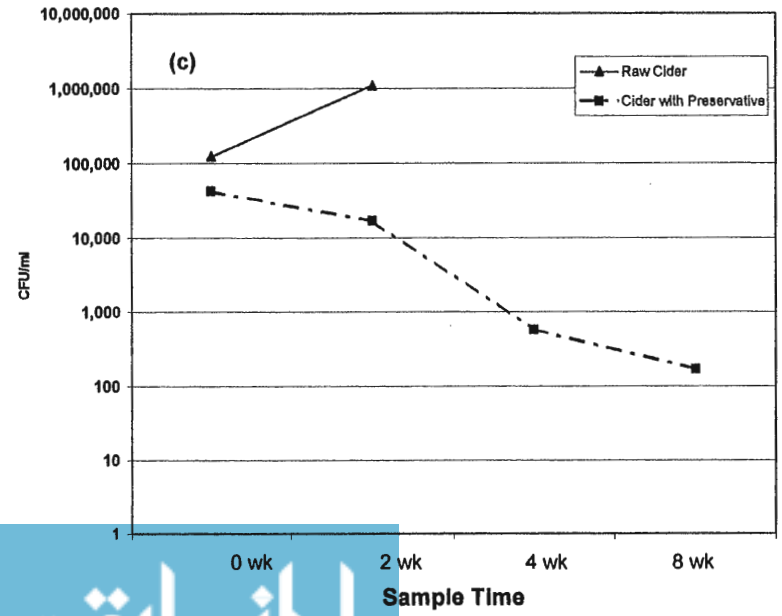
FIGURE 13c. Producer D - Changes in aerobic bacteria during cider storage, 2000-2001 season.

FIGURE 13d. Producer D - Changes in yeasts and molds during cider storage, 2000-2001 season.

Cider was held at 7 C. Ciders noted as "raw" were unpasteurized without preservative. Ciders noted as "with preservative" were not pasteurized. Storage studies were not performed on raw cider samples beyond two weeks of storage because of gas production.







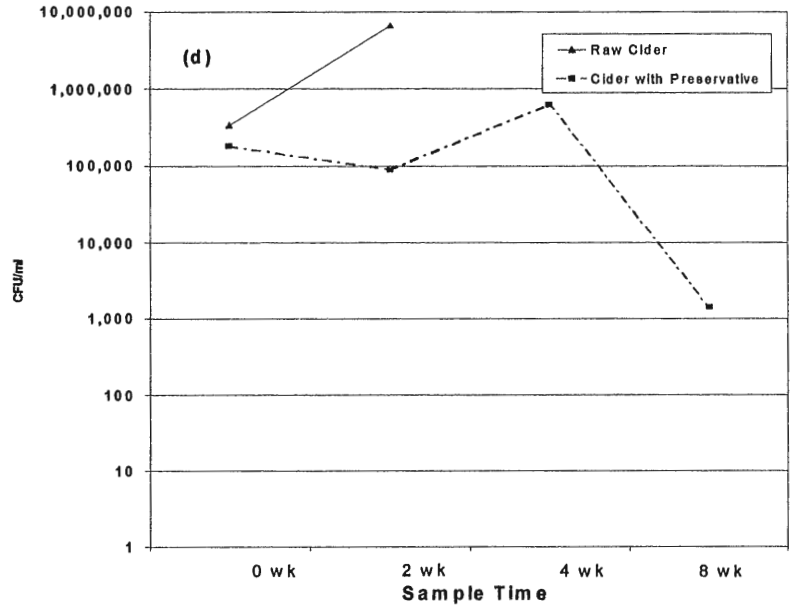
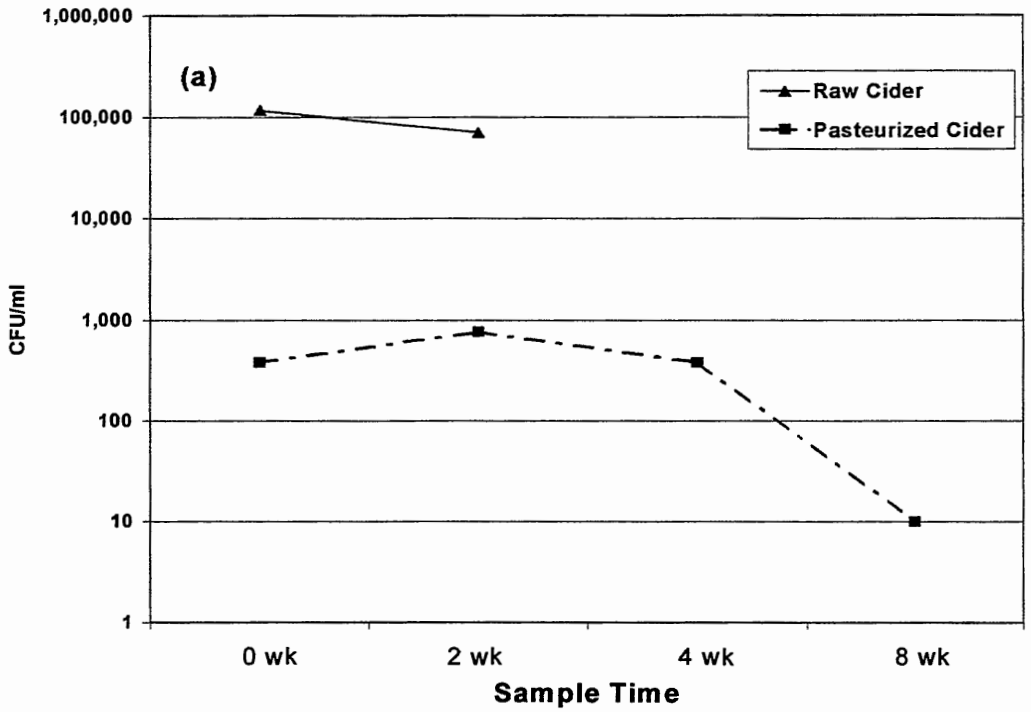
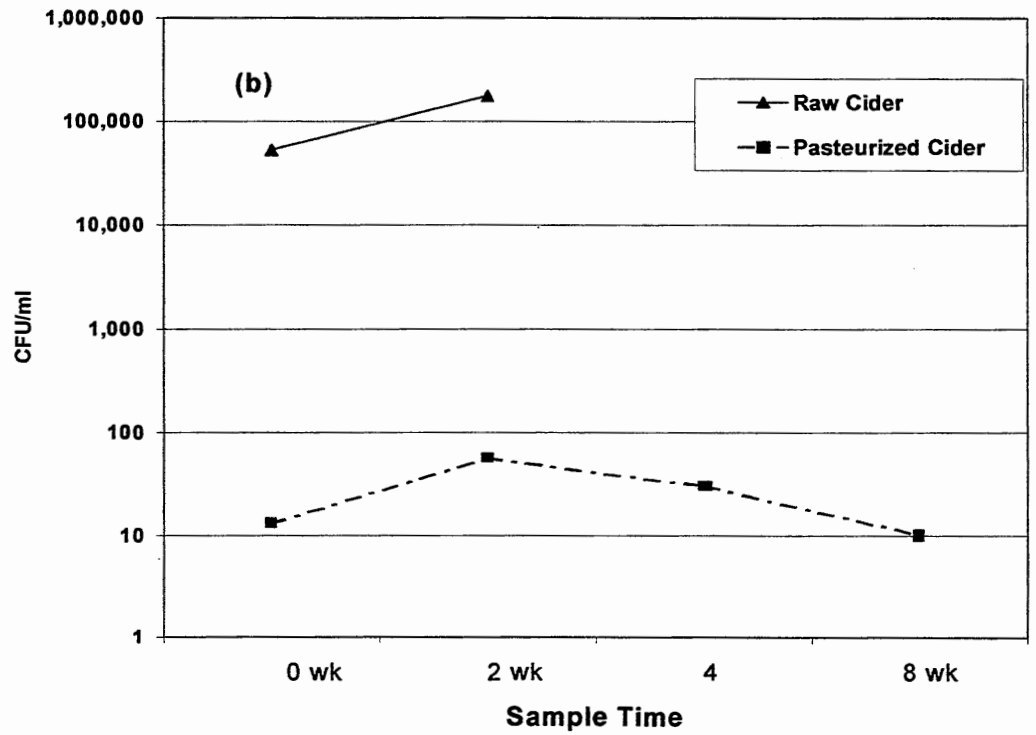


FIGURE 14a. Producer E - Changes in aerobic bacteria during cider storage, 2000-2001 season.

FIGURE 14b. Producer E - Changes in yeasts and molds during cider storage, 2000-2001 season.

Cider was held at 7 C. Ciders noted as "raw" were unpasteurized without preservative. Ciders noted as "pasteurized" contained 0.1% potassium sorbate. Storage studies were not performed on raw cider samples beyond two weeks of storage because of gas production.


(a)
Raw Cider
Pasteurized Cider
1,000,000
100,000
10,000
1,000
100
10
1
CFU/ml
0 wk
2 wk
4 wk
8 wk
Sample Time


(b)
Raw Cider
Pasteurized Cider
1,000,000
100,000
10,000
1,000
100
10
1
CFU/ml
0 wk
2 wk
4
8 wk
Sample Time

Similar observations have been made regarding the unpredictability of the cider fermentation process. The bacteria or yeasts present on the incoming apples largely determine what metabolic processes will take place in the cider (Downing, 1989). If acetic acid bacteria are found to dominate, vinegar will be produced through the conversion of ethanol to acetic acid and further to carbon dioxide and water (Jay, 1997). If, on the other hand, sufficient levels of wild yeasts are present a hard cider could be the end product via the conversion of sugars to ethanol (Deak et al., 1996).

Generally, yeasts and molds were found at higher levels in unpasteurized cider in comparison to pasteurized cider. These high levels may indicate that unpasteurized cider could more likely ferment into hard cider.

Pasteurization significantly reduced yeasts and molds; the initial counts found in pasteurized ciders were low. In these ciders, bacteria were found at higher levels than yeasts and molds late in storage. Acetic acid bacteria may be more likely to produce vinegar as the final product in these ciders.

All raw cider samples showed considerable gas production within two to three weeks of storage. Samples were not tested beyond this point, which was considered to be unpalatable to the consumer. Although the microbial counts in unpasteurized cider were not so high that they suggested obvious spoilage, the appearance and odor of this cider indicated otherwise. After about 5 to 7 weeks of storage, there was an obvious separation of solids and development of an off-odor. The aesthetics of the product would probably deter a consumer from drinking it. A

similar phenomenon was observed in pasteurized cider (with preservative) at about 10 to 12 weeks of storage.

These results indicate that the addition of preservatives to cider lengthens the shelf life by about 2 to 4 weeks, depending on the initial load of microorganisms in the raw cider. Pasteurization further increases the shelf life of cider by an additional 5 weeks. These values are reported averages and any post-processing contamination could reduce the shelf life of the product. Table 10 summarizes these results.

Table 10: Estimated Length of Shelf Life of Apple Cider

Raw Cider	With Preservative, Unpasteurized	Pasteurized With Preservative
2-3 wks	**5-7 wks**	**10-12 wks**

HACCP

HACCP plans were created for the five cider producers in the study. All HACCP plans presented to the processors included a flow diagram, product identification, hazard analysis, critical limits, verification activities, record-keeping procedures, standard operating procedure (SOP) example, and a list of good manufacturing practices (GMPs). All HACCP plans can be found in the appendix.

The plans were similar in regard to general hazards and prevention steps. Two to three critical control points (CCPs) were chosen, depending on the processor and the production methods. Selection of CCPs was based on the likelihood of the hazard occurring, the ability to control the hazard or lower the chances of its occurrence, and the degree of harm that the hazard posed to consumers if it was not

prevented. It is important not to confuse these CCPs that affect safety with control points. A control point is any point during food production where loss of control does not lead to a health risk (Marriot, 1997).

The pasteurization step was a CCP for producers A, B, and E. Points in the process prior to pasteurization were not chosen as CCPs because, when operating effectively, pasteurization and prevention steps leading up to pasteurization should reduce any previously identified hazards to a minimal level. Observations made during this study revealed that operators utilizing pasteurization equipment lacked appropriate methods to verify adequate pasteurization. Therefore, further studies need to be performed to determine a standard time and temperature combination to destroy 5 logs of the target microorganism.

Bottling was chosen as a CCP for all producers. Bottling was performed by hand in most instances and could be a potential source of contamination after the main kill step (pasteurization). All processors were advised to minimize the time that bottles were left open to the air, to ensure all bottles were properly sealed, and to expedite the cooling process once the cider was bottled. One producer was in the process of incorporating an automatic filling system, which could further reduce the risk of contamination.

Common CCPs for those producers that did not pasteurize consisted of receiving and inspection of apples, and bottling. Receiving and inspection was chosen as a necessary point of control because of the hazards associated with using drop apples. It is important to note that most processors receive apples from outside sources, to which no previous specifications were made and for which no

audits were performed. The receiving step becomes pertinent in these cases when a particular load of apples from another processor could contain a mix of both drop apples and tree-picked apples, and no differentiation can be made between these two groups. Specifications for tree-picked apples need to be made prior to outside source purchases.

On several occasions, producer D stored apples outdoors in uncovered cardboard boxes. This environment provided a significant opportunity for contamination of apples and further growth of bacteria. Cardboard boxes are not recommended for apple storage as they can collect moisture and become moldy themselves. Apples should be stored in clean, washable containers within a cooler (<40°F or <4.5°C) at all times to reduce the risk of bacterial growth. Temperature control is useful during the washing of apples. The infiltration of microorganisms into the core, stem, and calyx of the apple, previously determined in a study by Buchanan et al. (1998), can be reduced when the apples are colder than the water in which they are being washed.

An additional CCP for producer E was the addition of preservative. It was observed on several occasions that potassium sorbate had not been fully dissolved in the cider prior to bottling. This may result in an inconsistent concentration of preservative in each bottle. Potassium sorbate does pose a potential chemical hazard at elevated concentrations. As a prevention tool, the processor was advised to let the potassium sorbate fully dissolve before bottling the cider.

Verification activities and record keeping are vital contributions to the efficacy of a HACCP plan. Producers need to keep a record logbook for all CCPs and

corrective actions. Records are to be held on-site for 1 to 2 years to provide for sufficient information for the processor's verification activities (FDA, 2001). Although examples for standard operating procedures and sanitation standard operating procedures were provided to the producers, each individual producer must design his/her own as a tool of reference for all employees.

Inoculation Study

Cider was inoculated with four bacterial strains, two of which were *E.coli* O157:H7, one *Klebsiella spp.*, and one *Enterobacter spp.* The survival time was measured under four conditions: with and without the presence of yeasts at room temperature (25°C) or under refrigeration (4°C). Yeasts were utilized to mimic the natural environment of the cider and to observe the effect of natural microflora on the survival of the bacterial test strains. Different inoculation levels were also used, to determine the effect of the initial bacterial load on survival time in the cider. Table 11 (high inoculation levels) and table 12 (low inoculation levels) show the average survival times of each strain under the four tested conditions. The averages are the results of studies performed in duplicate or triplicate. The pH of the cider was 3.63 and fluctuated very little throughout the study.

All strains held at 4°C had similar survival times in cider, ranging from 10 to 13.6 days. Bacteria were detectable for 3.5 to 6 days longer when inoculated at 10^6/ml compared to 10^3/ml. The presence of yeast at 4°C had little effect on survival time of the bacteria.

Table 11. Average Survival Time (Days) of Bacterial Strains Inoculated at 10^6/ml of Apple Cider

Strain	4°C	4°C With Yeasts	25°C	25°C With Yeasts
1	13.6	10.5	3.6	15
2	11.3	9.5	12.6	2.5
3	10.3	8.0	2.0	2.5
4	10.0	8.5	22	3.0

Table 12. Average Survival Time (Days) of Bacterial Strains Inoculated at 10^3/ml of Apple Cider

Strain	4°C	4°C With Yeasts	25°C	25°C With Yeasts
1	8.5	9.0	2.5	2.0
2	5.5	5.0	1.0	1.0
3	4.0	5.0	1.5	1.0
4	7.0	9.0	1.5	2.0

All strains slightly increased in numbers at 4°C within 1-5 days when inoculated at 10^6/ml; no such increase was observed in samples inoculated with 10^3 CFU/ml. Similar results were reported by Zhao et al. (1993), who found that *E.coli* O157:H7 increased in numbers within 2 to 4 days post-inoculation when initial levels were at 10^5 CFU/ml of cider.

This increase in population was not consistently observed in cider incubated at room temperature or when yeasts were present. Fluctuations in populations were seen for strain 1 (non pathogenic *E.coli* O157:H7), 2 (pathogenic *E.coli* O157:H7, and 4 (*Klebsiella spp.*) for 22, 31, and 61 days, respectively. Strain 4 survived in room temperature apple cider without yeasts for an average of 22 days, but its survival time was much lower (4 days) when yeasts were present. Counts for this organism dropped to 10 CFU/ml but rebounded to 7,000 CFU/ml within 3 days.

Other studies of the fate of *E.coli* O157:H7 in trypticase soy broth adjusted to different pH values with HCl or lactic acid revealed that the minimum pH for growth at 37°C was between 4.0 and 4.5 or 4.6, respectively (Glass et al., 1992). Hence, the acidity of apple cider at pH < 4.0 could likely suppress the growth of *E.coli* O157:H7. Small increases in detectable populations may be the result of resuscitation of injured cells rather than growth (Zhao et al., 1993).

Mixed results were observed in samples held at 25°C. Bacteria were detectable for 0.5 to 19.5 days longer when inoculated at 10^6/ml compared to 10^3/ml. Strains 1, 2, and 4 were found to survive longer at 25°C than at 4°C. These findings are in contrast to those of Zhao et al. (1993) and Dingman (1999); *E.coli* O157:H7 survived longer in cider held at 4 to 8°C when compared to cider held at

room temperature. However, Dingman (1999) found that *E.coli* O157:H7 could survive in apple cider 14 days at 25°C, which is similar to the 15 and 12.6 days of survival observed for strain 1 and 2. The results of the present study also correspond to a study by Ryu et al. (1998) in which it was reported that *E.coli* O157:H7 survived in cider held at room temperature for more than 56 days.

The presence of yeasts did not affect the survival time of bacteria inoculated at 10^3/ml, however, major differences were observed for the bacteria inoculated at 10^6/ml. Strain 1 survived over 400% longer in the presence of yeasts. On the contrary, strains 2 and 4 experienced respective 500 and 700% increases in survival times when yeasts were absent. Duffy et al. (1999) reported that the presence of a competitive microflora could protect pathogens such as *E.coli* O157:H7 against the inhibitory effects of acid. It seems that the bacterial strains varied in their responses to the products from the yeasts, with survival either enhanced or inhibited by the presence of yeasts.

Since contamination of cider with *E.coli* O157:H7 is not a common event and levels of contamination are not expected to be as high as 5 log CFU/ml or even 2 log CFU/ml, the effects of storage are an important component in a risk assessment program (Duffy et al., 2001). The results of the inoculation study revealed a general decline in all tested bacterial strains in apple cider over time under any storage conditions. However, the reduction in populations during storage could not be used as a sole method of approaching the 5-log reduction required by the FDA.

Identification of Common Microorganisms in Cider

On each sampling date, 2 colonies each from TSA, VRB/Petrifilm, and PDA plates were chosen for further testing from each sample grouping (apples, raw cider, cider with preservative, final cider). The colonies were chosen because of their frequent appearance in each processor's product. These organisms were restreaked for purity and identification tests were performed to determine what types of microorganisms were commonly found in the sampled apple products. A gram stain was performed on all chosen colonies. All of the gram-negative colonies exhibited similar cell morphology (rods, either long and narrow or short and fat); biochemical tests were performed on most of these colonies. Based on the biochemical test results, 10 colonies classified as *Enterobacteriaceae* were tested further using the BBL Crystal E/NF identification kit. A tentative identification was then assigned to each colony. Tables 13 and 14 present the results of these tests.

The BBL Crystal E/NF identification kit was only partly successful in identifying various microorganisms in cider and on apples. Family or genus of microorganisms could be determined, but species could not be identified in most cases because of atypical biochemical reactions.

Several gram-positive microorganisms were also isolated from cider and apples. Most appeared to be *Bacillus spp* or *Streptomyces spp*. Yeasts isolated from samples were identified based on their cell shape and size.

Most bacteria found in apples and cider were members of the *Enterobacteriaceae*. Yeasts were also prevalent on apples and in unpasteurized

Table 13. Tentative Identification of Typical Colonies found on Apples

Colony ID	Colony Morphology on TSA	Gram Rxn	Cat.	Ind.	Oxid.	Cit.	M-R	V-P	Possible ID
A1	shiny, cream	-	+	-	-	+	+	-	*Enterobacter***
A3	shiny, orange	+							Yeast
B1	cone, white	+							Yeast
B2	shiny, translucent	-	+	-	+	+	-	-	Coliform
C1	shiny, yellow	-	+	-	-	+	+	-	Coliform
C2	shiny, cream	-	+	-	-	+	-	-	*Klebsiella***
D1	shiny, cream	-							Coliform
D2	shiny, yellow	-	+	-	-	+	+	-	*Enterobacter***
E1	soupy, cream	+							Yeast
E2	hard, rugged edge, white	+	+	-	+	-	-	-	*Streptomyces*

Cat. = Catalase
Ind. = Indole
Oxid. = Oxidase
M-R = Methyl – Red
Cit. = Citrate
V-P= Voges – Proskauer

** = Identification made through BBL Crystal E/NF Kit

Table 14. Tentative Identification of Typical Colonies found in Cider

Colony ID	Colony Morphology on TSA	Gram Rxn	Cat.	Ind.	Oxid.	Cit.	M-R	V-P	Possible ID
A1	hard, raised, white	+	+	-	+	-	-	-	*Streptomyces*
A2	shiny, pink	+							Yeast
A3	shiny, white	-	+	+	-	-	-	-	*Acetobacter*
B1	dull, cream	-	+	-	-	+	-	-	*Rahnella***
B2	rugged edge, cream	+	+	-	+	-	-	-	*Bacillus*
B3	shiny, cream	-	+	-	+	+ (L)	-	-	Enterobacteriaceae
C1	shiny, yellow	-	+	-	-	+ (L)	-	-	Coliform
C2	shiny, translucent	-	+	-	-	-	+	-	Enterobacteriaceae
C3	dull, red	+							Yeast
D1	shiny, yellow	-	+	-	-	+	-	-	Coliform
D2	shiny, cream	+	+	-	-				irregular, non-sporing
D3	shiny, translucent	-	+	+	-	-	-	-	*Acinebacter***
E1	shiny, cream	-	+	+	-	+	-	-	*Klebsiella***
E2	shiny, light pink	-	+	+	-	-	-	-	*Acetobacter*
E3	dull, white	+							Yeast

Cat. = Catalase
Ind. = Indole
Oxid. = Oxidase
M-R = Methyl – Red
Cit. = Citrate
V-P= Voges – Proskauer

(L) = represents those samples that had a late response

** = Identification made through BBL Crystal E/NF Kit

cider. It is not uncommon to find *Enterobacteriaceae* in fruit products as they are widespread and can be found throughout the environment. However, if high levels are present it could mean contamination from a fecal source has occurred and the likelihood of finding coliforms or *E.coli* would be much greater.

General Observations

Cider processing in Iowa is slowly disappearing. During this study, two producers indicated they had stopped processing, and three processors that participated in the in-depth study sold large portions of their orchards. It is likely that more producers will be cutting back on production for the upcoming 2001-2002 season.

At the moment, cider producers' choice of methods for reducing the concentrations of pathogens by 5-logs is pasteurization; UV light treatment, ozone, and irradiation are also under investigation, but all of these methods require a large investment of money. Inexpensive methods such as freezing/thawing and storage of cider at 35°C have been reported to produce a 5-log reduction in *E.coli* O157:H7 (Ulijas et al., 1999), but the practicality of those methods is questionable.

It is estimated that the cost of incorporating pasteurization could range from $9,500 to $35,000/year, depending on the production rate (Carbone, 2001). That estimate includes the fixed costs of the equipment as well as installation and operating costs. Most small producers (< 20,000 gal/year) are concerned with the costs of pasteurization as well as possible quality changes that may be imparted in the cider. Producer A pasteurized at 163°F (73°C) for 0.5 to 1.0 sec. and believed

that no flavor or color changes were imparted by pasteurization. However, Producer B pasteurized at 178°F (81°C) for 18.7 sec. and believed that a noticeable flavor and color change had occurred. Not only do times and temperatures of pasteurization play a large role in quality changes of the cider, but they also determine the degree of killing of microbial contaminants.

The educational component of this project has played an important role in keeping cider processors up-to-date with new regulations. Those who attended "cider school" have expressed great interest in methods to lower their possible risk of *E.coli* O157:H7 contamination and have worked together to reduce the likelihood of an outbreak in Iowa. The producers who were involved in the in-depth study were cooperative and accepting of new ideas. This continued attitude will allow for a smooth and successful transition into the new FDA regulations.

CONCLUSIONS

The results of this study indicate that most cider producers in Iowa are incorporating methods (apple sanitizers, temperature control, filtering, addition of preservatives, pasteurization) to lower the risk of *E.coli* O157:H7 contamination. Most producers appear to be using sound orchard management practices. The number of producers that use drop apples is low (18%) and it is hoped will continue to decrease. Specific recommendations for improvement are using chlorinated processing water and auditing outside apple suppliers.

It was anticipated that incorporating HACCP plans during the 2000-2001 season would help reduce microbial loads. Although microbial loads in apples and cider differed very little between the two years of the study, a slight trend towards reduced counts in the 2000-2001 season was observed, especially in coliforms. It is possible that the operators did not have sufficient time to assimilate the information or make processing changes prior to the start of the season. Despite the lack of significant change in microbial counts, there were improvements in good manufacturing and sanitation practices during the course of the study.

The microbial loads on apples were generally high, with coliforms being a natural part of the microflora. *E.coli* was detected in only a small percentage of the apples tested. It is essential that producers use sound fruit and maintain proper storage conditions. Observations of the operations indicated that various steps in apple handling (washing, storage, culling) could be improved. As the microbial loads on the incoming apples decrease, further processing steps should be more effective in keeping contamination low.

Microbial loads in cider varied throughout the production seasons. A wide variety of microorganisms were isolated from cider, most of which were yeasts or *Enterobacteriaceae*. The microbial levels generally decreased through the cider process with the various types of cider ordered as follows: nonpasteurized with no preservatives > nonpasteurized with preservatives > pasteurized cider.

The survival study on *E.coli* O157:H7, *Klebsiella spp.*, and *Enterobacter spp.* showed that the test strains could survive in apple cider but showed a wide range of rates of decline. On two occasions, *E.coli* O157:H7 survived for 21 to 31 days, which is a large portion of the cider's shelf life. Therefore, it should not be expected that pathogen populations would decline during short cider storage.

Pasteurization has proven to be an effective means of reducing microbial loads in cider and lengthening the shelf life. However, good manufacturing practices should continue to be incorporated after the pasteurization step to ensure that post-processing contamination does not occur. The cider producers utilizing pasteurization equipment lacked appropriate methods to verify adequate pasteurization. Pasteurization equipment, particularly thermometers and flow diversion devices, should be inspected, tested, and calibrated regularly to ensure proper function of all components. It is ultimately up to the processor to validate and verify that their process indeed meets the standard for 5-log reduction of the pertinent microorganism.

In conclusion, no *E.coli* were found in any cider samples from Iowa processors. Although *E.coli* were detected on a small percentage of apples, the various processing steps employed in cider production proved effective in keeping

the levels of *E.coli* in the cider below the level of detection. These findings do not mean that *E.coli* is completely absent from the cider produced in Iowa; however, its incidence appears to be low. Further compliance with HACCP will help lower the risk of a possible *E.coli* O157:H7 outbreak in apple cider.

Future Studies

More questions need to be researched to provide a solid base of information about microbial loads in cider, their length of survival, 5-log destruction methods, and effects of preventative programs such as HACCP. Future studies could include the following:

- Verification of time/temperature combinations, necessary to eliminate the pertinent microorganism by 5-logs, in various pasteurization units being used in production.
- Sensory studies based on consumer acceptance of cider treated with UV light, ozone, and pulsed-electric fields.
- The effect of waxing apples prior to storage on spoilage and microbial loads.
- More data comparing the heat and acid tolerance of *E.coli* O157:H7, *L. monocytogenes*, and *Crytosporidium parvum* in regard to apple cider.
- The effect of fully incorporating HACCP into a cider facility on microbial loads.

APPENDIX A

HACCP PLANS

Producer A: Product Description

HACCP Plan No. 1 Product Category: Pasteurized 100% Fruit Juice

1. Common name?
Pasteurized 100 % Apple Cider

2. How is it to be used?
Consumed as purchased (ready-to-drink)

3. Type of package?
Plastic bottling (high density polyethylene)

4. Length of shelf life, at what temperature?
Approx. 60 days if not opened
Maximum acceptable storage temperature 40°F
Recommend <38°F

5. Where will it be sold?
Retail and Wholesale (In Iowa)

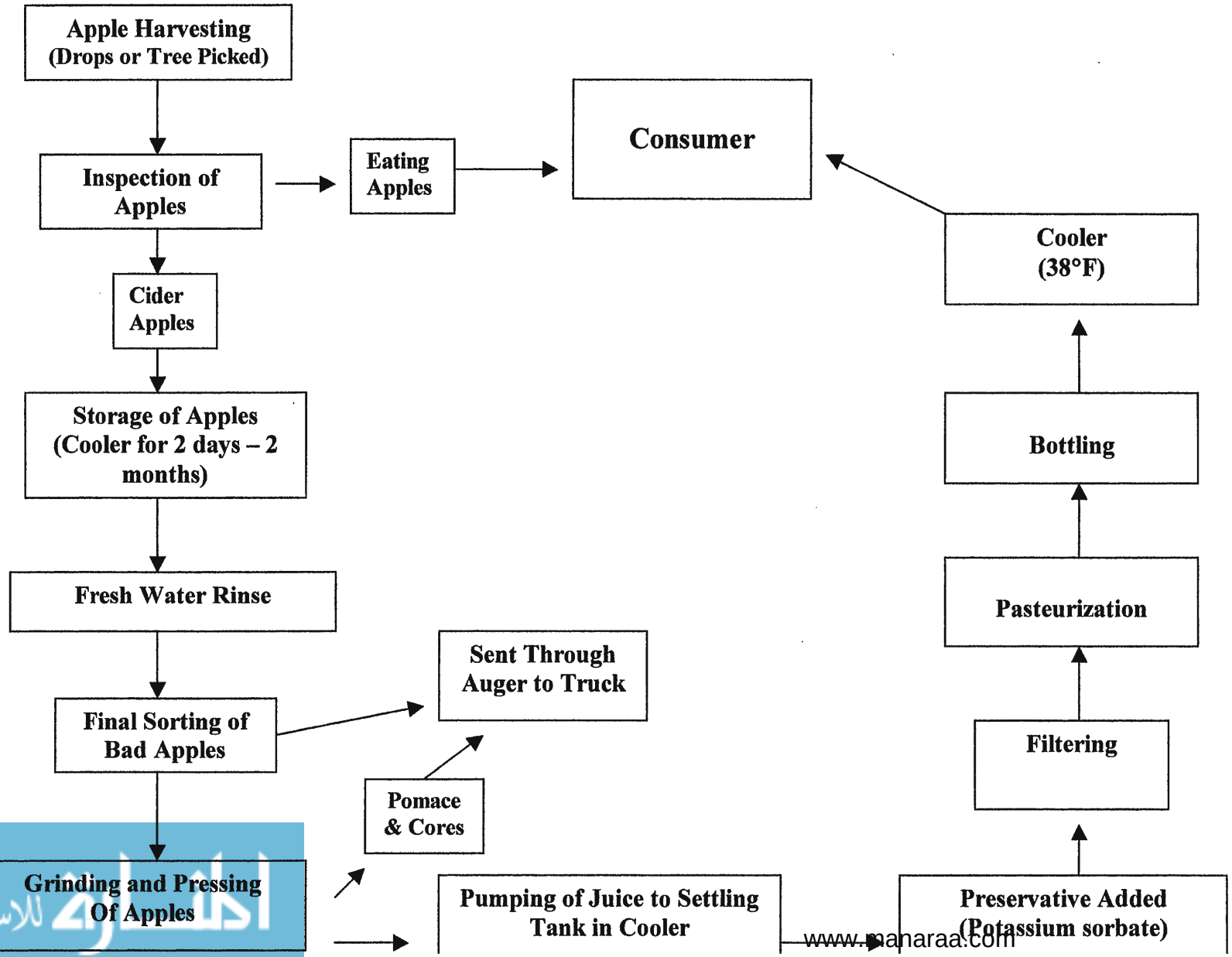
6. Labeling instruction?
Label should say "Keep Cold"
Ingredients, nutrition facts, net content, "Use By" / "Sell By" and/or Date of Production are recommended

7. Is special distribution control needed?
Lot code or date of production needed for traceability
Distribution and storage under acceptable refrigeration (maximum recommended temperature 40°F)
Use of temperature monitoring devices recommended

Approved by: _____ Date approved: _____

List Product Ingredients:
Apple, Potassium Sorbate

PRODUCER A: FLOW DIAGRAM FOR APPLE CIDER PRODUCTION



Producer A: Hazard Analysis
HACCP Plan No. _1_____ Process category _____
_____

Product: Pasteurized 100% Apple Cider

Hazard Analysis and Identification of Critical Control Points

Processing Step		**Potential Hazards Introduced**	**What Control Measures Can be Applied To Prevent The Hazard?**		**Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?**	**CCP #**
Raw Ingredients Apples: Harvesting/ Receiving and Inspection	B	Pathogens	-No drops -Inspection of apples (own and suppliers)	B	NO (Pasteurization at later step)	
	P	Wood	-Certified Supplier audits	P	NO	
	C	Pesticides	-Grate separates large debris -Remove visibly spoiled and damaged apples	C	NO	
Storage of Apples	B	Pathogens Mold growth Cross-contamination of bad to good apples	-Storage inside cooler 35-40°F, no outside, uncovered storage -Re-inspection weekly to remove bad apples	B	NO (Pasteurization at later step)	
	C	None		C	NO	
	P	Rodents, Insects	-Pest Control (GMP)	P	NO	
Final Sorting	B	None	-Good employee hygiene and GMP's	B	NO	
	C	None	-Removal of visibly spoiled and damaged apples	C	NO	
	P	None		P	NO	
Fresh Water Rinse	B	Pathogens, Parasites	-Potable Water -Guaranteed testing by municipal utilities	B	NO	
	C	Metals, Pesticides, Nitrites		C	NO	
	P			P	NO	
Grinding and Pressing	B	Contamination from dirty equipment	-Ensure proper cleaning of equipment (SOP)	B	NO	
	C	Cleaning residues	-Properly rinse equipment after cleaning and/or before start up	C	NO	
	P	Metal debris from machine	-Filter Cider	P	NO	

Processing Step		Potential Hazards Introduced	What Control Measures Can be Applied To Prevent The Hazard?		Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?	CCP #
Pumping Cider to Settling Tank in Cooler	B	None	-Ensure clean tubing and tank (SOP) -Proper cooler temperature (35 – 40F)	B	NO	
	C	None		C	NO	
	P	None		P	NO	
Add Preservative (Pot.Sorbate)	B	None	-Good record keeping with SOP, monitoring and maintenance of weighing scales	B	NO	
	C	Chemical limits exceeded		C	NO	
	P	None		P	NO	
Filtering 25 & 50 mesh bags	B	None	-Check filter daily for visible signs of damage -Replace as necessary	B	NO	
	C	None		C	NO	
	P	None		P	NO	
Pasteurization	B	Pathogens destruction	-Flash Pasteurization at 163 F for 5 seconds	B	YES	1
	C	None		C	NO	
	P	None		P	NO	
Bottling	B	Pathogens	-Trough is covered or enclosed -SOP for flavor adding -Capping performed in timely manner - containers not allowed to sit open -Visual inspection of bottles for foreign materials -Checked for proper sealing (SOP)	B	YES	2
	C	Introduction of chemical hazards		C	NO	
	P	Introduction of physical hazards		P	NO	
Cooling	B	Improper cooling allowing pathogen growth	-Bottles are sent to cooler immediately after sealing -Monitor cooler temperature 35-40°F	B	NO	
	C	None		C	NO	
	P	None		P	NO	

Processing Step		**Potential Hazards Introduced**	**What Control Measures Can be Applied To Prevent The Hazard?**		**Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?**	**CCP #**
Storage	B	Improper refrigeration temperature promotes bacterial growth	-Monitor storage and transportation temperature 35 – 40 F	B	NO	
	C	None		C	NO	
	P	None		P	NO	

Approved by: _____ Date approved: _____

Producer A: Critical Limits

Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 1 (B) Pasteurization	Pathogenic Bacteria (Destruction)	Product Target Time/Temp: 163°F for .5 – 1.0 sec. Minimum temp: 160°F (alarm will sound)	Temperature of cider at exit of holding tube	Temperature recorder at end of hold tube Low temperature manual divert valve	Continuous recording Temp. should also be recorded manually every restart as a comparison record	Pasteurizer operator	Cider will be manually diverted if temp. at end of holding tube is low; cider will be re-pasteurized If divert valve does not work; production will be stopped and the portion of cider in the holding tube and final holding tank will be re-pasteurized Verification of temperature gauge and alarm system to ensure deviation does not occur again Questionable cider may be tested for pathogen survivors before being shipped If seals are broken or gauge is not working properly; Stop processing if necessary, recalibrate pump and/or reseal
			Seals on pumps and tubing and flow rate gauge	Visual check	At start up and once during processing	Pasteurizer operator	

Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 2 (B)(P) **Bottling**	(B) Pathogens	Cap immediately after filling	Length of time containers are exposed to open air environment	By capping immediately once bottle is full of cider	Every container	Operator	Dispose of bottled cider
		Proper Seal achieved	Proper sealing of container	Visual	Every container	Operator	Reseal if necessary
		Bottles sent to cooler in a timely manner	Length of time bottles are setting at room temperature	Time	Get estimate time for every pallet of bottled cider	Operator	Dispose of bottled cider if has set at room temperature longer than 1 hour
	(P) Environmental debris	No visible debris	Physical contamination	Visual	Every container	Operator	Dispose of bottled cider

Producer A: Verification Activities and Recording Procedures

Verification Activities	Record-Keeping Procedures
CCP #1 Pasteurization 1. Verification of thermometers daily before start-up and calibration if necessary 2. Calibration of divert valves and alarm system at least once a week 3. Holding tube length and diameter are tested once per season with a tracer test to validate the residence time 4. Checks pump flow rate gauge daily Records are reviewed and initialed and dated on a weekly basis	- Pasteurization log which include temperature data, any deviations, etc. - Pasteurizer operator compares continuous temp. records and the manual temp. recording data on a daily or weekly basis - Calibration records for the thermometers, divert valve, alarm system, etc - Flow verification log (pump flow rate info) - Corrective action logs - Production manager will review and initial records daily
CCP #2 Bottling 1. Ensure bottles are capped and sent to cooler in a timely manner Records are reviewed and initialed and dated on a weekly basis	- Log verifying bottling equipment and flavoring equipment was cleaned and sanitized before use - Bottles capped timely and sealing documentation - Log documenting discarded product due to biological and/or physical contamination - Corrective action logs

Verification may also include microbial tests for coliforms and *E.coli.*
All deviations of CCPs will be recorded and corrective actions will be taken.
Corrective actions will also be recorded and reviewed.

Producer B: Product Description

HACCP Plan No. 1 Product Category: 100% Apple Cider

1. Common name?
100 % Apple Cider (Pasteurized)

2. How is it to be used?
Consumed as purchased (ready-to-drink)

3. Type of package?
Plastic bottling (high density polyethylene)

4. Length of shelf life, at what temperature?
Approx. 60 days if not opened
Maximum acceptable storage temperature 42°F
Recommend <38°F

5. Where will it be sold?
Retail (10%), Wholesale (90%)

6. Labeling instruction?
Label should say "Keep Cold"
Ingredients, nutrition facts, net content, "Use By" / "Sell By" and Date of Production are recommended

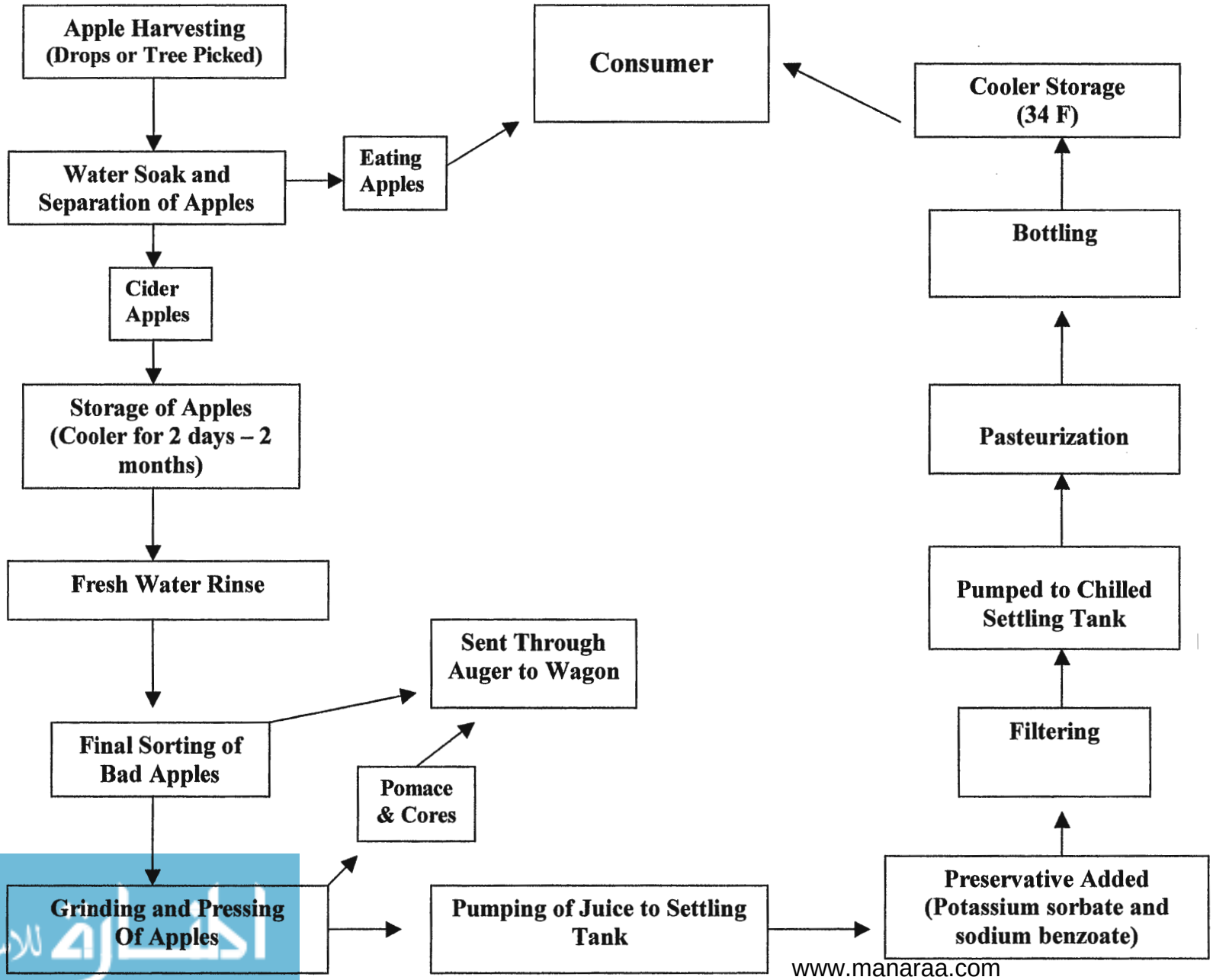
7. Is special distribution control needed?
Lot code or date of production control needed for traceability
Distribution and storage under acceptable refrigeration (maximum recommended temperature 42°C)
Use of temperature monitoring devices recommended

Approved by: _____ Date approved: _____

List Product Ingredients:

Apple, Potassium Sorbate, Sodium Benzoate

PRODUCER B: FLOW DIAGRAM FOR APPLE CIDER PRODUCTION



Producer B: Hazard Analysis
HACCP Plan No. _____ Process category _____

Product: Pasteurized 100% Apple Cider

Hazard Analysis and Identification of Critical Control Points

Processing Step		**Potential Hazards Introduced**	**What Control Measures Can be Applied To Prevent The Hazard?**		**Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?**	**CCP#**
Raw Ingredients Apples, Receiving and Inspection	B	Pathogens	-Inspection of apples (own and supplier audits)	B	NO	
	P	Wood	**-No drops** -Grate separates large debris	P	NO	
	C	Pesticides	-Remove visibly spoiled and damaged apples -Past. At later step	C	NO	
Water Soak	B	Pathogen contam. From water	-Change water every day	B	NO	
	C	Improper Chlorine levels	-Monitor chlorine use Use pH strips	C	NO	
	P	Debris	-Past. At later step	P	NO	
Storage of apples	B	Pathogens Mold growth Cross-contamination of good and bad apples	-Storage inside cooler 35-42°F, no outside, uncovered storage **-Re-inspection weekly to remove bad apples**	B	NO	
	C	None		C	NO	
	P	Rodents, Insects	-Pest Control (GMP)	P	NO	
Fresh water rinse	B	Pathogen contam. From water	-Potable Water Only	B	NO	
	C	Over chlorination etc.	-Guaranteed testing by municipal utilities	C	NO	
	P	None		P	NO	
Grinding and Pressing	B	None		B	NO	
	C	None		C	NO	
	P	Metal debris from machine	-Filter Cider	P	NO	

Pumping Juice to holding tank	B C P	None None None	-Clean tubing and tank -Covered tank recommended	B C P	NO NO NO	
Processing Step		**Potential Hazards Introduced**	**What Control Measures Can be Applied To Prevent The Hazard?**		**Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?**	**CCP#**
Add Preservatives (Pot. Sorbate and Sodium Benzoate)	B C P	None Chemical Limits exceeded None	-Good record keeping with SOP, monitoring and maintenance of weighing scales	B C P	NO NO NO	
Filtering	B C P	None None None	-Check filter daily for visible signs of damage -Change filters appropriately	B C P	NO NO NO	
Overnight Settling in Chilled Tank	B C P	Pathogens if improper holding temperature and not properly enclosed None None	-Monitor temperature -Tank is properly covered and sealed	B C P	NO NO NO	
Pasteurization	B C P	Pathogen destruction None None	-Proper time and temperature	B C P	YES NO NO	#1
Bottling	B C P	Pathogens Introduction of chemical hazards Introduction of physical hazards	-Bottling system placed where contamination from drips does not occur - Bottling system is covered or enclosed -Capping performed in timely manner - containers not allowed to sit in open environment -Visual inspection of bottles for foreign materials -Checked for proper sealing (SOP)	B C P	YES NO YES	#2
Cooling	B C	Improper cooling allowing pathogen growth None	-Bottles are sent to cooler immediately after sealing -Monitor cooler temperature 35-42°F	B C P	NO NO NO	

	P	None				
Storage	B C P	Improper refrigeration temperature promotes bacterial growth None	-Monitor storage temperature 35-42 F	B C P	NO NO NO	

Approved by: _____ Date approved: _____

Producer B: Critical Limits

Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 1 (B) Pasteurization	Pathogenic Bacteria (Destruction)	Product Target Time/Temp: 178-180°F for 18.7 sec. Minimum temp: 171°F	Temperature of cider at exit of holding tube	Temperature recorder at end of hold tube, dual automatic low temperature divert valve and also one manual divert valve	Continuous recording	Pasteurizer operator	Cider will automatically be diverted if temp at end of holding tube is low; cider will be re-pasteurized If divert valve does not work; product will be manually diverted and the portion of cider in the holding tube and final holding tank will be re-pasteurized Verification of temperature gauge and alarm system to ensure deviation does not occur again Questionable cider may be tested for pathogen survivors before being shipped If seals are broken or flow rate gauge is not working properly; Stop processing, recalibrate pump and/or reseal
			Seals on pumps and tubing and flow rate gauge	Visual check	At start up and once during processing	Pasteurizer operator	

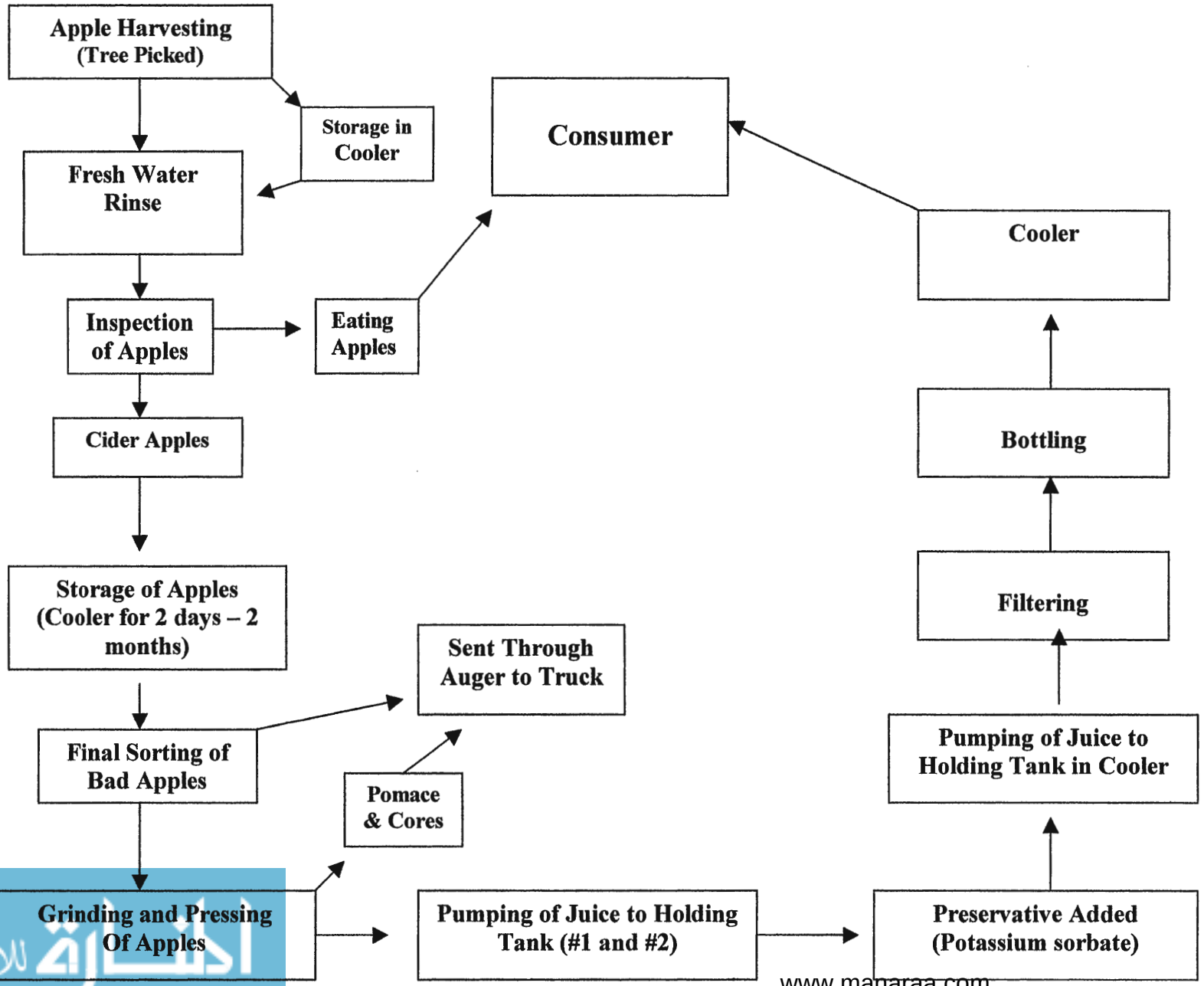
Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 2 (B, P) Bottling	(B) Pathogenic Bacteria	Cap immediately after filling	Length of time containers are exposed to open air environment	By capping immediately once bottle is full of cider	Every container	Operator	Re-pasteurize or dispose of bottled cider
		Ensure proper cleaning and sanitizing of equipment	Cleaning regime	SSOP	Every clean-up	Operator	Re-clean and rinse
		Proper seal achieved	Proper sealing of container	Visual	Every container	Operator	Reseal if necessary
		Bottles of cider sent to cooler in a timely manner	Length of time bottles of cider sit at room temperature after pasteurization	Time (starting immediately after cider is pasteurized)	Every 30 minutes	Operator	Send bottles of cider to the cooler immediately once 30-minute timer goes off. If 30 min. time limit not reached: may need to re-pasteurize
	(P) Environmental debris	No visible debris	Physical contamination	Visual	Every container	Operator	Dispose of bottled cider

Producer B: Verification Activities and Record Keeping

Verification Activities	Record-Keeping Procedures
CCP #1 Pasteurization – Manager reviews records weekly 1. Maintenance calibrates divert valves and alarm system weekly 2. Verification of thermometers daily before start-up 3. Holding tube length and diameter are tested once per season with dye tracer test to validate the residence time 4. Manager checks pump flow rate gauge daily and enters data in pasteurization log	- Pasteurization log which includes temperature data - Calibration records for the thermometers, divert valves, alarm system, etc - Flow verification log (pump flow rate info) - Corrective action logs - All records will be initialed and dated
CCP #2 Bottling – Manager reviews records weekly	- Log verifying bottles were sent to cooler as soon as possible - Bottles capped timely and sealing documentation - Log documenting any discarded product due to biological and physical contamination

Verification may also include microbial testing for coliforms and *E.coli.*
All deviations of CCPs will be recorded and corrective actions will be taken.
Corrective actions will also be recorded and reviewed.

PRODUCER C: FLOW DIAGRAM FOR APPLE CIDER PRODUCTION



Producer C: Hazard Analysis
HACCP Plan No. _____ Process category _____

Product: 100% Apple Cider

Hazard Analysis and Identification of Critical Control Points

Processing Step		Potential Hazards Introduced	What Control Measures Can be Applied To Prevent The Hazard?		Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?	CCP#
Raw Ingredients Apples, Receiving and Inspection	B	Pathogens	-Inspection of apples (own and suppliers)	B	YES	1
	P	Wood	-No drops -Grate separates large debris	P	NO	
	C	Pesticides	-Remove visibly spoiled and damaged apples	C	NO	
Fresh Water Rinse	B	Contamination from water	-Potable water source -Tested annually for microbial and chlorination levels	B	NO	
	C	None		C	NO	
	P	None		P	NO	
Storage of apples	B	Pathogens Mold growth Cross-contamination of good and bad apples	-Storage inside cooler 35-42°F, no outside or uncovered storage -Re-inspection weekly to remove bad apples	B	NO	
	C	None		C	NO	
	P	Rodents, Insects	-Pest Control (GMP)	P	NO	
Final Sorting	B	None	-Good employee hygiene and GMP's	B	NO	
	C	None	-Removal of visibly spoiled and damaged apples	C	NO	
	P	None		P	NO	
Grinding and Pressing	B	None		B	NO	
	C	None		C	NO	
	P	Metal debris from machine	-Filter Cider	P	NO	
Pumping Juice to holding tank	B	None	- EnsureClean tubing and tank (SSOP)	B	NO	
	C	None		C	NO	
	P	None		P	NO	
Add Preservatives (Pot.Sorbate)	B	None		B	NO	
	C	Chemical Limits exceeded	-Good record keeping with SOP, monitoring and maintenance of weighing scales	C	NO	
	P	None		P	NO	

Processing Step		Potential Hazards Introduced	What Control Measures Can be Applied To Prevent The Hazard?		Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?	CCP#
Overnight Settling in Holding Tank inside Cooler	B	Pathogens if improper holding temperature and not properly enclosed	-Monitor temperature 35-42°F -Tank is properly covered and sealed	B	NO	
	C	None	-Ensure tank is properly cleaned and sanitized (SSOP)	C	NO	
	P	None		P	NO	
Filtering	B	None	-Check filter daily for visible signs of damage	B	NO	
	C	None	-Change filters appropriately	C	NO	
	P	None		P	NO	
Bottling	B	Pathogens	-Bottling system is covered or enclosed	B	YES	2
	C	Introduction of chemical hazards	-Capping performed in timely manner - containers not allowed to sit in open environment	C	YES	
	P	Introduction of physical hazards	-Visual inspection of bottles for foreign materials -Checked for proper sealing (SOP)	P	YES	
Cooling	B	Improper cooling allowing pathogen growth	-Bottles are sent to cooler immediately after sealing	B	NO	
			-Monitor cooler temperature 35-42°F	C	NO	
	C	None		P	NO	
	P	None				
Storage	B	Improper refrigeration temperature promotes bacterial growth	-Monitor storage temperature	B	NO	
				C	NO	
	C	None		P	NO	
	P	None				

pproved by: _____ Date approved: _____

Producer C: Critical Limits

Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 1 (B) **Receiving and/or Inspection**	Pathogenic Bacteria (Prevention and Reduction)	- No Drops - No domestic manure in orchards - Removal of Rotten or Spoiled Apples - Auditing for Outside Apple Sources	Apple harvesting and shipments	Visual and thorough record keeping	Each harvest or shipment	Receiving Manager	-No Drops Policy -Reject Drops
CCP 2 (B)(P) **Bottling**	(B) Pathogens	Cap immediately after filling	Length of time containers are exposed to open air environment	By capping immediately once bottle is full of cider	Every container	Operator	Dispose of bottled cider
		Proper Seal achieved	Proper sealing of container	Visual	Every container	Operator	Reseal if necessary
		Bottles sent to cooler in a timely manner	Length of time bottles sit at room temperature	Time	Bottles should sit out no longer than 30 minutes	Operator	Send bottles of cider to the cooler (ensure temperature inside cooler)
	(P) Environmental debris	No visible debris	Physical contamination	Visual	Every container	Operator	Dispose of bottled cider

Producer C: Verification Activities and Recording Procedures

Verification Activities	Record-Keeping Procedures
CCP #1 – Apple Receiving and Inspection 1. No drops allowed 2. Proper inspection and removal of spoiled apples All records will be reviewed and initialed and dated weekly	- Documentation at harvest (tree picked vs. drops) - Valid records for sorting and separation of spoiled apples - Audits of outside apple suppliers - Log documenting any shipments of apples that are rejected
CCP #3 – Bottling 1. Verify that bottling system is properly enclosed 2. Ensure bottles and capped and sent to cooler in a timely manner All records will be reviewed and initialed and dated weekly	- Bottles capped timely and sealing documentation - Log documenting discarded product due to biological and physical contamination - Verification of thermometers in cooler (calibration if necessary)

Verification of overall process may also include microbial testing for coliforms/*E.coli*

All deviations of CCPs will be recorded and corrective actions will be taken.
Corrective actions will also be recorded and reviewed.

Producer D: Product Description

HACCP Plan No. 1 Product Category: 100% Apple Cider

1. Common name?
100 % Apple Cider

2. How is it to be used?
Consumed as purchased (ready-to-drink)

3. Type of package?
Plastic bottling (high density polyethylene)

4. Length of shelf life, at what temperature?
Approx. 40 days if not opened
Maximum acceptable storage temperature 42°F
Recommend ≤38°F

5. Where will it be sold?
Retail

6. Labeling instruction?
- Label should say "Keep Cold"
- Ingredients, nutrition facts, net content, "Use By" / "Sell By" and Date of Production are recommended
- Warning label required for wholesale

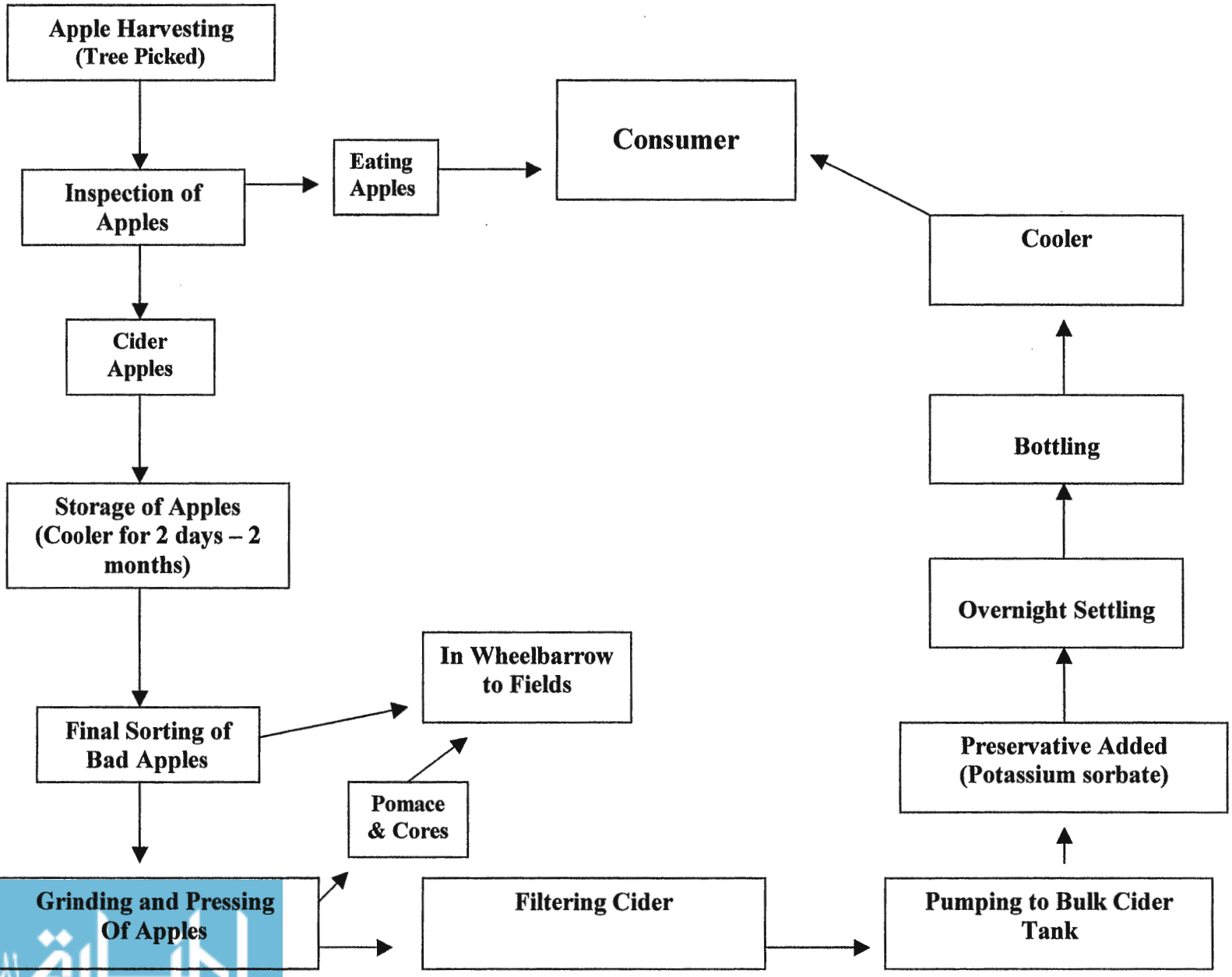
7. Is special distribution control needed?
Lot code or date of production control needed for traceability
Distribution and storage under acceptable refrigeration (maximum recommended temperature 42°F)
Use of temperature monitoring devices recommended

Approved by: _____ Date approved: _____

List Product Ingredients

Apples, Potassium Sorbate

PRODUCER D: FLOW DIAGRAM FOR APPLE CIDER PRODUCTION



Producer D
HACCP Plan No. __1_____ Process category _____

Product: !00% Fresh Unpasteurized Apple Cider

Hazard Analysis and Identification of Critical Control Points

Processing Step		Potential Hazards Introduced	What Control Measures Can be Applied To Prevent The Hazard?		Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?	CCP#
Raw Ingredients Apples, Receiving and Inspection	B	Pathogens	-Inspection of apples (own and suppliers)	B	YES	1
	P	Wood	-No drops -Grate separates large debris	P	NO	
	C	Pesticides	-Remove visibly spoiled and damaged apples	C	NO	
Storage of apples	B	Pathogens Mold growth Cross-contamination of good and bad apples	-Storage inside cooler 35-42°F, no outside or uncovered storage Re-inspection weekly to remove bad apples	B	YES	2
	C	None		C	NO	
	P	Rodents, Insects	-Pest Control (GMP)	P	NO	
Final Sorting	B	None	-Good employee hygiene and GMP's	B	NO	
	C	None	-Removal of visibly spoiled and damaged apples	C	NO	
	P	None		P	NO	
Grinding and Pressing	B	Contamination from dirty equipment	-Ensure equipment is properly cleaned (SSOP)	B	NO	
	C	Chemical residues from cleaning	-Properly rinse equipment after cleaning	C	NO	
	P	Metal debris from machine	-Filter Cider	P	NO	
Filtering (stainless steel filter)	B	None	-Check filter daily for visible signs of damage	B	NO	
	C	None		C	NO	
	P	None		P	NO	
Pumping Cider to Cooled Settling Tank	B	Contamination from dirty equipment	-Ensure clean tubing and tank (SSOP)	B	NO	
	C	None		C	NO	
	P	None		P	NO	

Processing Step		Potential Hazards Introduced	What Control Measures Can be Applied To Prevent The Hazard?		Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?	CCP#
Add Preservative (Pot.Sorbate)	B	None	-Good record keeping with SOP, monitoring and maintenance of weighing scales	B	NO	
	C	Chemical limits exceeded		C	NO	
	P	None		P	NO	
Overnight Settling in Holding Tank	B	Pathogens if improper holding temperature and not properly enclosed	-Monitor temperature -Tank is properly covered and sealed	B	NO	
	C	None		C	NO	
	P	None		P	NO	
Bottling	B	Pathogens	-Trough is covered or enclosed -Capping performed in timely manner - containers not allowed to sit in open environment longer than 5 minutes -Visual inspection of bottles for foreign materials -Checked for proper sealing (SOP)	B	YES	3
	C	Introduction of chemical hazards		C	NO	
	P	Introduction of physical hazards		P	NO	
Cooling	B	Improper cooling allowing pathogen growth	-Bottles are sent to cooler in a timely manner -Monitor cooler temperature 35-42°F	B	NO	
	C	None		C	NO	
	P	None		P	NO	
Storage	B	Improper refrigeration temperature promotes bacterial growth	-Monitor storage temperature	B	NO	
	C	None		C	NO	
	P	None		P	NO	

pproved by: _____ Date approved: _____

Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 1 (B) **Receiving and Inspection**	Pathogenic Bacteria (Prevention and Destruction)	- No Drops - No domestic manure in orchards - Removal of Rotten or Spoiled looking Apples	Apple harvesting and shipments	Visual	Each harvest or shipment	Receiving Manager	No Drops Policy Reject Drops
CCP 2 (B)(P) **Storage of Apples**	(B) Pathogens and mold growth	Storage inside cooler 35-42°F	Cooler temperature maintained	Temperature gauge – visual	Daily	Manager	Correct cooling problem, recalibrate and monitor hourly for next 6 hours. Move to other cooler if temperature has not been maintained within 6 hrs.
		Remove spoiled apples weekly	Apple Spoilage	Weekly Removal	Weekly	Manager	
	(P) Rodents and Insects	No outside or uncovered storage	Spoiled Apples	Visual	Weekly	Manager	Discard any spoiled apples
CCP 3 (B)(P) **Bottling**	(B) Pathogens	Cap within 5 minutes of filling	Length of time containers are exposed to open air environment	By capping bottle immediately after it is filled	Every container	Operator	Dispose of bottled cider
		Proper Seal achieved	Proper sealing of container	Visual	Every container	Operator	Reseal if necessary
	(P) Environmental debris	No visible debris	Physical contamination	Visual	Every container	Operator	Dispose of bottled cider

Producer D: Verification Activities and Recording Procedures

Verification Activities	Record-Keeping Procedures
CCP #1 – Apple Receiving and Inspection All records will be reviewed and initialed and dated weekly	- Documentation at harvest (tree-picked vs drops) - Valid records for sorting and separation of spoiled apples - Audits of outside apple suppliers - Log documenting any shipments of apples that are rejected
CCP #2 – Storage of Apples Weekly culling of apples in storage Daily verification of cooler temperature before start-up and calibration if necessary. All records will be initialed and dated	- Cooler log which includes temperature data - Calibration records for thermometers - Log verifying that apples were stored under proper conditions and were inspected weekly
CCP #3 – Bottling All records will be reviewed and initialed and dated weekly	- Log verifying trough was covered - Bottles capped timely and sealing documentation - Log documenting discarded product due to biological and physical contamination - Verification of thermometers in cooler (calibration if necessary)

Verification of our overall process may also include microbial testing for coliforms/*E.coli*
All deviations of CCPs will be recorded and corrective actions will be taken.
Corrective actions will also be recorded and reviewed.

Producer E: Product Description

HACCP Plan No. 1 Product Category: 100% Apple Cider

1. Common name?
Pasteurized 100% Apple Cider

2. How is it to be used?
Consumed as purchased (ready-to-drink)

3. Type of package?
Plastic bottling (high density polyethylene)

4. Length of shelf life, at what temperature?
Approx. 40 days if not opened
Maximum acceptable storage temperature 42°F
Recommend ≤38°F

5. Where will it be sold?
Retail

6. Labeling instruction?
- Label should say "Keep Cold"
- Ingredients, nutrition facts, net content, "Use By" / "Sell By" and Date of Production are recommended
- Warning label required for wholesale

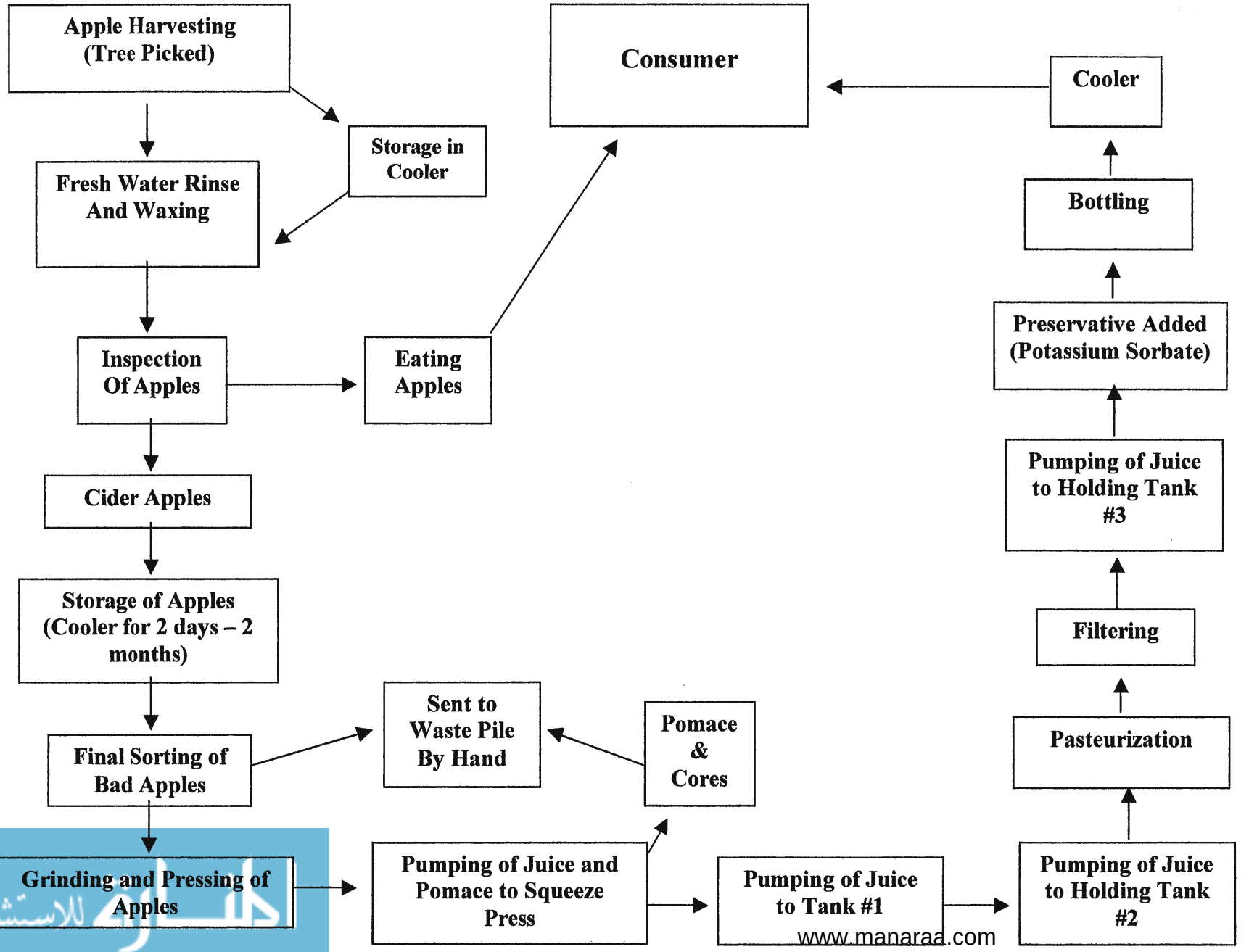
7. Is special distribution control needed?
Lot code or date of production control needed for traceability
Distribution and storage under acceptable refrigeration (maximum recommended temperature 42°F)
Use of temperature monitoring devices recommended

Approved by: _____ Date approved: _____

List Product Ingredients

Apples, Potassium Sorbate

PRODUCER E: FLOW DIAGRAM FOR APPLE CIDER PRODUCTION



Producer E: Hazard Analysis

HACCP Plan No. _1_____ Process category _____

Product: Pasteurized 100% Apple Cider

Hazard Analysis and Identification of Critical Control Points

Processing Step		Potential Hazards Introduced	What Control Measures Can be Applied To Prevent The Hazard?		Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?	CCP#
Raw Ingredients Apples: Harvesting/ Receiving and Inspection	B P C	Pathogens Wood Pesticides	-No drops -Inspection of apples (own and suppliers) -Certified Supplier audits -Grate separates large debris -Remove visibly spoiled and damaged apples	B P C	NO (Pasteurization at later step) NO NO	
Fresh Water Rinse and Waxing of Apples	B C P	Pathogens, Parasites Metals, Pesticides, Nitrites None	- Potable Water - Guaranteed testing by municipal utilities - Food grade wax only	B C P	NO NO NO	
Storage of Apples	B C P	Pathogens Mold growth Cross-contamination of bad to good apples None Rodents, Insects	-Storage inside cooler 35-40°F, no outside or uncovered storage -Re-inspection weekly to remove bad apples -Pest Control (GMP)	B C P	NO (Pasteurization at later step) NO NO	
Final Sorting	B C P	None None None	-Good employee hygiene and GMP's -Removal of visibly spoiled and damaged apples	B C P	NO NO NO	
Grinding and Pressing	B C P	Contamination from dirty equipment Cleaning residues Metal debris from machine	-Ensure proper cleaning of equipment (SOP) -Properly rinse equipment after cleaning and/or before start up -Filter Cider	B C P	NO (Pasteurization at later step) NO NO	

Processing Step		Potential Hazards Introduced	What Control Measures Can be Applied To Prevent The Hazard?		Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?	CCP#
Pumping Cider to Press	B	Contamination from dirty equipment	-Ensure clean tubing and press (SOP)	B	NO	
	C	Cleaning residues	- Properly rinse equipment after cleaning and/or before start up	C	NO	
	P	None		P	NO	
Pumping Cider to Holding Tanks	B	Contamination from dirty equipment	- Ensure clean tubing (SOP)	B	NO	
	C	Cleaning residues	- Properly rinse equipment after cleaning and/or before start up	C	NO	
	P	None		P	NO	
Pasteurization	B	Pathogen destruction	-Flash Pasteurization at 162 F for 11 seconds	B	YES	1
	C	None		C	NO	
	P	None		P	NO	
Filtering	B	None	-Check filter daily for visible signs of damage	B	NO	
	C	None	-Replace as necessary	C	NO	
	P	None		P	NO	
Bottling	B	Pathogens	-Bottling tube is cleaned and sanitized (SOP)	B	YES	2
	C	Introduction of chemical hazards	-Capping performed in timely manner - containers not allowed to sit open -Preservative is fully dissolved before bottling	C	NO	
	P	Introduction of physical hazards	-Visual inspection of bottles for foreign materials -Checked for proper sealing (SOP)	P	NO	
Add Preservative (Pot.Sorbate)	B	None	-Good record keeping with SOP, monitoring and maintenance of weighing scales -Allow preservative to fully dissolve	B	NO	
	C	Chemical limits exceeded		C	NO	
	P	None		P	NO	
Cooling	B	Improper cooling allowing pathogen growth	-Bottles are sent to cooler immediately after sealing -Monitor cooler temperature 35-40°F	B	NO	
	C	None		C	NO	
	P	None		P	NO	

Processing Step		**Potential Hazards Introduced**	**What Control Measures Can be Applied To Prevent The Hazard?**		**Is The Potential Safety Hazard Significant and Reasonable Likely To Occur?**	**CCP#**
Storage	B C P	Improper refrigeration temperature promotes bacterial growth	-Monitor storage and transportation temperature 35 – 40 F	B C P	NO NO NO	

Approved by: _____ Date approved: _____

Producer E: Critical Limits

Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 1 (B) **Pasteurization**	Pathogenic Bacteria (Destruction)	Product Target Time/Temp: 162°F for 11 sec. Minimum temp: 160°F (alarm will sound)	Temperature of cider at exit of holding tube	Temperature recorder at end of hold tube Low temperature manual divert valve	Continuous recording Temp. should also be recorded manually every restart as a comparison record	Pasteurizer operator	Cider will be manually diverted if temp. at end of holding tube is low; cider will be re-pasteurized If divert valve does not work; production will be stopped and the portion of cider in the holding tube and final holding tank will be re-pasteurized Verification of temperature gauge and alarm system to ensure deviation does not occur again Questionable cider may be tested for pathogen survivors before being shipped If seals are broken or gauge is not working properly; Stop processing, recalibrate pump and/or reseal
			Seals on pumps and tubing and flow rate gauge	Visual check	At start up and once during processing	Pasteurizer operator	

Critical Control Point (CCP)	Hazard(s) to be Addressed in HACCP Plan	Critical Limits for each Control Measure	Monitoring				Corrective Action
			What	How	Frequency	Who	
CCP 2 (B) (P) (C) **Bottling**	(B) Pathogens	Cap immediately after filling	Length of time containers are exposed to open air environment	By capping immediately once bottle is full of cider	Every container	Operator	Dispose of bottled cider
		Proper Seal achieved	Proper sealing of container	Visual	Every container	Operator	Reseal if necessary
		Bottles sent to cooler in a timely manner	Length of time bottles are setting at room temperature	Time	Get estimate time for every pallet of bottled cider	Operator	Dispose of bottled cider if has set at room temperature longer than 1 hour
	(P) Environmental debris	No visible debris	Physical contamination	Visual	Every container	Operator	Dispose of bottled cider
	(C) Preservative concentration per bottle of cider	Preservative is fully dissolved before bottling	Dissolved potassium sorbate granules	Visual	Every container	Operator	Pour cider back into tank and fill bottle once preservative is fully dissolved

Producer E: Verification and Record Keeping

Verification Activities	Record-Keeping Procedures
CCP #1 Pasteurization 1. Verification of thermometers daily before start-up and calibration if necessary. 2. Calibration of divert valves and alarm system at least once a week 3. Holding tube length and diameter are tested once per season with a tracer test to validate the residence time of the cider 4. Check pump flow rate gauge daily	- Pasteurization log, which includes temperature data, any deviations, etc. - Pasteurizer operator compares continuous temp. records and the manual temp. recording data on a daily basis - Calibration records for the thermometers, divert valve, alarm system, etc - Flow verification log (pump flow rate info) - Corrective action logs - Production manager will review and initial records daily
CCP #2 Bottling 1. Manager reviews records weekly	- Log verifying bottling tubing was clean and sanitized before use - Bottles capped timely and sealing documentation - Documentation of weight/volume used for preservative (dissolved fully before bottling) - Log documenting discarded product due to biological and/or physical contamination - Corrective action logs

Verification of overall process may also include microbial testing for coliforms/*E.coli.* All deviations of CCPs will be recorded and corrective actions will be taken. Corrective actions will also be recorded and reviewed.

APPENDIX B

RAW DATA

Producer A 1999-2000 Results (CFU/Apple or CFU/ml)

23-Sep			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
No Preservative	n/a	n/a	n/a
With Preservative	930	20,000	600
Final Cider	270	10 est	<10 est
Final Cider (2wk check)	290	15 est	<10 est

11-Nov			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
Apple 1	10,000,000	760,000	140,000
Apple 2	2,600,000	37,000	110,000
No Preservative	5,800	53,000 (27% molds)	200 est
With Preservative	2,200 est	61,000 (11% molds)	15 est
Final Cider	100 est	10 (0% molds) est	<1 est
Final Cider (2wk check)	70 est	3,500 (1% molds)	<1 est

2-Dec			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
Apple 1	510,000	45,000	500 est
Apple 2	7,800,000	1,400,000	17,000
No Preservative	2,600	23,000 (31% molds) est	25
With Preservative	1,700 est	80,000 (1% molds)	75
Final Cider	15 est	5 (0% molds) est	<1 est
Final Cider (2wk check)	35 est	<100 est (0% molds)	<1 est

16-Dec			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
Apple 1	430,000	93,000	15,000
Apple 2	50,000	160,000	1,900 est
Apple 3	100,000	860,000	33,000
Apple 4	91,000	230,000	1,800 est

Producer A 2000-2001 Results (cfu/ml cider, cfu/apple)

Sept. 28				
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**	**E. coli**
Apple 1 unwashed	1,700,000	72,000	<100 est	<100 est
Apple 2	530,000	81,000	<100 est	<100 est
Apple 3	2,400,000	130,000	50 est	<100 est
Apple 4	860,000	2,500,000	2,100	<100 est
No Preservative	5,200	15,000 (16% molds)	150 est	<10 est
No Preservative (2 wk)	2,300	18,000 est (26% molds)	<10 est	<10 est
With Preservative	9,200	24,000 (16 % molds)	55 est	<10 est
With Preservative (2wk)	2,200 est	9,700 (11% molds)	<10 est	<10 est
Final Cider	410 est	<10 est (0% molds)	<10 est	<10 est
Final Cider (2 wk)	160 est	10 est (0% molds)	<10 est	<10 est

Nov. 1					Non gas producers
TEST:	Aerobic Bacteria	Yeasts & Molds	Coliforms	E.coli	on petrifilm
Apple 1	12,000,000	6,200,000	<100 est	1,000 est	43,000
Apple 2	9,200,000	11,000,000	150 est	<100 est	160,000
Apple 3	2,600,000	5,300,000	100 est	<100 est	150,000
Apple 4	3,100,000	39,000,000	<100 est	<100 est	120,000
Apple 5	6,000,000	11,000,000	1400 est	<100 est	500,000
Apple 6	2,600,000	3,000,000	800 est	<100 est	51,000
Apple 7	6,700,000	4,900,000	750 est	1,000 est	1,200,000
Apple 8	18,000,000	10,000,000	2,800	50 est	33,000
No Preservative	n/a	n/a	n/a	n/a	n/a
With Preservative	6,500	120,000 (3% molds)	55 est	<10 est	830
With Preservative (2wk)	1100 est	54,000 (1% molds)	<10 est	<10 est	5 est
With Preservative (4 wk)	200 est	28,000 (2 % molds)	<10 est	<10 est	<10 est
With Preservative (6 wk)	50 est	100,000 (0% molds)	<10 est	<10 est	<10 est
Cider (Final)	150 est	15 est (0% molds)	<10 est	<10 est	<10 est
Final Cider (2wk)	990 est	150 est (0% molds)	<10 est	<10 est	<10 est
Final Cider (4 wk)	50 est	<100 est (0% molds)	<10 est	<10 est	<10 est
Final Cider (6 wk)	120 est	<10 est (0% molds)	<10 est	<10 est	<10 est

Nov. 29					Non gas producers
TEST:	Aerobic Bacteria	Yeasts & Molds	Coliforms	E.coli	on petrifilm
Apple 1	1,100,000	960,000	<100 est	<100 est	850 est
Apple 2	10,000 est	230,000	<100 est	<100 est	3,500
Apple 3	390,000	990,000	<100 est	<100 est	36,000
Apple 4	620,000	940,000	<100 est	<100 est	3,000
Apple 5	3,100,000	6,400,000	1,200 est	<100 est	87,000
Apple 6	15,000,000	12,300,000	15,000	50 est	250,000
Apple 7	1,800,000	3,500,000	450 est	<100 est	30,000
Apple 8	3,600,000	5,900,000	1,800 est	<100 est	11,000
No Preservative	6,200	120,000 (7 % molds)	180 est	<10 est	210 est
No Preservative (2 wk)	2,900	100,000 (10% molds)	70 est	<10 est	50 est
With Preservative	8,700	63,000 (11 % molds)	20 est	<10 est	25 est
With Preserv. (2wk)	9,600	46,000 (22% molds)	<10 est	<10 est	1,500
With preserv. (4 wk)	550	300,000(5% molds)	<10 est	<10 est	<10 est
With Preserv. (8 wk)	5 est	370,000 (3 % molds)	n/a	n/a	n/a
Final Cider	340	730 (1 % molds)	<10 est	<10 est	5 est
Final Cider (2wk)	160 est	300 (14 % molds)	<10 est	<10 est	<10 est
Final Cider (4 wk)	110 est	<100 est (0% molds)	<10 est	<10 est	<10 est
Final Cider (8 wk)	10 est	<10 est (0 % molds)	n/a	n/a	n/a

Dec. 1					Non gas producers
TEST:	Aerobic Bacteria	Yeasts & Molds	Coliforms	E.coli	on petrifilm
Apple 1	3,000,000	9,900,000	<100 est	<100 est	300,000
Apple 2	660,000	2,400,000	<100 est	<100 est	63,000
Apple 3	310,000	580,000	<100 est	<100 est	200 est
Apple 4	800,000	1,900,000	<100 est	<100 est	200 est
Apple 5	1,700,000	2,000,000	23,000	<100 est	78,000
Apple 6	26,000,000	7,100,000	3,400	<100 est	500,000
Apple 7	26,000,000	7,600,000	2,900	<100 est	200,000
Apple 8	4,500,000	3,400,000	3,300	<100 est	81,000
No Preservative	n/a	n/a	n/a	n/a	n/a
With Preservative	4,100	46,000 (11% molds)	90 est	<10 est	30 est
With Preserv. (2wk)	930	33,000 (18 % molds)	<10 est	<10 est	850
With preserv. (4 wk)	280	70,000 (6 % molds)	<10 est	<10 est	<10 est
With Preserv. (8 wk)	5 est	49,000 (8 % molds)	n/a	n/a	n/a
With Preserv. (10 wk)	35 est	4,000 (100 %molds)	n/a	n/a	n/a
Final Cider	120 est	20 est (25% molds)	<10 est	<10 est	<10 est
Final Cider (2wk)	120 est	640 (0% molds)	<10 est	<10 est	<10 est
Final Cider (4 wk)	75 est	<100 est	<10 est	<10 est	<10 est
Final Cider (8 wk)	10 est	<100 est	n/a	n/a	n/a
Final Cider (10 wk)	20 est	<10 est	n/a	n/a	n/a
Final Cider (12 wk)	40 est	<10 est	n/a	n/a	n/a

Producer B 1999-2000 Results (cfu/ml cider, cfu/apple)

29-Sep			
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**
No Preservative	n/a	n/a	n/a
With Preservative	55,000	190,000 (3% molds)	<100 est
Cider (Final)	100 est	50 est (0% molds)	<1 est
Final Cider (2wk)	130 est	<10 est (0% molds)	<1 est

27-Oct				
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**	**E.coli**
Apple 1	2,500,000	1,700,000	100,000	300 est
Apple 2	150,000	520,000	600 est	<100 est
No Preservative	20,000	25,000 (8% molds)	150 est	<10 est
With Preservative	7,700	28,000 (6% molds)	35 est	<10 est
Cider (Final)	35	<10 est (0 % molds)	<10 est	<10 est
Final Cider (2wk)	20 est	<10 est (0% molds)	n/a	n/a

11-Jan			
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**
Apple 1	420,000	250,000	73,000
Apple 2	900,000	270,000	4,000
Apple 3	2,000,000	330,000	180,000
Apple 4	700,000	200,000 est	40,000
No Preservative	n/a	n/a	n/a
With Preservative	17,000	16,000 est (100% molds)	37
W/Preserv. (2wk)	2,000 est	2,300 est (100% molds)	4 est
W/Preserv. (4wk)	2,500	3,500 (7% molds)	<1 est
Final Cider	1,600	500 est (0% molds)	2 est
Final Cider (2wk)	1,200 est	<100 est (0% molds)	<1 est
Final Cider (4wk)	610	<100 est (0 % molds)	<1 est
Final Cider (7wk)	140 est	<10 es (0% molds)t	<1 est

Producer C 1999-2000 Results (cfu/ml, cfu/apple)

7-Oct			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
No Preservative	10,000	17,000 est (36% molds)	2,200
Final Cider	210	1,100 est (9% molds)	40
Final Cider (2wk)	35 est	<100 est (0% molds)	<10 est

14-Oct			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
No Preservative	n/a	n/a	n/a
Final Cider	11,000	8,000 est (4% molds)	1,300 est
Final Cider (2wk)	26,000	<100 est (0% molds)	<10 est

31-Oct			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
Apple 1	200,000	150,000 est	300 est
Apple 2	650,000	900,000 est	16,000 est
No Preservative	16,000	52,000 (55% molds)	40 est
Cider (Final)	3,900	5,000 (0% molds)	38 est
Check on Final Cider (2wk)	800 est	<100 est (0% molds)	6 est

16-Dec			
TEST:	**Total Aerobic**	**Yeasts & Molds**	**Coliforms**
Apple 1	360,000	160,000 est	2,400
Apple 2	160,000 est	20,000 est	1,000 est

Producer C 2000-2001 Results (cfu/ml cider, cfu/apple)

Sept. 28				
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**	***E. coli***
Apple 1	710,000	170,000	150 est	<100 est
Apple 2	48,000	150,000	<100 est	<100 est
Apple 3	1,800,000	980,000	<100 est	<100 est
No Preservative	110,000	230,000 (3% molds)	75 est.	<10 est
No preservative (2wk)	200,000	480,000 (1% molds)	50 est	<10 est
Final Cider	16,000	79,000 (1 % molds)	<10 est	<10 est
Final Cider (2wk)	450 est	47,000 (1 % molds)	<10 est	<10 est

Oct. 11					Non Gas producers
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**	***E.coli***	**on petrifilm**
Apple 1	15,000,000	5,200,000	<100 est	<100 est	<100 est
Apple 2	12,000,000	1,200,000 est	34,000	<100 est	<100 est
Apple 3	270,000	30,000	<100 est	<100 est	<100 est
No Preservative	12,000	66,000 (28 % molds)	140 est	<10 est	2,700
No Preservative (2wk)	21,000	14,000,000 (0 % molds)	50 est	<10 est	1,300
Final Cider	9,900	88,000 (0 % molds)	<10 est	<10 est	<10 est
Final Cider (2wk)	680	9,500 (0% molds)	<10 est	<10 est	<10 est
Final Cider (4 wk check)	600 est	60,000 (99% molds)	<10 est	<10 est	<10 est

Producer D 1999-2000 Results (cfu/ml, cfu/apple)

12-Oct			
TEST:	Total Aerobic	Yeasts & Molds	Coliforms
No Preservative	220,000	61,000 (1% molds)	210,000
Final Cider	4,000	73,000 (1% molds)	5 est

1-Dec			
Bottled 12/8			
TEST:	Total Aerobic	Yeasts & Molds	Coliforms
Apple 1	40,000 est	50,000	3,000
Apple 2	26,000,000 est	3,400,000	2,000,000
No Preservative	340,000	70,000 (27% molds)	140,000
No Preserv. (2 wk check)	150,000	100,000 (10% molds)	34,000
Final Cider	220	6,100 (1% molds)	<10 est
Final Cider (2wk check)	130 est	7,500 (0% molds)	<1 est

11-Jan			
Made 1/11			
TEST:	Total Aerobic	Yeasts & Molds	Coliforms
Apple 1	4,900,000	310,000	9,900
Apple 2	850,000	240,000	1,300
Apple 3	2,400,000	1,200,000	1,700,000
Apple 4	12,000,000	2,600,000	590,000
No Preservative	110,000	20,000 (95% molds) est	5,400
No Preserv. (2wk check)	17000	20,000 (72% molds) est	2 est
No. Preserv (4 wk check)	13,000,000	980,000 (9% molds)	5 est
Final Cider (4 wk check)	20,000	3,900 (0% molds)	6 est
Final Cider (6 wk check)	300 est	17,000 (0% molds)	14 est

TEST	TSA	PDA	Coliforms
Made 12/27 Bottled 1/7			
Final Cider	850	3,000 (32% molds)	30
Final Cider (4 wk check)	15,000	3,500 (7 % molds)	<1 est
Final Cider (6 week check)	390,000	1,900 (3 % molds)	<1 est

Producer D 2000-2001 Results (cfu/ml cider, cfu/apple)

Nov. 12					
TEST:	Aerobic Bacteria	Yeasts & Molds	Coliforms	E. coli	Non gas producers on petrifilm
Apple 1	660,000	2,800,000	<100 est	<100 est	700 est
Apple 2	510,000	630,000	<100 est	<100 est	36,000
Apple 3	1,400,000	420,000	<100 est	<100 est	37,000
Apple 4	26,000,000	9,400,000	11,000	<100 est	450,000 est
Apple 5	30,000,000 est	4,700,000	26,000	<100 est	300,000 est
Apple 6	40,000,000 est	12,000,000	2,000	<100 est	340,000 est
Apple 7	51,000	33,000	<100 est	<100 est	1,600 est
No Preservative	240,000	550,000 (34% molds)	380	<10 est	40,000 est
No preserv. (2wk check)	1,100,000	6,600,000 (2% molds)	60 est	<10 est	1200
Final Cider	110,000	390,000 (19% molds)	180 est	<10 est	430
Final Cider (2wk check)	23,000	160,000 (6% molds)	<10 est	<10 est	280
Final Cider (4 wk check)	1,200	360,000 (5% molds)	<10 est	<10 est	5 est

Made 12/1 Bottled 12/8					
TEST:	Aerobic Bacteria	Yeasts & Molds	Coliforms	E.coli	Non gas producers on petrifilm
Apple 1	5,900,000	910,000	6,500 est	<100 est	130,000
Apple 2	13,000,000	580,000	<100 est	<100 est	38,000
Apple 3	480,000	870,000	<100 est	<100 est	3,700
Apple 4	13,000,000	1,580,000	100 est	<100 est	250,000 est
No Preservative	n/a	n/a	n/a	n/a	n/a
Final Cider	13,000	140,000 (3 % molds)	<10 est	<10 est	480
Final Cider (3wk check)	11,000	16,000 (6 % molds)	<10 est	<10 est	390
Final Cider (5 wk check)	400	1,500,000 (0 % molds)	<10 est	<10 est	<10 est

Made 12/22 Bottled 12/30					
TEST:	Aerobic Bacteria	Yeasts & Molds	Coliforms	E.coli	Non gas producers on petrifilm
No Preservative	5,400	120,000 (25 % molds)	5 est	<10 est	100 est
No Preserv. (2 wk check)	gassy	gassy	gassy	gassy	gassy
Final Cider	3,500	5,600 (61 % molds)	<10 est	<10 est	<10 est
Final (4 wk check)	110 est	3,000 (29 % molds)	<10 est	<10 est	<10 est
Final (8 wk check)	170 est	1,400 (100% molds)	n/a	n/a	n/a
Final (12 wk check)	270 est	4,300 (24% molds)	n/a	n/a	n/a

Producer E 2000-2001 Results (cfu/ml cider, cfu/apple)

Oct. 2				
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**	**E. coli**
Apple 1	170,000	25,000 est	<100 est	<100 est
Apple 2	540,000	20,000 est	<100 est	<100 est
Apple 3	130,000	45,000 est	<100 est	<100 est
No Preservative	230,000	63,000 (4 % molds)	4,000	10 est
No Preserv. (2wk check)	120,000	220,000 (5% molds)	1,100	5 est
Final Cider	210 est	25 est (20% molds)	<10 est	<10 est
Final Cider (2 wk check)	1,000	<100 est (0% molds)	<10 est	<10 est

Oct. 20				
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**	**E.coli**
Apple 1	94,000	25,000	<100 est	<100 est
Apple 2	1,500,000	190,000	<100 est	<100 est
Apple 3	1,700,000	150,000 est	<100 est	<100 est
Apple 4	850,000	40,000	<100 est	<100 est
No Preservative	110,000	76,000 (5 % molds)	930	<10 est
No Preserv. (2 wk check)	gassy	gassy	gassy	gassy
Final Cider	410	5 est (0% molds)	<10 est	<10 est
Final Cider (2 wk check)	300	<10 est (0% molds)	<10 est	<10 est

Dec. 1					Non gas producers
TEST:	**Aerobic Bacteria**	**Yeasts & Molds**	**Coliforms**	**E.coli**	**on petrifilm**
Apple 1	410,000	640,000	<100 est	<100 est	9,900
Apple 2	500,000	700,000	<100 est	<100 est	4,800
Apple 3	330,000	610,000	<100 est	<100 est	1,300 est
Apple 4	470,000	710,000	<100 est	<100 est	3,000
No Preservative	7,000 est	19,000 est(47% molds)	60 est	<10 est	70 est
No Preserv. (2 wk check)	20,000	130,000 (9 % molds)	<10 est	<10 est	35 est
Final Cider	520	<10 est	<10 est	<10 est	<10 est
Final Cider (2 wk check)	530	<10 est	<10 est	<10 est	<10 est
Final Cider (4 wk check)	460	50 est (0% molds)	<10 est	<10 est	<10 est
Final Cider (8 wk check)	<10 est	<100 est	n/a	n/a	n/a
Final Cider (10 wk check)	<10 est	<10 est	n/a	n/a	n/a
Final Cider (14 wk check)	5 est	<10 est	n/a	n/a	n/a

Producer A 1999-2000 ENVIRONMENTAL SWAB EVALUATIONS

LOCATION	Yeasts & Molds	Aerobic Bacteria	Coliforms	OBSERVATION
Side of Pasteurized Tank (per cm^2)	<1	<1	<1	
Meshy belt of Press (after cleaning) per cm^2	<1	<1	<1	
Nozzle of jug filler (side of) per cm^2	<1	0.5	<1	
Drip from Pasteurized Tank (per ml)	150	370	<10	Was taken care of
Unused lid (entire area of lid)	<10	<10	<1	
Steel tray in Press (after cleaning) per cm^2	0.25	0.5	<1	Had some visible cider/ apple debris
Water from apple rinse (1) per ml	<10	400	2	
Water from apple rinse (2) per ml	<10	50	1	

Assessment: Equipment seemed to be fairly clear of debris. Very low counts of microorganisms were observed on the equipment. Therefore, methods for cleaning and sanitizing are sufficient. The drip from the pasteurized cider tank had higher counts, but at later visits this problem was observed to be taken care of. The water used in the fresh water rinse may be questionable due to aerobic bacteria counts found on one occasion.

Producer A 2000-2001 ENVIRONMENTAL SWAB EVALUATIONS				
LOCATION	Yeasts and Molds	Aerobic Bacteria	Coliforms	OBSERVATIONS
Paddle used to stir flavoring (before use) per cm2	1.5 est	1.5 est	<1 est	Stored on the apple press
Water from Hose in Processing Room (per ml)	<10 est	780	<1 est	
Apple Belt (From Washer to Elevator) per cm2	1 est	<1 est	<1 est	After cleaning
Water from Hose in Processing Room (per ml)	<10 est	480	<1 est	
Paddle used to stir flavoring (before use) per cm2	<1 est	<1 est	<1 est	
Tip of Bottler Tube	<1 est	<1 est	<1 est	
Water from Hose in Processing Room (per ml)	<10 est	2200	<1 est	
Water from Faucet in Processing Room (per ml)	<10 est	280	<1 est	
Water from Hose in Processing Room (per ml)	<10 est	240	<1 est	

ASSESSMENT: The equipment seemed fairly clear from debris and from the results of the swabs, was clean of bacteria and yeasts and molds. The processing room, bottling room, and coolers are well maintained and free from clutter. However, the water used in processing had various levels of bacteria detected. Due to the levels of bacteria found in the water, there were increased levels of bacteria found on the apples that were washed in the water. It would be recommended to test the levels of chlorine on a more regular basis (perhaps every 3-4 months) to ensure that the proper level of chlorine is being maintained. If levels are low, chlorinating of the water should be performed. Doing this would ensure that the water itself would not contaminate anything that comes in contact with the water.

Producer B 1999-2000 SWAB EVALUATIONS

LOCATION	Yeasts and Molds	Aerobic Bacteria	Coliforms	OBSERVATION
Tip of Bottler Spout (after cleaning)	5 est	10 est	<1 est	
Water tank where apples soak (counts per ml)	11,000	59,000	9,000	Needs to be changed regularly
Capper machine (where caps slide down)	55 est	15 est	<1 est	
Tip of Bottler Spout (during use)	20 est	5 est	<1 est	
Drip from lip of Pasteurizer (counts per ml)	1,500	340,000	120	Need to move bottling system
Tip of Bottler Spout (after cleaning)	130 est	40 est	<1 est	

Assessment: The water tank in which the apples are washed in had extremely high levels of microorganisms. The water needs to be changed every day and possibly more often if apples are abnormally dirty. The chlorination levels also need to be regulated. The drip from the pasteurizer also had high microbial loads. The drips were condensation from the tank that were dripping directly above the bottling system and capping machine. The drip needs to be fixed or the bottling system needs to be moved to prevent contamination into the final product. The bottling spouts also need to be cleaned and sanitized before and after use to prevent post-processing contamination. Overall, sanitation regimes need to be increased to keep microbial loads in the processing environment low.

Producer C 1999-2000 ENVIRONMENTAL SWAB EVALUATIONS				
LOCATION	Yeasts and Molds	Aerobic Bacteria	Coliforms	OBSERVATIONS
Water (processing room) count per ml	<10 est	<10 est	<1 est	Chlorinated rural water
Water from apple rinse (count per ml)	<10 est	> 2,500 est	<1 est	
Apple elevator (after cleaning) count per cm2	34	8 est	< 0.1 est	
Steel Roller in Press (during production) per cm2	87	10 est	1 est	
Sponge in Press (during production)	3 est	4 est	< 0.1 est	
Steel Holding Tank (after cleaning) per cm2	< 0.5 est	< 0.5 est	< 0.05 est	
Bottler Tip	5 est	10 est	<1 est	
Unused lid (count per lid)	<10 est	10 est	<1 est	

ASSESSMENT:
The equipment appeared to be in clean of debris. High counts were only observed in the water from the apple rinse. Contamination most likely came from the equipment rather than the water itself. Based on the results the cleaning regimes seem to be effective. The processing room was well-kept and the storage room was clean and organized. Restrooms and cleaning facilities were well-kept and assessable. Doors were open during production and chemicals/paint were stored above the apple grader. One employee was found to be smoking in the processing room during production. The operator was notified about the possible improvements and the chemicals/ paints were removed from the area.

Producer C 2000-2001 ENVIRONMENTAL SWAB EVALUATIONS				
LOCATION	Yeasts and Molds	Aerobic Bacteria	Coliforms	OBSERVATIONS
Water (processing room) count per ml	<10 est	30 est	<1 est	Chlorinated rural water
Water from apple rinse (count per ml)	<10 est	<10 est	<1 est	Incorporated a sanitizer in rinse
Apple elevator (after cleaning) count per cm^2	<1 est	1 est	< .1 est	
Unused lid (count per lid)	5 est	10 est	<1 est	

ASSESSMENT: All of the equipment was clean of debris and appeared in good condition. Microbial counts were all low. Changes in the process from the previous year consisted of incorporating a sanitizer rinse on the apples prior to storage.

Producer D 1999-2000 SWAB EVALUATIONS

LOCATION	Yeasts and Molds	Aerobic Bacteria	Coliforms	OBSERVATION:
Belt of Press (during production) per cm2	8.4	140	25	
Steel filter (during production) per cm2	31	174	75	
Inside of Steel Bottling Trough (before use) per cm2	.5 est	.8 est	<1 est	
Steel Paddle (used to stir preserv.) per cm2	8 est	4 est	<1 est	Had some visible apple/cider debris on it
Tip of Bottler (after cleaning)	190	25	<1 est	

Assessment: The equipment was found to be fairly clear of debris – however, a more intensive cleaning program could be incorporated. The paddle used to stir preservatives and the bottler system needs to be cleaned and sanitized before and after use. The filter and press had high loads due to the presence of cider (during production). The bottler tip had extremely high levels of yeasts and molds present; attention should be paid to this area when cleaning and perhaps brushing would need to be incorporated.

Producer D 2000-2001 ENVIRONMENTAL SWAB EVALUATIONS				
LOCATION	Yeasts and Molds	Aerobic Bacteria	Coliforms	OBSERVATIONS
Press (after cleaning) per cm2	1 est	.5 est	<.1 est	
Lip of steel holding tank (per cm2)	51	32	.14 est	
Hole in top of steel holding tank (per cm2)	21 est	7 est	.10 est	Used to put the bottling tube through
Tip of Bottler (after use)	1300	1000	<1 est	

ASSESSMENT: The press equipment was clean of debris and the low microbial results indicate that the cleaning is sufficient. However, the lid and lip of the cider holding tank did have higher microbial loads. These are hard to clean areas and need special attention during cleaning. Brushing may need to be incorporated to get these areas clean. The tip of the bottler also had high loads. Perhaps before and after use and after any breaks, it would be beneficial to rinse with water and sanitize.

Producer E 2000-2001 ENVIRONMENTAL SWAB EVALUATIONS				
LOCATION	Yeasts and Molds	Aerobic Bacteria	Coliforms	OBSERVATIONS
Lid (unused) count per lid	95 est	35 est	<1	Lid did appear to have some dried cider on it
Water from faucet in processing room (per ml)	<10	10 est	<1	
Hole in top of steel cider holding tank (after cleaning) per cm 2	0.18	0.18	<1	
Tip of funnel for pomace collection (after cleaning) per cm 2	>5100 est	>5100 est	<1	
Press (after cleaning) per cm 2	>6500 est	>6500 est	<1	
Water from bucket used to dip bottling tube (per ml)	<10	<10	<1	Contained chlorine
Bottom edge of bottling tube (per cm2)	<10	<10	<1	
Lid (unused) count per lid	<10	75 est	<1	

ASSESSMENT: The steel equipment and the holding tanks seemed fairly clear from debris. The water used in processing is of good quality. The processing room, bottling room, and coolers are well maintained and free from clutter. Two pieces of equipment are of definite concern; the funnel and the press. Both contained very high levels of microorganisms even after cleaning. Although, no coliforms were present, the high levels of bacteria that were found on the equipment could contaminate any cider that comes in contact with it, especially the initial batch. Special attention should be paid to these areas when cleaning and perhaps incorporating scrubbing and a sanitizing step with rinsing afterwards may be beneficial. Rinsing should also be performed before start-up. A low count of microorganisms was found on the unused lids, which should ideally be free from bacteria and yeasts and molds. Care should be taken to keep the lids in a sealed container or covered when not being used.

2000 Cider Survey

HARVESTING

1) Is manure fertilizer used in the orchard? YES NO

2) Is there a deer fence around the orchard? YES NO

3) Do you use drop apples in cider? YES NO

4) Are drop apples separated from tree picked apples? YES NO

5) Are apples from another supplier used? YES NO
If yes, are records kept documenting the source (drop vs tree-picked) of the supplier? YES NO

6) Do you provide hand wash stations and easily accessible toilets to field workers? YES NO

PROCESSING

7) Are outside windows or doors open during processing? YES NO

8) What is the water source used for processing? Well water Rural water Municipal water

9) Is your water source chlorinated? YES NO

10) Is your water source tested regularly for microbial counts? YES NO
Is your water source tested for chorine content? YES NO

11) Are apples stored inside a cooler? YES NO What Temperature?_____

12) Are rotten apples discarded at any point during storage? YES NO When_____

13) Are drop apples used in cider? YES NO

14) Are apples washed prior to processing? YES NO

15) Are brushes used on the apples? YES NO

16) Are apples sanitized prior to processing? YES NO
If yes, what is the sanitizer and concentration used?_____

17) Is an auger system used to dispose of pomace? YES NO
If yes, is the auger system enclosed? YES NO
If no, how is pomace removed?_____

18) Do you pasteurize? YES NO
If yes, what time and temperature do you use?_____

19) Is a preservative used? YES NO What kind and concentration?_____

20) Is the cider filtered? YES NO Through steel or mesh?_____

21) How long is cider allowed to settle before bottling?
1 Day 3 days 5 days 1 week more than 1 week

22) What temperature is cider held at during settling?_____

23) What type of bottling system is used? By hand Automatic

24) If bottling is done by hand, is the trough covered? YES NO

25) After bottling, approximately how long are bottles allowed to sit at room temperature before being transported to the cooler? _____

26)Are only new containers and caps used to bottle cider? YES NO

27)Is a date code or other method to identify lots used? YES NO

28)Is the "unpasteurized warning statement" used on the labels? YES NO

CLEANING/SANITATION

29) Is the processing equipment rinsed prior to startup? YES NO

30) Is processing equipment cleaned after each use? YES NO

31) Is a cleaner and/or sanitizer used on the equipment? YES NO
What kind and concentration? _____

32) Is a pest management system enforced? YES NO Through what means? _____

_____

EQUIPMENT

33)Are press cloths used that are specifically designed for cider production? YES NO
Are the press cloths cleaned and/or sanitized after use? YES NO

34)What are the press racks made of? Food-grade plastic Wood Other

35)Are press racks made of properly maintained? YES NO

36)Are press racks and cloths stored off floors in a well-ventilated area? YES NO

STANDARD OPERATING PROCEDURES

37) Has a written HACCP plan been developed? YES NO
If yes, are records maintained? YES NO

38)Are good manufacturing practices summarized and implemented? YES NO

39)Have written standard operating procedures (SOPs) been developed? YES NO

40)Have you attended the Cider School? YES NO

41)Are you currently certified? YES NO

42)Have you been inspected yearly? YES NO

43)Where do you sell your cider? On site Farmer's Market Retail Store Other

44)Has your cider sales volume increased or decreased over the past 2 years? _____

45)Have you considered, or actually begun pasteurizing your cider in the past 2 years? _____

Comments: _____

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