HUMAN HEALTH RISK ASSESSMENT AND RISK RANKING ASSOCIATED WITH EXPOSURE TO CHEMICAL AND MICROBIAL HAZARDS VIA CONSUMPTION OF APPLE AND APPLE JUICE PRODUCTS IN THE UNITED STATES

By

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ABSTRACT

HUMAN HEALTH RISK ASSESSMENT AND RISK RANKING ASSOCIATED WITH EXPOSURE TO CHEMICAL AND MICROBIAL HAZARDS VIA CONSUMPTION OF APPLE AND APPLE JUICE PRODUCTS IN THE UNITED STATES

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Food hazard risk assessment and risk ranking enable policy makers to develop appropriate regulatory and other risk management approaches to control the most important hazards in food products. The objective of this study was to determine the microbial and chemical hazards associated with three different types of apple products and rank their health burden on humans using various risk metrics. This research included data from foodborne illness outbreaks associated with apple and apple juice products from 1991 through 2015. Risk analysis and ranking of chemical hazards were conducted using FDA-iRisk 2.0. Microbiological hazard risk assessment and ranking were conducted using average numbers of outbreak-associated foodborne illness cases per year, adjusted for under-diagnosis. The risk ranking metrics used were disability adjusted life years (DALY) per year, DALY per consumer or eating occasion, and mean risk of illness. Apple cider consumption was associated with the greatest total DALY per year, with 13.18 DALY per year reflecting approximately 73% of the total DALYs per year predicted for consumption of all foods assessed in this study. The primary health concern with apple juice or fresh apples was inorganic arsenic, which accounted for 4.84 and 0.14 DALYs per year for apple juice and fresh apples, respectively. The results demonstrated that apple and apple juice products have relatively low DALYs associated with their consumption in the US.

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1. INTRODUCTION

Identifying the most predominant food hazards in specific foods, such as microbial pathogens and toxic chemicals, has been a major development in the food science field during the past several decades. However, a critical question facing the field today is determining which food hazards in specific foods represent the greatest risk to consumers. Ranking the risk of food hazards is a basic step that government, policy makers and other food industry decision makers need to do when designing strategic plans to protect the public against risks associated with eating foods containing contaminants such as toxic chemicals and pathogenic microbes (Morris et al., 2011).

Risk ranking methodology using either quantitative or qualitative approaches has been developed in recent years to rank food safety risks across all foods or for ranking health effects associated with specific food:hazard pairs (Romero et al., 2013). Using robust risk ranking methodology enables policy makers to develop appropriate regulatory and other risk management approaches to control the most important hazards in specific food products. These risk-ranking approaches also allow comparison of vastly different hazards having either acute or chronic health effects. Comparison of different hazards based on disability adjusted life years (DALYs) is a common approach to evaluate the effect of risk on human health (Devleesschauwer et al., 2014). For example, during the last four years (2011-2015) several important studies have been published using risk rankings based on DALYs. The World Health Organization used DALYs to estimate the global burden of foodborne illness during the time between 2007 and 2015 (WHO, 2015a). Murray et al. (2013) calculated DALYs for 291 illnesses and injuries in 21 countries from 1990 through 2010. Another key study assessed the cost of

illness and loss of quality-adjusted life years (QALYs; which are similar to DALYs) to rank the burden of disease associated with 14 top foodborne pathogens in the United States (Batz et al., 2012).

Apple juice is a popular drink of consumers in the United States (US), where approximately 2.6 billion liters of apple juice were consumed in 2012 (USDA, 2012a). However, chemical hazards such as heavy metals and patulin can be associated with apple products. Patulin is a mycotoxin synthesized in apples by *Penicillium expansum* and the US Food and Drug Administration (US FDA) has established an action level of 50 µg patulin/kg for apple juice, apple sauce and apple juice concentrate (when diluted to single strength) in the US. Surveillance indicates that patulin concentrations in commercial juice often exceed this action level (Harris et al., 2009).

Apple juice products that have not been treated by pasteurization or other pathogen-destruction technologies have been associated foodborne illness outbreaks caused by microbial pathogens in the past two decades. The major pathogens associated with these outbreaks include pathogenic *Escherichia coli* (*E. coli* O157:H7 and O111) and *Cryptosporidium parvum* (CDC, 2014).

Whereas the most commonly occurring pathogenic microbes and toxic chemicals in US apple products are well characterized, the relative risk to public health presented by these various food hazards in different apple products is not well understood. This research aims to fill this knowledge gap.

2. REVIEW OF LITERATURE

2.1. The Importance of Risk Ranking of Foodborne Diseases as a Policy Decision Tool

The estimation of foodborne diseases is critical in predicting the future burden of these diseases in people. It is important to understand the trend of foodborne diseases that are associated with specific foods to set strategic goals to monitor progress in reducing foodborne illness and food contamination and to set goals for improvement (Scallan et al., 2011a; Newsome et al., 2009).

Ranking the risk of hazards associated with specific foods or food ingredients is a decision making tool that government, policy makers and other food industry decision makers need to establish effective policies. This decision-making tool is needed when designing strategic plans to protect the public against risks associated with eating foods that are potentially contaminated with toxic chemicals or pathogenic microbes. It is important to describe and compare risks among different populations because there likely are differences in risk among population groups (e.g. adults, children). Improving our understanding of these differences can provide evidence for investigating the reasons why one group may have a higher risk of foodborne illness (or exposure to toxic chemicals) compared to other groups, and collectively this type of analysis can help to identify methods to assess the risks of specific food hazards (Newsome et al., 2009; FDA, 2014a).

2.2 Foodborne Illness in the United States

Ingestion of food contaminated with pathogenic microorganisms, toxic chemicals, or other food hazards can cause different foodborne diseases in humans. It is possible for food to be contaminated at any stage of food production from primary production through consumption. Certain diseases that result from consumption of contaminated food have a high likelihood to lead to disability or mortality (WHO, 2016b). Food contamination can be caused by microbial pathogens (foodborne infections) or by poisonous chemicals or harmful toxins such as are derived from poisonous mushrooms. However, most of the documented foodborne illnesses that have been described (more than 250) are infections caused by microbial pathogens including pathogenic bacteria, viruses, and parasites (CDC, 2015a).

Foodborne illnesses have a negative impact on public health and contribute to the cost of healthcare significantly in the US. It has been estimated that approximately 17% of people in the US (48 million) are sick annually because of foodborne illnesses and that these diseases lead to approximately 3,000 deaths and 128,000 hospitalizations annually. It has been estimated that more than 95% of foodborne illnesses are caused by only 15 pathogens, as presented in Table 1. These 15 pathogens also account for approximately 98% of deaths caused by foodborne pathogens in the US each year and account for 84% of the annual economic costs associated with foodborne illness (Hoffmann et al., 2015).

Table 1. The 15 pathogens that cause 95% of foodborne illnesses in the United States each year.

Pathogens	Mean Incidence			Percentage		
	Cases	Hospitalizations	Deaths	Cases	Hospitalizations	Deaths
Campylobacter, all species	845,024	8,463	76	9.0	15.1	5.6
Clostridium perfringens	965,958	438	26	10.3	0.8	1.9
Cryptosporidium, all species	57,616	210	4	0.6	0.4	0.3
Cyclospora cayetanensis	11,407	11	0	0.1	0.0	0.0
Listeria monocytogenes	1,591	1,455	255	0.0	2.6	18.9
Norovirus	5,461,731	14,663	149	58.2	26.2	11.0
Salmonella, all non-typhoidal species	1,027,561	19,336	378	10.9	34.6	28.0
Shigella, all species	131,254	1,456	10	1.4	2.6	0.7
STEC 0157	63,153	2,138	20	0.7	3.8	1.5
STEC non-0157	112,752	271	0	1.2	0.5	0.0
Toxoplasma gondii	86,686	4,428	327	0.9	7.9	24.2
Vibrio vulnificus	96	93	36	0.0	0.2	2.7
Vibrio parahaemolyticus	34,664	100	4	0.2	0.1	0.6
Vibrio, other non-cholera species	17,564	83	8	1.0	1.0	2.1

Table 1 (cont'd)

Pathogens	Mean Incidence			Percentage		
	Cases	Hospitalizations	Deaths	Cases	Hospitalizations	Deaths
Yersinia enterocolitica	97,656	533	29	1.0	1.0	2.1
16 other identified pathogen causes	473,362	2,283	29	5.0	4.1	2.1
Total	9,388,075	55,961	1,351	100	100	100

Source: Scallan et al., 2011a. Foodborne illness acquired in the United States - major pathogens.

Foodborne illnesses can be acute or chronic diseases. Acute illnesses occur suddenly, typically have a short duration, and can have severe clinical signs. The majority of foodborne illnesses caused by microbial pathogens and some chemical hazards cause acute clinical signs such as diarrhea, vomiting and dysentery. In most cases, people recover from these acute illnesses in a few days or weeks without treatment. However, certain acute foodborne illnesses may also lead to chronic sequelae such as Guillain-Barré Syndrome (CDC, 2016a; Lindsay, 1997; Hahn, 1998).

On the other hand, chronic diseases such as obesity, heart disease, cancer, arthritis and diabetes are the leading causes of disability and death in the US. In 2012, the US Centers for Disease Control and Prevention (CDC) reported that 117 million US adults had one or more chronic illnesses. Also, of the top ten causes of death in the US, seven of them are because of chronic diseases. Forty-eight percent of all deaths are due to cancer and heart disease (CDC, 2016a).

More research is needed to assess the chronic disease burden attributable to foodborne hazards. Until now, there has been limited evidence concerning the impact of acute foodborne

illness on chronic disease risk, and there are no published data from food and health agencies in the US about the relative contribution of foodborne hazards to total chronic disease burden. Moreover, recent estimates of foodborne illnesses in the US (Scallan et al., 2011) did not include the health burden due to chronic conditions, long-term disabilities and latent negative effects from acute foodborne diseases. Finally, a recent report by the US Department of Agriculture (USDA, 2015) indicated that it is expected that "more chronic conditions may be included in future estimates of the burden of foodborne illness" (USDA, 2015).

2.2.1 The Risk Analysis of Microbial and Chemical Foodborne Diseases

More than 200 known illnesses caused by many agents have been demonstrated to be transmittable via contaminated food. These agents include both infectious (e.g. pathogenic bacteria) and noninfectious (e.g. toxic chemicals) hazards. While many agents responsible for causing foodborne illnesses are known, a large proportion of illnesses in the US are believe to be caused by unknown agents (Scallan et al., 2011b; Tables 2 and 3).

Bacteria, viruses, parasites, prions, toxins, and metals are main causes of foodborne diseases. The clinical signs of foodborne illness can be acute or chronic and range from moderate gastroenteritis to life-threatening conditions such as renal failure, neurological and hepatic syndromes (Mead et al., 1999).

The analysis of risk for acquiring foodborne diseases from microbial and chemical hazards is a critical challenge for food scientists and policymakers in the US and globally. Quantifying an accurate number of foodborne illnesses that are caused by chemical or microbial hazards every year and identifying their negative effects (acute and chronic) on public health are two fundamental challenges that are associated with determining the magnitude of the risk of foodborne diseases. Relatively few studies have attempted to comprehensively quantify these

risks (e.g. Scallan et al., 2011a; Table 1), and these studies have primarily focused on risks associated with microbial pathogens. Conversely, relatively little research has been conducted to quantify the risk of foodborne illnesses associated with chemical hazards. One of the main reasons for this lack of attention is that there are far fewer outbreaks associated with chemical hazards compared to microbial hazards. Another potential reason for the limited attention to chemical hazards is that their health impacts often are not acute, but rather impact chronic disease risk.

Through the last century in the US, the nature of public health, food and foodborne diseases have changed greatly. While technological advances like convenient canning and pasteurization have contributed to reducing some illness, new types of foodborne diseases have been recognized (Mead et al., 1999; Scallan et al., 2011a).

Foodborne illness surveillance is considered to be complicated because of three basic elements. The first element is underreporting of foodborne illnesses, where milder cases of illness are mostly undiscovered during routine surveillance because persons suffering from relatively mild foodborne illnesses often do not seek medical attention. Secondly, many pathogens that are transmitted via food are, at the same time, further spread through water or by person-to-person transmission. Lastly, some foodborne diseases caused by pathogens or different agents have not yet been identified and, as a result, the relative contribution of these illnesses to overall foodborne illness burdens is difficult to estimate (Mead et al., 1999; Scallan et al., 2011a).

There are fundamental factors that affect the accuracy of foodborne illness estimates, such as new systems of surveillance, identification of new foodborne illnesses, and continuing changes in the global food supply. Consequently, in order to update and improve food safety

policies and regulations and improve foodborne illness prevention, there is a need for new and more accurate estimates of foodborne illnesses and the food-hazard combinations that are major contributors to these illnesses (Mead et al., 1999).

Currently, annual foodborne illnesses in the US are estimated from reported illnesses taking into account adjustments for under-reporting and under-diagnosis (Scallan et al., 2011a). The actual numbers of foodborne diseases in US are unknown. There is a huge gap between the annual reported illnesses that are attributed to foodborne illness outbreaks and the estimated foodborne illnesses that take into account under-reporting and under-diagnosis. For example, during 1998-2014 estimates from CDC outbreak data indicate that known pathogens caused 18,211 outbreaks, 358,391 illnesses, 13,715 hospitalizations, and 318 deaths (CDC, 2015b). However, Mead et al. (1999) estimated that these known pathogens cause 14 million illnesses, 60,000 hospitalizations, and 1,800 deaths annually.

Because only small numbers of microbial foodborne diseases are diagnosed and reported, it is important to develop more accurate estimates of the annual total incidence of microbial foodborne disease in US to evaluate the health burden (Scallan et al., 2011a).

Estimates of the annual numbers of foodborne illnesses attributable to microbial pathogens in the US were published in the US in 1999 (Mead et al., 1999) and 2011 (Scallan et al., 2011a). The major outcomes of these studies are presented in Table 2 and Table 3. However, the estimates by Scallan et al. (2011a) are not directly comparable to those of Mead et al. (1999) due to differences in methodology employed.

Table 2. Studies that estimated the total incidence of foodborne disease in United States.

Study	**Year**	**The study results**
Food-related illness and death in the United States. (Mead et al., 1999).	1999	1. In the United States every year, foodborne illnesses cause approximately: 76 million illnesses, 325,000 hospitalizations, and 5,000 deaths. 2. Each year, known pathogens cause 14 million illnesses, 60,000 hospitalizations, and 1,800 deaths. 3. *Salmonella*, *Listeria*, and *Toxoplasma* are three pathogens that were responsible for 1,500 deaths annually. 4. Unknown agents cause 62 million illnesses, 265,000 hospitalizations, and 3,200 deaths.
Foodborne illness acquired in the United States-major pathogens. (Scallan et al., 2011a)	2011	1. 9.4 million foodborne illnesess are caused by only 31 major pathogens each year in the U.S. 2. The estimated foodborne diseases cause 55,961 hospitalizations and 1,351 deaths. 3. "Scarce data precluded estimates for other known infectious and noninfectious agents, such as chemicals".
Foodborne illness acquired in the United States-unspecified agents. (Scallan et al., 2011b)	2011	1. "Unspecified agents are major contributors to the total number of episodes of acute gastroenteritis and foodborne diseases". 2. These agents cause 38.4 million episodes of foodborne gastroenteritis each year in the United States, resulting in 78,878 hospitalizations, and 1,686 deaths. 3. The total effect of contaminated food consumed in the United States from the combination of the estimates of unspecified agents and major known pathogens are 47.8 million episodes of illness, 127,839 hospitalizations, and 3,037 deaths (Table 3).

Table 3. Estimated annual number of episodes of domestically acquired foodborne illnesses, hospitalizations, and deaths caused by 31 pathogens and unspecified agents transmitted through food in the United States.

Category	Illnesses per year	Hospitalizations per year	Deaths per year
Major known pathogens	9,388,075	55,961	1,351
Unspecified agents	38,392,704	71,878	1,686
Total	47,780,779	127,839	3,037

Source: Foodborne Illness Acquired in the United States-Unspecified Agents (Scallan et al., 2011b).

2.3 Food Safety Associated with Fresh Apples and Apple Juice Products

2.3.1 Microbiological Hazards

Fresh apples contain the necessary nutrients to support the growth of different pathogens that can cause foodborne illness such as *E. coli* O111, *E. coli* O157:H7, *Cryptosporidium parvum* and *Salmonella* spp. However, external barriers like the rind and the peel serve as effective barriers to prevent pathogens from entering the interior of apples and growing there, provided these barriers are intact. Consequently, until now no reported outbreaks of foodborne illness associated with consumption of fresh apples have been reported. Nevertheless, injured or cut apple slices are appropriate environments for pathogenic microbes to grow because the external barriers are broken (Abdul et al., 1993; Janisiewicz et al., 1999).

Processing of apples to manufacture juice or cider has the potential to introduce and distribute microbial pathogens, that otherwise would have been only present as surface contaminants, throughout the product. For perceived quality and health-related reasons, many consumers have a preference for consuming unpasteurized apple juice (cider) compared to

thermally processed, shelf-stable commercial juice. However, this unpasteurized juice can cause increased risk of consumer illness associated with pathogens such as *Cryptosporidium parvum and E. coli* O157:H7. Outbreaks caused by these pathogenic microbes have been associated on numerous occasions with the consumption of unpasteurized apple cider. The ultimate sources of pathogen contamination of these products can include contaminated water, feces from animals or humans, or the processing environment. As a result, grinding and pressing apples to produce juice is commonly associated with pathogenic microbes (Mihajlovic et al., 2013).

2.3.1.1 Cryptosporidium spp.

Cryptosporidium is a microscopic parasite that can infect animals and humans and cause diarrhea in humans. The parasite has an outer shell that keeps it protected in the environment. After exposure, the clinical signs of infected people can be observed after seven to ten days. Usually, infected humans recover without any medical care. The illnesses typically caused by *Cryptosporidium* infection are mild in nature (CDC,2010).

Contaminated water (recreational or drinking water) is one of the most common sources of exposure of persons to *Cryptosporidium*. However, food contaminated by infected handlers is believed to be responsible for some reported outbreaks of different species of *Cryptosporidium* (CDC, 2010; CDC, 2015c).

According to Morris et al. (2011), *Cryptosporidum* ranked 12th among the 15 leading pathogens causing foodborne illnesses in the US in terms of economic burden and 11th based on the total number of cases in the US. The estimated number of *Cryptosporidium*-associated illnesses is 57,600 cases each year (Table 1; Scallan et al., 2011a). The estimated annual economic burden associated with these illnesses is $51.8 million (Hoffman et al., 2015).

Since 1993, *Cryptosporidium* spp. has been associated with several foodborne illness outbreaks caused by consuming unpasteurized apple cider in United States. Between 1993 and 2015, *Cryptosporidium* caused eight outbreaks that resulted in 410 cases, 18 hospitalizations and no deaths (Table 4). Six of these outbreaks were associated with *Cryptosporidium parvum*, and the other two were attributed to generic *Cryptosporidium*. In five of these outbreaks, *Cryptosporidium* was the sole pathogen present in the product. However, two of the outbreaks were associated with both *Cryptosporidium parvum* and *E. coli* O111, and one outbreak was associated with multiple strains of *Cryptosporidium*.

From 1992 to 2015, no outbreaks due to *Cryptosporidium* were reported from the consumption of fresh apples or shelf stable apple juice. This illustrates the importance of pasteurization and compliance with the FDA juice HACCP regulation for reducing the risk of pathogens in juice products (Lindsay, 1997; Millard et al., 1994).

Based on information presented in Table 4, most illnesses that were associated with unpasteurized apple cider were observed between 1993 and 2004 (94% of total observed cases), while the period from 2004 to 2015 only included 6% of the total observed cases. Comparing the period since 2004 to the 1993 to 2004 time frame, the rate numbers of outbreaks per year did not change but the number of cases per outbreak decreased dramatically. This is significant because enforcement of the provisions of the FDA juice HACCP regulation began during the time frame of 2002-2004 (Vojdani et al., 2008).

The three largest outbreaks were in 1993, 2003 and 2004, respectively. In Maine, a *Cryptosporidium* outbreak was reported in 1993 from drinking unpasteurized apple cider by students and school staff who attended a school agriculture fair for one day. This outbreak resulted in 213 cases from the students and staff (Millard et al., 1994). In 2003, *Cryptosporidium*

parvum caused an outbreak in Ohio that resulted in 144 cases associated with consuming commercial apple cider. Although this commercial apple cider was covered by the FDA juice HACCP rule, it was processed in a manner that did not meet HACCP requirements because the ozone treatment used by the establishment to control pathogens was inadequate to meet the performance standard required by the juice HACCP regulation (5-log reduction in the pertinent pathogens of concern). In 2004, an outbreak of *Cryptosporidium parvum* and *E. coli* 0111 in New York was associated with drinking fresh pressed apple cider from an orchard that sold the cider by direct sales to consumers. This outbreak resulted in 212 illnesses associated with drinking unpasteurized apple cider. Among these illnesses, those attributed to each pathogen (*Cryptosporidium parvum* and *E. coli* O111) were not reported, so it is not possible to disaggregate the cases by pathogen. The cider may have been contaminated with *Cryptosporidium* by incoming fruit, personnel, the processing environment or equipment, and was not pasteurized after processing to control pathogens. The facility was exempt from compliance with the FDA juice HACCP regulation as it was only selling cider directly to consumers (Vojdani et al., 2008).

Table 4. Reported outbreaks, cases, hospitalizations and deaths due to *Cryptosporidium parvum* foodborne illnesses associated with apple cider, 1991-2015.

Year	Outbreaks	Cases	Hospitalizations	Deaths	State	Vehicle
1993	1	213	0	0	Maine	Unpasteurized apple cider
1996	1	31	0	0	New York	Unpasteurized apple cider
2003	1	144	3	0	Ohio	Unpasteurized apple cider
2011	1	4	0	0	Ohio	Unpasteurized apple cider
2013	1	8	0	0	Ohio	Unpasteurized apple cider
2013	1	10	1	0	Iowa	Unpasteurized apple cider
Total	6	410	4			

Sources: (CDC, 2015b; Millard et al., 1994; Vojdani et al., 2008).

2.3.1.2 Escherichia coli O157:H7

Escherichia coli O157:H7 *(E. coli* O157:H7) is a toxin-producing strain of *E. coli. E*. coli has six pathotypes; these are Shiga toxin-producing *E. coli* (STEC), enterotoxigenic *E. coli* (ETEC), enteroaggregative *E. coli* (EAEC), enteropathogenic *E. coli* (EPEC), enteroinvasive *E. coli* (EIEC) and diffusely adherent *E. coli* (DAEC). These six pathotypes of *E. coli* can cause diarrhea, and can be transmitted to humans by contaminated food, water, or through contact with people or animals (CDC, 2015d; Vojdani et al., 2008). STEC causes a very severe illness that can include damage to kidneys and other organs and death in severe cases (Tarr, 1995).

Each year in US, it is estimated that *E. coli* O157:H7 causes 63,153 illnesses, 2,138 hospitalizations, and 20 deaths (Table 1; Scallan et al., 2011a). Moreover, the annual economic

burden associated with *E. coli* O157:H7 illnesses in the US is estimated at $271 million (Hoffman et al., 2015).

The first reported outbreak of *E. coli* O157:H7 was in 1982 when it was recognized for the first time as a pathogen during the investigation of an outbreak of hemorrhagic colitis associated with hamburger consumption from fast-food chain restaurants. Since 1982, *E. coli* O157:H7 has been reported to be one of the main pathogens that causes foodborne diseases associated with contaminated fresh food (Scallan et al., 2011a; Rangel et al., 2005).

E. coli O157:H7 is ranked as one of the leading causes of multi-state outbreaks of foodborne diseases in the US each year. For example, from 1982 to 2002 it was reported by Rangel et al. (2005) that 350 outbreaks were caused by *E. coli* O157:H7 with illnesses reported in 49 states. Among these outbreaks, 183 (52%) were foodborne and ground beef consumption was associated with 41% of these outbreaks (Rangel et al., 2005). In 2006, an *E. coli* O157:H7 outbreak associated with consumption of fresh spinach caused 199 illnesses and 3 deaths (FDA, 2006)

From 1991 to 2015, *E. coli* O157:H7 was implicated in 10 outbreaks associated with consumption of apple cider in the US (Table 5). These outbreaks included 164 reported illnesses with cases in 10 states, 38 hospitalizations and no deaths (Table 5). All of these outbreaks were associated with consumption of unpasteurized apple cider and no cases were reported with consumption of fresh apples or shelf stable apple juice (Besser et al.,1993; Janisiewicz et al., 1999; CDC, 1997; Cody et al., 1999). However, the incidence of illnesses associated with unpasteurized apple cider decreased dramatically from 1991 to 2000, a time frame that accounted for 84% of cases, to the time frame of 2000 to 2015 (16% of cases). Many factors, such as

HACCP implementation, may have contributed to this reduction of reported illnesses (Danyluk et al., 2012).

In October 1996, a multi-state outbreak of *E. coli* O157:H7 illnesses associated with drinking unpasteurized apple cider manufactured by the Odwalla Company was instrumental in driving changes in food safety policies concerning unpasteurized juices in the US. This outbreak resulted in 66 illnesses and the death of one infant (16 months of age). The illnesses occurred in three different states (Washington, California and Colorado) as well as British Columbia in Canada. Among these reported cases, more than a dozen persons developed hemolytic uremic syndrome (HUS). The outbreak investigation found that three lots of incoming apples used for apple cider production were suspected to be the source of contamination. Two lots of the apples were procured from an orchard where deer were frequently observed. Deer are known to be a potential reservoir for *E. coli* O157:H7. One lot of apples reportedly included waxed spoiled apples (CDC, 1997).

Table 5. Reported outbreaks and illnesses of *E. coli* O157:H7 associated with apple cider in the United States, 1991-2015.

Year	Outbreaks	Cases	Hospitaliza-tions	Deaths	State	Vehicle
1991	1	23	Not reported	0	Massa-chusetts	Apple cider, unpasteurized
1996	2	20	Not reported	0	Connecti-cut, Washing-ton	Apple cider, unpasteurized
1996	1	66	25	1	California, Colorado, Washing-ton	Apple cider, unpasteurized
1997	1	6	Not reported	0	Indiana	Apple cider, unpasteurized
1999	1	25	6	0	Oklahoma	Apple cider, unpasteurized
2007	1	9	1	0	Massa-chusetts	Apple cider, unpasteurized
2008	1	5	2	0	Iowa	Apple cider, unpasteurized
2010	1	7	4	0	Maryland	Apple cider, unpasteurized
2012	1	3	0	0	Michigan	Apple cider, unpasteurized
Total	**10**	**164**	38	0	12 States	

Sources: Besser et al.,1993; CDC, 1997; CDC, 2015b; Cody et al., 1999; Danyluk et al., 2012; Janisiewicz et al., 1999).

2.3.1.3 Escherichia coli O111

E. coli O111 is one of the most widely recognized of the non-O157 STEC serogroups (O26, O45, O103, O111, O121, and O145) that are connected with human illness in the US. In 2011, *E. coli* O111 and the other strains of non-O157 STEC were regulated by the Food Safety and Inspection Service of the U.S. Department of Agriculture (USDA) as adulterants in raw beef

products (CDC, 2012; Kalchayanand et al., 2013). More interesting, when the estimated cases associated with non-O157 STEC is compared estimated cases associated with *E. coli* O157, non-O157 STEC strains are associated with more foodborne illness cases than *E. coli* O157 (Scallan et al., 2011a).

As is the case with *E. coli* O157:H7, infections with *E. coli* O111 can be caused by consuming food or water that is contaminated with the pathogen. Moreover, *E. coli* O111 is able to infect animals, and the pathogen can be transmitted to humans who come into contact with these infected animals (CDC, 2012). The first outbreak implicating *E. coli* O111 as the main cause of severe gastroenteritis was reported in the United Kingdom in 1940 (Campos, 1994). In the US, the largest outbreak of *E. coli* O111 occurred in 2008 in a restaurant in Oklahoma. This outbreak caused illnesses in 341 persons of different ages that resulted in one death, 70 hospitalizations and 26 cases of hemolytic uremic syndrome (Bradley et al., 2012).

In the US, two outbreaks caused by *E. coli* O111 in 2004 and 2011 were associated with consumption of unpasteurized apple cider. These two outbreaks caused 226 illnesses and, coincidentally, both outbreaks also had co-contamination of the implicated product with *Cryptosporidium* (Table 6). The 2004 outbreak at a cider mill in New York resulting in 212 illnesses was the larger of the two outbreaks and already has been discussed in the *Cryptosporidium* section. The other outbreak of *E. coli* O111 with *Cryptosporidium* occurred in 2011 in Minnesota and caused 14 illnesses with no reported hospitalizations or deaths (CDC, 2015b).

Table 6. Reported outbreaks and illnesses caused by *E. coli* O111 and *Cryptosporidium* spp. associated with apple cider, 1991-2015.

Year	Outbreaks	Cases	Hospitalizations	Deaths	State	Vehicle
2004	1	212	14	0	New York	Unpasteurized apple cider
2011	1	14	0	0	Minnesota	Unpasteurized apple cider
Total	2	226	14	0	2	

Source**:** CDC (2015b).

2.3.1.4 Salmonella spp.

In the US, it is estimated that *Salmonella* spp. (non-typhoidal) causes 1,027,561 cases, 19,336 hospitalizations, and 378 deaths annually (Scallan et al., 2011a). From 1998 to 2014, the Foodborne Outbreak Online Database maintained by CDC reports that *Salmonella* caused 61,630 illnesses, 6,952 hospitalizations and 79 deaths (CDC, 2015b). In addition, the estimated annual economic burden from these illnesses in the US is approximately $3.67 billion per year (Hoffmann et al., 2015)

Salmonella lives in the intestines of humans and animals, and people are commonly exposed to *Salmonella* via water, food and surfaces that can be contaminated with feces. Handling birds or reptiles is another common source of exposure to *Salmonella*. The clinical signs of non-typhoidal *Salmonella* mostly occur from 12 to 72 hours after human infection. Salmonella causes mild diarrhea, vomiting, and fever in most healthy people (CDC, 2016b).

During the time period of 1990 to 2015, it was reported that two *Salmonella* outbreaks were associated with apple cider in the US (CDC, 2015b). These outbreaks occurred in 1999 and 2011 and caused 18 illnesses, 7 hospitalizations and no reported deaths (Table 7). Both outbreaks were similar in the number of illnesses caused (8 and 10 cases; CDC, 2015b).

Table 7. Reported outbreaks and illnesses caused by *Salmonella* spp. associated with apple cider, 1991-2015.

Year	Outbreaks	Cases	Hospitalizations	Deaths	State	Vehicle
1999	1	8	2	0	Illinois	Unpasteurized apple cider
2011	1	10	5	0	Pennsylvania	Unpasteurized apple cider
Total	2	18	7	0		

Source: (CDC, 2015b)

2.3.1.5 Listeria monocytogenes

Listeria monocytogenes is a facultative intracellular bacterium that can cause severe infections and death in humans. *Listeria* is able to grow at low temperatures (e.g. at typical refrigeration temperatures), can survive on different types of surfaces, and can be found in soil and water. Even though listeriosis infections only account for approximately 1,600 illnesses per year in the US, *Listeria monocytogenes* ranks as the third leading cause of death from foodborne illness in the US, causing about 19% of all U.S. foodborne illness deaths annually (Scallan et al., 2011a). All people are potentially susceptible to infection with *Listeria*. However, the elderly, pregnant women, infants, and adults with weakened immune systems are much more susceptible than the general population (Data, 2015).

Every year, *Listeria monocytogenes* causes approximately 280 cases of listeriosis in U.S. However, cases associated with foodborne illness outbreaks reported by CDC (1998 to 2014) accounted for only 766 cases, 521 hospitalizations and 116 deaths (CDC, 2015b). Human exposure to *Listeria monocytogenes* is primarily via consumption of contaminated food. Some animals such as cattle and poultry can carry *Listeria* and contaminate dairy and poultry products

and meats. Manure that is used as fertilizer or *Listeria* in soil can contaminate vegetables and fruits. Sliced food such as deli meat, soft cheeses and other deli food can be contaminated after the slicing process. Foods produced from unpasteurized milk can be contaminated by *L. monocytogenes* (Data, 2015).

Each year, the total estimated economic burden of *Listeria*-associated illness in the US is $2.8 billion. Of these estimated costs, $2.1 billion are a consequence of deaths from *L. monocytogenes* and $600 million is associated with infections in newborns and prenatal infections (Hoffmann et al., 2015).

In 2014, the CDC reported a multi-state outbreak of *Listeria monocytogenes* infections associated with eating caramel apples in 12 US states. This outbreak resulted in 35 illnesses, 34 hospitalizations and 7 deaths (Table 8). The outbreak investigation indicated that the probable source of this outbreak was commercially produced prepackaged caramel apples. Further investigation indicated that the apples, packed by Bidart Brothers in California, were the source of *Listeria contamination*. Among affected individuals, 28 confirmed they had consumed commercially produced, prepackaged caramel apples before becoming ill. Eleven of the reported illnesses affected pregnant women. The range of ages of sick persons in this outbreak was from 7 to 92 years and 33% from them were female. The range of dates for these illnesses (listeriosis) was from October 17, 2014 until January 6, 2015 (CDC, 2015e).

2.3.1.6 Campylobacter spp

Campylobacter is a bacterial species that causes illnesses in humans and animals. *Campylobacter jejuni* is the most common species responsible for causing human illnesses, but other *Campylobacter* species also can cause human illnesses. *Campylobacter* is one of the leading causes of foodborne illnesses in the US. The majority of *Campylobacter*-associated

illnesses are not recognized as being associated with outbreaks, but rather occur as sporadic cases (Hoffmann et al., 2015).

In the US, *Campylobacter* is estimated to be the third leading cause of foodborne illnesses annually. FoodNet reports that 14 cases of *Campylobacter*-associated foodborne illness are diagnosed annually per 100,000 persons in the US (CDC, 2014). However, numerous cases are not diagnosed or reported every year. It is estimated that *Campylobacter* causes 845,024 cases, 8,463 hospitalizations, and 76 deaths each year in the US (Scallan et al., 2011a). However, cases associated with foodborne illness outbreaks reported by CDC (1998-2014) accounted for only 7,860 cases, 310 hospitalizations and one death (CDC, 2015b). Furthermore, the annual economic burden associated with these estimated illnesses is approximately $1.93 billion per year (Hoffmann et al., 2015).

In 2014, the CDC reported an outbreak of *Campylobacter jejuni* illnesses associated with consumption of unpasteurized apple cider in Arizona. This outbreak resulted in six illnesses, one hospitalization and no reported deaths (CDC, 2015b).

Table 8. Reported outbreaks of *Listeria monocytogenes* and *Campylobacter jejuni* associated with fresh apple, apple cider and apple juice in the United States, 1991-2015.

Pathogen	Year	Outbreaks	Cases	Hospitaliza-tions	Deaths	State	Vehicle
Listeria monocytogenes	2014	1	35	34	7	Multiple	Caramel apples (Fresh apples)
Campylobacter jejuni	2014	1	6	1	0	Arizona	Unpasteur-ized apple cider

Source: CDC (2015b).

Table 9 includes a summary of all outbreaks of foodborne illness caused by microbial pathogens associated with fresh apples, apple juice or unpasteurized apple cider since 1991 in the US. It is notable that the majority of outbreaks and illnesses were associated with *E. coli* O157 or O111, or *Cryptosporidium parvum*. It also is important to note that all outbreaks except for the *Listeriosis* outbreak associated with caramel apples were associated with consumption of apple cider that had not been pasteurized or otherwise treated to destroy pathogenic microorganisms.

Table 9. Summary of reported outbreaks of foodborne illnesses in the United States caused by the selected pathogens associated with fresh apples, apple juice and apple cider, 1991-2015.

Contaminated ingredient	Pathogen	Outbreaks	Cases	Hospitalizations	Deaths
Unpasteurized apple cider	*E. coli* O157:H7	10	164	13	0
Unpasteurized apple cider	*E. coli* O111	2	226	14 (With *C. parvum*)	0
Unpasteurized apple cider	*Cryptosporidium parvum*	8	402	31 (With *E. coli* O111)	0
Unpasteurized apple cider	*Salmonella* spp.	3	41	7	0
Fresh apples (caramel apples)	*Listeria monocytogenes*	1	35	34	7
Unpasteurized apple cider	*Campylobacter jejuni*	1	6	1	0
Total		25	874	100	7

2.4 Chemical Hazards Associated with Apple, Apple Juice and Apple Cider in the United States

2.4.1 Arsenic

Arsenic is a toxic metalloid that is found in the environment from different sources. Arsenic occurs in the earth from both natural and anthropogenic sources including disintegration of arsenic-containing rocks, volcanic ejections, sullying from mining and purifying metals, and past or current utilization of arsenic-containing pesticides (FDA, 2013a; Ratnaike, 2003).

Arsenic is found in two forms, inorganic and organic. Both forms are detected in a variety of foods, but the organic form is reported to be rare in water. Arsenic concentrations in foods usually range from 20 to 140 parts per billion (ppb) and the highest concentrations of total arsenic are found in rice and its products, seafood, seaweed, mushrooms and some meats (FDA,

2013). The US Environment Protection Agency (EPA) decreased the allowable level of total arsenic in drinking water from 50 ppb to 10 ppb in 2001 (Ratnaike, 2003).

In the US, arsenic is considered to be a major concern in drinking water. Inorganic arsenic (arsenite and arsenate) is the primary form that is found in drinking water (Ratnaike, 2003; FDA, 2013a). The concentration of arsenic is commonly less than 10 ppb in groundwater and natural surface waters in the US. However, arsenic concentrations can exceed these concentrations in areas having high soil concentrations of arsenic or in contaminated areas. For example, arsenic concentrations in drinking water ranging between 14 to 166 ppb have been observed in Millard County, Utah (Ratnaike, 2003).

2.4.1.1 Arsenic Toxicity and Carcinogenicity in Humans

Chemically, arsenic has two oxidation states: a trivalent form, arsenite (As_2O_3; called arsenic III) and the second state is a pentavalent form, arsenate (As_2O_5; called arsenic V). However, arsenic III is considered to be 60 times more toxic than arsenic V. Arsenic toxicity is due to its ability to inactivate around 200 enzymes that have roles in DNA replication and repair in addition to cellular energy pathways (Ratnaike, 2003; CDC, 2013a).

Arsenic is responsible for both acute and chronic toxicities that cause adverse health effects for many millions of people around the world. Most cases of acute arsenic poisoning have occurred as a consequence of accidental ingestion of arsenic-containing compounds such as certain pesticides. Mostly, the clinical characteristics are related to the gastrointestinal system (Ratnaike, 2003). Chronic arsenic exposure affects various systems but most cases of chronic toxicity in humans' result in skin lesions that come from oral exposure. The features of these skin lesions are hyperkeratosis, hypopigmentation and hyperpigmentation (Hughes, 2002).

Inorganic arsenic is categorized as a human carcinogen based on several epidemiological studies (Hughes, 2002). Inorganic forms of arsenic (tri- and pentavalent arsenic) exhibit greater carcinogenicity than organic forms. Exposure to inorganic arsenic has been connected with cancer, skin injuries, cardiovascular illness, neurotoxicity, and diabetes in people (FDA, 2013a; EFSA, 2014a).

In northeastern Taiwan, two long-term observational studies have been conducted on Taiwanese exposed to arsenic from drinking water. Cohort studies reported by Chen et al. (2010) during an 11-year period monitored the chronic toxicity due to arsenic exposure for 8,086 residents in northeastern Taiwan (FDA, 2011; Chen, 2010). These studies are considered the best that illustrate human carcinogenicity from arsenic exposure (FDA, 2011). The findings from these studies proved a fundamental health concern associated with lifetime exposure to arsenic and showed a clear development of bladder and lung cancers as a result drinking contaminated water with arsenic (FDA, 2011). Also, the results of these studies showed that 72% of Taiwanese who developed skin cancer in this cohort had hyperkeratosis and 90% of them had hyperpigmentation (FDA, 2011).

Children are more vulnerable to the effects of arsenic exposure than adults for two main reasons. The first reason is that, based on their body weight, children have much greater food consumption than that of adults. Secondly, the dietary patterns of children are generally characterized to have less variety than those of adults (FDA, 2013a; EFSA, 2014; Conklin and Chen, 2012).

2.4.1.2 How Residents Are Exposed to Arsenic via Apple Juice in United States

Generally, there are two main sources of arsenic contamination – natural geological occurrence and anthropogenic sources such as mining or application of arsenic-containing

agricultural chemicals. These sources can contaminate drinking water as a result of arsenic leaching into aquifers (Ratnaike, 2003). Human exposure to arsenic occurs most commonly through consumption of drinking water and foods. While dermal and inhalation exposures to arsenic are possible, consumption of water, foods, and beverages are the main routes of human exposure.

In the US, there has been recent concern about exposure to arsenic through consumption of apple juice and other juice products (Hooper, 2012). Apples and processed apple products may be contaminated by arsenic through different routes. Beside potential contamination from arsenic that occurs naturally in the soil, there has been particular concern regarding historical use of arsenic-containing pesticides. Lead arsenate was a primary pesticide used in apple and other fruit orchards in the US and other countries for decades starting in the early 1900s. Lead arsenate was used in many countries such as the US, Canada, UK, France, China, etc., and was primarily used to control coddling moth in apple orchards. Lead arsenate use in the US was largely replaced by DDT during the 1950s, although some uses are documented into the 1960s. All insecticidal uses of lead arsenate in the US were officially banned on August 1, 1988 (Peryea, 1998).

A second source of concern with respect to arsenic concentrations in juice is country of origin, in that the US imports large quantities of apple juice concentrate from China and other countries (Hooper, 2012). The majority of apple juice consumed in the US is manufactured using this imported apple juice concentrate that is reconstituted with water. Therefore, most commercial apple juice available for sale in the US could have arsenic contamination arising from the original apples, via the juice concentrate, as well as arsenic contributed by the water used to reconstitute the juice (Hooper, 2012).

There are several reasons why food safety regulatory agencies such as FDA and European Food Safety Authority (EFSA) have conducted detailed studies regarding the potential toxicity of arsenic associated with apple juice consumption. The first reason is that apple juice consumption is extremely prevalent in the population, ranking as the second most consumed juice in the US (Hooper, 2012). Approximately 1.2 billion liters of apple juice are produced each year globally. The top two producing countries of apple juice in the world are China and Poland, respectively. In the US, it was reported in 2002 that the annual average apple juice consumption per person was 42.8 L (Hooper, 2012). The second reason is that apple juice is a primary ingredient in a variety of other juices and fruit drinks such as cider, berry, orange, grape and other blended juices (Hooper, 2012).

In 2010, FDA convened a meeting on arsenic risk assessment with experts from organizations including Food and Agriculture Organization of the United Nations (FAO), World Health Organization (WHO) and JECFA. The outcomes of this meeting included the statement that "food can be a major contributor to inorganic arsenic exposure" (FDA, 2013). The European Food Safety Authority suggested that exposure to inorganic arsenic through human foods needs to be decreased (FDA, 2013).

For several years, the US FDA has assessed arsenic concentrations in apple juice in order to manage its risk and protect the public health from its negative effects. Between 2005 and 2011, FDA analyzed 160 samples of apple juice and apple juice concentrate (FDA, 2011a). FDA has utilized four routine surveillance programs – the Total Diet Study, Radionuclides in Food Program, Monitoring of Imports and Targeted Domestic Assignments Program, and Toxic Elements in Food and Foodware. Through these programs, FDA has reported inorganic and total arsenic concentrations in samples of apple juice (FDA, 2011a). In the past, FDA inorganic

arsenic risk assessment focused on short-term exposures. However, FDA started assessing lifetime cancer risk associated with inorganic arsenic because its associated disease burden is correlated with lifetime exposure (FDA, 2013). Furthermore, most of the chronic risk that arises from inorganic arsenic exposure from apple juice is estimated to be from childhood exposure (FDA, 2013a).

2.4.1.3 The Estimation of Dietary Intake and Risk from Short Term Exposure to Inorganic Arsenic in Apple Juice

The risk associated with inorganic arsenic exposure via apple juice consumption can be estimated by calculating its chronic daily intake (CDI). The equation used to estimate CDI (mg/kg-day) of inorganic arsenic includes the following parameters; the concentration (C) of inorganic arsenic in consumed apple juice, the average daily intake (DI) of apple juice (L/day), and the average body weight (kg BW) of persons who consume apple juice. Until now, National Health and Nutrition Examination Survey (NHANES) data use calculated CDI (Tvermoes et al., 2014). This equation is used only for short term exposure.

CDI (mg/kg-day) = C (mg/L) * DI (L/day) / BW (kg).

2.4.2 Lead

Lead (Pb) is a toxic heavy metal that occurs naturally in the environment in small quantities. Because of its low melting point, lead can be shaped and combined with different metals to formulate alloys. Thus, lead has been used widely by humans for thousands of years for various products such as pigments, pipes, ammunition, storage batteries, glazes, paints, vinyl products, cable covers, weights, shot and radiation shielding (WHO, 2010). Generally, human exposure to small amounts of lead has no meaningful impact on public health. However, the real concern is

exposure to large quantities of lead because it may lead to lead poisoning (EPA, 2006, FDA, 2011b).

Lead has organic and inorganic forms, and all of these forms are toxic. Inorganic lead is found in soil, dust, old paint and several products. The organic form (tetra-ethyl lead) was used extensively as an additive in gasoline. The combustion process of gasoline containing organic lead releases lead into the air. Organic lead is quite dangerous and poses more risk and is more toxic to the central nervous system of humans compared to the inorganic form (WHO, 2010).

2.4.2.1 Lead Toxicity and its Health Effects

Lead toxicity has been recognized since 2,000 BC, and can affect every system in the human body. The primary routes of lead exposure in humans are oral and inhalation (WHO, 2010). Inorganic lead can accumulate in the skeletal system, and bone holds around 90% of the total body burden of lead in humans. Thus, fast release of stored lead from bone can result in a health risk for people who have skeletal diseases (EPA, 2006).

Lead toxicity affects the nervous, renal and vascular systems (FDA, 2011; WHO, 2010). The degree of toxicity differs greatly from one organ system to another. For example, in the brain, acute lead poisoning can cause fatal encephalopathy while exposure to low amounts of lead can change the functions of the nervous system. In the kidney, chronic exposure may result in nephrosclerosis (Patrick, 2006).

More importantly, the adverse effects of lead depend upon many factors such as duration and amount of exposure and age of the individual. Exposure to high concentrations of lead may cause severe effects that need urgent medical attention (FDA, 2010). However, the health effects from chronic lead exposure are of greater concern for the developing fetus, infants and young children because they are at high risk from the toxicity of lead for many reasons such as

exposure to lead during critical developmental stages, the potential for relatively high exposures in relation to body weight, and the potential for life-long health effects (WHO, 2010; FDA, 2011b). Furthermore, chronic exposure of children to lead is linked with impaired cognitive abilities/function and behavior difficulties in addition to other problems (FDA, 2011b).

Finally, no safe level of lead has been identified in studies that have assessed lead exposure by measuring blood lead concentrations (CDC, 2016). As a result, blood testing in children is the best way to know if they have lead poisoning because these children may not appear to be sick or have obvious clinical signs (CDC, 2015f; FDA, 2010).

2.4.2.2 How Do Humans Become Exposed to Lead?

Humans become exposed to lead by different routes such as inhaling dust, drinking water and eating food (FDA, 2010). Lead is ubiquitous in the environment in small amounts that arise from human activities or that occur naturally. Plants absorb lead from the soil and therefore can contribute to lead consumption from plant-based foods (Tvermoes et al., 2014). Lead incorporated into plant tissues cannot be removed by processes such as washing (FDA, 2011b). As a result, several food products consumed in the diet contain small concentrations (typically parts per billion concentrations) of lead (FDA, 2011b).

Millions of people in the US are at high risk of exposure to potentially toxic levels of lead each year. It estimated that more than three million US workers are at risk from occupational exposure to lead every year (WHO, 2010). Furthermore, millions of children who are living in more than four million households are exposed to high amounts of lead every year in the US. There are at least one half million US children between one and five years of age who have blood lead concentrations in excess of the reference concentration of lead (5 µg/dL) that is recommended by the CDC (Tvermoes et al., 2014).

In the US, the Centers for Disease Control and Prevention and the Food and Drug Administration have regular programs for assessing lead exposure in children and testing lead concentrations in food. Since 1995, the CDC has implemented a national program (Childhood Lead Poisoning Prevention Program) for collecting childhood blood lead surveillance data in the US and the agency receives approximately 2.5 million tests of children's blood every year from different states. This program gathers data from state and local health departments. The main goal of CDC's program is reducing lead concentrations in the blood of US children to less than 10 µg/dL (CDC, 2013b). In order to assess the concentration of blood lead in US children, CDC had reported data collected from 1997 to 2014 (Figure 1). Between 1997 and 2014, the data in Figure 1 illustrate that the percentage of children having blood lead concentrations in excess of 10 µg/dl has decreased dramatically from 7.6% in 1997 to less than 1% in 2014 (CDC, 2013b; CDC, 2015f).

Since 1970, FDA has been working with the food industry to conduct the Total Diet Study to assess the concentrations of lead and other contaminants in food products. The Total Diet Study is an annual FDA program for surveillance of the most consumed food products in the US for food safety purposes. The tests are carried out on various food products including fruit juices, canned fruit and vegetable and baby foods (FDA, 2011b). Historically, FDA has found various foods including potatoes and carrots that contained lead at concentrations exceeding 15 ppb. These foods are often used as baby food ingredients and, therefore, these relatively high concentrations are a concern. In 2006, FDA established the recommended maximum lead level in candy to be 100 ppb (0.1 ppm) because candy is frequently consumed by children (FDA, 2011b).

In 2010, FDA tested 13 samples of specific products including apple juice, grape juice, pears, mixed fruit, peach slices and fruit cocktail to determine lead concentrations. FDA tested these 13 samples because these products were cited in an action taken by the Environmental Law Foundation (ELF), a California-based private advocacy organization, which alleged that several manufacturers of these food products were not in compliance with California's Proposition 65 requirements (FDA, 2011b) because the products contained excessive lead. In its testing, FDA found that almost all of the 13 tested samples contained lead but it was present in only small amounts that were lower than the tolerable intake levels FDA has established for various age/sex groups (FDA, 2011b).

In 2014, FDA published a summary of results of the Total Diet Study from 2006 through 2011 (FDA, 2014b). This summary report assessed 382 chemical elements in different foods and beverages including apples and apple juice. The lead concentrations did not exceed the FDA standard for tolerable intake levels for any of the products tested (FDA, 2014b).

FDA is still concerned about the lead concentration in foods or other sources because high lead exposures can affect immune function, the central nervous and the kidneys. Consequently, FDA has established legal limits for lead concentrations in bottled water, and some listed food ingredients such as juice, candy and sugar. Moreover, FDA declared in its "Guidance for Industry: Juice HACCP Hazards and Controls" that 50 ppb is the maximum level of lead in juice, and concentrations above this level can be considered to be a health hazard (FDA, 2010). Finally, FDA is working to decrease the lead concentration in food products as far as possible, especially in foods that are mostly consumed by children (FDA, 2011b).

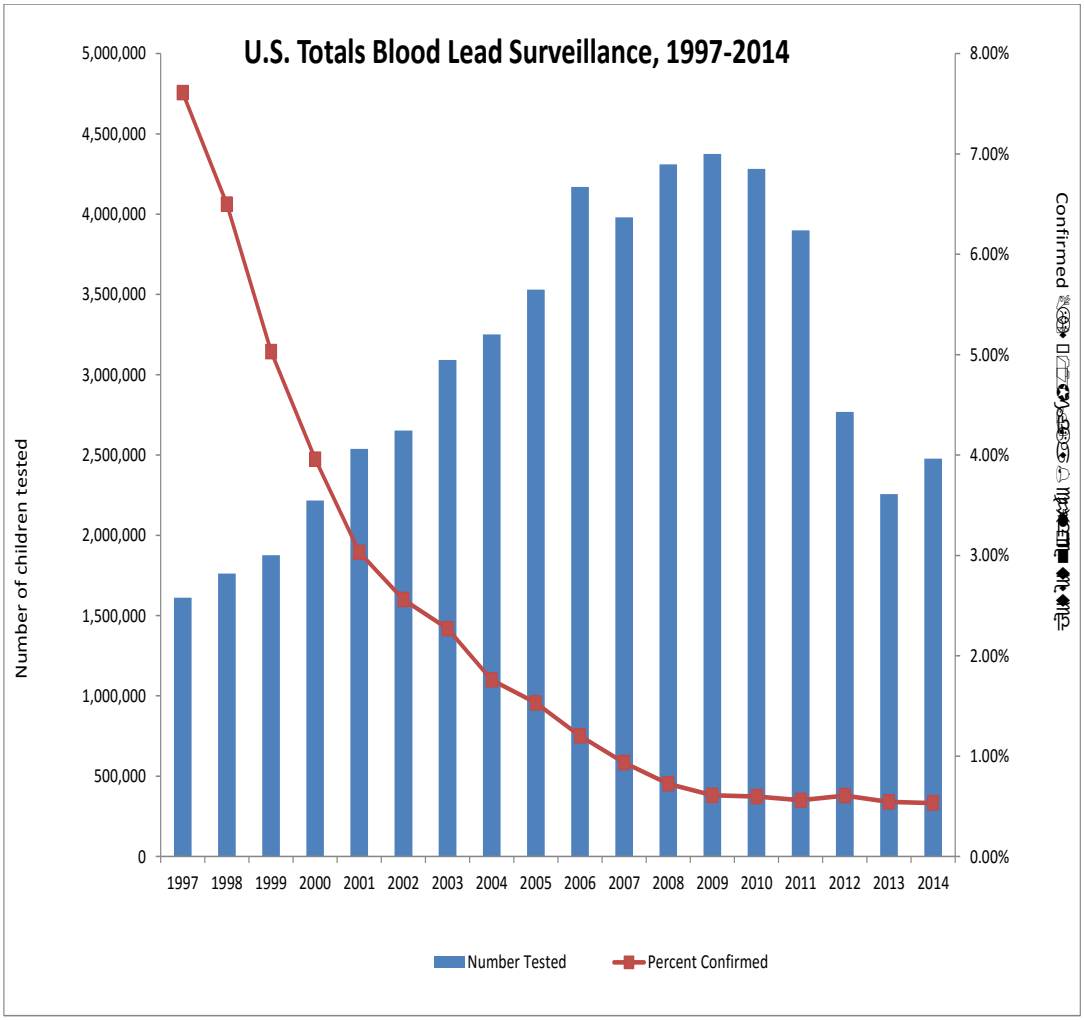

Figure 1. CDC surveillance summary of US children having blood total lead concentrations in excess of 10 ug/dL, 1997-2014 (CDC, 2016c).

2.4.3 Patulin

Patulin is a toxic secondary chemical metabolite that is produced by different species of fungi (molds) including *Penicillium*, *Byssochylamys* and *Aspergillus* (Martins, 2002; FDA, 2001). These molds can grow on different types of foods such as fruits, cheeses and grains

(Harwig et al., 1973; FDA, 2001). The primary source of patulin in fruits is from growth of *Penicillium expansum* (Jackson, 2003; Moss, 2008). *Penicillium expansum* occurs naturally in soil and is essentially ubiquitous in apple orchards and on apple handling equipment such as wooden bins (Jackson, 2003; Moss, 2008; FDA, 2001). Patulin has been reported in several tested foods including pears, apples and apple juice (Evan, 1999; FDA, 2001). Its presence is generally restricted to the area of decay (rot) caused by *Penicillium* infection of the fruit (Jackson, 2003; Moss, 2008). Patulin is reduced slightly during typical thermal processing conditions used in food manufacturing processes. As a result, patulin remains in apple juice after pasteurization. Direct exposure from consumption of moldy apples is unlikely as people typically would avoid consuming decayed fruit (FDA, 2001). For that reason, exposure to patulin is generally restricted to processed foods or ingredients such as apple juice, apple sauce or apple juice concentrate.

However, some processes can destroy patulin. For example, patulin cannot be found in alcoholic fruit beverages or vinegars because it is destroyed during the fermentation process (IARC, 1986; WHO, 1990; Harrison, 1989). In most grains and fruits, the rot can be physically removed before consumption (FDA, 2001). The nature of some foods such as cheese helps protect them from patulin formation. Cheese contains high concentrations of cysteine that can form adducts with patulin and thereby reduce its toxicity (Harwig et al., 1973; FDA, 2001).

The potential toxicity of patulin has been extensively assessed by the International Agency for Research on Cancer (IARC, 1986), WHO (1990), and the US FDA (FDA, 1994; FDA, 2001). In its toxicological assessment of patulin, FDA concluded that patulin is toxic based on oral doses of 1.5 mg/kg BW that caused early death in rats (FDA, 2001). Acute exposure to patulin affects several tissues and organs such as the gastrointestinal tract, liver, kidney, and components

of the immune system (Moake, 2005). Chronic exposure to patulin may cause mutagenicity, genotoxicity, clastogenicity, and embryotoxicity. Moreover, high dosages of patulin may cause neurotoxicity, immunosuppression and immunotoxicity (Moake, 2005; Moss, 2008).

However, there is no strong evidence for patulin carcinogenicity (FDA, 2001). Due to demonstrated toxicity of patulin in animal studies, FDA determined that persons who are exposed to patulin at relatively high concentrations may be at risk from this exposure (FDA, 2001). The risk assessment of patulin toxicity conducted by FDA resulted in the establishment of a No Observable Adverse Effect Level (NOAEL) for patulin exposure of 0.3 mg/kg BW/week. FDA established 0.43 µg/kg BW as the provisional tolerable daily intake (PTDI) of patulin in apple cider and juice (FDA, 2001). Based on this analysis, FDA established an action level of 50 µg/kg for patulin in apple juice, apple juice concentrate and apple sauce (FDA, 2001). This maximum level of patulin in apple juice (50 µg/kg) is in agreement with concentrations recommended by the World Health Organization and the European Union (FDA, 2001).

Following Good Agricultural Practices (GAP) and Good Handling Practices (GHP) can minimize the likelihood of patulin contamination. For example, culling apples and not using rotten or spoiled apples can help minimize patulin contamination during the process of juice production. These practical steps can manage patulin exposure successfully (USDA, 2016a).

Since 2003, FDA surveillance has resulted in multiple recalls of apple juice products due to patulin concentrations that exceeded 50 µg/kg (Harris, 2009). Other countries also have aggressively enforced regulations to reduce the patulin residues in apple juice products. For example, patulin surveillance data from the United Kingdom demonstrated that apple juice products containing patulin concentrations in excess of 50 µg/kg decreased from 26% of samples to 2% during a six-year period following establishment of the action level (Evan, 1999).

The US FDA has conducted and reported surveillance data on patulin concentrations in apple juice and apple cider in the US on multiple occasions. Between 1973 and 1977, FDA surveyed patulin concentrations in 176 samples of apple juice sold in U.S markets and found patulin concentrations ranging from 10 to 440 ug/kg (Ware, 1975; Stoloff, 1976). From 1994 through 2000, FDA conducted a risk assessment of patulin in apple juice that included results of analyses of 1,525 samples obtained from US marketplaces. In this survey, 56.2% of the apple juice samples contained detectable patulin, and 12.6% of samples contained patulin concentrations exceeding 50 μg/kg (Roach, 2002).

Harris et al. (2009) assessed patulin prevalence and concentrations in apple cider and juice marketed in Michigan. This study included an assessment of 493 end product samples of apple cider produced by Michigan cider mills from 2002 to 2004. Harris et al. (2009) also determined patulin concentrations in various brands of apple juice purchased from retail grocers in Michigan in 2005 and 2006. The results from this study demonstrated that 18.7% from all apple cider samples from mills contained detectable patulin (>4 μg/kg) while 2.2% of cider mill samples contained patulin concentrations exceeding 50 μg /kg (Harris et al., 2009). For the apple juice samples obtained from retail grocery stores (159 samples), 23% contained detectable patulin while 11.3% contained patulin concentrations in excess of 50 μg /kg (Harris et al., 2009).

Powell and Bourquin (2011) published an abstract that summarized FDA patulin surveillance analyses on all samples of domestic and imported 100% apple juice the agency tested between 1994 and 2008. The results from this study demonstrated that 50.2% of all samples contained detectable patulin, while 4.4% contained patulin concentrations 50 μg/kg.

2.5 Supply and Consumption of Apples in the United States

The US is the second leading country (after China) in production, import and supply of fresh apples. From 2014 to 2015, the total supply of fresh apples in the US was 8,306.7 million pounds and 96% (7,946.6 million pounds) of this amount was produced domestically (USDA, 2016b). While large amounts of apples are consumed as a fresh fruit, processed apple products such as apple juice and cider are in high demand in the US (USDA, 2016b). In 2015, the total production of apples in the US was 10.1 billion pounds and 91% of this production was from four states (Table 10) – Washington, New York, Michigan and Pennsylvania (USDA, 2015).

Table 10. Leading states in apple production in the US in 2015.

State	Production (million pounds)
Washington	6,688
New York	1,100
Michigan	999
Pennsylvania	525

Source: (USDA, 2015).

2.5.1 NHANES and Apple Consumption

The National Health and Nutrition Examination Survey (NHANES) is a national data collection survey program designed to provide information for evaluation of health and dietary conditions of the US population (CDC, 2015g). This program began in the 1960s and is a prime program of CDC's National Center for Health Statistics (NCHS). NHANES uses survey tools to gather data from the targeted populations. The NHANES surveys are distinctive because the data come from the combination of physical examinations and interviews (CDC, 2015g). In 1999,

NHANES became a continuous program to collect representative samples from approximately 5,000 US participants annually (CDC, 2015g).

One of the fundamental outcomes of the NHANES program is providing food consumption data for US adults and children using self-reported assessment of consumption of over 6,000 different food and beverage items during two non-consecutive days. Since NHANES food consumption surveys characterize food consumption of the population at all ages, the population average is reasonably representative of average per capita exposure over a lifetime (Carrington et al., 2013). However, NHANES food consumption surveys also have several limitations. First, the surveys are conducted at a particular point of time and do not characterize consumption during childhood and adulthood for the same individual. In addition, surveys based on self-reported food consumption over two days are not adequate for estimating individual consumption or the variation in long-term consumption in a population. For these reasons, it is not possible to estimate lifetime consumption rates at specific population percentiles using NHANES survey data (Carrington et al., 2013).

2.5.2 Consumption of Apple Products in the US

Fresh apples are among the most consumed fruits by the US population, although there has been some reduction in apple consumption over time. For example, during 1980 to 1990, the annual consumption of fresh apples (per person) ranged between 19.2 to 21.1 pounds per capita, but by 2011 to 2012 apple consumption decreased to 15.3 pounds per capita (USDA, 2015).

Apple juice is one of most consumed juices in the US. In 2012, USDA reported that 2.6 billion liters of apple juice were consumed annually in the US. The majority (two thirds) of the apple juice sold in the US is manufactured using imported apple juice concentrate from China and considerable amounts of apple juice concentrate are imported from other countries such as

Canada, Brazil, Chile and Argentina. However, US apple juice production from domestic apples only accounts for approximately 17% of apple juice consumption (USDA, 2011; USGS, 2011). China is the leading global producer and exporter of apple juice concentrate. In the US and many other countries, apple juice concentrate is the main ingredient used for production of apple juice and other beverages (Gale, 2010; USDA, 2010).

Adjusted for body weight (Table 11), children consume much greater quantities of apple juice than adults in the US (NHANES, 2009; Exposure Factors Handbook, 2011).

There are no published data on consumption of unpasteurized (other otherwise treated to kill pathogenic microorganisms) apple cider in the US (Perez, 2016). However, the majority of the reported foodborne illness outbreaks associated with apples or processed apple products in the US are associated with consumption of unpasteurized apple cider. More information on production and consumption of unpasteurized juices would be useful to facilitate development of risk assessment models for unpasteurized apple juice products.

Table 11. Average apple juice consumption by age group in the US.

Age	Average apple juice consumption (g/day)	Mean body weight (kg)	Consumption per weight (g/kg body weight/day)
0-6	203	12.1	16.9
7-70+	318	64.8	5.1

Source: (Tvermoes et al., 2014; US EPA, 2011).

2.6 FDA-iRISK

FDA-iRISK is a web-based system designed to analyze data concerning microbial and chemical hazards in food and return an estimate of the resulting health burden on a population level. FDA-iRisk integrates data for seven elements: food, hazard, population, process model,

consumption model, dose response model and health effect (Figure 2) after the user defines them using the built in templates, and generates results from these inputs through Monte Carlo simulations (Figure 3). As is standard in quantitative risk analyses, FDA-iRisk defines risk by connecting probability and consequence through specific risk scenario-risk assessment models (Figure 4).

FDA-iRISK depends on using two scenarios to estimate the probability of consequences of consuming contaminated food on the population. These scenarios are Monte Carlo simulation and specified risk scenarios (FDA, 2012; Chen et al., 2013). FDA-iRISK has a model structure for microbial hazards and model structure for chemical hazards. The consumption model for chemicals is based on consumption per unit body weight per unit time, whereas the consumption model for microbial or other acute hazards is per serving.

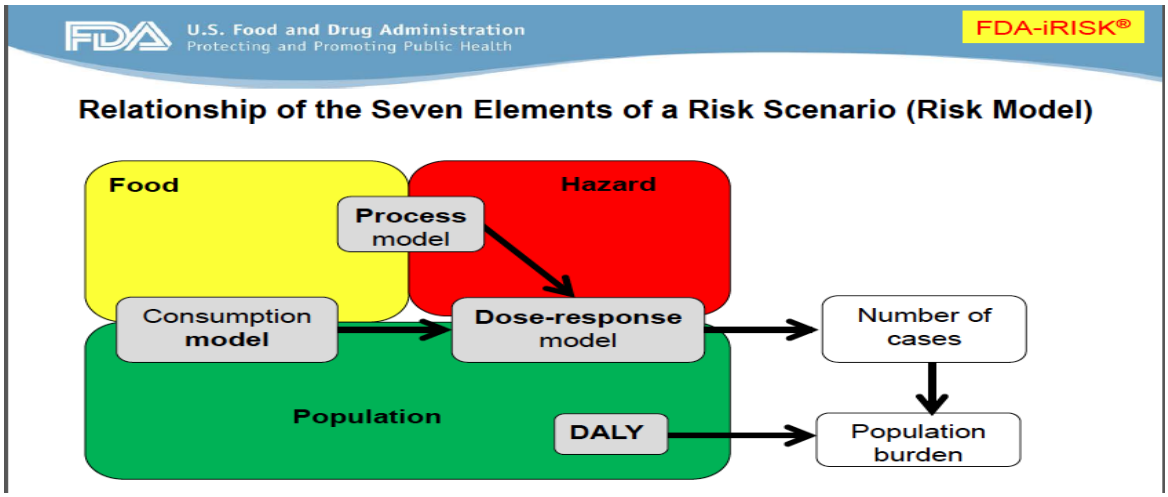

Figure 2. The relationship between the seven elements of a risk scenario (FDA, 2012)

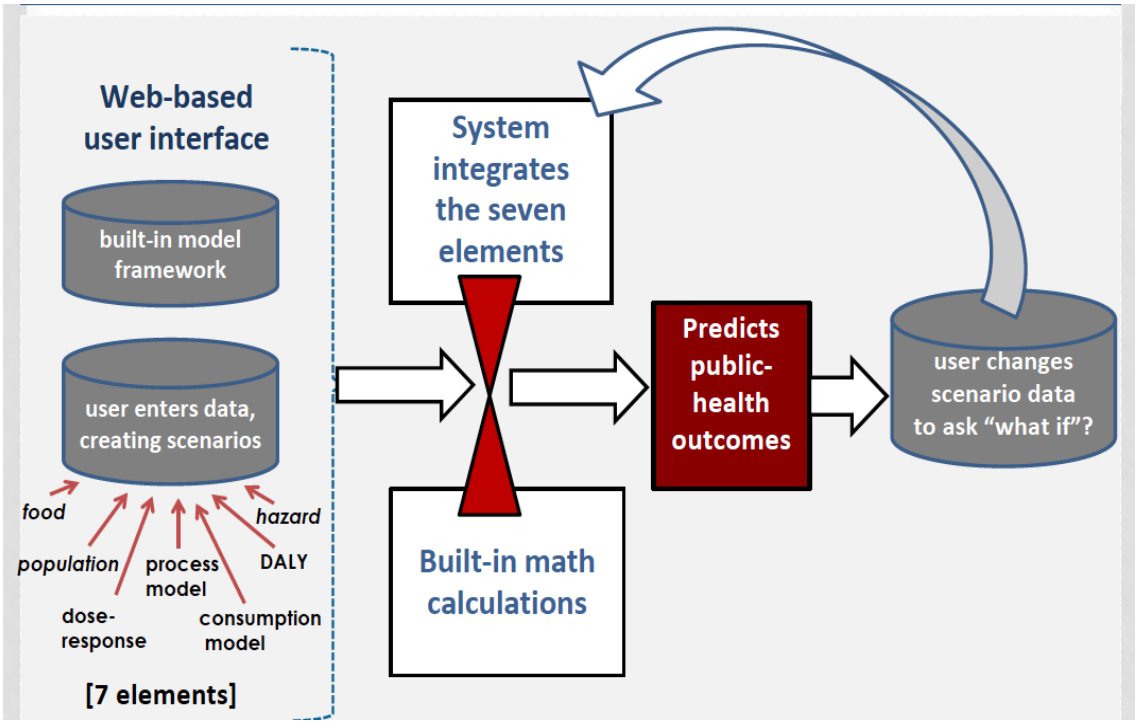

Figure 3. Summary: Overarching approach of FDA-iRISK (FDA, 2012).

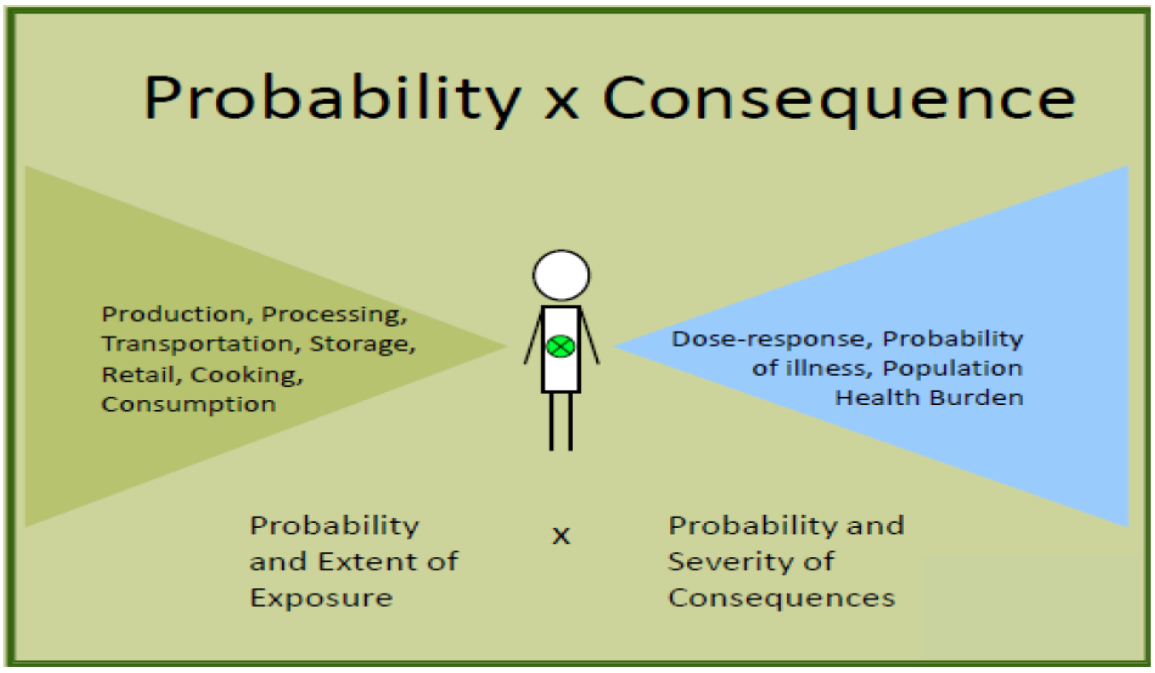

Figure 4. The definition of risk implemented in FDA-iRISK (FDA, 2012).

2.7 Disability Adjusted Life Years (DALYs)

The use of Disability Adjusted Life Years (DALYs) is a common approach to evaluate the effect of risk on human health (Devleesschauwer, 2014). The WHO describes a DALY in the following manner: “One DALY can be thought of as one lost year of healthy life. The sum of these DALYs across the population, or the burden of disease, can be thought of as a measurement of the gap between current health status and an ideal health situation where the entire population lives to an advanced age, free of disease and disability” (WHO, 2016).

DALY estimates are driven by number of factors including the likelihood of occurrence and/or concentration of various hazards as well as the level of consumption of the food product by consumers (WHO, 2015a). DALYs are calculated as “the sum of the Years of life lost to mortality (YLL) due to premature mortality in the population and the years lost due to morbidity (YLD) for people living with the health condition or its consequences” (WHO, 2015a). The summarized equation of DALYs (WHO, 2015a) is DALY = YLL + YLD. The equation for calculating cumulative DALYs in a population can be further expanded to the following (WHO, 2015a).

DALY = YLL + YLD

Years of life lost due to early mortality (YLL) is estimated as:

$$YLL = \sum_{l} n_l \times e_l$$

Where:

(n) = the summation of all fatal cases due to the health outcomes of a specific disease(i), and

(e) = the expected individual life span in a specified population minus the observed age of death.

Years of life lost due to morbidity (YLD) is estimated as:

$$YLD = \sum_{l} n_l \times t_l \times w_l$$

Where:

(n) = the number of cases in the population,

(t) = the duration of the illness until remission or death, and

(w) = the severity weight of a specific disease or disability weight.

3. RATIONALE AND SPECIFIC AIMS

3.1 Rationale

Identifying the most predominant hazards in specific foods, such as microbial pathogens and toxic chemicals, has been a major development in the food science field during the past several decades. However, a critical question facing the field today is determining which food hazards represent the greatest risk to consumers of specific foods (Morris et al., 2011). Ranking the risk of food hazards is a basic step that government, policy makers and other food industry decision makers need when designing strategic plans to protect the public against risks associated with eating foods contaminated with toxic chemicals and pathogenic microbes (Morris et al., 2011)

Apple juice is a popular drink for consumers in the US, where approximately 2.6 billion liters of apple juice were consumed in 2012 (USDA, 2012). However, there are major foodborne hazards associated with apple products such as arsenic and patulin. For example, concern exists regarding the concentration of arsenic (As) in apple juice (FDA, 2016). Patulin is a mycotoxin produced in apples by *Penicillium expansum* and the US Food and Drug Administration has established an action level of 50 µg patulin per kg for apple juice, apple sauce and apple juice concentrate (when diluted to single strength) in the US. Surveillance indicates that patulin concentrations in commercial juice often exceed this action level (Harris, 2009).

Apple juice products that have not been treated by pasteurization or other pathogen-destruction technologies have frequently been associated with foodborne illness outbreaks caused by microbial pathogens in the past two decades (CDC, 2014). The major pathogens

associated with these outbreaks include pathogenic *E. coli* (O157:H7 and O111) and *Cryptosporidium parvum*.

Whereas the most commonly occurring pathogenic microbes and toxic chemicals in US apple products are well characterized, the relative risk to public health presented by these various food hazards in different apple products is not well understood. This research aims to fill this knowledge gap. At the conclusion of this research, the relative risk associated with each of these hazards will be well characterized. The results of this work will provide policy makers, other food industry professionals, and consumers with information needed to minimize the potential public health impact of these hazards.

3.2 Specific Aims

The overall goal of this research is to rank the most common food safety hazards associated with fresh apples and apple juice products with respect to their potential public health impact on US citizens. The specific objectives of this research are:

1. To identify the most common microbial and chemical hazards associated with fresh apples and apple juice products in the US.

2. To determine the likelihood of foodborne illness caused by microbiological hazards associated with fresh apples and apple juice products in the US (apple juice and unpasteurized apple cider).

3. To determine the concentrations of potential chemical hazards associated with fresh apples and apple juice products in the US.

4. Using the information collected in aims 1-3, to conduct a quantitative risk assessment using FDA iRisk to rank the health impacts of these chemical and microbial hazards associated

with fresh apples, apple juice and unpasteurized apple cider using DALYs as the primary ranking metric.

4. MATERIALS AND METHODS

This research focused on comparing the relative public health impact of the major food safety hazards commonly associated with apple juice and cider products and fresh apples consumed in the United States. Hazard identification and determination of the prevalence and concentrations of selected hazards was accomplished by conducting a comprehensive literature review encompassing peer-reviewed publications, government documents (e.g. FDA, CDC and EPA publications), international organizations (e.g. EFSA, FAO, WHO) and "grey" literature from reliable sources. This literature review focused on food safety hazards identified in apples and apple juice and cider products during the time frame of 1991 to 2015. Both microbiological and chemical hazards are considered in this analysis. Data generated from this literature review were used to determine estimates of food safety hazard prevalence and/or concentrations in these products in the US. Rather than point estimates, we determined, where possible, the relative empirical distributions of selected hazards in these products. The parameters of chemical hazards were entered into the FDA-iRISK 2.0 model using the online website (https://irisk.foodrisk.org).

FDA-iRisk is a computational application to estimate and compare disability adjusted life years (DALYs) associated with exposure to identified food safety hazards. As will be described in more detail later in this section, FDA-iRisk was used to calculate DALYs associated with chemical hazards in this study. A general overview of the inputs used for FDA-iRisk in this research are outlined in Table 12. Because there is no published literature adequately describing the prevalence and distribution of microbial pathogens in apples and apple juice products that are the focus of this research, the DALYs associated with each of these pathogens were calculated

manually based on the numbers of cases associated with reported foodborne illness outbreaks after adjustment for under-diagnosis and under-reporting using the scaling factors reported by Scallan et al. (2011a).

Briefly, DALY is a parameter used to estimate and rank the negative health outcomes (morbidity and mortality) due to illnesses or diseases that are attributable to chemical or microbiological hazards. In this research, the focus was on estimating DALYs for the major chemical and microbiological hazards associated with the consumption of fresh apples and apple juice and cider.

Table 12. Description of the seven elements of FDA-iRISK based on the goals of the present research.

#	The Seven Elements		Description
1	Food:		Consumed fresh apples, apple cider and apple juice in the US.
2	Hazard	Chemical	Lead, inorganic arsenic, and patulin found in consumed fresh apples, apple cider and apple juice in US from 1991 to 2015.
		Microbial	Microbial pathogens implicated in foodborne illness outbreaks associated with consuming contaminated fresh apples, apple cider and apple juice in US from 1991 to 2015. [FDA-iRisk was not used for these calculations because prevalence and distributions of microbial pathogens in these food products are not available in the published literature.]
3	Population of consumers		In this study we used two population groups (0-6 years of age and 7-70+ years of age).
4	Consumption model		FDA-iRISK provides two consumption models for consumers of interest. One is acute exposure (eating occasions per year) and the other is chronic exposure (amount per day in life stages). Chronic exposure models were used for chemical hazards in this research.
5	Process model		The function of the process model is to serve to connect models for hazard prevalence and distribution with consumption models for the food items. The process model also can be used to account for changes in hazard parameters (concentration, prevalence, etc.) due to processing steps.
6	Dose response		Dose response provides an estimate of the probability of adverse effects of the chemical or microbial hazards at varying doses.
7	DALY		Used to estimate the burden of disease associated with health effects from the microbial and chemical hazards by combining morbidity and mortality outcomes in one measure. DALY was chosen because “It is an established WHO metric with international application” and “it is consistent with the Global Burden of Disease project” (WHO, 2015).

4.1 Apple and Apple Juice Consumption

The US population in 2015 was 321,418,820, with 27,944,646 infants and children 0-6 years of age and 293,474,174 persons aged 7 years and older (United States Census, 2016). Consumption patterns of apples and apple juice for the two age groups (0-6 years and 7 years and older) were derived from US EPA's *What We Eat in America (WWEIA) - Food Commodity*

Intake Database (FCID), 2005-2010 (available online at http://fcid.foodrisk.org). This database translates food consumption as reported eaten in NHANES (1999-2010) survey cycles) and CSFII (Continuing Survey of Food Intake by Individuals, 1994-96/1998) surveys into consumption of EPA-defined food commodities.

The WWEIA-FCID database (Foodrisk, 2016) was queried to produce estimates of total consumption of apples and apple juice for the two age groups. The commodity code used for apples was "apple, fruit with peel". Two commodity codes were used to estimate apple juice consumption, "apple, juice" and "apple, juice-baby food". For the US population aged 0-6 years (WWEIA-FCID data based on 9,106 persons), the average daily consumption of apple juice and apple, juice-baby food was 4.46 grams per kg body weight per day (Foodrisk, 2016).

Because no consumption data for apple juice and apple cider separately are available, we assumed that a small percentage of total apple juice consumption was contributed by unpasteurized apple cider. The assumptions were based on approximate numbers of cider mills in the US and their approximate volumes of annual production. It was also considered that young children were much less likely to consume apple cider compared to older children and adults. For the US population aged 0-6 years, we assumed that 1% of total apple juice consumption was apple cider. For the US population aged 7 years and older, we assumed that apple cider accounted for 5% of total apple juice consumption. Apple cider consumption was calculated by applying these percentages to total apple juice consumption data, and the apple juice consumption distributions were corrected to account for estimated cider consumption.

Based on these assumptions and total apple juice consumption from the WWEIA-FCID database, for this age group we estimated the average daily consumption of apple juice to be 4.4

g/kg BW/day (calculated by multiplying 4.46 * 0.99) and we estimated the average daily consumption for apple cider to be 0.045 g/kg BW/day (calculated by multiplying 4.46 * 0.01).

For the US population aged 7 years and older (WWEIA-FCID data based on 40,237 persons), the average daily consumption of apple juice and apple, juice-babyfood was 0.26 grams per kg bodyweight per day (Foodrisk, 2016). We estimated the average daily consumption of apple juice to be 0.247 g/kg BW/day (calculated by multiplying 0.26 * 0.95) and we estimated the average daily consumption of apple cider to be 0.013 g/kg BW/day (calculated by multiplying 0.26 * 0.05).

The average daily consumption of apple, fruit with peel for the US population aged 0-6 years was 1.05 g/kg BW/day (Foodrisk, 2016). The average daily consumption of apple, fruit with peel for the US population aged 7 years and older was 0.28 g/kg BW/day (Foodrisk, 2016). These average consumption data for the two age groups are summarized in Table 13.

Table 13. Summary of the average consumption values (g/kg BW/day) of apple products in two age groups in the US, and estimates of annual consumers of these products based on the total US population.

Parameter	**0-6 year**			**7-70 + year**		
	Fresh apple	Apple juice and juice-baby food	Apple cider	Fresh apple	Apple juice and juice-baby food	Apple cider
Average Daily Consumption: (g/kg BW/day)	1.05	4.40	0.045	0.28	0.247	0.013
Annual Consumers	**27,944,646**			**293,474,174**		

Sources: Foodrisk (2016) and US census data.

Data from the WWEIA-FCID database (Foodrisk, 2016) also were used to develop empirical distributions of consumption of apples, apple juice and apple cider. In addition to average consumption for the overall population and consumers of each food commodity, the database provides consumption estimates for five percentile increments of the population. We used these data to calculate the empirical distributions of consumption for each commodity as follows:

1. The proportion of consumers of each food was calculated by dividing the commodity eaters by total filtered population.
2. The non-consumers were calculated by subtracting the proportion of consumers from the number one (1- the proportion of consumers).
3. Every decimal fraction of population was multiplied by the proportion of consumers and then adding the result to the proportion of non-consumers.

Appendix 1 presents the output data from the WWEIA-FCID database used to calculate the empirical distribution of apple consumption by infants and children aged 0-6 years. Using the data in Appendix (Table38), we generated the empirical distribution of consumption values for apples by this age group. The results are presented in Appendix; Table 39. Appendices 40 through 47 provide similar data used for deriving empirical distributions of consumption of apples, apple juice and apple cider by all age groups in this study. These empirical distributions were input into FDA-iRisk to serve as the consumption models used in estimating health effects of chemical hazards.

4.2 Chemical Hazards Risk Assessment

Risk assessment modeling using FDA-iRISK requires the user to input data about the food and the relevant hazards. With respect to the foods being assessed, the user is required to provide consumption data and information for the process models with respect to the initial prevalence and/or concentrations of the hazard, the unit mass of the food, and processing steps that can influence hazard levels. User-provided information for the hazard models includes dose response and the health metric (e.g. DALY) associated with each hazard. Table 14 outlines the data structure for this research to assess chemical hazards in apples, apple juice and apple cider. This table includes our definitions, calculations and key assumptions for each of the parameters. We utilized the FDA-iRISK tool with fresh apple, apple juice and apple cider for the three chemicals.

Table 14. Guideline of input data for chemical hazards in FDA-iRisk.

Element	Model	Parameter	Note
Food	**Consumption and Population group**	**1. Annual Consumers**	Assumed to be the entire population of the US (US Census Data).
		2. Average Daily Consumption:	Apple consumption model derived from WWEIA-CFID (Foodrisk, 2016).
		3. Body Weight	Not needed in this model because the consumption model used g/kg BW/day.
		Unit	µg/kg-day
	Process	**1.Initial Units Are Contaminated**	Yes for all three chemical hazards assessed – inorganic arsenic, lead and patulin.
		2. Initial Unit Mass	The size of the tested contaminated food. We assumed it to be 100 g for every product (fresh apples, apple juice and apple cider).
		3. Initial Prevalence	It was assumed that the initial prevalence was 1 for all hazards (all the samples are contaminated). However, hazard concentration data were input using empirical models for inorganic arsenic in apple juice and patulin in apple juice and apple cider.
		4. Initial Concentration (Inorganic Arsenic, Lead, Patulin)	Based on the available data, we used the following: 1. Inorganic Arsenic: we used a fixed value for fresh apple and apple cider, but for apple juice we used an empirical distribution based on FDA surveillance data. 2. Lead: we used a fixed value with the three products. 3. Patulin: we used a fixed value of zero for fresh apple (assumed no voluntary consumption of apples molded with *Penicillium expansum*) and an empirical distribution for apple juice and cider.
		Unit	µg/kg
		Stages	We did not use process stages.

Table 14 (cont'd)

Element	Model	Parameter	Note
Hazard	Dose Response	**Exposure Type:**	Chronic for the all three chemical hazards.
		Dose Unit:	µg/kg-day
		Response Type	1. The response type for inorganic arsenic is Linear by Slope Factor. 2. The response type for lead is Threshold Linear. 3. The response type for patulin is Threshold.
		Probability of Adverse Effect Given Response	100% for all three chemical hazards.
	Health Metric	**DALY**	Based on the available data: 1. Inorganic Arsenic: DALY for inorganic arsenic health endpoints for lung and bladder cancers are from the 2013 FDA draft risk assessment for inorganic arsenic (see Tables 23, 24, and 25). 2. For Lead (see Table 26).

4.2.1 Determining the Concentration of Chemical Hazards

4.2.1.1 Inorganic Arsenic Concentration in Apple Juice

The concentration of inorganic arsenic was calculated as a cumulative empirical distribution using FDA surveillance data for total and inorganic arsenic (FDA, 2011), and these data were used in FDA-iRisk as a process model for the distribution of inorganic arsenic concentrations in apple juice. The FDA surveillance data used to calculate the empirical distribution are presented in Appendix 11 and the resulting empirical distribution is presented in Table 15.

Table 15. Calculated cumulative empirical distribution of inorganic arsenic concentrations in apple juice.

Probability	Concentration (µg/kg)
0	0
0.28	1.4
0.30	2.8
0.31	3
0.32	3.1
0.33	3.5
0.34	3.8
0.38	3.9
0.39	4
0.40	4.1
0.41	4.2
0.43	4.3
0.44	4.5
0.45	4.6
0.47	4.7
0.48	4.8
0.52	4.9
0.54	5
0.60	5.1
0.62	5.2
0.65	5.3
0.69	5.4
0.70	5.5
0.72	5.6
0.76	5.8
0.78	6.5
0.79	6.6
0.80	6.7
0.81	6.8
0.83	6.9
0.85	7
0.87	7.2
0.89	7.5
0.90	7.7
0.91	7.8
0.93	7.9
0.94	8.1
0.96	8.2
0.97	8.3
0.99	8.4
1.00	9.8

4.2.1.2 Inorganic Arsenic Concentration in Apple Cider

The inorganic arsenic concentration in apple cider was calculated using surveillance data on total arsenic concentrations in apple cider (78 samples) reported by Cao and Bourquin (2016). A fixed value was used in FDA-iRisk. The average concentration of total arsenic determined by Cao and Bourquin (2016) (0.37 µg/L) was multiplied by the average ratio of inorganic arsenic to total arsenic (0.748) observed in the surveillance study on apple juice by FDA (2011). The resulting value was 0.37 µg/L * 0.748 = 0.28 µg inorganic arsenic per liter of apple cider. For the purposes of this research, it was assumed that 1 liter of apple cider weighed 1 kg, so the value used in FDA-iRisk was 0.28 µg/kg.

4.2.1.3 Inorganic Arsenic Concentration in Fresh Apples

Because of a lack of data on inorganic arsenic concentrations in fresh apples, we assumed that apples contain inorganic arsenic at concentrations similar to that in apple cider (0.28 µg/kg).

4.2.1.4 Lead Concentration in Apple Juice

The concentration of lead in apple juice was calculated from the average of values (samples = 41) from three available studies on lead concentrations in apple juice (Table 16). The final calculated value was 2.6 µg/L. For the purposed of this research, 1 liter of apple juice was assumed to weigh 1 kg, so a fixed value of 2.6 µg/kg was used in the FDA-iRisk model.

Table 16. Calculation of mean concentration of lead in apple juice in the US.

No of samples (n)	Mean (µg/L)	Data Source
15	3.46	Hooper et al (2012)
9	2.8	Tvermoes et al. (2014)
17	1.5	Cao and Bourquin (2016)
41	2.6	Total

4.2.1.5 Lead Concentration in Apple Cider

The value of lead concentration in apple cider was obtained from a study of lead in Michigan apple cider (n=78 samples) reported by Cao and Bourquin (2016). This is the only available study in US that reports lead concentrations in apple cider. Based on this research, a fixed value of 1.8 µg/kg was used in the FDA-iRisk model.

4.2.1.6 Lead Concentration in Fresh Apples

Because of a lack of available data on lead concentrations in fresh apples, we assumed that average lead concentration in fresh apples is equal to its value in apple cider reported by Cao and Bourquin (2016). This assumes that the lead content of apples is equal to that in expressed juice during apple cider production. The value for apple cider was used because these data are representative of lead content in US-grown apples, which is the main source of apples consumed in the US. It is worth mentioning here that Michigan is one of the top four states in the US in apple production (USDA, 2015).

4.2.1.7 Patulin Concentration in Apple Juice

The concentration of patulin in apple juice was calculated as an empirical cumulative distribution based on data reported by Powell and Bourquin (2011), which was a summary of 3,061 patulin measurements in apple juice samples conducted by FDA from 1994-2008 (Table 17). The primary data from this research was used to calculate the empirical cumulative distribution for patulin in apple juice, which is reported in Table 18. This distribution was used in FDA-iRisk as a process model for patulin in apple juice.

Table 17. Occurrence of patulin in commercial apple juices sold in the United States, 1994-2008.

		Frequency of detection (%)		Concentration (ppb)	
	n	**Detectable**	**At or above 50 ppb**	**Average**	**Range**
All Years	**3061**	**50.2**	**4.4**	**24.4 ± 1.4**	**0.4 – 1031**
1994-1998	619	56.2	4.9	25.3 ± 2.7	1-708
2002	366	63.9	6.0	30.4 ± 5.6	0.7 – 1031
2003	376	46.8	5.9	29.1 ± 3.9	0.8 – 344
2004	469	49.0	3.0	24.0 ± 3.9	0.8– 502
2005	437	50.6	5.3	20.0 ± 2.4	1 – 322
2006	299	44.5	4.0	20.3 ± 2.2	0.8– 145
2007	267	39.0	1.1	14.4 ± 1.4	0.4 – 75
2008	228	39.0	3.5	25.1 ± 4.4	1.1 – 241

Source: Powell and Bourquin (2011) and Bourquin, L.D. (personal communication).

Table 18. Calculated cumulative empirical distribution of patulin concentrations in apple juice.

Probability	Concentration (µg/kg)
0	0
0.49853	0
0.5	0.8
0.55	3
0.6	4.8
0.65	6.6
0.7	9
0.75	11.5
0.8	15
0.85	20
0.9	29.5
0.92	35
0.94	43
0.96	52.5
0.97	64.6
0.98	86
0.99	163.1
1	1030.5

4.2.1.8 Patulin Concentration in Apple Cider

The concentration of patulin in apple cider was calculated as an empirical cumulative distribution based data reported by Harris et al. (2009), which included patulin analyses of 394 samples of apple cider produced by mills in Michigan during 2002-2004. The mean value of patulin concentration from Harris et al. (2009) is shown in Table 19. The primary data from this research was used to calculate the empirical cumulative distribution for patulin in apple cider, which is reported in Table 20. This distribution was used in FDA-iRisk as a process model for patulin in apples.

Table 19. The utilized data on patulin concentrations in apple cider in the US.

No of samples (n)	Mean (µg/L)	Source
394	36.9	Harris et al. (2009)

Table 20. Calculated cumulative empirical distribution of patulin concentrations in apple cider.

Probability	Concentration (µg/kg)
0	0
0.81212	0
0.81414	4.6
0.85	7.3
0.9	12.8
0.92	19.7
0.94	30.1
0.96	36.1
0.97	42.7
0.98	68.5
0.99	159
1	467.4

4.2.1.9 Patulin Concentration in Fresh Apples

Because of lack of data about the concentration of patulin in raw apples, we assumed that the patulin concentration in fresh apple is zero. We believe this assumption is reasonable because it is unlikely that people will consume apples having rot caused by *Penicillium expansum*, and patulin formation is restricted to the rotted area of *Penicillium expansum* infected fruit.

4.2.2 Dose-Response Models for Chemical Hazards

Dose-response models were developed for each of the health endpoints for input into the FDA-iRisk model. For inorganic arsenic exposure, the dose-response model used for each health endpoint (bladder, lung and non-melanoma skin cancers) was linear by slope factor. The slope

factors were derived by averaging the slope factors for men and women determined by Oberoi et al. (2014) for use in the model in this research. The resulting slope factors are presented in Table 21, and the dose response relationships for bladder, lung and non-melanoma skin cancers are presented in Figures 5, 6 and 7, respectively.

Table 21. Dose response slope factors for inorganic arsenic.

Model	Exposure	Response
Inorganic Arsenic - Bladder Cancer	Chronic	Linear by Slope Factor Dose unit: µg/kg-day (Slope:0.00001625; 100%)
Inorganic Arsenic - Lung Cancer	Chronic	Linear by Slope Factor Dose unit: µg/kg-day (Slope:0.00001655; 100%)
Inorganic Arsenic - Non-melanoma Skin Cancer	Chronic	Linear by Slope Factor Dose unit: µg/kg-day (Slope:0.000015; 100%)

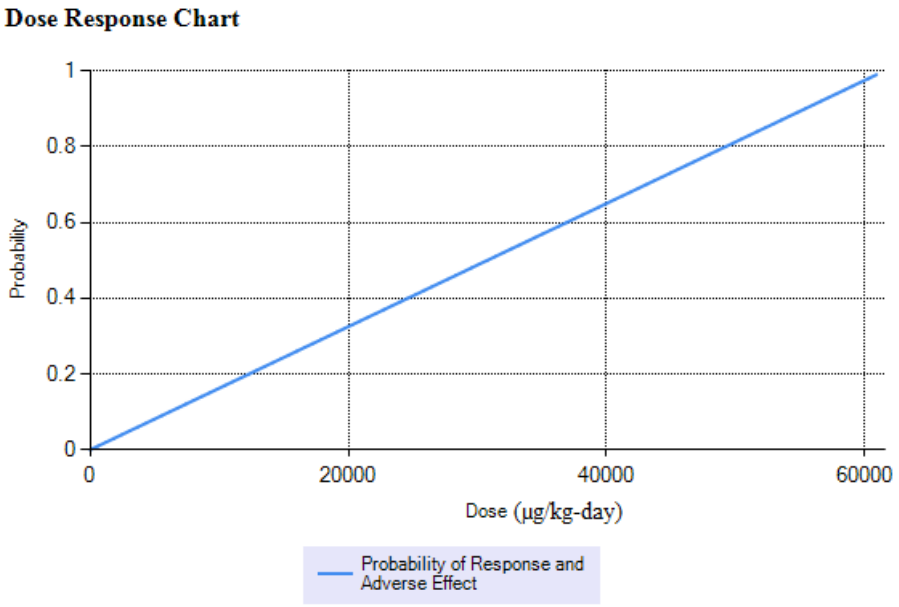

Figure 5. Dose response relationship of the effects of inorganic arsenic dose on probability of bladder cancer (µg/kg-day).

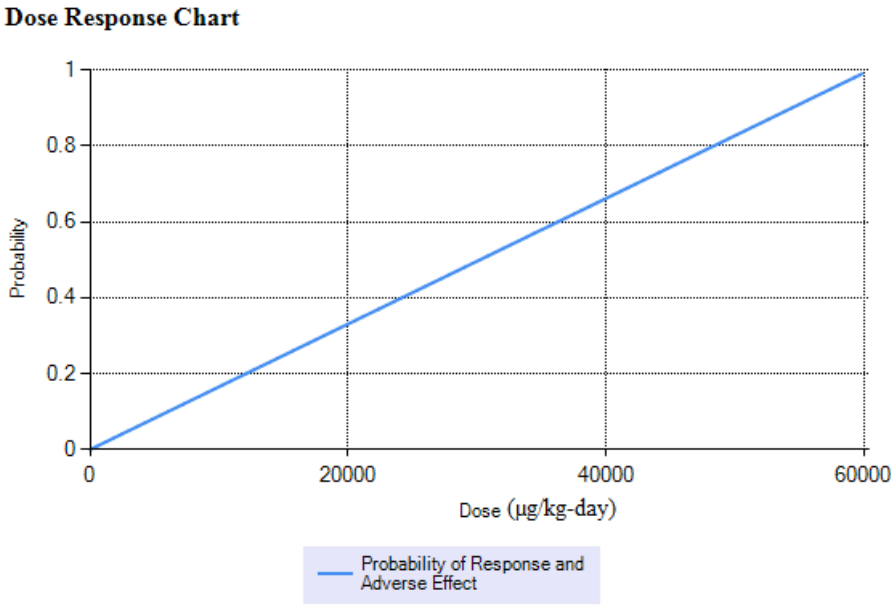

Figure 6. Dose response relationship of the effects of inorganic arsenic dose on probability of lung cancer (µg/kg-day).

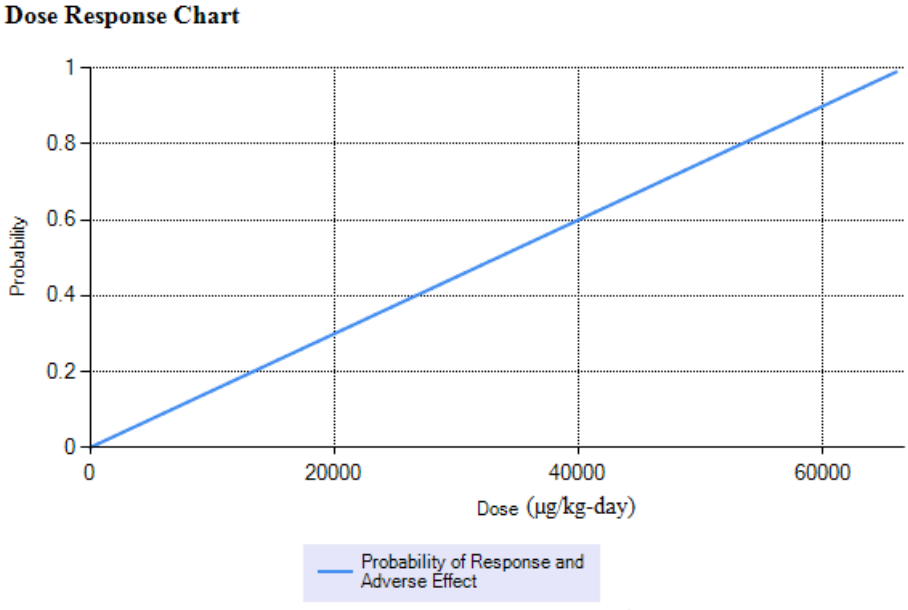

Figure 7. Dose response relationship of the effects of inorganic arsenic dose non-melanoma skin cancer (µg/kg-day).

For lead exposure, threshold linear dose response models were used for each health endpoint (hypertensive heart disease, reduced intelligence quotient). The parameters were based on evaluations by the Joint FAO/WHO Expert Committee on Food Additives (JECFA; WHO, 2011). In its most recent meta-analysis of epidemiological data, the chronic dietary exposure to lead corresponding to a decrease of 1 IQ point was estimated to be 0.6 µg/kg BW/day (WHO, 2011). Based on the averaged median reference slope estimates for blood lead levels versus systolic blood pressure from four epidemiology studies, the dietary exposure corresponding to an increase in systolic blood pressure of 1 mm Hg was estimated to be 1.3 µg/kg BW/day (WHO, 2011).

The health metrics used in this model were for DALY/case associated with hypertensive heart disease and mild mental retardation, which are health conditions that would not be experienced at low chronic exposures to dietary lead. Therefore, we used a threshold linear dose response model for each health metric, wherein the slope estimates for the health effects from WHO (2011) were multiplied by a factor of 30 to establish the minimum threshold for lead to exert health effects associated with these metrics. Therefore, the threshold values used in the model were 39 and 18 µg lead/kg BW/day. These dose response models are summarized in Table 22 and Figures 9 and 10.

Table 22. Dose response models for lead.

Model	Exposure	Response
Hypertensive heart disease for adults	Chronic	Threshold Linear Dose unit: µg/kg bw/day (Risk at Reference Point: 1, Reference Point: 100, Threshold: 39; 100%)
Reduced intelligence quotient (IQ)	Chronic	Threshold Linear Dose unit: µg/kg bw/day (Risk at Reference Point: 1 , Reference Point: 50, Threshold: 18; 100%)

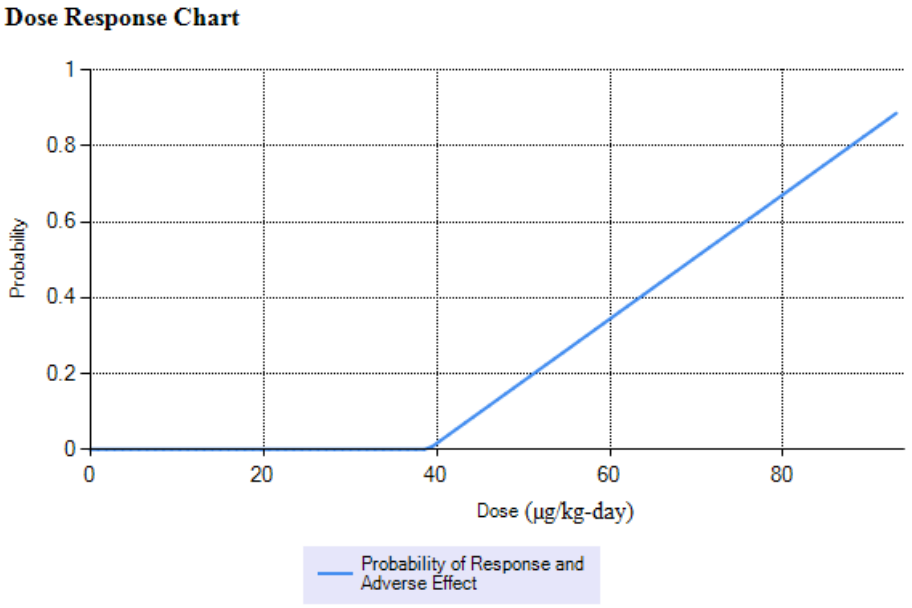

Figure 8. Dose response relationship of the effects of lead dose on probability of hypertensive heart disease (30-point increase in systolic blood pressure) in adults.

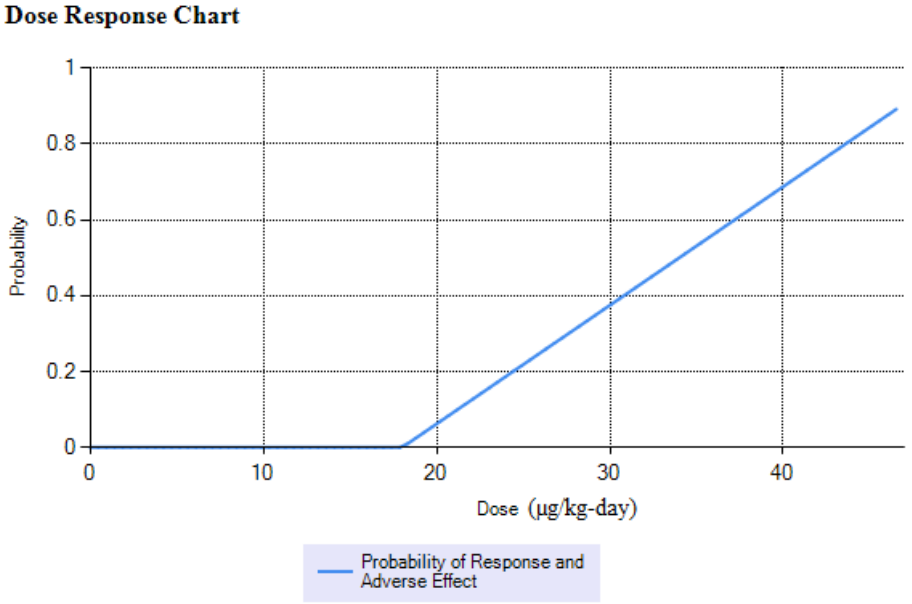

Figure 9. Dose response relationship of the effects of lead dose on probability of a 30-point reduction in intelligence quotient in children.

4.2.3 Estimating DALYs per Case for Chemical Hazards

4.2.3.1 Estimated DALY for Inorganic Arsenic

We estimated the values of DALYs associated with health endpoints for each of the three chemicals based on a review of the literature. For inorganic arsenic, DALY were estimated for bladder, lung and non-melanoma skin cancers. The DALYs for bladder and lung cancers were derived from the FDA-iRISK sample risk assessment for inorganic arsenic in apple juice as illustrated in Tables 36 and 37, respectively. The individual morbidity sequelae, the disability weight of each, and the duration of each were from the bladder cancer tables of the Australian Burden of Disease study (Mathers et al., 1999). The estimates in Table 23 use an average age of diagnosis of bladder cancer in the US of 73 (American Cancer Society, www.cancer.org). Based on this, the 5-year survival rate for bladder cancer among people 65 to 74 years of age (Mathers et al., 1999) was used to determine the fraction of cases that were assumed to have normal life expectancy. The remaining cases were assumed fatal, with a duration given by the life expectancy at age 75-76 and average survival time being 2.25 years (FDA-iRisk, 2016). Life expectancy for this example was as reported in Arias (2011). Calculated DALY/case for lung cancer (Table 24) was derived using similar methodology (FDA-iRisk, 2016; Mathers et al., 1999; Arias, 2011).

The calculated DALY per case for non-melanoma skin cancer (Table 25) was derived by dividing the total value of DALY per 100,000 persons attributed to non-melanoma skin cancer by Murray et al. (2013) and by the total number of non-melanoma skin cancer procedures per 100,000 population estimated by Rogers et al. (2015).

Table 23. Weighted DALY per case for bladder cancer.

Health End Point	Duration	Unit	Severity	DALY	Fraction of Cases	Weighted DALY
Diagnosis and primary therapy	0.12	Y	0.27	0.032400	1	0.032400
Disseminated carcinoma	0.92	Y	0.64	0.58880	0.284	0.16722
In remission	1	Y	0.18	0.18000	0.284	0.051120
Mortality (at age 74-75)	11.7	Y	1	11.700	0.284	3.3228
State after intentionally curative primary therapy	4.88	Y	0.18	0.87840	0.716	0.62893
Terminal stage	0.08	Y	0.93	0.074400	0.284	0.021130
Total DALY/case						4.22

Table 24. Weighted DALY per case for lung cancer.

Health End Point	Duration	Unit	Severity	DALY	Fraction of Cases	Weighted DALY
Diagnosis and chemotherapy small cell cancer	0.17	Y	0.68	0.11560	0.43	0.049708
Diagnosis and primary therapy for non operable non-small cell cancer	0.5	Y	0.76	0.38000	0.43	0.16340
Diagnosis and primary therapy for operable non-small cell cancer	0.5	Y	0.44	0.22000	0.14	0.030800
Disease free after primary therapy for non-small cell cancer	5	Y	0.47	2.3500	0.04	0.094000
Disseminated non-small cell cancer	0.5	Y	0.91	0.45500	0.53	0.24115
Mortality (age 71-72)	14.3	Y	1	14.300	0.96	13.728
Relapse/terminal stage small cell cancer	0.08	Y	0.93	0.074400	0.43	0.031992
Small cell cancer in remission	0.08	Y	0.54	0.043200	0.43	0.018576
Terminal stage non-small cell cancer	0.08	Y	0.93	0.074400	0.53	0.039432
Total DALY/case						14.4

Table 25. Weighted DALY per case for non-melanoma skin cancer.

Disease	**Health endpoint**	The adjusted procedure rate per 100,000 beneficiaries	Estimated DALY/case.	Source
Non-melanoma Skin Cancer	Basal cell carcinoma (BBC)	4280	*12/6558 = **0.00183**	1. Adjusted non-melanoma skin cancer procedure rate from Rogers et al. (2015). 2. DALY/100,000 population from Murray et al. (2013).
	Squamous cell carcinoma (SCC)	3278		
Total cases per 100,000	-	**6558**		

*The total value of DALY per 100,000 persons attributed to non-melanoma skin cancer by Murray et al. (2013) is 12. DALY/case for non-melanoma skin cancer was estimated by dividing 12 by the total number of non-melanoma skin cancers estimated by Rogers et al. (2015).

4.2.3.2 Estimated DALY for Lead

Consistent with the approach used in the Global Burden of Foodborne Disease research, we estimated the DALY per case for exposure to lead for two health endpoints – reduced intelligence quotient (IQ) for children and hypertensive heart disease for adults (Table 26). The DALY per case for reduced IQ was estimated to be 11.96 by dividing the total global DALY burden for reduced IQ associated with lead exposure (WHO, 2009) by the number of annual cases of lead-associated intellectual disabilities (Prüss-Ustün et al., 2011). The DALY per case for hypertensive heart disease was estimated to be 0.15324 based on DALY/100,000 population from Murray et al. (2013).

Table 26. Estimated DALY per case for lead exposure.

Health Condition	Health Endpoint	Estimated DALY/case	Source
Mild mental retardation	Reduced intelligence quotient (IQ) for children (under age 6 years.)	*11.96	1. Total cases: Prüss-Ustün et al. (2011). 2. Total DALYs: WHO (2009).
Cardiovascular Diseases	Hypertensive heart disease for adults	0.15324	DALY/100,000 population from Murray et al. (2013).

* 7,189,000/600,000 from Prüss-Ustün et al. (2011) and WHO (2009).

4.2.3.3 Estimated DALY and Dose Response for Patulin

The Bradford Hill criteria are a group of guidelines used to assess the evidence of biomedical causation (Perrio and Shakir, 2007), Based on an analysis using these criteria, we did not estimate the burden disease associated with patulin exposure because the human health effects associated with patulin exposure are not well understood at all, and have never been documented. The current patulin action level in the US is based on results of studies conducted in animal models (FDA, 2001), and these effects have never been demonstrated in humans.

4.3 Microbial Hazards Risk Assessment

Since the prevalence and concentrations of microbial pathogens associated with apple products were unknown, we did not use FDA-iRisk to model the number of illnesses or DALY associated with these pathogens. Instead, these parameters were estimated using known numbers of illnesses associated with foodborne illness outbreaks attributed to apple or apple cider

products, adjusting these known cases for under-diagnosis, and directly calculating DALY per year using disability weights from the published literature.

Table 27 summarizes illnesses associated with foodborne illness outbreaks attributed to consumption of apple cider or apple products since 1991. All outbreaks were attributed to consumption of apple cider except for the single outbreak due to *Listeria monocytogenes*, which was attributed to consumption of caramel apples. It was determined that apples were the source of the *Listeria* in the caramel apple outbreak (CDC, 2015). The two outbreaks associated with *E. coli* O111 also were attributed to products containing *Cryptosporidium parvum*, so these two pathogens are considered together for those outbreaks.

The total numbers of observed foodborne illness cases listed in Table 27 were adjusted for under-diagnosis using multipliers from Scallan et al. (2011a), and then were divided by 25 (years covered by the literature review to identify foodborne illness outbreaks) to obtain an adjusted number of cases attributable to each pathogen per year. These values are reported in Table 28.

Table 27. Reported outbreaks associated with microbial hazards attributed to the consumption of unpasteurized apple cider or apple products in US (1991-2015).

Pathogen	Number of outbreaks	Number of cases	Number of Deaths	Number of Hospitalizations
E. coli O157:H7	10	164	1	13
E.coli O111 + *Cryptospori-dium parvum*	2	226	0	14
Cryptospori-dium parvum	8	402	0	31
Salmonella spp.	3	41	0	7
Listeria monocytogenes	1	35	7	34
Campylobacter jejuni	1	6	0	1
Total	25	874	8	100

The source of data is CDC (2016) http://www.cdc.gov/chronicdisease/overview/

Table 28. Estimated illnesses caused by microbial pathogens associated with the consumption of unpasteurized apple cider and apple products in the US (1991-2015).

Pathogen	Apple product	Number of Cases	[a] Multiplier for under-diagnosis	Estimated Cases	[b] Duration of time frame (years)	[c] Adjusted cases per year
E. coli O157:H7	Cider	164	26.1	4,280	25	171
E. coli O111 + *Cryptosporidium parvum*	Cider	226	102.7	23,210	25	928
Cryptosporidium parvum	Cider	402	98.6	39,638	25	1,586
Salmonella (non-typhoidal)	Cider	41	29.3	1,200	25	48
Campylobacter	Cider	6	30.3	183	25	7.3
Listeria monocytogenes	Apple (caramel apples)	35	2.1	73.5	25	2.9
Total		874				

[a] The multiplier is a factor that accounts for under-diagnosis of foodborne illness cases from Scallan et al. (2011a). The multipliers for under-diagnosis of *E. coli* O111 and *Cryptosporidium parvum* were averaged for illnesses associated with the two pathogens simultaneously.
[b] The period of this study (1991-2015).
[c] Adjusted cases per year = (number of cases * multiplier)/25.

We estimated DALYs per year for the six pathogens associated with unpasteurized apple cider and fresh apples by using QALY (Quality Adjusted Life Years, which is equivalent to DALY in this case) loss per 1,000 cases for these microbial pathogens from Table 3 of Batz et al. (2014). These values were divided by 1,000 to estimate DALY/case, and then multiplied by the

adjusted cases per year (Table 29) for each pathogen to obtain estimates of total DALY per year associated with each pathogen.

Table 29. Estimated DALY per year attributable to microbial hazards associated with the consumption of unpasteurized apple cider and apple products in the US (1990-2015).

Pathogen	Adjusted cases per year	QALY per 1,000 cases	[a] Estimated DALY/year
E. coli O157:H7	171	26.3	4.50
E. coli O111+ *Cryptosporidium parvum*	928	[b] 2.4	2.23
Cryptosporidium parvum	1,586	3.5	5.55
Salmonella (nontyphoidal)	48	16.3	0.78
Campylobacter spp	7.3	15.7	0.12
Listeria monocytogenes	2.9	5892.4	17.32
Total			30.50

[a] Calculated by multiplying the adjusted cases per year by QALY/1000 cases derived from Table 3 of Batz et al. (2014).
[b] Calculated from the average of the two values for these pathogens from Table 3 of Batz et al. (2014) [(1.3+3.5)/2=2.4].

4.3.1 Calculation of Eating Occasions per Year for the Acute Consumption Model

We calculated eating occasions (EO) per year using data estimates for mean consumption of apple juice and the number of persons in each age group. These estimates assumed that a serving of apple juice is 4 ounces, or 113.4 grams. Using this approach, the total number of eating occasions for both age groups for apple cider were estimated to be 806,748,159 per year where

population group EO/year consumption for 0 to 6 years of age for apple cider was 58,149,629 and for 7+ years of age for apple cider was 748,598,530.

5. RESULTS

Foodborne illness outbreaks associated with six microbial pathogens were attributed to the consumption of unpasteurized apple cider or apple products in the US during the period of time covered by this risk assessment (1991 - 2015). Five pathogens – *Cryptosporidium parvum*, *E. coli* O157:H7, *E. coli* O111, *Salmonella* (non-typhoidal) and *Campylobacter* were associated with consumption of unpasteurized apple cider (Table 30), while one outbreak associated with *Listeria monocytogenes* (Table 31) was recently attributed to consumption of caramel apples. The apples were conclusively associated with the caramel apple *Listeriosis* outbreak (CDC, 2015).

Collectively, 839 cases of foodborne illness associated with these five pathogens in apple cider were reported during the 25-year period of 1991 to 2015 (Table 30). Adjusting these reported illnesses by factors for under-diagnosis from Scallan et al. (2011a), we estimated the total number of cases associated with these outbreaks at 68,584 illnesses. Three pathogens – *Cryptosporidium parvum*, *E. coli* O111, and *E. coli* O157:H7 accounted for 94% of all reported illnesses in apple cider-associated outbreaks, while illnesses associated with *Salmonella* spp. and *Campylobacter* accounted for fewer than 6% of these illnesses.

Table 30. Ranking of microbial pathogens associated with the consumption of unpasteurized apple cider based on reported and estimated cases in the US (1991-2015).

Microbial Pathogens	Apple Product	Total Reported Cases (1991-2015)	Estimated Cases (1991-2015)
Cryptosporidium parvum	Cider	402	39,638
E. coli O111 + *Cryptosporidium parvum*	Cider	226	23,210
E. coli O157:H7	Cider	164	4,280
Salmonella (nontyphoidal)	Cider	41	1,200
Campylobacter	Cider	6	182
Total		839	68,584

Table 31. Total reported illnesses and estimated cases of foodborne illness associated with *Listeria monocytogenes* in apple products (caramel apples) in the US (1991-2015).

Microbial Pathogen	Apple product	Total Reported Cases (1991-2015)	Estimated Cases (1991-2015)
Listeria monocytogenes	Caramel apples	35	73.5

T

able 32 shows ranking of the health risk associated with microbial pathogens in unpasteurized apple cider by adjusted cases per year, mean risk of illness, estimated DALYs per eating occasion (EO), and total estimated DALYs per year. Respectively, *Cryptosporidium parvum, E. coli* O157:H7, *E. coli* O111 + *Cryptosporidium parvum*, *Salmonella* spp. and *Campylobacter* ranked first through fifth, respectively, for DALYs per year and DALYs per eating occasion. For adjusted cases per year and mean risk of illness, the rankings were similar except that *E. coli O111 + Cryptosporidium parvum* was ranked second and *E. coli* O157:H7 was ranked third.

Based on DALY per year, *Cryptosporidium parvum, E. coli* O157:H7 and *E. coli* O111 + *Cryptosporidium parvum* accounted for 93% of total DALY with values of 5.55, 4.50 and 2.23, respectively. The other two pathogens reflect only 7% of the health burden associated with foodborne illness outbreaks attributed to apple cider.

Table 32. Ranking the health risk associated with microbial pathogens in unpasteurized apple cider by adjusted cases per year, mean risk of illness, estimated DALYs per eating occasion (EO), and total estimated DALYs per year.

Microbial Pathogen	[a] Adjusted Cases Per Year (rank)	[b] Mean Risk of Illness ($*10^{-9}$) (rank)	[c] Estimated DALYs Per Eating Occasion ($*10^{-9}$) (rank)	Estimated DALYs Per Year
Cryptosporidium parvum	1,586 (1)	1,965 (1)	6.88 (1)	5.55
E. coli O157:H7	171 (3)	212 (3)	5.58 (2)	4.50
E. coli O111 + *Cryptosporidium parvum*	928 (2)	1,151 (2)	2.76 (3)	2.23
Salmonella (nontyphoidal)	48 (4)	59.5 (4)	0.97 (4)	0.78
Campylobacter	7.3 (5)	9.05 (5)	0.14 (5)	0.12
Total	2,740	-	16.3	13.18

[a] The first number shown is the value for each parameter and the second number in parenthesis is the rank within each column.
[b] Mean Risk of Illness = Adjusted Cases Per Year divided by the total number of eating occasions for both age groups for apple cider (eating occasions for unpasteurized apple cider estimated to be 806,748,159 per year).
[c] DALYs Per Eating Occasion = DALYs per year divided by the total number of eating occasions for both age groups for apple cider.

The ranking summary of the risk of inorganic arsenic hazards by total estimated annual illnesses, mean risk of illness, DALYs Per Consumer and total DALYs per year is presented in Table 33. No illnesses were predicted by the FDA-iRisk model for lead in any commodity. All inorganic arsenic scenarios:food pairs had the same risk ranking for each of the four measures presented in Table 33.

The rankings in Table 33 indicate that inorganic arsenic in apple juice is ranked first based on its adverse health for the measure of illness and DALYs. The DALY value per year of inorganic arsenic in apple juice was 4.84 and this value reflected 97% of the total DALY

attributable to all apple products that were assessed. The total estimated annual illness (0.753 cases) associated with inorganic arsenic in apple juice reflects 97% of the total cases for the total health burden predicted by the model.

Table 33. Ranking summary of the risk of inorganic arsenic hazards by total estimated annual illnesses, mean risk of illness, DALYs Per Consumer and total DALYs per year.

Hazard:Food Pair [a]	Total Estimated Annual Illnesses	[b] Mean Risk of Illness ($*10^{-9}$)	[c] DALYs Per Consumer ($*10^{-9}$)	Total DALYs Per Year
Inorganic Arsenic in Apple Juice	0.753	2.34	15.0	4.84
Inorganic Arsenic in Fresh Apples	0.0213	0.0663	0.426	0.137
Inorganic Arsenic in Apple Cider	0.00156	0.00484	0.0311	0.00999
Total	0.776	-	15.46	4.987

[a] No illnesses were predicted by the FDA-iRisk model for lead in any commodity or for patulin in fresh apples or apple cider.
[b] Mean Risk of Illness = Total Estimated Annual Illnesses /Annual Consumers (assumed to be equal to US population = 321,418,820).
[c] DALYs Per Consumer = Total DALYs Per Year /Annual Consumers.

Tables 34, 35 and 36 present the ranking of health risk for hazard:food pairs associated with the consumption of apple products by DALYs per year, DALYs per person (for chemical hazards) or eating occasion (for microbiological hazards), or mean risk of illness, respectively. The rankings based on DALYs per year (Table 34) and DALYs per person or eating occasion (Table 35) were similar, except that *Cryptosporidium parvum* in unpasteurized apple cider and inorganic arsenic in apple juice were ranked first and second for DALYs per year, and this order was inverted for DALYs per consumer or EO. However, the rankings were dramatically changed when hazard:food pairs were sorted by mean risk of illness (Table 36). For example, inorganic arsenic in apple juice ranked sixth in mean risk of illness, whereas *Cryptosporidium*

parvum and *E. coli* O111 + *Cryptosporidium parvum* in unpasteurized apple cider were ranked first and second, respectively, using this parameter. Furthermore, inorganic arsenic in apple juice ranked second in DALYs per year and first in DALYs per consumer, but ranked only sixth in mean risk of illness.

Table 34. Ranking the health risk of hazard food pairs associated with the consumption of apple products by DALYs per year.

Hazard: Food Pair	Estimated DALYs Per Year	Ranking
Cryptosporidium parvum in unpasteurized apple cider	5.55	1
Inorganic arsenic in apple Juice	4.84	2
E. coli O157:H7 in unpasteurized apple cider	4.50	3
E. coli O111 + *Cryptosporidium parvum*	2.23	4
Salmonella in unpasteurized apple cider	0.78	5
Inorganic arsenic in fresh apples	0.137	6
Campylobacter in unpasteurized apple cider	0.115	7
Inorganic arsenic in apple cider	0.00999	8
Total	18.16	

Table 35. Ranking the health risk of hazard food pairs associated with the consumption of apple products by DALYs per consumer or eating occasion.

Hazard:Food Pair	DALYs Per Consumer or Eating Occasion ($*10^{-9}$)	Ranking
Inorganic arsenic in apple juice	15.0	1
Cryptosporidium parvum in unpasteurized apple cider	6.88	2
E. coli O157:H7 in unpasteurized apple cider	5.58	3
E. coli O111 + *Cryptosporidium parvum*	2.76	4
Salmonella in unpasteurized apple cider	0.969	5
Inorganic arsenic in fresh apples	0.426	6
Campylobacter in unpasteurized apple cider	0.142	7
Inorganic arsenic in apple cider	0.0311	8

Table 36. Ranking the health risk of hazard food pairs associated with the consumption of apple products by mean risk of illness.

Hazard: Food Pair	Mean Risk of Illness ($*10^{-9}$)	Ranking
Cryptosporidium parvum in unpasteurized apple cider	1,965	1
E. coli O111 + *Cryptosporidium parvum* in unpasteurized apple cider.	1,151	2
E. coli O157:H7 in unpasteurized apple cider	212	3
Salmonella (nontyphoidal) in unpasteurized apple cider	59.5	4
Campylobacter in unpasteurized apple cider	9.05	5
Inorganic Arsenic in Apple Juice	2.34	6
Inorganic Arsenic in Fresh Apples	0.0663	7
Inorganic Arsenic in Apple Cider	0.00484	8

A summary analysis ranking of hazard:food combinations (Table 37) demonstrated that apple cider consumption was associated with the greatest total DALY per year, with 13.18

DALY per year reflecting approximately 73% of the total DALYs per year predicted for consumption of all foods. Apple juice consumption was associated with most of the remaining DALYs per year, with fresh apples contributing little to the total. *Cryptosporidium parvum*, *E. coli* O157:H7 and *E. coli* O111+ *Cryptosporidium parvum* were the most important potential hazards associated with apple cider while the health concern with apple juice was primarily associated with exposure to inorganic arsenic hazard.

Table 37. Ranking the health risk of apple products based on all associated hazards.

Food	Associated Hazards	DALYs per Year for each hazard:food pair	DALYs per Year for each food	Ranking
Apple Cider	*Cryptosporidium parvum*	5.55	13.18	1
	E. coli O157:H7	4.50		
	E. coli O111+ *Cryptosporidium parvum*	2.23		
	Salmonella (nontyphoidal)	0.782		
	Campylobacter	0.115		
	Inorganic Arsenic	0.00999		
Apple Juice	Inorganic Arsenic	4.84	4.84	2
Fresh Apples	Inorganic Arsenic	0.137	0.137	3
Total			18.157	

6. DISCUSSION

In risk management, protecting human health from the adverse effects of different food safety hazards is a public policy goal. Using risk assessment and risk ranking approaches enables policy makers to prioritize specific approaches to manage the risk associated with exposure to food hazards. In this study we used one main approach (disability-adjusted life years) and another minor approach (mean risk of illness) to rank the health burden from the selected microbial and chemical hazards. Disability-adjusted life year (DALY) estimates include both mortality and morbidity associated with illnesses, and are particularly suited for comparing the different health effects (e.g. gastrointestinal infections, cancer, etc.) caused by different hazard exposures.

This risk assessment demonstrated that six foodborne hazards (five pathogens and one chemical) in contaminated apple products are predicted to result in around 2,741 illnesses each year (Tables 32 and 33) and approximately 18.2 DALYs per year (Table 37) from the consumption of apple products. There was no difference in ranking of the chemical and microbial hazards when DALY-based measures were used to rank individual hazard:food pairs (not combined ranking) by different scenarios of the risk ranking (DALY per year or DALY per consumer or eating occasion). In contrast, ranking hazard:food pairs by mean risk of illness (Table 36) changed the ranking of hazards compared to DALY-based parameters. This difference in ranking was driven by the relatively high risk of illness caused by pathogenic microorganisms (*Cryptosporidium parvum*, *E. coli* O111, and *E. coli* O157:H7) associated with consuming unpasteurized apple cider, but relatively low DALY per case associated with these illnesses.

For the microbial hazards, illnesses were reported for six pathogens that were found to be associated with the consumption of apple products. Five of these pathogens have been associated with consuming unpasteurized apple cider. Recently, *Listeria monocytogenes* was associated with consuming fresh apples in the form of caramel apples. Although the trace back investigation in the 2014 caramel apple listeriosis outbreak clearly implicated the apples as the *Listeria* source (CDC, 2015), we did not include this listeriosis outbreak in the summary tables for the risk assessment. This is due to the fact that none of the illnesses in the caramel apple outbreak were clearly associated with consuming only the apples. Further research by Glass et al. (2015) suggests that this listeriosis outbreak was facilitated by proliferation of *Listeria* bacteria in the microenvironment created when caramel apples are produced (i.e. puncturing the apple flesh with a stick and then coating the apple with caramel). Normally, pathogen infiltration into apple flesh is prevented by the external barriers of the fruit (Abdul-Raouf et al., 1993). Had we included the DALYs per year estimated for *Listeria monocytogenes* in the summary analysis (17 DALY per year), this would have reflected more than 55% of the total DALY for the five pathogens associated with apple cider consumption.

More interesting is that no reported cases of foodborne illness caused by pathogenic microorganisms have been attributed to pasteurized apple juice during the time frame of this research (1991-2015). This observation stresses the importance of applying appropriate process controls to destroy pathogenic microorganisms that can be present in raw juices and the application of HACCP-based food safety management systems in food industry (Panisello et al., 2000).

We predict that additional foodborne illness outbreaks will be associated with unpasteurized apple cider in the future, as the lack of a pathogen destruction step in fresh cider manufacturing

places very high demands on fruit quality and cleanliness and effective sanitation programs to minimize the likelihood of foodborne illness outbreaks associated with this product. As noted in the results, most (>90%) of the illnesses associated with consumption of unpasteurized apple cider in the US have been due to infections with *Cryptosporidium parvum*, *E. coli* O157:H7 and *E. coli* O111. It is notable that few deaths have been reported in association with the outbreaks caused by the shiga toxin-producing strains of *E. coli*, which is somewhat in contrast with outbreaks associated with these pathogens in other food vehicles.

It is instructive to compare the DALYs per year metrics for microbial pathogens in apple cider with that reported for other pathogen:food pairs in the United States. Morris et al. (2011) reported risk ranking of pathogen:food pairs in the US based on annual quality adjusted life year (QALY) loss (similar to DALY in this research) and found that *Campylobacter* in poultry products caused the greatest QALY loss at 9,541 per year. Combining the DALYs per year for all microbial pathogens in unpasteurized apple cider in this study results in a total of 13.18, which would not rank in the top 50 hazard: food pairs in the United States according to Morris et al. (2011), where norovirus in dairy products was ranked 50th in QALY loss at 109 per year.

The DALY per year associated with pathogens in apple cider also is relatively small compared to the total QALY loss per year attributable to these pathogens (Morris et al., 2011; Batz et al., 2014). For example, the 5.55 DALY per year associated with *Cryptosporidium parvum* in apple cider would account for 2.8% of QALY loss per year attributed to this pathogen from all sources, while the 4.50 DALY per year associated with *E. coli* O157:H7 in apple cider would account for only 0.27% of total QALY loss per year for this pathogen (Morris et al., 2011; Batz et al., 2014). However, these relatively low DALY per year estimates for pathogens in apple cider do not mean that our results are unimportant, as susceptible populations such as

children are particularly susceptible to infection with these pathogens. Given the relatively low consumption of apple cider relative to shelf-stable apple juice in the US and the fact that all foodborne illness outbreaks with apple juice products have been associated with cider consumption, it would be interesting to compare the risks associated with consumption of unpasteurized cider with that of other untreated foods that are common vehicles for foodborne illness outbreaks (e.g. raw milk, seed sprouts, raw seafood or shellfish products).

While the primary concerns with unpasteurized apple cider are microbial pathogens, health effects associated with chemical hazards appear to be more important with apple juice consumption (Table 37). Among the chemical hazards assessed in this research, inorganic arsenic was associated with the greatest DALY per year for apple juice consumption. This is can be explained as the result of the high consumption of apple juice, particularly by children, and the relatively widespread occurrence (albeit at low levels) of inorganic arsenic in apple juice products. Inorganic arsenic can contaminate apple juice by two primary sources – naturally occurring arsenic in soils and anthropogenic arsenic added by human activities such as use of lead arsenate pesticides (Ratnaike, 2003). Inorganic arsenic in apple juice could arise from these sources that contribute to arsenic in the fruit itself (if the juice is manufactured directly from fruit), or could be contributed by imported apple juice concentrate during apple juice manufacturing (Hooper and Shi, 2012). Apple juice concentrate is the main ingredient used in producing apple juice in the US. Another potential source of inorganic arsenic in apple juice comes from the water used to reconstitute apple juice concentrate during apple juice production. This concern would depend on arsenic concentrations in local groundwater, as aquifers very considerably in inorganic arsenic concentrations.

There are several assumptions and limitations associated with this risk assessment and risk ranking exercise for hazards in apples and apple juice products. These factors add to the uncertainty of our estimates in this study. First, due to a lack of data for several factors, assumptions had to be made with regarding to selection of disability weights used to calculate DALY for chemical hazards and for the concentrations and distributions of certain hazards in the food products (e.g. lead). In the case of DALY for microbial hazards, we elected to use the average QALY loss estimated for pathogens by Batz et al. (2014).

No data were available on consumption of unpasteurized apple cider. Therefore, we assumed that 5% of total apple juice consumption by persons aged 7 and older was apple cider, and 1% of apple juice consumed by infants and children up to age 6 was apple cider. This assumption resulted in our calculating there are 806,748,159 eating occasions for apple cider per year based on a four-ounce serving size. This translates to 25.2 million gallons of unpasteurized apple cider consumed per year. Assuming the average cider mill manufactures 7,850 gallons per year – the average annual production reported by Bobe et al. (2007) – this level of production could be achieved by 3,200 facilities producing apple cider. Given that there are currently approximately 130 licensed cider mills in the state of Michigan (Michigan Apple Committee, personal communication), our assumption regarding the volume of unpasteurized apple cider consumption may be an overestimate.

Finally, because there are no published data on the prevalence or concentrations of microbial pathogens in unpasteurized apple cider, we were not able to model the exposure assessment in FDA-iRisk. As an alternative, we used foodborne illnesses associated with outbreaks attributed to unpasteurized apple cider to predict the numbers of illnesses associated with this product each year. These predictions took into account a scaling factor for under-diagnosis reported by

Scallan et al. (2011a). However, since it is likely that several outbreaks and sporadic cases of foodborne illness associated with any food product are not detected or reported, our estimates of foodborne illnesses per year associated with apple cider are likely to be underestimated compared to the actual numbers.

The results of this work provide valuable information that can be used by policy makers, other scientists for additional research, the food industry, and consumers in order to minimize the potential public health adverse effects of these hazards. Based on this work, four important recommendations are suggested:

1. Applying DALY as a legal standard (maximum level) for health burden associated with specific foods or hazard:food pairs should be explored as a tool to guide food safety policy decisions by competent authorities.
2. The US FDA should establish a maximum level of lead in food, as current FDA guidance on allowable lead levels is ambiguous.
3. Improved statistics regarding consumption of unpasteurized apple cider and other juices are needed.
4. We recommend expanded use of DALYs to rank the risk of chemicals such as inorganic arsenic and lead in the most commonly consumed foods in the United States.
5. More research is needed to more clearly establish what health effects can be definitively associated with patulin consumption by humans.

APPENDIX

Table 38. Raw data from the WWEIA-FCID database of average daily consumption (g/kg BW/day) of fresh apples by infants and children aged 0-6 years (Foodrisk, 2016).

Decimal fraction of consumers	Commodity Eaters Only	Total Filtered Population
	N=1,410	N=9,106
	Mean=6.67	Mean=1.05
5%	1.32	0
10%	2.03	0
15%	2.56	0
20%	3.2	0
25%	3.54	0
30%	4.08	0
35%	4.54	0
40%	4.95	0
45%	5.56	0
50%	6.08	0
55%	6.59	0
60%	7.08	0
65%	7.54	0
70%	8.15	0
75%	8.92	0
80%	9.33	0
85%	10.28	1.31
90%	11.82	4.64
95%	14.87	7.85
100%	38.95	38.95

Source: What We Eat In America - Food Commodity Intake Database 2005-2010 U.S. Environmental Protection Agency - Office of Pesticide Programs University of Maryland 2012 – 2016.

Table 39. Empirical distribution of daily consumption (g/kg BW/day) of apples by infants and children aged 0-6 years in the US.

Decimal Fraction of Population (Probability)	Consumption (g/kg BW/d)
0	0
0.845157039	0
0.852899187	1.32
0.860641335	2.03
0.868383483	2.56
0.876125631	3.2
0.883867779	3.54
0.891609928	4.08
0.899352076	4.54
0.907094224	4.95
0.914836372	5.56
0.92257852	6.08
0.930320668	6.59
0.938062816	7.08
0.945804964	7.54
0.953547112	8.15
0.96128926	8.92
0.969031408	9.33
0.976773556	10.28
0.984515704	11.82
0.992257852	14.87
1	38.95

Table 40. Raw data from the WWEIA-FCID database of average daily consumption (g/kg BW/day) of fresh apples by persons aged 7 years and older (Foodrisk, 2016).

	Commodity Eaters Only	Total Filtered Population
N	5,046	40,237
Mean	2.34	0.28
5%	0.28	0
10%	0.67	0
15%	1.01	0
20%	1.23	0
25%	1.42	0
30%	1.6	0
35%	1.75	0
40%	1.85	0
45%	2	0
50%	2.13	0
55%	2.26	0
60%	2.39	0
65%	2.54	0
70%	2.7	0
75%	2.88	0
80%	3.12	0
85%	3.5	0
90%	3.93	1.08
95%	5.17	2.35
100%	23.56	23.56

Source: What We Eat In America - Food Commodity Intake Database 2005-2010, US. Environmental Protection Agency - Office of Pesticide Programs University of Maryland 2012 – 2016.

Table 41. Empirical distribution of daily consumption (g/kg BW/day) of apples by persons aged 7 years and older in the US.

Decimal Fraction of Population	Average Daily Consumption (g/kg BW/day)
0	0
0.874593036	0
0.880863384	0.28
0.887133733	0.67
0.893404081	1.01
0.899674429	1.23
0.905944777	1.42
0.912215125	1.6
0.918485474	1.75
0.924755822	1.85
0.93102617	2
0.937296518	2.13
0.943566866	2.26
0.949837215	2.39
0.956107563	2.54
0.962377911	2.7
0.968648259	2.88
0.974918607	3.12
0.981188955	3.5
0.987459304	3.93
0.993729652	5.17
1	23.56

Table 42. Raw data from the WWEIA-FCID database of average daily consumption (g/kg BW/day) of apple juice by infants and children aged 0-6 years (Foodrisk, 2016).

	Commodity Eaters Only	Total Filtered Population
N	4,749	9,106
Mean	8.31	4.46
5%	0.03	0
10%	0.04	0
15%	0.06	0
20%	0.08	0
25%	0.13	0
30%	0.2	0
35%	0.32	0
40%	1.15	0
45%	3.09	0
50%	4.42	0.03
55%	5.9	0.06
60%	7.26	0.13
65%	8.59	0.32
70%	10.24	2.75
75%	11.92	5.46
80%	13.97	7.92
85%	17.02	10.88
90%	21.9	14.69
95%	30	22.34
100%	175.62	175.62

Table 43. Empirical distribution of daily consumption (g/kg BW/day) of apple juice by infants and children aged 0-6 years in the US.

Decimal Fraction of Population	Average Daily Consumption (g/kg BW/day)
0	0
0.47847573	0
0.504551944	0.0297
0.530628157	0.0396
0.556704371	0.0594
0.582780584	0.0792
0.608856798	0.1287
0.634933011	0.198
0.661009225	0.3168
0.687085438	1.1385
0.713161652	3.0591
0.739237865	4.3758
0.765314079	5.841
0.791390292	7.1874
0.817466506	8.5041
0.843542719	10.1376
0.869618933	11.8008
0.895695146	13.8303
0.92177136	16.8498
0.947847573	21.681
0.973923787	29.7
1	173.8638

Table 44. Empirical distribution of daily consumption (g/kg BW/day) of apple cider by infants and children aged 0-6 years in the US.

Decimal Fraction of Population	Average Daily Consumption (g/kg BW/day)
0	0
0.47847573	0
0.504551944	0.0003
0.530628157	0.0004
0.556704371	0.0006
0.582780584	0.0008
0.608856798	0.0013
0.634933011	0.002
0.661009225	0.0032
0.687085438	0.0115
0.713161652	0.0309
0.739237865	0.0442
0.765314079	0.059
0.791390292	0.0726
0.817466506	0.0859
0.843542719	0.1024
0.869618933	0.1192
0.895695146	0.1397
0.92177136	0.1702
0.947847573	0.219
0.973923787	0.3
1	1.7562

Table 45. Raw data from the WWEIA-FCID database of average daily consumption (g/kg BW/day) of apple juice by persons aged 7 years and older (Foodrisk, 2016).

	Commodity Eaters Only	Total Filtered Population
N	9,270	40,237
Mean	1.33	0.26
5%	0.01	0
10%	0.01	0
15%	0.01	0
20%	0.02	0
25%	0.02	0
30%	0.03	0
35%	0.03	0
40%	0.04	0
45%	0.05	0
50%	0.07	0
55%	0.1	0
60%	0.15	0
65%	0.3	0
70%	0.75	0
75%	1.44	0
80%	2.2	0
85%	3.06	0.02
90%	4.4	0.06
95%	6.6	1.33
100%	70.79	70.79

Table 46. Empirical distribution of daily consumption (g/kg BW/day) of apple juice by persons aged 7 years and older in the US.

Decimal Fraction of Population	Average Daily Consumption (g/kg BW/day)
0	0
0.769615031	0
0.781134279	0.0095
0.792653528	0.0095
0.804172776	0.0095
0.815692025	0.019
0.827211273	0.019
0.838730522	0.0285
0.85024977	0.0285
0.861769019	0.038
0.873288267	0.0475
0.884807515	0.0665
0.896326764	0.095
0.907846012	0.1425
0.919365261	0.285
0.930884509	0.7125
0.942403758	1.368
0.953923006	2.09
0.965442255	2.907
0.976961503	4.18
0.988480752	6.27
1	67.2505

Table 47. Empirical distribution of daily consumption (g/kg BW/day) of apple cider by persons aged 7 years and older in the US.

Decimal Fraction of Population	Average Daily Consumption (g/kg BW/day)
0	0
0.769615031	0
0.781134279	0.0005
0.792653528	0.0005
0.804172776	0.0005
0.815692025	0.001
0.827211273	0.001
0.838730522	0.0015
0.85024977	0.0015
0.861769019	0.002
0.873288267	0.0025
0.884807515	0.0035
0.896326764	0.005
0.907846012	0.0075
0.919365261	0.015
0.930884509	0.0375
0.942403758	0.072
0.953923006	0.11
0.965442255	0.153
0.976961503	0.22
0.988480752	0.33
1	3.5395

Table 48. FDA surveillance data for total and inorganic arsenic in apple juice (FDA, 2011).

	Total Arsenic Analysis	Arsenic Speciation Analysis		
Sample ID	Total As Concentration (µg/kg, ppb)	Inorganic As Concentration (AsIII + AsV) (µg/kg, ppb)	DMA Concentration (µg/kg, ppb)	MMA Concentration (µg/kg, ppb)
561799	5.6	5.2	TR	0
561800	36	8.3	TR	19
592030	7.5	5.4	TR	0
606077	4.1	TR	TR	0
606078	6.6	3.9	0	0
615659	1.3	TR	0	0
629367	6.9	5.0	TR	0
629368	10	8.1	TR	0
645508	30	8.4	TR	20
645509	5.5	TR	0	TR
645510	1.4	TR	0	0
657385	TR	0	TR	0
658160	2.6	TR	0	0
658161	5.1	TR	0	0
658162	7.2	4.0	TR	TR
659338	6.8	4.8	0	0
661782	6.3	5.5	0	0
663975	5.9	4.9	TR	0
665626	3.5	TR	TR	0
665627	3.5	TR	TR	0
665628	TR	TR	0	0
669327	9.6	9.8	TR	0
669328	6.5	4.5	0	0
669329	TR	0	0	0
669330	7.1	6.9	TR	0
669331	4.8	4.3	TR	0
669332	6.1	5.8	0	0
669333	7.9	7.8	TR	0
669997	5.4	4.6	TR	0
669998	5.3	3.9	0	0
669999	TR	TR	0	0
670000	3.2	2.8	TR	0
670001	5.4	4.1	TR	0
671312	7.5	5.2	TR	0
671313	5.0	4.7	TR	0

Table 48 (cont'd)

	Total Arsenic Analysis	Arsenic Speciation Analysis		
Sample ID	Total As Concentration (µg/kg, ppb)	Inorganic As Concentration (AsIII + AsV) (µg/kg, ppb)	DMA Concentration (µg/kg, ppb)	MMA Concentration (µg/kg, ppb)
671314	TR	0	TR	0
671560	5.8	5.3	0	0
671561	1.4	TR	TR	0
688172	8.4	6.9	TR	0
688173	6.2	4.9	TR	0
688921	5.5	4.2	TR	0
689740	6.7	5.8	TR	0
689741	5.8	3.9	0	0
689742	4.3	TR	TR	0
693283	9.2	7.2	TR	0
693284	9.9	7.9	TR	0
694020	TR	TR	0	0
694021	5.4	5.1	TR	0
695975	5.4	4.7	TR	0
695976	4.6	TR	TR	0
695977	4.9	3.9	TR	0
695978	TR	TR	TR	0
695979	7.1	6.5	TR	0
695980	7.5	6.6	TR	0
697132	6.8	5.1	TR	0
697133	6.5	4.9	0	0
697134	6.7	5.1	0	0
701815	3.5	TR	TR	0
701816	8	TR	TR	TR
702418	1.9	TR	TR	0
702419	6.1	5.4	0	0
702420	6.0	5.3	0	0
702586	6.3	5.4	TR	0
702587	8.5	7.5	TR	0
708245	2.9	TR	0	0
708246	7.8	5.4	0	0
708247	7.0	5.1	0	0
714078	2.5	TR	0	0
714317	6.1	5.3	0	0
714318	2	7.0	TR	0

Table 48 (cont'd)

	Total Arsenic Analysis	Arsenic Speciation Analysis		
Sample ID	Total As Concentration (µg/kg, ppb)	Inorganic As Concentration (AsIII + AsV) (µg/kg, ppb)	DMA Concentration (µg/kg, ppb)	MMA Concentration (µg/kg, ppb)
714319	11	8.2	TR	0
714721	8.9	8.4	TR	0
714722	4.6	TR	TR	0
715042	5.4	TR	TR	0
715043	5.6	3.8	TR	0
716089	8.4	6.8	TR	0
717104	6.7	5.1	0	0
717105	9.0	6.5	TR	0
718082	9.4	8.2	TR	0
718083	7.7	7.2	TR	0
718084	4.2	3.5	0	0
718085	2.8	3.0	0	0
718086	3.6	3.1	0	0
720876	2.6	TR	TR	TR
720877	11	5.6	TR	4.4
722786	7.1	5.0	0	0
722787	9.6	7.5	TR	0
722788	8.2	5.8	0	0
722789	9.1	7.0	0	0
722790	7.8	6.7	TR	0
722819	6.5	5.6	TR	0
724360	9.6	7.7	0	0
724361	3.6	2.8	TR	0
724362	5.5	4.9	TR	0

Source: FDA, 2011.

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