A TWO-PHASE PILOT STUDY OF BROCCOLI:

1. THE EFFECT OF REFRIGERATED STORAGE ON BETA-CAROTENE, VITAMIN C, AND FOLATE;

2. ON-SITE NUTRITION EDUCATION STUDY TO PROMOTE THE CONSUMPTION OF BROCCOLI.

A THESIS SUBMITTED TO THE GRADUATE DIVISION OF THE UNIVERSITY OF HAWAI'I IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

IN

NUTRITIONAL SCIENCES

DECEMBER 2004

By:
Andreana A. Valiere

Thesis Committee:
Alvin Huang, Chairperson
Dian A. Dooley
Stuart Nakamoto
ABSTRACT

Fruits and vegetables are providers of many nutrients in the human diet including many vitamins, minerals, and many other important compounds. A high intake of fruits and vegetables is clearly associated with a decreased risk of chronic diseases, such as various cancers and cardiovascular disease. The current evidence from epidemiologic studies as well as clinical trials for beta-carotene, vitamin C, and folate are reviewed.

Fruits and vegetables are highly perishable products and tend to lose some nutrients after harvest. In addition, the nutritive value of fruits and vegetables varies tremendously even among the same fruit or vegetable depending on a variety of factors. Most consumers rely on supermarkets for their produce; hence, they have no control over the factors relating to the nutritive value of their produce prior to their produce selection. Especially in an island state like Hawaii, all of the non-local produce has to endure a great deal of traveling before it sees a supermarket shelf. Hence, the effect of refrigerated storage on the beta-carotene, vitamin C, and folate content of broccoli purchased at the supermarket is explored in the first experiment.

Since the 1970's, the supermarket has been increasingly sought after as a prime location to promote healthy eating through various programs. Supermarkets have had mixed results as the sites for nutrition intervention studies; however, consumers do seem receptive to information presented at the point of purchase. The purpose of the second part of this pilot study is to ascertain the effect of presenting nutritional messages displayed at the point of purchase on consumers' purchasing of broccoli in the produce section of a supermarket.
# TABLE OF CONTENTS

LIST OF TABLES .................................................................................................................. vii
LIST OF FIGURES ............................................................................................................... viii

CHAPTER I
BETA-CAROTENE, VITAMIN C & FOLATE LITERATURE REVIEW ...................................... 1
Introduction .......................................................................................................................... 1
Beta-Carotene ...................................................................................................................... 2
  Beta-Carotene & Cancer .................................................................................................. 4
Vitamin C ............................................................................................................................ 9
  Vitamin C & Cardiovascular Disease .............................................................................. 11
  Vitamin C & Cancer ...................................................................................................... 16
Folate ................................................................................................................................. 17
  Folate & Cardiovascular Disease .................................................................................. 18

CHAPTER II
THE EFFECT OF REFRIGERATED STORAGE ON THE VITAMIN C, BETA-
CAROTENE, AND FOLATE CONTENT OF BROCCOLI ...................................................... 21
Literature Review ............................................................................................................... 21
  Introduction ..................................................................................................................... 21
  Nutrient Loss in Produce .............................................................................................. 22
  Purpose .......................................................................................................................... 27
Methods ............................................................................................................................. 28
  Location Selection ........................................................................................................ 27
  Sampling ......................................................................................................................... 28
  Nutrient Analysis .......................................................................................................... 29
  Statistical Analysis ........................................................................................................ 30
Results & Discussion ......................................................................................................... 30
  Fresh Broccoli Results ............................................................................................... 30
  Analysis Test Results ................................................................................................. 33
  Refrigerated Storage Results ...................................................................................... 34
Conclusion/Implications .................................................................................................. 41
# LIST OF TABLES

<table>
<thead>
<tr>
<th>Table</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Epidemiological Studies of Vitamin C Concentrations in Blood and Cardiovascular Disease</td>
<td>11</td>
</tr>
<tr>
<td>Interpreting Vitamin C Concentrations in Plasma &amp; Leukocytes</td>
<td>12</td>
</tr>
<tr>
<td>Prospective Cohort Studies of Vitamin C Intake and Cardiovascular Disease</td>
<td>13</td>
</tr>
<tr>
<td>Clinical Trials Using Vitamin C Supplementation</td>
<td>15</td>
</tr>
<tr>
<td>Epidemiological Studies on Folate and Cardiovascular Disease</td>
<td>20</td>
</tr>
<tr>
<td>Ascorbic Acid Retention after Refrigerated Storage of Raw Vegetables</td>
<td>25</td>
</tr>
<tr>
<td>Beta-Carotene Retention after Refrigerated Storage of Raw Vegetables</td>
<td>26</td>
</tr>
<tr>
<td>Supermarket Intervention Studies</td>
<td>46</td>
</tr>
<tr>
<td>Demographic Characteristics of Respondent</td>
<td>55</td>
</tr>
<tr>
<td>Broccoli Sales During the Intervention Compared to Control Weeks</td>
<td>59</td>
</tr>
</tbody>
</table>
## LIST OF FIGURES

<table>
<thead>
<tr>
<th>Figure</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh Broccoli Vitamin C Content</td>
<td>31</td>
</tr>
<tr>
<td>Fresh Broccoli Beta-Carotene Content</td>
<td>32</td>
</tr>
<tr>
<td>Fresh Broccoli Folate Content</td>
<td>33</td>
</tr>
<tr>
<td>Broccoli Vitamin C Content After Refrigeration</td>
<td>36</td>
</tr>
<tr>
<td>Broccoli Beta-Carotene Content After Refrigeration</td>
<td>37</td>
</tr>
<tr>
<td>Broccoli Folate Content After Refrigeration – Initial Experiment</td>
<td>39</td>
</tr>
<tr>
<td>Broccoli Folate Content After Refrigeration – Second Experiment</td>
<td>39</td>
</tr>
<tr>
<td>Self-Reported Vegetable Intake Per Week</td>
<td>57</td>
</tr>
<tr>
<td>Self-Reported Broccoli Intake Per Week</td>
<td>58</td>
</tr>
<tr>
<td>Broccoli Sales Data</td>
<td>60</td>
</tr>
</tbody>
</table>
CHAPTER I
BETA-CAROTENE, VITAMIN C & FOLATE LITERATURE REVIEW

Introduction

A high intake of fruits and vegetables is clearly associated with a decreased risk of chronic diseases, such as various cancers, especially those of the respiratory and digestive tracts (1,2) and cardiovascular disease (CVD) (3,4). As a result of these established health benefits of fruits and vegetables, different United States (U.S.) government agencies have recommended intake levels for the adult public. For example, the United States Department of Agriculture (USDA) Food Guide Pyramid recommends five to nine servings of fruits and vegetables per day. Consumers are getting the message and as a result are increasing their intake of fruits and vegetables (5). The average number of servings for fruit has increased to 1.4 and vegetables to 3.8 (6) (combined total of 5.2 servings) from approximately 2.8 servings (combined total) in 1988 (7).

Although Americans are consuming more fruits and vegetables, their intake of cruciferous vegetables (e.g., broccoli, Brussel sprouts, cabbage, and cauliflower) remains low (5). According to an analysis of the 1994 – 1996 Continuing Survey of Food Intakes by Individuals (CSFII), iceberg lettuce, tomatoes, French fried potatoes, bananas, and orange juice comprised nearly 30% of all fruits and vegetables consumed (5). Less than one in five Americans surveyed ate a cruciferous vegetable during the two-day survey period. This is unfortunate because cruciferous vegetables may offer unique health benefits and contain a variety of nutrients and other substances, such as vitamin C, folate, and phytochemicals (e.g., carotenoids, glucosinolates, and phenolic compounds) (8-11).
A high intake of cruciferous vegetables like broccoli, has been shown to decrease the risk of a number of cancers (12-14).

**Beta-Carotene**

Carotenoids consist of a family of fat-soluble compounds found in plants that are able to absorb light, thus producing the red color found in tomatoes or the yellow color found in yellow peppers. There are more than 600 carotenoids, with beta-carotene being the most widely publicly recognized and most scientifically researched. Beta-carotene is found predominantly in fruits and vegetables with orange, yellow, red, and even green pigments (see Appendix A, p. 85). The green pigment masks the carotenoid's yellow-orange color. It has been estimated that the average U.S. dietary intake of beta-carotene ranges from 0.5 to 6.5 mg per day (15).

About 50 of the 600 carotenoids have been shown to have pro-vitamin A activity (16). Beta-carotene's structure allows it to be cleaved into two molecules of vitamin A. This makes beta-carotene the carotenoid with the highest pro-vitamin A activity. Many populations throughout the world rely on beta-carotene foods and supplements for their vitamin A needs (17). This is currently the only universally accepted function of beta-carotene, because the other functions are still controversial (18). Beta-carotene has been shown to be an antioxidant *in vitro*; however, there have been mixed results *in vivo* and the extrapolation of this to human health is not currently understood well. Beta-carotene has been found to be a quencher of the reactive oxygen species, singlet oxygen (16,17). This singlet oxygen has been found in the skin of people exposed to the sun (17).
Specifically, in those with a genetic condition called erythropoietic protoporphyria, which causes extreme photosensitivity in their skin with painful symptoms resulting from sun exposure, high doses of beta-carotene supplements have historically been prescribed to reduce the photosensitivity. Beta-carotene is lipid soluble, and also has been found to act as an antioxidant that protects human lipoproteins, low-density lipoprotein (LDL) and high-density lipoprotein (HDL), by quenching free radicals in these lipoproteins or within cell membranes (19). Several mechanisms resulting from \textit{in vitro} studies have been proposed to explain beta-carotene’s capacity to quench free radicals: addition of the radical to beta-carotene, hydrogen abstraction, and/or the transfer of electrons (20-23). Beta-carotene has also been shown to work with vitamin E by regenerating the antioxidant form of vitamin E after vitamin E has quenched a reactive oxygen species and protect vitamin E from oxidation in lipids (20). In cell membranes, beta-carotene, vitamin E and vitamin C have been shown to work synergistically to protect lipids from oxidative damage (20,21). Niki et al. specified that the antioxidant ability of beta-carotene is higher at low oxygen pressures within the human body (20). There have also been several studies conducted regarding beta-carotene and various immune function indices with differing results (17). Bendich showed that beta-carotene could increase lymphocyte production in the rat independent of the rat’s pro-vitamin A status (24). Thus, beta-carotene may possibly have many roles in the human body; however, we are just beginning to understand the potential depth and complicated interrelationships between beta-carotene and other molecules.
Beta-Carotene & Cancer

Plasma levels of carotenoids have been shown to be the best indicators of fruit and vegetable intake (18), and this has been shown to be inversely proportional to disease risk for certain cancers (25). Although studies have shown that carotenoids may contribute to a possible decrease in risk for a variety of cancers (cervical, breast, colon/rectum, stomach and esophageal), lung cancer seems to show the strongest and most consistent inverse association with carotenoids from food sources, specifically beta-carotene (26). According to a review by Johnson (25), eleven of fourteen prospective epidemiologic studies published between 1981 and 1991 have shown a significant inverse relationship between beta-carotene (measured as beta-carotene intake and/or plasma level of beta-carotene) and the risk of obtaining lung cancer.

Based on the results from the observational epidemiologic studies, several large scale randomized clinical trials were planned to test this association between beta-carotene and cancer. A few of these trials have shown conflicting results and pointed to the opposite relationship where an increase in beta-carotene intake correlates with an increase in cancer rates (27,28). For example, the Beta-Carotene and Retinol Efficacy Trial (CARET) was a large, multi-center, randomized, double-blind, placebo-controlled primary prevention trial (27). The CARET trial included 18,314 smokers, former smokers and workers with exposure to asbestos. Omenn et al. tested the effect of a 30 mg beta-carotene supplement combined with 25,000 IU of retinol as retinyl palmitate [5 times the then Recommended Dietary Allowance (RDA)] per day on the incidence of
lung cancer. After four years of follow-up, the results of this study showed that the group treated with the combination of beta-carotene and retinol supplements did not prove beneficial. Instead, it may have increased the risk of death from lung cancer and cardiovascular disease in the smokers and those exposed to asbestos. As a consequence of these results, the intervention portion of the trial was stopped 21 months early, although the follow-up was expected to continue until 2001.

The Alpha-Tocopherol, Beta Carotene Cancer Prevention Study (ATBC) was a large scale (29,133 participants) randomized, double-blind, placebo-controlled clinical trial, which aimed to test whether or not daily supplementation with alpha-tocopherol, beta-carotene, or both would decrease the incidence of lung cancer and other cancers in Finnish male smokers (age 50-69 years) (28). These men all had no prior history of any cancers except nonmelanoma skin cancer. Each group of participants were randomly assigned to one of four groups: alpha-tocopherol (50 mg per day), beta-carotene (20 mg per day), both alpha-tocopherol, and beta-carotene or placebo. The intervention lasted for five to eight years for each participant and the entire study lasted from 1985 to 1993. There was a higher incidence of lung cancer in the strictly beta-carotene group and total mortality was eight percent higher than the subjects that were not given beta-carotene. There was also no evidence of any association between the combining of beta-carotene and alpha-tocopherol on the incidence of lung cancer. These results suggest a harmful effect of beta-carotene on the incidence of lung cancer in male smokers.
Another large scale (22,071 participants), randomized, double-blind, placebo-controlled clinical trial using beta carotene supplementation was a component of the Physicians’ Health Study (29). This study was longer that the CARET and ATBC clinical trials, with the intervention and follow-up lasting for an average of twelve years (1982-1995). In one portion of this study 50 mg of supplemental beta-carotene was given on alternating days to male physicians in the U.S. (40-84 years old). Eleven percent of the male physician participants were current smokers, 39% were former smokers, and all of them were healthy. The results of the study showed that there were practically no differences in the occurrence of malignant neoplasms, cardiovascular disease, or overall rate of mortality between those physicians supplemented with beta-carotene and those given placebo. Therefore, this large clinical trial that lasted twelve years did not show any benefit or harm inflicted by the beta-carotene supplementation.

More recently, a large clinical trial involving women (approximately 20,000) in the United States, the Women’s Health Study, saw no significant differences in the incidence of cancer, CVD or total mortality, in women who were supplemented with 50 mg of beta-carotene on alternating days for two consecutive years (30). These results were consistent for the smokers (13% of the total participants), as well as overall treatment versus placebo. These women were only followed for two years after supplementation. However, another clinical trial in China found a significant benefit of beta-carotene supplementation (31).
Recently, there have been a few epidemiological studies that have shown an association between intake of carotenoid-containing fruits and vegetables and an increase in lung function in smokers (32,33). Schunemann et al. reviewed the NHANES III survey data and found that the greater the intake of foods with antioxidant activity, the healthier the lung function was of smokers (32). The hypothesis extrapolated from these recent epidemiological studies is explained below.

In both the CARET and ATBC clinical intervention studies, the researchers have shown a direct relationship between beta-carotene supplementation and lung cancer occurrence, lung cancer mortality, and overall mortality. In both of these trials large doses of supplemental beta-carotene were used, rather than beta-carotene from food sources. As stated earlier, the average intake of beta-carotene from food sources is only about 0.5 to 6.5 mg per day (15). Therefore, the doses given in these clinical trials were at least three times the maximum amount of beta-carotene consumed through food sources. A few years of supplementation may also have been too brief to reduce the development of cancers in individuals exposed to many years of tobacco carcinogens (28). In addition, these clinical trials used subjects who were already at high risk for lung cancer (smokers and asbestos workers). In high risk individuals beta-carotene may have a different effect than in healthy individuals. These people may have already been further along in the cancer process, such that the supplementation may not be effective (17). Beta-carotene is known to act as a prooxidant or antioxidant depending on the specific conditions. Therefore, it is plausible that large doses of beta-carotene may act as
a prooxidant in those at high risk for lung cancer. Yeum et al. suggests that it is the resulting carotenoid oxidative products formed when beta-carotene comes in contact with smoke that are the actual carcinogens responsible for the proliferation of cancer (34).

Bendich suggests that if lung function is increased in smokers supplemented with beta-carotene, as suggested by some researchers (32,33,35), then this increase in lung function could cause a greater exposure to tobacco carcinogens (17). This hypothesis would also suggest a dose-response relationship between lung cancer incidence and degree of smoking. In fact, this relationship was found in the ATBC trial.

There are currently two large studies underway involving beta-carotene. The Women’s Health Initiative (WHI) has over 150,000 postmenopausal women enrolled since 1993 (36). Baseline information from these participants indicates that 44% take a supplement that includes beta-carotene and 5% use a supplement of beta-carotene alone. This means that there are about 7,500 participants consuming supplemental beta-carotene; the information resulting from these participants should be helpful in elucidating the association between beta-carotene and disease. In addition, the Physician’s Health Study II is currently in progress with 15,000 U.S. male physicians over the age of 55 years (37). This is the only ongoing study which still uses high doses of beta-carotene supplements (50 mg on alternating days) in humans.

These two ongoing studies should enhance the knowledge of the relationship between beta-carotene and chronic disease, which is quite complex and not clearly understood. The functions of beta-carotene need to be clearly elucidated before
researchers can understand beta-carotene’s role in chronic disease. As a result of the conflicting evidence of beta-carotene supplements, and the possibility of a harmful effect, the Dietary Reference Intake (DRI) committee does not recommend the use of beta-carotene supplements for the general population (18). They recommend that the general population consume beta-carotene only in its natural food form.

**Vitamin C**

Vitamin C is an essential micronutrient that is soluble in water. It is required in the human diet as well as by other mammals that lack the enzyme gulonolactone oxidase (38). The presence of this enzyme gives most mammals, like cats, the ability to synthesize vitamin C from glucose. Vitamin C is found in plants, most notably in fruits and vegetables (see Appendix A, p. 86). Vitamin C is essential because it is a cofactor for several enzymes, which take part in the biosynthesis of collagen, carnitine, and neurotransmitters (16). Without vitamin C available for hydroxylation of proline and lysine, and subsequent failure of fibroblasts to generate mature collagen, a deficiency called scurvy results. The symptoms of scurvy include weakness, anemia, weak gums structures, as well as weakening of all collagen structures, development of mouth ulcers, and poor wound healing. Scurvy can be prevented by as little as 10 mg of vitamin C per day (3). The RDA for vitamin C used to be based on preventing scurvy and was set at 60 mg per day, which would prevent scurvy for roughly one month without additional vitamin C (39). However, now the RDA (75 mg per day for adult women and 90 mg per day for adult men) is based upon vitamin C’s ability to quench free radicals in neutrophils (38). This amount of vitamin C is near the maximal concentration that can be maintained in neutrophils without excess spillage of ascorbate into urine. In addition, a Tolerable
Upper Intake Level (UL) has been set for vitamin C, which is the maximum amount that will cause no adverse effects in most healthy individuals. For adults the UL is two grams per day. This is based on the occurrence of osmotic diarrhea and gastrointestinal side effects at an intake (from food and supplements) larger than 2 grams. According to the Third National Health and Nutrition Examination Survey (NHANES III) data from 1988 – 1994, the median dietary intake of vitamin C for adult males was 105 mg/day (120 mg/day including supplements) and 90 mg/day for adult women (108 mg/day including supplements) (38). Although most Americans consume enough vitamin C to meet their bodies’ needs, those at risk for possible deficiency are the elderly, individuals with poor diets, especially if they are alcoholics and smokers and those with infections and/or diseases such as diabetes mellitus and some cancers that increase the rate of vitamin C turnover (16).

The name “vitamin C” refers to both ascorbic acid and dehydroascorbic acid. In vivo vitamin C subsists predominantly in its reduced form, ascorbic acid (40). The oxidized form, dehydroascorbic acid, is readily reduced back to ascorbic acid intracellularly by one of several cofactors (e.g., glutathione, nicotinamide adenine dinucleotide, or nicotinamide adenine dinucleotide phosphate). This recycling from ascorbic acid to dehydroascorbic acid and back to ascorbic acid is what makes vitamin C an effective antioxidant in aqueous parts of the cell. There is extensive evidence of vitamin C administration resulting in a decrease of several markers of oxidative stress (38). Oxidative stress has been anticipated to be related to the etiology of numerous chronic diseases, such as: cancer, CVD, cataracts, age-related macular degeneration, central neurodegenerative diseases (e.g., Parkinson’s disease and Alzheimer’s disease),
and aging (38,41). Of all of the chronic diseases related to oxidative stress, the available evidence is strongest for CVD (38). Vitamin C has been shown to be associated with a decrease in the risk of CVD, certain cancers, and cataracts; however, only CVD will be discussed in the following section.

**Vitamin C & Cardiovascular Disease**

The available evidence of research studies on vitamin C intake suggests an association between increased vitamin C intake and a decreased risk of cardiovascular disease, certain cancers, and cataracts (38). Several different researchers have looked at vitamin C concentrations in the blood and the relationship to CVD (Table 1).

### Table 1: Epidemiological Studies of Vitamin C Concentrations in Blood and CVD

<table>
<thead>
<tr>
<th>Study/Reference</th>
<th>Population</th>
<th>Disease Outcome(s)</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basel Prospective Study</td>
<td>2,974 Swiss men</td>
<td>CAD stroke</td>
<td>Non-significant ↓ in risk of CAD &amp; stroke with plasma vitamin C &gt; [23 μmol/L (0.4 mg/dl)]</td>
</tr>
<tr>
<td>Eichholzer 1992 (42)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gale 1995 (43)</td>
<td>730 British elderly adults</td>
<td>Stroke</td>
<td>30% ↓ risk of death from stroke with plasma vitamin C &gt; [28 μmol/L (0.5 mg/dl)] vs. &lt; [12 μmol/L (0.2 mg/dl)]</td>
</tr>
<tr>
<td>Nyyssonen 1997 (44)</td>
<td>1,605 Finnish men</td>
<td>Coronary heart disease (CHD)</td>
<td>60% ↓ risk of CHD of men with ↑ plasma vitamin C (&gt; [11.4 μmol/L (0.2 mg/dl)])</td>
</tr>
<tr>
<td>NHANES II Simon 1998 (45)</td>
<td>6,624 U.S. adults</td>
<td>CHD stroke</td>
<td>26% ↓ relative risk of CHD &amp; stroke with serum vitamin C of [63-153 μmol/L (1.1-2.7 mg/dl)] vs. [6-23 μmol/L (.1-.4 mg/dl)]</td>
</tr>
<tr>
<td>Singh 1995 (46)</td>
<td>Indian men &amp; women</td>
<td>Coronary artery disease (CAD)</td>
<td>2 x ↓ risk of CAD (top vs. bottom quartile of plasma vitamin C concentrations)</td>
</tr>
</tbody>
</table>
Table 2: Interpreting Vitamin C Concentrations in Plasma & Leukocytes

<table>
<thead>
<tr>
<th></th>
<th>Plasma µmol/L (mg/dl)</th>
<th>Mixed Leukocytes nmol/10^8 cells (µg/10^8 cells)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adequate</td>
<td>&gt; 23 (&gt; 0.4)</td>
<td>&gt; 114 (&gt; 20)</td>
</tr>
<tr>
<td>Low</td>
<td>11.4-23 (0.2-0.4)</td>
<td>57-114 (10-20)</td>
</tr>
<tr>
<td>Deficient</td>
<td>&lt; 11.4 (&lt; 0.2)</td>
<td>&lt; 57 (&lt; 10)</td>
</tr>
</tbody>
</table>

Source: Jacob R. (47)

Four out of the five studies listed in Table 1 have found a significant inverse association between CVD risk and blood vitamin C concentration. Table 2 gives guidelines on how to interpret blood vitamin C concentrations. Similar to the association of beta-carotene as a reliable marker for vegetable consumption, plasma ascorbate has been found to be a valid marker of fruit intake (41). Additionally, five out of the ten epidemiological studies listed in Table 3 have found an inverse association between vitamin C intake and risk of cardiovascular disease. One of the five studies mentioned looked at the carotid artery wall of the heart, specifically, and found this thickness to be negatively associated with vitamin C intake (51). However, the levels of vitamin C that were attributed to this negative association (728 mg/day for women and 982 mg/day for men) were rather large and would require the consumption of many servings of fruits and vegetable per day or approximately a one gram supplement of vitamin C.
<table>
<thead>
<tr>
<th>Study/Reference</th>
<th>Population</th>
<th>Disease Outcome(s)</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enstrom 1986 (48)</td>
<td>3,119 Alameda County, CA residents</td>
<td>CVD</td>
<td>No association with vitamin C intakes &amp; reduction of risk for CVD</td>
</tr>
<tr>
<td>Enstrom 1992 (49)</td>
<td>11,000 U.S. adults</td>
<td>CVD</td>
<td>45% ↓ in CVD in men &amp; 25% ↓ in women with &gt; 300 mg/day vitamin C</td>
</tr>
<tr>
<td>Gale 1995 (43)</td>
<td>730 British elderly adults</td>
<td>Stroke CAD</td>
<td>50% ↓ in risk of stroke with &gt; 45 mg/day vitamin C vs. &lt; 28 mg/day vitamin C Nonsignificant ↓ (20%) in CAD</td>
</tr>
<tr>
<td>Knekt 1994 (50)</td>
<td>5,000 Finnish men &amp; women</td>
<td>CAD</td>
<td>↓ risk of CAD in women with &gt; 91 mg/day vitamin C vs. women with &lt; 61 mg/day Men - no association</td>
</tr>
<tr>
<td>Atherosclerosis Risk in Communities Study Kritchevsky 1995 (51)</td>
<td>Men &amp; women &gt; 55 years</td>
<td>CAD</td>
<td>↓ intima thickness in women with &gt; 728 mg/day vitamin C &amp; men with &gt; 982 mg/day vs. women with &lt; 64 mg/day &amp; men with &lt; 56 mg/day</td>
</tr>
<tr>
<td>Iowa Women’s Health Study Kushi 1996 (52)</td>
<td>34,486 postmenopausal women</td>
<td>CHD</td>
<td>No association with vitamin C intakes &amp; reduction of risk in CHD mortality</td>
</tr>
<tr>
<td>Established Populations for Epidemiologic Studies of the Elderly Losonezy 1996 (53)</td>
<td>&gt; 11,000 adults &gt;/65 years</td>
<td>CHD</td>
<td>No association with vitamin C intakes &amp; reduction of risk in CHD mortality</td>
</tr>
<tr>
<td>Nurses’ Health Study Osganian 2003 (54)</td>
<td>85,118 nurses in U.S.</td>
<td>CHD</td>
<td>Significant inverse association (27%) between total intake of vitamin C (highest quintile – &gt;/ 360 mg vitamin C vs. lowest – &gt;/ 93 mg vitamin C) &amp; risk of CHD (RR = 0.73). With those who did not use supplements the association was not significant. Vitamin C supplementation alone was associated with a significantly ↓ risk (28%) of CHD.</td>
</tr>
<tr>
<td>Western Electric Study Pandey 1995 (55)</td>
<td>1,556 U.S., middle-aged men</td>
<td>CAD</td>
<td>25% ↓ risk of CAD with &gt; 113 mg/day vitamin C vs. &lt; 82 mg/day</td>
</tr>
<tr>
<td>U.S. Health Professionals Follow-up Study Rimm 1993 (56)</td>
<td>39,910 male health professionals</td>
<td>CHD</td>
<td>No association between ↑ intakes of vitamin C (92 – 1162 mg/day) &amp; reduction in risk of CHD</td>
</tr>
<tr>
<td>Sahyoun 1996 (57)</td>
<td>725 elderly Massachusetts adults</td>
<td>CVD</td>
<td>62% ↓ risk of CVD with &gt; 388 mg/day vitamin C vs. &lt; 90 mg/day</td>
</tr>
</tbody>
</table>
Although many epidemiologic studies point to an inverse association between CVD risk and vitamin C intake, the evidence remains inconclusive because epidemiology identifies only associations and cannot demonstrate causality. Therefore, it is necessary to look at the evidence from clinical trials. Table 4 lists five clinical trials that have used vitamin C supplementation either alone or in combination with other antioxidants. The mechanism most often studied in relation to atherosclerosis is the oxidation of LDL cholesterol, which is then taken up by macrophages, which develop into foam cells (38,63,64). The subsequent events result in the release of cytokines and oxygen free radicals, which promotes the bursting of plaques in the arteries and a possible heart attack or stroke (65). Vitamin C has been found to be the best water-soluble antioxidant in vitro for the protection of plasma lipoproteins from free radical damage (66). Vitamin C has also been found to enhance endothelial vasodilation (67), restore coronary microcirculation in smokers (68), shield vascular smooth muscle cells from oxidative damage in vitro (69), and impede various pro-inflammatory mechanisms in vitro, in animal models and in human forearm models (70). Given all of these possible mechanisms vitamin C, in theory, should help protect against CVD. However, the available evidence from clinical trials does not entirely support this conclusion (Table 4). In addition, most of the attention in clinical trials so far has been placed on vitamin E because it is a fat-soluble antioxidant and is known to neutralize lipid peroxidation. It has been suggested that supplementation with both vitamin C and vitamin E might produce better results because vitamin C is believed to restore vitamin E from its oxidized state back to its reduced state (65). Therefore, there are a few large scale clinical trials underway which combine vitamin C and vitamin E or compare vitamin C, vitamin E, and
beta-carotene with a placebo (e.g., the Supplementation Vitamins Minerals and Antioxidant trial (SUVIMAX), Women’s Antioxidant Cardiovascular Disease Study (WACS), and Physician’s Health Study II) (38,65).

<table>
<thead>
<tr>
<th>Study/Reference</th>
<th>Population</th>
<th>Duration (years)</th>
<th>Daily Dose</th>
<th>Disease Outcome</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>(Randomized) Brown 2001 (58)</td>
<td>146 CAD patients</td>
<td>3</td>
<td>Statin drugs, statins + 800 IU α-toc, 1000 mg vitamin C, 25 mg selenium &amp; β-carotene, or antioxidants alone.</td>
<td>CAD</td>
<td>Nonsignificant ↓ in CAD progression with antioxidants alone vs. placebo. Greater progression of stenosis &amp; significantly more cardiovascular events with statins + antioxidants than with statins alone.</td>
</tr>
<tr>
<td>Heart Protection Study (59) (Randomized, Secondary Prevention)</td>
<td>20,536 high-risk adults in U.K. (40-80 years)</td>
<td>&gt;/5</td>
<td>20 mg β-carotene, 600 mg α-toc., or 250 mg vitamin C</td>
<td>Total mortality</td>
<td>No significant difference in cardiac outcomes.</td>
</tr>
<tr>
<td>ASAP study (Randomized) Salonen 2000 (60)</td>
<td>520 hypercholesterolemic men &amp; women (45-69 years)</td>
<td>6</td>
<td>136 IU α-DL-tocopherol &amp; 250 mg vitamin C</td>
<td>CAD</td>
<td>↓ in intima-media thickness progression in common carotid artery - vitamin E and C groups for men &amp; women, and in combination group (men).</td>
</tr>
<tr>
<td>(Randomized) Tardif 1997 (61)</td>
<td>80 patients</td>
<td>6 months</td>
<td>Probucol vs. β-carotene, 700 IU α-toc. &amp; 500 mg vitamin C</td>
<td>Restenosis</td>
<td>Probucol significantly ↓ risk of restenosis while vitamin combination did not.</td>
</tr>
<tr>
<td>(Randomized, Secondary Prevention) Waters 2002 (62)</td>
<td>423 postmenopausal women</td>
<td>&gt;/2</td>
<td>800 IU vitamin E and 1000 mg vitamin C</td>
<td>CHD</td>
<td>No ↓ in CHD. Positive trend towards ↑ death, nonfatal MI &amp; stroke.</td>
</tr>
</tbody>
</table>

a = tocopherol
**Vitamin C & Cancer**

Although studies have shown that vitamin C may contribute to a possible decrease in risk of a number of cancers (cervical, pancreatic, lung, esophageal and mouth and pharynx), the risk for stomach cancer (gastric cancer) seems to show the strongest and most consistent inverse association with vitamin C intake from food, than any other form of cancer (26). The mechanisms proposed by both epidemiological and intervention researchers for vitamin C to protect against gastric cancer include scavenging of reactive oxygen species and reactive nitrogen species in the gastric mucosa or hindering the formation of carcinogenic N-nitroso compounds (71-73). Another possible mechanism for decreasing cancer risk is the enhancement of the immune response and hastening of the detoxification of carcinogens by vitamin C (74). Vitamin C is known to be secreted in the gastric juice, and patients with gastritis and *Helicobacter pylori* infection, which are both risk factors for gastric cancer, have decreased ascorbate in their gastric juice (75,76). In addition, vitamin C supplementation has been shown to increase gastric ascorbate. However, the evidence from two intervention studies has not shown a reduction in gastric cancer incidence (73).

In conclusion, it is unclear at this time whether or not supplementation with vitamin C alone is helpful in preventing CVD or other chronic diseases. Long-term high intakes of vitamin C may simply be a marker for a more healthful lifestyle. Also, vitamin C may have different effects in different subject populations, e.g., healthy subjects versus subjects with CVD or congestive heart failure. Whether a subject is well-nourished or not may also make a difference. Furthermore, there may be undiscovered roles of vitamin C in assisting with protecting against chronic diseases. Thus, vitamin C may be
one small piece of a much larger puzzle. The results from the ongoing clinical trials mentioned above will represent over 80,000 subjects. This should provide some insight into these unanswered questions about vitamin C.

**Folate**

Folate or folacin is the name for a group of compounds that are essential, water-soluble micronutrients, which function as coenzymes for the metabolism of amino acids and nucleic acid synthesis, and thus critically important for growth and development. Folic acid or pteroylglutamic acid, is the simplest form of folate compounds (monoglutamate), and this is also the form used in supplements and fortified foods and occurs rarely in food (3). In foods, such as green leafy vegetables, orange juice, and legumes, folate is found primarily as pteroylpolyglutamates (polyglutamates). The RDA for adult males and adult nonpregnant and non-lactating females is 400 µg dietary folate equivalents (DFE) (77). It is recommended that women of child-bearing age consume 400 µg/day supplementally in addition to consuming food folate, to reduce the risk of neural tube defects. Using DFEs compensates for the significantly lower (about 50%) bioavailability of food folate compared to supplemental or fortified folic acid. From 1988 to 1994 the median intake of folate among U.S. adults was estimated as 250 µg/day. This was prior to the mandatory fortification of grains in 1998; therefore, this amount is estimated to be between 80 to 100 µg more for women and even higher for men. In addition, food composition databases used to calculate folate intake underestimate folate data because of inconsistencies of traditional analytical methods for folate analysis. The current RDA is
based on adequate erythrocyte folate, which indicates the amount of folate stored in tissues. A deficiency in folate results in an increase in blood homocysteine concentration and may result in megaloblastic or macrocytic anemia (78). Deficiency is not common in the U.S. but may be seen with the elderly, alcoholics, pregnant women, and premature infants. The adult UL is set at 1,000 μg/day of folate from supplemental or fortified food because adequate folic acid can mask a vitamin B₁₂ deficiency (77). The evidence is strong for folate reducing the risk of neural tube defects; and, the evidence is promising, yet still being gathered, for folate reducing the risks of cardiovascular disease, certain cancers, and psychiatric and mental disorders (77). Folate’s role in reducing the risk of CVD will be discussed in the next section.

**Folate & Cardiovascular Disease**

Folate in the form of methyltetrahydrofolate, is required as a coenzyme for methionine synthase in the metabolism of methionine. Without adequate folate, homocysteine will accumulate (78). Elevated homocysteine has been established as a risk factor for vascular disease through many observational and experimental studies (77,79). Although the mechanism is still uncertain, there have been many possible hypotheses to explain the role of homocysteine in CVD (80). These hypotheses include homocysteine being toxic to endothelial cells and stimulating the growth of smooth muscle cells, resulting in atherosclerotic lesions (81), and increasing the adhesiveness of platelets and affecting clotting factors (82). Furthermore, homocysteine has been found to be significantly elevated in folate deficient subjects (83,84), as well as inversely correlated with serum
and plasma folate concentration of normal subjects (85-89). In a recent meta-analysis of clinical trials using folic acid supplementation to lower homocysteine levels in the blood, it was determined that folic acid supplements reduced homocysteine levels by roughly 25% [Homocysteine Lowering Trialists’ Collaboration (HLTC)] (90). In addition, these effects were more prominent if the subjects had higher pre-treatment blood homocysteine levels or lower pre-treatment blood. This inverse correlation between dietary folate and homocysteine is linear up to 300 μg of folate, where it then reaches a plateau (77).

There have not been many studies that have directly looked at the relationship between folate and cardiovascular disease (79). Table 5 lists the various epidemiological studies that have analyzed the relationship between CVD and folate. In five out of the eight studies listed there is a significant correlation between reduction in risk of CVD and higher blood folate levels or higher dietary intake of folate (supplemental or food sources) (93-97).

In summary, there is ample evidence linking increased blood homocysteine with low dietary folate; however, the relationship between folate and CVD although promising, has not been proven to date. Large-scale, randomized clinical trials involving folic acid supplementation are needed to help explain this relationship further. Although the mechanism for folate’s involvement in CVD seems convincing with respect to homocysteine metabolism, folate may be acting in other ways to help reduce CVD risk. Folate could also be a marker of other variables such as fruit and vegetable intake.
Several clinical trials involving folic acid supplementation are needed to further the knowledge related to folate’s role in CVD.

<table>
<thead>
<tr>
<th>Study/Reference</th>
<th>Population</th>
<th>Disease Outcome(s)</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Physicians’ Health Study (Nested case-control) Chasan-Taber 1996 (91)</td>
<td>333 male physicians, U.S. CHD</td>
<td><strong>Nonsignificant relationship between extreme quintiles of plasma folate and incidence of CHD.</strong></td>
<td></td>
</tr>
<tr>
<td>(Nested case-control) Folsom 1998 (92)</td>
<td>232 males &amp; females in 4 U.S. communities CHD</td>
<td><strong>Nonsignificant relationship between extreme quintiles of plasma folate and CHD risk.</strong></td>
<td></td>
</tr>
<tr>
<td>Nutrition Canada Survey (Cohort) Morrison 1996 (93)</td>
<td>165 males &amp; females in Canada CHD</td>
<td><strong>Significant ↑ risk of fatal CHD for lowest serum folate quintile vs. highest quintile.</strong></td>
<td></td>
</tr>
<tr>
<td>(Case-control) Pancharuniti 1994 (87)</td>
<td>101 males with early onset CAD from Georgia Heart Clinic in La Grange, GA CAD</td>
<td><strong>Nonsignificant relationship between plasma folate and CAD.</strong></td>
<td></td>
</tr>
<tr>
<td>Health Professionals Study (Cohort) Rimm 1996 (94)</td>
<td>1416 male health professionals, U.S. Myocardial Infarction (MI), CHD</td>
<td><strong>Significantly ↓ risk of CHD for highest quintile of folate intake vs. lowest quintile.</strong></td>
<td></td>
</tr>
<tr>
<td>Nurses’ Health Study (Cohort) Rimm 1998 (95)</td>
<td>939 females nurses, U.S. MI, CHD</td>
<td><strong>Significant inverse relationship between dietary folate and risk of CHD. (Greatest for women who consumed alcohol).</strong></td>
<td></td>
</tr>
<tr>
<td>(Case-control) Robinson 1998 (96)</td>
<td>750 males &amp; females in Europe Vascular Disease</td>
<td><strong>Significant inverse relationship between red cell folate levels and risk of vascular disease.</strong></td>
<td></td>
</tr>
<tr>
<td>(Case-control) Verhoef 1996 (97)</td>
<td>130 males &amp; females in Boston MI</td>
<td><strong>Nonsignificant relationship between plasma folate and risk of MI.</strong></td>
<td></td>
</tr>
</tbody>
</table>

Source: Elcholzer et al. (79)
CHAPTER II
THE EFFECT OF REFRIGERATED STORAGE ON THE VITAMIN C,
BETA-CAROTENE, AND FOLATE CONTENT OF BROCCOLI

Literature Review

Introduction

Fruits and vegetables are providers of many nutrients in the human diet, including but not limited to: vitamin C (ascorbic acid), vitamin A (as beta-carotene and other pro-vitamin A carotenoids), folate, vitamin B₆, magnesium, iron, thiamin, niacin, dietary fiber, carbohydrates, some protein, and even a minimal amount of fat (in most fruits and vegetables). However, they are highly perishable products and tend to lose some of these nutrients after harvest (98). In addition, the nutritive value of fruits and vegetables varies tremendously even among the same fruit or vegetable depending on a variety of factors including the following: genetics (cultivars/varieties), cultural factors (soil, irrigation, pruning, growth regulators, fertilizers, etc.), light, temperature, seasons and climate, maturity and ripeness, and post-harvest handling [mechanical damage, storage and transportation (temperature, humidity)]. Especially in an island state like Hawaii, all of the non-local produce has to endure a great deal of traveling before it sees a supermarket shelf. It takes about ten to fourteen days to get from the farm where it is harvested, to the shelves of a supermarket in Hawaii. Therefore, there are many confounding factors when considering the nutritive value of a fruit or vegetable. However, these factors are out of the control of the average consumer. Most consumers rely on supermarkets for their produce; hence, they have no control over the factors relating to the nutritive value of
their produce prior to their produce selection. Once they have selected their produce, they need to properly store their produce at refrigerated temperatures in order to protect the produce from further nutrient degradation. Most consumers have the mistaken impression that fresh produce is superior in nutrient content to processed produce, but this is not always the case due to circumstances like post-harvest handling or other factors affecting the nutrient content of raw produce (4). In addition, produce that is frozen or canned is usually processed immediately after harvest, which can increase nutrient retention (4).

**Nutrient Loss in Produce**

Respiration of fruits and vegetables pertains to the enzymatic oxidation of sugars to carbon dioxide, water, and release of energy (98). Respiration is an indicator of their physiological activity and possible storage life. The respiration rate varies for different fruits and vegetables and also is directly related to the temperature at which it is stored. For example, broccoli’s respiration rate is much lower at 4-5°C (32-37 mg/kg/h) than it is at room temperature (20-21°C) (278-320 mg/kg/h). This indicates that broccoli’s storage life is lengthened by refrigeration and shortened at room temperature. The effects of temperature have been studied extensively. Researchers have found that lower temperatures reduce respiration and increase storage length of vegetables (98). Other factors such as reducing oxygen content with controlled or modified atmosphere have also been found to reduce respiration (98). Broccoli is in the group of fruits and vegetables with the highest respiration rate. Other produce in this category include asparagus, mushrooms, peas, spinach and sweet corn. Broccoli is known to have a short
shelf-life of three to four weeks in air at 0°C (99,100) and two to three days in air at 20°C (101).

Different vegetables tend to lose more of certain nutrients than others and this varies with different storage parameters. Vitamin C has been analyzed in several different vegetables upon different storage conditions because it is known to be particularly susceptible to degradation (4). Vitamin C is adversely affected by oxygen, light, heat, enzymes, pH, and heavy metal ions (98,102). Table 6 compares different studies looking at various vegetables and their vitamin C content after several storage parameters. Overall, most non-cruciferous vegetables tend to lose vitamin C at different rates over time even when refrigerated, and cruciferous vegetables tend to retain more vitamin C when refrigerated (103,104,106-108). Out of the cruciferous vegetables, broccoli has been studied the most to determine the effect of refrigerated storage on the content of vitamin C. Although some researchers have found the content of vitamin C to decrease with refrigerated storage (4,106); most have found that even over a two or three week refrigerated storage period, broccoli has a nonsignificant change from baseline levels of vitamin C immediately after harvest (103-105,107,108). Therefore, broccoli seems to be a good source of vitamin C even after a couple weeks after harvest and especially compared to other vegetables that may rapidly lose vitamin C.

Broccoli is known to be a good source of carotenoids, particularly the pro-vitamin A carotenoid beta-carotene. Furthermore, Vitamin C (ascorbic acid) and beta-carotene are the most abundant antioxidant contributors naturally found in vegetables (4). However, carotenoid pigments are subject to degradation by oxygen, light, enzymes, and acid conditions (98,102), but whether or not beta-carotene is lost in vegetables as a result
of refrigerated storage and post-harvest handling needs further investigation. Table 7 lists the known studies looking specifically at vegetables and the effect of refrigerated storage on the beta-carotene content. Compared to vitamin C, not many different vegetables have been analyzed. The focus so far has been only on broccoli, green beans, and carrots. Overall, these vegetables have shown high retention of beta-carotene with some even increasing their beta-carotene content over time (4,107,108). For example, Paradis et al. found a significant increase in the beta-carotene content of broccoli even after 28 days after harvest under refrigeration at 4°C (107). On the contrary, Howard et al. found two differing results between broccoli harvests from different years (4). In 1994, Howard reported broccoli to retain 119% of its original beta-carotene content after three weeks of storage at 4°C; in 1995, he found broccoli to retain only 36% of its beta-carotene under the same storage conditions. He reported that this extreme difference possibly resulted from differences in environmental conditions between the two years. However, more research is needed to confirm the effect of refrigerated storage on the beta-carotene content of raw vegetables.

Folate is an important nutrient in health and some vegetables are a good source of folate, especially broccoli. However, folate is a complicated nutrient due to its many forms and difficulty testing for it (109). Folate is sensitive to acid conditions, light (predominantly ultraviolet light), heat, and oxygen (78,102). Significant losses of folate in vegetables from processing and cooking have been recognized by investigators as far back as 1943 (110). However, the effect of refrigerated storage on the folate content of raw vegetables has not been studied much. Mullin found that spinach and Swiss Chard stored at 4°C for fourteen days incurred no serious loss of total folacin (TFA) (111).
<table>
<thead>
<tr>
<th>Reference</th>
<th>Vegetables Analyzed</th>
<th>Replicates (n)</th>
<th>Storage Parameters</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Albrecht 1990</td>
<td>Broccoli, Brussel sprouts, cauliflower, green cabbage, Savoy cabbage, green beans, and potato (red and white)</td>
<td>7</td>
<td>All: 3 weeks (DAH) at 2°C &amp; 95-100% RH</td>
<td>75-98% retention after 3 weeks in cruciferous vegetables and 16-75% retention in noncruciferous vegetables (green beans = 16% and potatoes = 75%).</td>
</tr>
<tr>
<td>Albrecht 1991</td>
<td>Broccoli, Brussel sprouts, cauliflower, turnip, asparagus, green beans, green peas and spinach</td>
<td>7</td>
<td>All: 3 weeks (DAH) at 2°C &amp; 95-100% RH</td>
<td>&gt; 95% retention after 3 weeks for broccoli and brussel sprouts; 65-70% retention for green peas, spinach and turnip; 5-30% retention for asparagus and green beans.</td>
</tr>
<tr>
<td>Eheart &amp; Odland 1972</td>
<td>Broccoli</td>
<td>72</td>
<td>Both: 7 days (8 DAH) at 2°C &amp; 95-100% RH</td>
<td>Overall no significant change after 7 days; although, different varieties (3) behaved differently during storage.</td>
</tr>
<tr>
<td>Hudson 1986</td>
<td>Broccoli</td>
<td>702</td>
<td>Wholesale vs. retail vs. stored 3 days at 6.5°C &amp; 90-95% RH</td>
<td>17% average loss between wholesale level &amp; retail level (about 9 DAH).</td>
</tr>
<tr>
<td>Paradis 1996</td>
<td>Broccoli (2 harvests)</td>
<td>4</td>
<td>28 DAH at 4°C</td>
<td>Increase after 14 days with nonsignificant decrease after 28 days (both harvests).</td>
</tr>
<tr>
<td>Reference</td>
<td>Vegetables Analyzed</td>
<td>Replicates (n)</td>
<td>Storage Parameters</td>
<td>Results</td>
</tr>
<tr>
<td>--------------</td>
<td>---------------------</td>
<td>----------------</td>
<td>--------------------</td>
<td>-----------------------------------------------------------------------------------------------------------------------------------------</td>
</tr>
<tr>
<td></td>
<td>Green Beans (1995)</td>
<td>3</td>
<td>3 weeks (DAH&lt;sub&gt;a&lt;/sub&gt;) at 4°C</td>
<td>Initial increase, then linear decrease. Average retention after 16 days = 90%.</td>
</tr>
<tr>
<td></td>
<td>Carrots (1994 &amp; 1995)</td>
<td>3</td>
<td>6 months (DAH&lt;sub&gt;a&lt;/sub&gt;) at 4°C</td>
<td>Initial increase, then decrease, then increased (after 6 weeks). Average retention after 84 days = 128% (1994 &amp; 1995).</td>
</tr>
<tr>
<td>Paradis 1996 (107)</td>
<td>Broccoli (2 harvests)</td>
<td>4</td>
<td>28 DAH at 4°C</td>
<td>Significant increase after 14 days (160% retention) and remained higher than starting value (both harvests).</td>
</tr>
<tr>
<td>Wu 1992 (108)</td>
<td>Broccoli</td>
<td>3</td>
<td>Both: 7 DAH for 3 days at 4°C (after RMF&lt;sub&gt;c&lt;/sub&gt; simulation)</td>
<td>No significant change after 10 DAH.</td>
</tr>
<tr>
<td></td>
<td>Green Beans</td>
<td>3</td>
<td></td>
<td>No significant change after 10 DAH.</td>
</tr>
</tbody>
</table>

<sup>a</sup> = days after harvest  
<sup>b</sup> = relative humidity  
<sup>c</sup> = retail market fresh simulation
While Chen found that spinach held at room temperature for ten hours did not affect TFA content but did cause an increase in the free folacin (FFA) content by 57% (112). TFA is defined as the folacin content after treatment with conjugase, and FFA is the folacin content without conjugase treatment. He attributes this increase to a rapid deconjugation of pteroylpolyglutamates from the initiation of the intrinsic conjugase enzyme as a result of physical cellular damage and the high temperature. Chen also discovered that when refrigerating the fresh spinach for seven days at 4°C the colder temperature was able to slow conjugase activity so the FFA increased only 30%, but the TFA decreased about 26% over the seven days. More research is needed on the folate content of fresh vegetables after refrigerated storage.

Although one study (106) has analyzed the vitamin C content in vegetables at the retail level and compared this to the vitamin C content immediately after harvest, most researchers only analyze nutrient content starting immediately after harvest. Produce consumers in Hawaii may receive their produce later than those in the continental United States; thus, research on the nutritional quality of produce in Hawaii supermarkets is needed to determine if these values coincide with the values in the USDA database.

**Purpose**

The purpose of this research was two-fold: 1) to compare broccoli in Hawaii to USDA reference values for vitamin C, folate, and beta-carotene; and 2) to determine how simulated consumer storage affects the content of vitamin C, folate, and beta-carotene in raw broccoli.
Methods

Location Selection

Two supermarkets from two different large supermarket chains in two different locations were chosen as the locations for the sampling of broccoli. The companies that own these supermarkets are the main suppliers of broccoli in Hawaii. The majority of broccoli in Hawaii comes from producers in California. The broccoli is then shipped to Hawaii and delivered by various produce distributors. This process takes roughly 10 days to get from a farm in California to a supermarket produce shelf in Hawaii.

Sampling

Approximately two pounds of broccoli (broccoli crowns with approximately two inches cut off stem base) were chosen for each sample, so the value would represent a composite of different broccoli crowns. Each sample was treated the same way throughout the experiment. Each sample was placed in a plastic produce bag available in all supermarkets’ produce sections. The samples were chosen from the supermarkets’ refrigerated produce storage section located in a large walk-in refrigerator in the back of each location. The samples were chosen from the center of broccoli boxes that had been delivered that same day. The day each sample was selected was considered day zero for the purpose of simulating the first possible day that consumers could purchase broccoli. Five samples were selected from each location to obtain averages for beta-carotene, vitamin C, and folate content of broccoli in Hawaii. These samples are referred to as “fresh broccoli” in the results section. They were collected on selected dates on Monday through Thursday in order to be received by the analysis company, Anresco Inc., during normal business hours to ensure consistent processing. These averages served as
indicators of how close the USDA database values were to those in broccoli on the
supermarket shelf at the freshest possible point of purchase for Hawaii residents. These
values did not serve as baseline values because these were not the samples that were
tested in the storage portion of this experiment. At a later date three samples were chosen
from each location out of the same box and stored at 4°C in a walk-in refrigerator for
either 3, 7, or 14 days. This was repeated three times (3 replicates for each storage
parameter). To further examine folate analysis, a second experiment involving folate
analysis only was performed. Three replicates were chosen from each location and stored
at 4°C in a walk-in refrigerator for either 7 or 14 days.

Analysis Test
To serve as an indicator of Anresco Inc.'s methods, two samples of broccoli
(approximately two pounds each) were taken from the same shipment container in the
refrigerated section of one supermarket’s produce storage section and processed by two
different methods. These samples were expected to produce extremely different nutrient
results. One sample was immediately shipped by one-day express mail to Anresco Inc.
where it was analyzed for vitamin C, beta-carotene, and folate. The other sample was
puréed and frozen at −20°C over night. The next morning the puréed sample was shipped
by one-day express mail to Anresco for analysis of vitamin C, beta-carotene, and folate.

Nutrient Analysis
After the appropriate length of storage, each sample was sent to Anresco Inc. in San
Francisco, California to be analyzed. Each sample was transferred from the refrigerator
to a sealed Ziploc bag and sent in a Styrofoam container with blue ice packs surrounding
the samples. Each container was sent with a temperature indicator that would signal if
the containers became too warm. No containers indicated that they were too warm after shipment. Each container was shipped by one-day express mail to California. All of the samples were only shipped Monday through Thursday and arrived the next morning in California. Anresco immediately transfers the samples into their freezer when they arrive. Then, the appropriate method of analysis was followed. The beta-carotene method of analysis Anresco Inc. used was AOAC 941.15. The vitamin C method of analysis used was AOAC 985.33. The folate method was AOAC 992.05.

Statistical Analysis

Data were analyzed with the statistical software program SPSS (ver. 10.0). T-tests for independent samples were used to compare the means of the fresh broccoli values of vitamin C, beta-carotene, and folate for supermarket A versus supermarket B as well as for comparing the fresh broccoli values to the USDA nutrient values. A p-value of <0.05 was considered significant for all tests.

Results & Discussion

Fresh Broccoli Results

The results of the five broccoli samples taken on different days from each supermarket are illustrated in Figures 1, 2, and 3. The mean content of vitamin C from all ten fresh broccoli samples obtained in Hawaii is 87.2 mg/100g (standard deviation (SD) = 19.1). This mean is not statistically different than the USDA reference value of 89.2 mg/100g (113). However, the mean vitamin C content was 104.2 mg/100g (SD = 7.4) for store A and 70.2 mg/100g (SD = 6.8) for store B; these means were significantly different with p < 0.01. The vitamin C content of the broccoli samples was consistent among the same
stores; however, store B was 33% lower (significant with \( p < .01 \)) on average than store A (Figure 1). This is in contrast to the findings of Hudson et al. who found that the difference in ascorbic acid in broccoli within stores was larger than the difference between stores (106).

If the mean beta-carotene content of all ten fresh broccoli samples (571.44 \( \mu g/100g \), \( SD = 201.7 \)) is compared to the USDA reference value of 383 \( \mu g/100g \) (113), then the beta-carotene content of fresh broccoli obtained in Hawaii is significantly higher than the USDA reference value (\( p < 0.05 \)). This implies that the broccoli samples obtained in Hawaii were older due to the increased beta-carotene synthesis. The beta-carotene content averaged 396.5 \( \mu g/100g \) (SD = 85.8) for store A and 746.4 \( \mu g/100g \) (SD = 87.2) for store B. Store B’s mean was 47% higher in beta-carotene compared to store A (significantly different with \( p < 0.01 \)). This large difference could be attributable to
increased storage length prior to supermarket display because beta-carotene is said to increase during post-harvest senescence from the continuation of physiochemical reactions (4,114). Also, this difference could be the result of adverse environmental conditions experienced by the broccoli crops (4), but this is unlikely because the broccoli was sampled over the course of one month so the samples represented different shipments. The variation between the samples was fairly consistent within the stores and is illustrated in Figure 2.

![Figure 2: Fresh Broccoli Beta-Carotene Content](image)

If the mean total folate content of all ten fresh broccoli samples (88.9 µg/100g, SD = 25.0) is compared to the USDA reference value of 63 µg/100g, then the folate content of fresh broccoli obtained in Hawaii is significantly higher than the USDA reference value (p < 0.01). The mean total folate content was 101.4 µg/100g (SD = 25.4) for store A and 76.4 µg/100g (SD = 19.3) for store B, and this difference is not
significantly different. Since folate compounds are sensitive to light, the samples from store B may have been exposed to more light in the shipment or trucking process. The variation seen between the samples is illustrated in Figure 3. The folate values from broccoli in store A were consistent with the exception of one high value of 145 μg/100g. On the contrary, the samples from store B were consistent with the exception of one low value of 50 μg/100g. With a higher vitamin C average, lower beta-carotene average, and higher folate average, the data from the fresh broccoli suggests that store A may have had fresher broccoli, or at least at the times sampled.

![Figure 3: Fresh Broccoli Folate Content](image)

Analysis Test Results

The results from the samples used to check Anresco Inc.’s nutrient analysis of broccoli are listed below. The pureed sample was expected to yield very different results than the fresh sample. The raw broccoli sample had a vitamin C value of 89.2 mg/100g, beta-
carotene value of 690 μg/100g, and folate value of 31.7 μg/100g, while the puréed sample had a vitamin C value of 2.2 mg/100g, beta-carotene value of 231 μg/100g, and folate value of 30.6 μg/100g. The vitamin C and beta-carotene were both notably affected by the processing. The puréeing and cold temperature were probably largely at fault for this loss of vitamin C. Vitamin C is known to be easily oxidized in air or in plant tissues by the enzyme ascorbic acid oxidase (115). Puréeing the sample would bring the broccoli into contact with oxygen as well as allow the ascorbic acid oxidase to come into contact with the vitamin and degrade it. Freezing can also cause cells to burst and allow ascorbic acid oxidase to destroy the ascorbic acid, especially since the sample was not blanched prior to freezing which would inactivate this enzyme. The beta-carotene was probably lost from light and the presence of large amounts of oxygen. Beta-carotene is susceptible to enzymatic and non-enzymatic oxidation and this loss is known to increase as the cellular structure is damaged (114). Puréeing is an obvious way to damage cells and thus induce the oxidation of beta-carotene. This experiment implied that there is a difference in the nutrient content of the raw broccoli and the processed broccoli for the two nutrients, vitamin C and beta-carotene, which according to the literature (114,115), are known to be vulnerable to this type of processing. Although these results imply Anresco Inc.’s methods are accurate, more samples would be needed to confirm this as well as analysis of a standard with a known value.

Refrigerated Storage Results

The broccoli refrigerated storage results are illustrated in Figures 4, 5, 6, and 7. Five out of the six samples exhibited a high retention of vitamin C with the samples varying from a retention of 62% - 121% and with an average retention of 92% (Figure 4). After
fourteen days of refrigerated storage 62% is high for retention of vitamin C, especially considering that after fourteen days of refrigerated storage it is roughly 24 to 28 days after harvest for broccoli transported to Hawaii. This is consistent with Albrecht who found high vitamin C retention in broccoli cultivars with an average range of 56 - 98% (103). Figure 4 illustrates how each sample changed in vitamin C content over the fourteen days of storage. Two samples even increased in their vitamin C content compared to their initial content. Some researchers have found an increase in vitamin C postharvest and then a linear decrease (4,105,108). As described earlier, most vegetables do not exhibit such high retention of vitamin C because it is especially vulnerable to degradation; however, broccoli and other cruciferous vegetables seem to retain more vitamin C than most other vegetables (103-105). Eheart and Odland attributed this difference in vitamin C retention to broccoli’s sulfhydryl groups (105). Albrecht found a high correlation between the total sulfur content of various cruciferous and non-cru-ciferous vegetables and the initial ascorbic acid content (103,104). Albrecht also found that the cruciferous vegetables analyzed had a higher initial ascorbic acid content and retained more vitamin C (103). This suggests that sulfur compounds may take part in initial ascorbic acid content as well as ascorbic acid retention although the method is unclear. In 1991, Albrecht tried to further clarify this effect of sulfhydryl groups but was unable to explain ascorbic acid retention mechanisms by the content of sulfhydryl groups alone (104). Favell has suggested that the differences seen in ascorbic acid retention from various vegetables may also be the result of differences in surface area, mechanical damage, endogenous enzyme activities, as well as sulfhydryl content (116). Broccoli
does seem to retain vitamin C more readily than other vegetables, but to explain this mechanism requires further investigation.

![Figure 4: Broccoli Vitamin C Content After Refrigeration](image)

The results of the beta-carotene analysis after the refrigerated storage of broccoli demonstrated no predictable pattern of beta-carotene retention (Figure 5). The average retention after fourteen days was rather high at 152%, with a range of 79% - 369%. Three out of the six samples exhibited a higher content of beta-carotene after the fourteen days of storage compared to their initial content. Other researchers have found that broccoli and other vegetables do retain a high quantity of beta-carotene (4,107,108). For example, Wu et al. found that the beta-carotene content did not significantly change in either green beans or broccoli after ten days of refrigerated storage (108). Some researchers have also found beta-carotene to increase initially postharvest and end up with higher beta-carotene content than the initial value (4,107). Howard et al. found this pattern of increased beta-carotene after three weeks of storage in carrots and one crop of
broccoli (1994); however, he found that in 1995 the broccoli exhibited a low retention of beta-carotene (36%) (4). He attributed this difference to adverse environmental conditions experienced by the broccoli grown in 1994. Beta-carotene is known to be quite stable in fruits and vegetables that are unprocessed and intact (114). Carotenoids are also known to continue to increase postharvest in many plants by biosynthesis as long as the plant is kept intact and protected so that the enzymes responsible for the biosynthesis are not inactivated. In some plants carotenoids can be degraded postharvest if the conditions promote it, such as high temperatures and exposure to significant amounts of light and oxygen. Broccoli demonstrated high retention of beta-carotene, thus it is a good contributor of vitamin A even after two weeks of refrigerated storage.

![Figure 5: Broccoli Beta-Carotene Content After Refrigeration](image)

The results of the folate analysis after the refrigerated storage of broccoli demonstrated a decrease in five out of the six samples initially tested after fourteen days.
The average retention after fourteen days was moderately high at 65%, with a range of 24% - 156%. 50% of the samples increased in folate content after seven days; however, all but one sample exhibited a decrease after the seven days of refrigeration. As a consequence of the unexplicable results, folate was reanalyzed in six more samples (Figure 7). The starting point of analysis was chosen as day 7 rather than day 3 because a larger difference is likely to be shown between day 7 and 14 as opposed to the difference between day 3 and day 7. In addition, the change from day 3 to day 7 had already been examined in the previous experiment. These results indicated a comparable average folate retention of 70%, with a range of 45% - 115%. There were two samples that ended up with higher retention than that observed initially, although the increase was only 15 or 14% versus the 56% increase in folate seen in the initial experiment. Overall, these results were similar to those in the initial experiment. These results are not consistent with other authors. For example, Mullin found that spinach and Swiss Chard stored at 4°C for fourteen days incurred no serious loss of total folacin (TFA) (111). While Chen found that spinach held at room temperature for ten hours did not affect TFA content (112). Chen has suggested that folacin degradation is predominantly due to oxidation and that the stability of folacin compounds can be enhanced by ascorbate or a nitrogen atmosphere. To confirm the results observed in both the initial and subsequent folate analysis a greater number of samples need to be tested as well as analyzing day 0 for a more accurate picture of the change in nutrient content over time. No absolute conclusions can be drawn from the folate analysis.
The results from the refrigerated storage portion of the experiment were not compared to the initial averages in the previous section because the broccoli was sampled
at different times and was from different batches. Furthermore, the samples used for the
initial averages were taken from broccoli immediately delivered to the supermarkets (day
0) versus broccoli sampled after 3, 7, or 14 days of refrigeration. The broccoli samples
for the storage portion were not analyzed for a day 0 marker. This was an error in
experimental design, and the refrigerated samples should have been divided into four
equal batches (day 0, 3, 7, and 14) rather than three (day 3, 7, and 14) to more efficiently
compare day 0 values with the day 0 values obtained in the initial averages portion of the
experiment. In addition, the initial averages and refrigerated storage portions of the
experiment were meant to each serve different purposes. The initial averages were meant
to serve as indicators of the nutrient content of raw broccoli from supermarkets in Hawaii
versus the standard USDA reference values, as well as to compare the variability in
nutrient content of one supermarket’s broccoli versus another. The refrigerated storage
portion of the experiment was meant to compare one sample of broccoli and its change in
nutrient content over time. Also, the means of the broccoli samples for day 3, 7, and 14
were not compared because these samples were all taken from different batches on
different days and therefore all have a unique starting value (day 0) that was not analyzed
so cannot be compared. This could have been rectified by having all of the samples for
the refrigerated storage portion of the experiment taken on the same day and from the
same batch of broccoli and dividing each sample into four equal batches (day 0, 3, 7, and
14). Then all of the samples would have a more accurate starting point and could be
compared to each other as well as the starting point compared to the initial averages
obtained in the first portion of the experiment. The samples would also each have an
additional point to better track the changes over time.
**Conclusion/Implications**

Overall, broccoli has shown to be a good source of vitamin C, beta-carotene, and folate, both initially after acquired from the supermarkets tested and after fourteen days of refrigerated storage. The average retention observed in broccoli after fourteen days was 92% for vitamin C, 152% for beta-carotene, and 68% for folate. The variability of these nutrients was also shown by the large differences in vitamin C, beta-carotene, and folate both between stores as well as when compared to the USDA reference value. The means of store A versus B were significantly different for both vitamin C and beta-carotene. The means were also significantly different from the USDA reference value for vitamin C in both store A (significantly higher) and store B (significantly lower), and beta-carotene in store B (significantly higher), as well as for folate in store A (significantly higher). However, after combining the samples from the two different locations, only beta-carotene and folate were significantly higher than the USDA value. This experiment has shown some interesting results; however, an improved experimental design is needed to draw absolute conclusions.

The nutrient content of raw fruits and vegetables, even among the same fruits or vegetables, will vary tremendously based upon several factors including but not limited to: genetics, growing conditions, light, temperature, seasons and climate, maturity and ripeness, and post-harvest handling. Furthermore, additional losses of nutrients will occur at different levels from home processing such as washing, cutting, and cooking. Consuming produce initially after purchase is one way to help ensure superior nutritional content than consuming produce after days of refrigerated storage. Moreover, it is up to
the consumer to select a variety of fruits and vegetables to satisfy his/her body’s nutritional requirements and include this as part of a balanced overall diet to ensure adequacy.
CHAPTER III
THE IMPACT OF NUTRITIONAL MESSAGES DISPLAYED AT THE POINT OF PURCHASE ON THE PURCHASING OF BROCCOLI IN A SUPERMARKET

Literature Review

Introduction

Since the 1970's, the supermarket has been increasingly sought after as a prime location to promote healthy eating through various programs (117). In fact, according to the Consumer Information Processing model, a theory used to explain rational and motivational ideas of consumers, the supermarket is the ultimate location to provide nutrition information at the point of purchase (118). Research has shown that consumers are receptive to nutrition information in the supermarket setting (119). Fruits and vegetables have been successfully promoted since the early 1990s in supermarkets across the nation through a partnership between a nonprofit consumer organization, the Produce for Better Health (PBH) Foundation, the National Cancer Institute (NCI) and the Department of Health and Human Services (DHHS) (119). Holben et al. found that consumers were more receptive to learning about phytochemicals in the supermarket environment rather than the classroom setting (120). Furthermore, consumers make roughly 80% of their purchase decisions at the point of purchase; (121,122) therefore, information presented at the point of purchase (POP) provides consumers with cues to help guide purchases. POP interventions try to impact consumers’ nutrition knowledge, attitudes, and/or behavior (123). Many consumers have favorable attitudes towards nutrition interventions in this setting, and would like to see more nutrition information in
supermarkets (123-125). Whether or not the programs are successful in achieving their goals, they have the capability of improving the images of the supermarkets involved and boosting consumer confidence in that particular store (126).

**Supermarkets as Nutrition Research Sites**

Table 8 reviews the literature of major intervention studies in supermarkets since 1989. Mayer reviewed studies in supermarkets prior to 1989 and found that four out of six studies reported no significant changes in the sales data of the targeted food groups (140). Glanz and Mullis reviewed 20 studies using point of choice interventions prior to 1988 but included not only studies in supermarkets, but also those in restaurants, cafeterias, and vending machines (141). Overall, they reported mixed results, with more impact on consumers’ knowledge and attitudes than on altering buying habits. Since 1989, there have been at least seventeen studies in supermarkets with about half of the studies reporting favorable outcomes (Table 8). Researchers have used different techniques for their methods of intervention, such as various POP activities (signs, displays, videotapes, interactive technology); advertising in newspapers and on the radio; nutrition experts present to answer consumer questions; and audio formats [in-store audio public service announcements (PSAs), take-home audiotapes]. Connell et al. used two different audio formats (in-store audio PSAs and a take-home audiotape) to promote messages regarding eating five fruits and vegetables per day (119). They found a significant increase in knowledge scores with the intervention shoppers (compared to baseline and to controls). Although the study was not designed to determine which audio format was more effective, they hypothesized that this result was attributable to the audiotapes. Narhinen et al. found that multiple methods of intervention were effective in boosting sales over
the short-term and different products were affected differently (134). The POP activities were effective at boosting sales of whatever products were being promoted at the time (whether or not they were healthful products). They felt that getting the media involved through newspaper advertisements, a newspaper article written about the study, and an interview by a local radio station really helped with consumers’ awareness of their intervention. Narhinén also emphasized that using supermarket sales data was an easy, cheap, and effective way of ascertaining the effects of interventions. Winett et al. found that a five- to seven-week intervention with an interactive information system was effective in increasing participants’ purchasing of high fiber items and decreasing high fat purchases; however, this effect was predominantly seen only during the intervention (139). This was the case for Narhinén, as well, who saw only short-term increases in sales of the promoted items (134). However, Anderson et al. found that their computer-based intervention was successful after four to six months (127). This showed that consumers are usually only affected by the interventions on a short-term basis.

**Purpose**

The ultimate aim of this pilot study was to increase consumers’ purchasing of broccoli in the produce section of a supermarket. There were two primary objectives for this study: 1) to determine whether or not the overall intervention was effective at influencing the consumer to purchase broccoli; and 2) to ascertain whether certain demographic variables correlated with the influence of nutritional messages on broccoli purchasing by supermarket shoppers.
<table>
<thead>
<tr>
<th>Reference</th>
<th>Population</th>
<th>Intervention</th>
<th>Outcome Measured</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anderson 2001 (127)</td>
<td>277 people from 1 supermarket</td>
<td>15 weeks of a computer based-intervention on nutrition behavior self-efficacy &amp; outcome expectations</td>
<td>Nutrition outcomes: intake of fat, fiber, &amp; fruits and vegetables (using FFQ &amp; receipts); &amp; social cognitive outcomes (using a survey)</td>
<td>1) Significant improvement in levels of fat, fiber, &amp; fruits and vegetables both after intervention &amp; at 4-6 month follow-up. 2) Higher levels of nutrition-related self-efficacy, physical outcome expectations &amp; social outcome expectations in intervention group.</td>
</tr>
<tr>
<td>Arsenault &amp; Glanville 1994 (123)</td>
<td>178 people from 1 supermarket in Canada</td>
<td>4 days of a display, brochures, an audio-visual presentation &amp; an R.D. present to promote Canada's guidelines for healthy eating</td>
<td>Program recognition &amp; customers' attitudes towards the program</td>
<td>1) Only 12% were aware of the intervention 2 weeks prior. 2) Attitudes were generally favorable towards the intervention.</td>
</tr>
<tr>
<td>Connell 2001 (119)</td>
<td>682 people, from 6 supermarkets in Eastern MA</td>
<td>2 audio formats: in-store audio PSAs (for 4 weeks) &amp; a take-home audiotape</td>
<td>Knowledge scores &amp; self-reported F/V intake</td>
<td>1) Knowledge scores increased significantly over baseline &amp; compared to controls. 2) Self-reported F/V intake increased significantly in both groups (significantly higher increase from baseline with intervention group.)</td>
</tr>
<tr>
<td>Cotugna &amp; Vickery 1992 (125)</td>
<td>1,050 people from 2 supermarkets</td>
<td>3 days (20 hours) of in-store videotapes with PSAs (5 1-minute PSAs) titled &quot;Learn to Eat Smart to Reduce Cancer Risk&quot;</td>
<td>Video viewing, video impact &amp; perception of stores' providing nutrition information</td>
<td>1) Only 26% of customers surveyed viewed the videotapes. 2) 43% of those who saw the videos said it was new information; 21% said they would change their eating habits as a result of viewing; and, 17% purchased foods recommended in tapes. 3) 40% of all surveyed would shop more in a supermarket with nutrition videos.</td>
</tr>
<tr>
<td>Study</td>
<td>Setting</td>
<td>Intervention Duration</td>
<td>Intervention Methods</td>
<td>Pre- vs. Post-Intervention Findings</td>
</tr>
<tr>
<td>------------------</td>
<td>--------------------------------------</td>
<td>------------------------</td>
<td>---------------------------------------------------------------------------------------</td>
<td>-------------------------------------------------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Dougherty 1990</td>
<td>3 supermarkets in New Jersey</td>
<td>6 month</td>
<td>Nutrition Lifeline, using: brochures at all 3 stores, videocassettes at 2 stores &amp; visits from an RD.</td>
<td>1) The degree of the intervention did not correlate with an increase in sales in applicable products. 2) Sales did not reveal any clear patterns of food-purchasing behavior change.</td>
</tr>
<tr>
<td>Dwivedi 1997</td>
<td>4 supermarkets in Ottawa-Carleton, Canada (206 people surveyed)</td>
<td>1 week</td>
<td>Using taste-test booths to promote a lower-fat cheese</td>
<td>1) Most (96%) of consumers who used the taste-test booth had a favorable opinion of it. 2) Sales of the low-fat cheese were significantly higher during the intervention as well as 1-2 weeks post vs. 1-2 weeks prior. 3) Most (87%) of consumers said they would choose other lower-fat foods.</td>
</tr>
<tr>
<td>Hunt 1990</td>
<td>3 supermarkets in Pawtucket, RI</td>
<td>Intervention from 1984-1988 to promote the Four Heart program using primarily shelf labels, signs, posters, brochures, recipe cards &amp; periodic promotions - contests, blood pressure and cholesterol screening and counseling and referral services.</td>
<td>Self-reported purchase behavior and awareness of labeled items.</td>
<td>1) Awareness of the shelf labels was significantly higher in 1988 vs. 1984; awareness was higher in females than males. 2) Shoppers reported more encouragement to purchase labeled foods in 1988 vs. 1984.</td>
</tr>
<tr>
<td>Kershner 1997</td>
<td>15 supermarkets in Anne Arundel County, MD</td>
<td>6 week campaigns using: newsletters, signs, recipes &amp; a weekly nutrition information booth staffed by an R.D. to promote community-based program for cancer risk reduction - &quot;Learn to Live&quot;</td>
<td>Program recognition, knowledge &amp; intended behavior change.</td>
<td>1) Recognition of the program logo averages 26% &amp; is highest in females (18 - 40 years). 2) &gt; 75% can recognize foods that may reduce cancer risk. 3) &gt; 20% who read program materials said they would make changes in shopping &amp; cooking.</td>
</tr>
<tr>
<td>Study</td>
<td>Setting</td>
<td>Intervention Duration</td>
<td>Intervention Details</td>
<td>Key Findings</td>
</tr>
<tr>
<td>---------------</td>
<td>----------------------------------</td>
<td>-----------------------</td>
<td>--------------------------------------------------------------------------------------</td>
<td>------------------------------------------------------------------------------</td>
</tr>
</tbody>
</table>
| Krista 1997   | 8 supermarkets in Iowa           | 8 month intervention  | Using: flyers, signs & food demonstrations to promote consumption of F/V             | 1) Significant difference in intervention shoppers' awareness of flyers vs. control.  
                                                                                       | 2) Insignificant increase in intervention shoppers' purchasing of F/V vs. control.  
                                                                                       | 3) No change in self-reported intake of F/V.                                    |
| Morris 1993   | 622 pre-test, 941 post-test from 20 supermarkets in Idaho | 3 months of in-store videos (4 60-90 s messages) on lower-fat cooking & brochures | Nutritional knowledge                                                        | 1) Knowledge increased significantly in consumers who watched 1 or more videos.  
                                                                                       | 2) The elderly, males, households with members under 18 and the less educated were less likely to participate in watching the nutritional videos. |
| Narhinen 2000 | 1 supermarket in Finland         | 11 weeks of various promotions - signs, newspaper advertisements/article, "heart week" activities, & an audiotape over the loudspeaker - to advertise low salt & low fat products | Direct & proportional sales of both single products & whole food groups during intervention & long-term follow-up | 1) Short-term increases in sales (37 - 49%) of actively promoted healthier products (not long-term).  
                                                                                       | 2) Proportional sales of some healthier products decreased significantly when reference products were actively promoted.  
                                                                                       | 3) Mean percentage salt content of weekly sales had declined in all food groups & mean percentage fat content had either declined or was unchanged.  
<pre><code>                                                                                   | 4) Supermarket environment was still affected by intervention at follow-up. |
</code></pre>
<table>
<thead>
<tr>
<th>Study</th>
<th>Setting</th>
<th>Intervention</th>
<th>Outcomes</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>O' Loughlin 1996 (135)</td>
<td>2 supermarkets &amp; 3 grocery stores in Montreal, Canada</td>
<td>4 month intervention to promote heart healthy foods using: displays, posters, bulletin boards, grocery bags w/logo, pamphlets, recipes, advertising - newspaper, community posters, &amp; special events - cholesterol screening, supermarket tours, lottery for cookbooks &amp; grocery certificates, &amp; taste testing.</td>
<td>Awareness of program &amp; use of program materials.</td>
<td>1) Awareness of the program was moderate (52% of those surveyed). 2) Self-reported use of the program was low (22% had taken or read material or participated in an activity).</td>
</tr>
<tr>
<td>Rodgers 1994 (117)</td>
<td>40 supermarkets (20 intervention &amp; 20 comparison matched controls) in Washington &amp; Baltimore</td>
<td>2 year intervention using - shelf labels, food guide booklets, produce signs &amp; monthly bulletins to promote &quot;Eat for Health&quot; for cancer risk reduction.</td>
<td>Changes in knowledge, attitudes, food purchasing behavior &amp; sales data on selected foods.</td>
<td>1) Not much change in knowledge, attitudes or food purchasing behavior; greatest positive change occurred in knowledge of the program and knowledge of a relationship between diet and cancer. 3) Limited success in changing food purchasing behaviors; largest effect seen in produce section (by sales data).</td>
</tr>
<tr>
<td>Scharf Silzer 1994 (136)</td>
<td>124 people in Ontario, Canada</td>
<td>2 hour Supermarket Safari tours to promote Canada's Guidelines for Healthy Eating.</td>
<td>Changes in food purchasing behaviors scores &amp; food preparation practices.</td>
<td>1) The tour group vs. comparison group had a significantly greater gain in skills/behaviors 1 month following the tours.</td>
</tr>
<tr>
<td>Scott 1991 (122)</td>
<td>1 supermarket in Australia</td>
<td>15 week intervention using materials displayed at POP, mass media campaign, taste testing &amp; cooking demonstrations to promote certain food groups</td>
<td>Awareness of program &amp; self-reported behavior change</td>
<td>1) Awareness of the program was high. 2) Some self-reported behavior change.</td>
</tr>
<tr>
<td>Study</td>
<td>Setting</td>
<td>Intervention details</td>
<td>Outcome</td>
<td>Conclusion</td>
</tr>
<tr>
<td>-----------------------------------</td>
<td>----------------------------------------------</td>
<td>--------------------------------------------------------------------------------------</td>
<td>-----------------------------------------------------------------------------------------------</td>
<td>---------------------------------------------------------------------------------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Shannon 1990</td>
<td>2 supermarkets in Bloomington, MN</td>
<td>Project used kiosks (unmanned) that contained &quot;The Shop Smart Game&quot; and related products had shelf labels to promote foods low in fat and sodium.</td>
<td>Awareness of the game, level of participation and self-reported purchasing changes.</td>
<td>1) Awareness of the game was high (68% of respondents). 2) Participation was low (22% of respondents). 3) Significant change in food purchasing for only 2/20 food categories.</td>
</tr>
<tr>
<td>Stenberg Nichols &amp; Schmidt 1995</td>
<td>20 grocery store in Idaho</td>
<td>3 month Intervention using 4 in-store videos to educate consumers about dietary fat and cholesterol.</td>
<td>Change in nutritional knowledge (using nutrition quiz scores).</td>
<td>Significant increase in nutrition knowledge in treatment groups vs. controls.</td>
</tr>
<tr>
<td>Winett 1991</td>
<td>49 people from 1 supermarket in VA</td>
<td>7 - 8 week intervention (5-7 wk baseline &amp; 5-8 wk follow-up) using interactive information system with instructional video programs, feedback on purchases, weekly programs &amp; ability to track user interactions &amp; intended purchases to promote lower fat &amp; higher fiber purchases.</td>
<td>Actual food purchases</td>
<td>1) Experimental participants compared to controls, significantly decreased high fat purchases &amp; increased high fiber purchases (but not significantly) during intervention, with some maintenance in follow-up.</td>
</tr>
</tbody>
</table>

a = food frequency questionnaire
b = fruits/vegetables
c = Registered Dietitian
Methods

Message Design

The three nutritional messages used in the intervention were:

1. It's a fact! Broccoli (1/2 cup) still contains most of your daily requirement for vitamin C, even after 2 weeks in your refrigerator.

2. It's a fact! Broccoli is rich in carotenoids. Carotenoids may reduce the risk of eye diseases, like cataracts and macular degeneration.

3. It's a fact! Broccoli is a good source of a B vitamin called folate. Folate can decrease the risk of heart disease and certain cancers.

The messages were each displayed individually on signs and were created using a consistent format with the same sign size, colors, background, and font. Balch et al. found through focus groups with consumers that longer messages about the “5 A Day” program were dismissed by consumers (1997). Consumers seemed to like the positive, short messages (142). Therefore, the messages were designed to be short and simple.

Each message emphasized the important concepts by enlarging certain words and using graphics for words that would not likely be misinterpreted, i.e. broccoli (see Appendix B, pp. 87-89).

Each message was chosen to pertain to vitamin C, carotenoids, or folate. The message concerning vitamin C contained two key pieces of information: 1) the content of vitamin C that broccoli can provide; and 2) information on refrigerated storage of broccoli and its effects on vitamin C. The former part of the message is from nutrient content data from the USDA database (113). The latter part of the message is based upon prior findings of this project (see Chapter II). The message regarding carotenoids
pertained to the role of carotenoids in degenerative eye diseases, specifically macular
degeneration and cataracts. This information is well established through past research
(16, 18, 23, 25, 38, 78). The message pertaining to folate centered on folate’s role in
preventing major chronic diseases. This information is also well established (16, 77, 78,
93-97), although the mechanisms have not yet been clearly elucidated (77).

Survey Design
The survey (see Appendix B, p. 90) was designed to capture information from consumers
in three different areas: 1) demographic information (sex, age range, ethnicity, education,
whether they are the primary shopper for their family/household, average money spent on
groceries per week, and how many people this amount feeds); 2) self-reported vegetable
intake, and specifically broccoli intake per week; and 3) the impact of the signs to see if
consumers noticed the signs, read the signs, and, if so, could they reiterate them, and
finally, to see if their decision to purchase broccoli was impacted by the signs. It is clear
from previous studies that consumers prefer brief interventions because of their busy
schedules (139). For this reason the survey instrument was limited to one page, which
only took approximately five minutes of the respondents’ time.

Intervention
A local supermarket in Honolulu, which is a part of a large supermarket chain, was
chosen as the pilot site. Permission was given to conduct the research at this location by
the supermarket’s district office. Notification and information were also given to the
produce manager and produce employees of this particular site regarding what exactly
would take place at this location. The produce manager was instructed whom to contact
if he had any questions regarding the study while the intervention was underway. The
intervention lasted a total of four weeks. The three different signs containing the nutritional messages were displayed above the fresh broccoli produce section. Each sign was displayed by itself for a week, specifically from Wednesday a.m. to the following Tuesday, because of the price change every Wednesday morning. An exception was made for sign 2, which was displayed for two weeks because the first weekend included a holiday when no surveying was conducted in an attempt to not skew the survey results. Each week the price of the broccoli was recorded in order to use a previous week with the same price to serve as the historical control when no intervention took place.

The survey period was done on three Saturdays and Sundays during the hours of 10 a.m. to 4:00 p.m. in the produce section. The survey was intended to be done on three consecutive Saturdays and Sundays; however, during the first week that sign 2 was displayed, no survey was performed on that weekend due to the Easter Sunday holiday. The inclusion criteria for those surveyed were consumers being 18 years of age or older and being seen purchasing raw broccoli. Those who met the criteria were approached and invited to participate in the survey. Those who agreed to participate were given the option to fill out the survey themselves or be administered the questionnaire orally. The participants were provided with a short information page regarding what information would be collected and whom they could contact if they have any questions about the study (see Appendix B, p. 91). The initial goal for the number of participants was n = 50 for each of the three signs (n_total = 150). However, during the first survey weekend this number was changed to 30 (n_total = 90) because of the scarcity of respondents. Although the survey was only two days out of the week, each sign was displayed for an entire
week. In addition, broccoli sales data were collected for each week, as well as for the historical control weeks with the same prices.

**Statistical Analysis**

Data were analyzed using a multiple regression with the statistical software program SPSS (ver. 10.0). Broccoli purchasing behavior was the dependent variable. Gender, age, ethnicity, education, amount spent on groceries, primary food shopper, vegetable consumption, broccoli consumption, and the nutritional message were the independent variables. A p-value of <0.05 was considered significant for all tests. Broccoli sales information was also collected from the supermarket’s sales data when the nutritional messages were displayed and compared to historical control periods of equal length when no messages were displayed.

**Approval**

This study was reviewed and approved by the University of Hawaii’s Committee on Human Studies in Honolulu, Hawaii.

**Results**

**Demographic Data**

The demographic characteristics of this sample (n = 90) are listed in Table 9. The majority of respondents were female (67%), and almost equally distributed between the age groups with 21% of the participants between the ages of 40-49, 19% between 30-39 years, and 18% between 50-59 years. The median age of the shoppers was between 30-59 years. Thirty eight percent of the sample identified themselves as Caucasian, followed
Table 9: Demographic Characteristics of Respondents

<table>
<thead>
<tr>
<th>Variables</th>
<th>$n_{sign 1}$</th>
<th>$n_{sign 2}$</th>
<th>$n_{sign 3}$</th>
<th>$n_{total}$</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>9</td>
<td>12</td>
<td>9</td>
<td>30</td>
<td>33%</td>
</tr>
<tr>
<td>Female</td>
<td>21</td>
<td>18</td>
<td>21</td>
<td>60</td>
<td>67%</td>
</tr>
<tr>
<td>Age</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>18-19</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>1%</td>
</tr>
<tr>
<td>20-29</td>
<td>1</td>
<td>6</td>
<td>5</td>
<td>12</td>
<td>13%</td>
</tr>
<tr>
<td>30-39</td>
<td>5</td>
<td>5</td>
<td>7</td>
<td>17</td>
<td>19%</td>
</tr>
<tr>
<td>40-49</td>
<td>9</td>
<td>7</td>
<td>3</td>
<td>19</td>
<td>21%</td>
</tr>
<tr>
<td>50-59</td>
<td>6</td>
<td>6</td>
<td>4</td>
<td>16</td>
<td>18%</td>
</tr>
<tr>
<td>60-69</td>
<td>4</td>
<td>2</td>
<td>4</td>
<td>10</td>
<td>11%</td>
</tr>
<tr>
<td>70-79</td>
<td>4</td>
<td>3</td>
<td>5</td>
<td>12</td>
<td>13%</td>
</tr>
<tr>
<td>80+</td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>3%</td>
</tr>
<tr>
<td>Ethnicity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caucasian</td>
<td>15</td>
<td>8</td>
<td>11</td>
<td>34</td>
<td>38%</td>
</tr>
<tr>
<td>Chinese</td>
<td>3</td>
<td>5</td>
<td>4</td>
<td>12</td>
<td>13%</td>
</tr>
<tr>
<td>Filipino</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>1%</td>
</tr>
<tr>
<td>Hawaiian/part Hawaiian</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>Japanese</td>
<td>7</td>
<td>7</td>
<td>7</td>
<td>21</td>
<td>23%</td>
</tr>
<tr>
<td>Korean</td>
<td>0</td>
<td>3</td>
<td>0</td>
<td>3</td>
<td>3%</td>
</tr>
<tr>
<td>Samoan/Polynesian</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>Southeast Asian</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>Spanish/Puerto Rican</td>
<td>0</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>4%</td>
</tr>
<tr>
<td>Other</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>2%</td>
</tr>
<tr>
<td>Mixed</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>12</td>
<td>13%</td>
</tr>
<tr>
<td>Education</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>≤ Junior High School</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>High School</td>
<td>5</td>
<td>4</td>
<td>1</td>
<td>10</td>
<td>11%</td>
</tr>
<tr>
<td>Junior College/some College</td>
<td>3</td>
<td>7</td>
<td>9</td>
<td>19</td>
<td>21%</td>
</tr>
<tr>
<td>Vocational Training</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>College</td>
<td>9</td>
<td>11</td>
<td>10</td>
<td>30</td>
<td>33%</td>
</tr>
<tr>
<td>≥ Graduate School</td>
<td>13</td>
<td>8</td>
<td>10</td>
<td>31</td>
<td>34%</td>
</tr>
<tr>
<td>Other</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>Primary Food Shopper</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>27</td>
<td>24</td>
<td>26</td>
<td>77</td>
<td>86%</td>
</tr>
<tr>
<td>No</td>
<td>3</td>
<td>6</td>
<td>4</td>
<td>13</td>
<td>14%</td>
</tr>
<tr>
<td>Dollars Spent per Week on Groceries</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt; $50</td>
<td>0</td>
<td>2</td>
<td>7</td>
<td>9</td>
<td>10%</td>
</tr>
<tr>
<td>$51-100</td>
<td>12</td>
<td>14</td>
<td>12</td>
<td>38</td>
<td>42%</td>
</tr>
<tr>
<td>$101-150</td>
<td>13</td>
<td>12</td>
<td>9</td>
<td>34</td>
<td>38%</td>
</tr>
<tr>
<td>$151-200</td>
<td>3</td>
<td>2</td>
<td>2</td>
<td>7</td>
<td>8%</td>
</tr>
<tr>
<td>&gt; $201</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>2%</td>
</tr>
<tr>
<td>Household Size</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>One</td>
<td>4</td>
<td>2</td>
<td>6</td>
<td>12</td>
<td>13%</td>
</tr>
<tr>
<td>Two</td>
<td>12</td>
<td>10</td>
<td>14</td>
<td>36</td>
<td>40%</td>
</tr>
<tr>
<td>Three</td>
<td>6</td>
<td>10</td>
<td>7</td>
<td>23</td>
<td>26%</td>
</tr>
<tr>
<td>Four or more</td>
<td>8</td>
<td>8</td>
<td>3</td>
<td>19</td>
<td>21%</td>
</tr>
</tbody>
</table>
by 23% Japanese, 13% Chinese, and 13% Mixed. This latter category had to be added when the results were tabulated because some respondents had chosen multiple ethnicity categories. This was not anticipated in the study design, but should have been, considering the mixed nature of the Hawaiian Islands’ population. This sample was highly educated with 34% completing graduate school, 33% completing a four-year degree in college, and 21% completing junior college or some college. The survey was done in an urban area surrounding the major research university in Hawaii that has the only medical and law schools in Hawaii; therefore, it was expected that this sample might be highly educated. Most (86%) of those surveyed indicated that they were the primary food shopper. Some respondents indicated that they shared the role of primary food shopper with their significant other, so in this situation their answer was changed to “no.”

The majority of respondents (42%) indicated that they spent an average of $51-$100 on groceries per week, followed by 38% who spent $101-$150 per week. This amount was most often for a household size of only two (42%), followed by a household size of three (36%), and four or more (21%). This latter category was collapsed into one (four or more). Originally respondents wrote in their response for this category and it ranged from a household size of four to eight. Some respondents may have misinterpreted this category as being only for produce rather than the total of grocery bills per week. One person indicated that his/her total was low because he/she bought items at Costco. Apparently, this person thought that the total spent on groceries per week was only for supermarket purchases.
Self-Reported Intake Results

Figure 8 illustrates the self-reported vegetable intake per week of those surveyed. Almost half (47%) of respondents indicated that they ate vegetables at 12-15 meals per week (about 2 meals containing vegetables per day), followed by 34% eating vegetables at 8-11 meals per week. Only 6% of this sample is eating vegetables at more than 2 meals per day.

Figure 9 illustrates the self-reported broccoli intake per week of the participants. The majority of respondents (37%) reported consuming broccoli only once per week, followed by 30% eating broccoli twice per week, 14% three times per week, 9% four times per week, 8% five or more times per week, and 2% reporting zero times per week.
**Intervention Results**

Only one-third (30/90) of those surveyed reported that they noticed the signs containing the nutritional messages. The breakdown for each sign was as follows: 12/30 persons (40%) noticed sign 1 (vitamin C sign), 7/30 (23%) sign 2 (carotenoid sign), and 11/30 (37%) for sign 3 (folate sign). Of the total respondents that noticed the signs only 14/30 (47%) actually read the signs [6/12 (50%) read sign 1, 2/7 (29%) read sign 2, and 6/11 (55%) read sign 3]. Also, some consumers were seen reading the signs but did not purchase broccoli and therefore did not qualify to be surveyed. Of the respondents who read the signs, 3/14 people (21%) had incorrect answers when they were asked to recall the messages (2 for sign 1 and 1 for sign 3). Overall, only 1/90 persons (1%) responded that the sign influenced their decision to purchase broccoli and that respondent was referring to sign 3.

The sales data from the intervention are listed in Table 10 and Figure 10. Each week's sales were compared to previous weeks with the same price. Sign 2 was actually
displayed for two weeks; however, the survey was conducted only during the second week. Each week shows an increase in sales versus the historical controls. The sales data from Sign 1 indicated that 143 pounds (19%) more broccoli was sold versus the control week, 117 (26%) more pounds sold in week 2a, 35 (10%) more pounds sold in week 2b and 337 (210%) more pounds of broccoli sold with sign 3. In both week 2b and week 3 broccoli had the same high price ($2.99/lb), yet week 3 seemed to show the largest increase in sales. The sales data point to the intervention being effective; however, the survey data did not support this conclusion.

<table>
<thead>
<tr>
<th>Dates</th>
<th>Price/lb. ($)</th>
<th>Total Sales ($)</th>
<th>Total Sold (lbs)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control 1</td>
<td>2/4/04 - 2/10/04</td>
<td>1.19</td>
<td>891</td>
</tr>
<tr>
<td>Sign 1</td>
<td>3/31/04 - 4/6/04</td>
<td>1.19</td>
<td>1,062</td>
</tr>
<tr>
<td>Control 2a</td>
<td>2/18/04 - 2/24/04</td>
<td>1.69</td>
<td>743</td>
</tr>
<tr>
<td>Sign 2</td>
<td>4/7/04 - 4/13/04</td>
<td>1.69</td>
<td>941</td>
</tr>
<tr>
<td>Control 2b</td>
<td>3/17/04 - 3/23/04</td>
<td>2.99</td>
<td>1007</td>
</tr>
<tr>
<td>Sign 2b</td>
<td>4/14/04 - 4/20/04</td>
<td>2.99</td>
<td>1112</td>
</tr>
<tr>
<td>Control 3</td>
<td>3/17/04 - 3/23/04</td>
<td>2.99</td>
<td>1007</td>
</tr>
<tr>
<td>Sign 3</td>
<td>4/21/04 - 4/27/04</td>
<td>2.99</td>
<td>2114</td>
</tr>
</tbody>
</table>

*a* = sign was up but no survey done
*b* = sign was up and survey done
Discussion

From the survey data collected it was determined that the majority of the respondents were female. It is common in supermarket interventions to have the majority of participants as females. For example, Connell et al. found that 81% of their participants in both the control and intervention groups were female (119). In addition, Morris et al. discovered that 91% of those who viewed their videotapes were females (133). The median age is also consistent with the results of Morris et al. and Connell et al. The ethnicity of this sample was not representative of the state of Hawaii. There were a greater number of Caucasians in this sample [38% versus 22.9% from state census data (143)], no native Hawaiians or other Pacific Islanders [0% versus 9.4% (143)], a lower number of persons reporting Mixed descent [13% versus 21.4% (143)] but a very similar total of persons reporting Asian descent [40% versus 41.6% (143)]. This sample was highly educated and this is in contrast to the results of Morris et al. who found the
majority (38%) of their participants had only finished only high school (133). Stenberg Nichols and Schmidt also found that 39% of their participants had finished only high school (138). It is common for supermarket study participants to indicate that they are the primary food shopper for their household. For example, Morris et al. reported that 90% of their participants were the primary food shopper (133). The amount reported on groceries per week was higher than the results given by Morris’. Morris et al. reported that 71% of the participants spent only $25-100 per week (133). This may be the result of a higher cost of living in Hawaii versus Idaho, and/or the result of inflation due to the ten-year difference between the two studies. With one person indicating an incorrect interpretation of the survey question requesting the amount spent on groceries per week, this question should have been more clearly written and contained examples for clarification.

The sample was not selected randomly because only those shoppers who purchased broccoli were chosen to be interviewed. Anytime the sample is not random, intrinsic biases may result (125). For example, by choosing only people who purchase broccoli, the sample may select for more health conscious people. This may lead to a sample that is not representative of the average population. Sampling bias may also result from choosing times and days to survey rather than randomly selecting the survey periods. People who shop on the weekend may not be representative of those who shop during the week. In summary, this sample consisted of more females than males, mostly Caucasians and Japanese, and highly educated persons.

The question requesting vegetable intake data could have been more clearly designed to decipher how many vegetables or servings of vegetables are eaten per day.
and for comparability to vegetable intake data reported in the literature. The USDA food guide pyramid currently recommends 3-5 servings of vegetables per day. This question was not designed to measure servings because of the presumed difficulty of respondents correctly estimating serving sizes. Therefore, it is difficult to compare the self-reported vegetable intake with recommended USDA servings because of the ambiguity of "meals vegetables eaten per day;" however, the observation can be made that only 6% of this sample is eating vegetables at more than 2 meals per day. Self-reported intake is another potential area for bias because respondents often over- or underestimate intake. The low self-reported broccoli intake data are consistent with the low cruciferous vegetable intake data from the CSFII survey done in 1994 – 1996 (5). These results could also be biased due to the self-reporting factor. Anytime data is collected regarding food intake respondents commonly over- or underestimate their responses (125).

A low awareness of the signs displayed at the POP was reported by respondents. Low awareness is commonly a problem for supermarket intervention studies. Most interventions have a low awareness by consumers. For example, Connell et al. reported that only 20% of those surveyed were aware of the intervention (119). In addition, Arsenault reported only 12% of shoppers were aware of the nutrition intervention just two weeks after the intervention (123). Furthermore, Kershner reported that on average only 26% of consumers were aware of the "Learn to Live" cancer prevention program logo and the program had been going on in the Maryland community for five years at the time of publication (131). The produce shoppers may have not noticed the signs because they were not placed at eye level. The signs were easily seen from a few feet away but as shoppers came closer to the broccoli shelves, the signs were then above their heads. This
may have contributed to the low awareness. With only one person out of ninety reporting that his/her broccoli purchasing was influenced by a sign, it is apparent that the intervention was not effective in influencing the purchasing of broccoli. Due to the ineffectiveness of the intervention, it is not relevant statistically to analyze which demographic variables were more influential in affecting broccoli purchasing.

The sales data seemed to indicate that some factors were influencing consumers’ purchasing of broccoli; however, the survey data did not support the conclusion that it was the signs causing this difference. There are many confounding variables that were not accounted for that could have influenced the purchasing of broccoli. For instance, the supermarkets advertising of broccoli through newspaper mailings was not recorded; therefore, there is no way of knowing if this could have influenced broccoli purchasing. Furthermore, there may have been some other vegetables like asparagus between which consumers rotate their broccoli purchasing. Therefore, the price of these other vegetables could have influenced the purchasing of broccoli. This sample was rather small \( n = 90 \) and surveys were collected only on the weekends. If more surveys were collected throughout the week it may have given a clearer picture as to what was influencing this purchasing behavior because shoppers during the week may not be representative of those shopping on the weekend. Also, the survey should have included a question that ascertained the primary factor influencing the consumer’s purchasing of broccoli. In addition, a larger sample of shoppers should have been obtained to confirm that the signs did not influence the purchasing behavior of consumers. Research shows that consumers do like nutrition information presented at the point of purchase (123-125); therefore, the signs could be influencing their purchases but those surveyed were not representative of
the entire week’s shoppers. Perhaps consumers during the week were fond of the unaided part of the intervention were information was presented but no survey was conducted. This could have caused an increase in sales during the week, but these consumers were not included in the sample surveyed.

It is interesting to note that in both week 2b and week 3 broccoli had the same high price ($2.99/lb), yet week 3 seemed to show the largest increase in sales. Therefore, this difference could not be due to price alone. This difference could be attributable to the sign’s content. The information contained in the sign was regarding cancer and heart disease. These diseases are very apparent in the general public and given an enormous amount of attention in the media. The public may have a sense of fear of these diseases, so saying that broccoli could reduce this risk could have had an impact on sales. In contrast, the information in sign 2 pertained to eye diseases like macular degeneration and cataracts. This information is only relevant to a smaller segment of the population and does not uniformly apply to the general population as cancer and heart disease do.

**Conclusion/Implications**

The demographics indicated that this sample was mostly female, between the ages of 30-59, of Caucasian, Japanese, and Chinese ethnicity, and highly educated. Although the sales data indicated that there was an increase in sales of broccoli during the intervention versus the historical control weeks, the survey data points to other factors besides the signs causing this increase. Overall, from the survey data, the intervention was not effective in influencing the purchasing of broccoli. Sampling bias, selection bias, and self-reporting bias (response bias) may have obscured the results.
Some of the components of this pilot study were effective while others were not. The supermarket environment seemed to be a favorable environment for nutrition research activities. Moreover, participants seemed to be receptive to the survey as well as to the signs displayed at the POP. However, in the future, the survey instruments and materials should be pilot tested to ensure understanding and readability by consumers. In addition, some questions on the survey were not effective in ascertaining the desired information. For instance, the broccoli and vegetable intake question should be redesigned to include servings with examples, and the question inquiring about the amount spent on groceries per week should be redesigned to better explain “groceries.” The fact that the survey was conducted on the weekends and only in a small sample of ninety (thirty for each sign) may have biased the data; therefore, in the future surveying should be conducted throughout the week and a larger number of participants should be obtained. The survey instrument did produce an extensive amount of data that would have been useful for statistical analysis; however, the intervention was clearly not effective in influencing broccoli purchasing so these variables were irrelevant.

Collecting the supermarket sales data was beneficial and had the additional bonus of being an easy, reliable, and noninvasive way to obtain data on sales; however, the confounding factors have to be considered as well. It may be advantageous to obtain sales data on other products as well to see if the same pattern in sales is viewed. Consumers may prefer to be left alone and simply presented with the information at the POP.

Although this pilot study was ineffective in influencing consumers’ purchasing of broccoli, other interventions in the supermarket environment may be influential.
Partnering with supermarkets can be a straightforward way to promote nutrition at the POP. Nutrition research in the supermarket can possibly be effective in impacting short-term purchasing behavior whether the research is performed in one supermarket or a part of a much larger state-wide campaign.
REFERENCES


72. Mirvish S. Experimental evidence for inhibition of N-nitroso compound formation as a factor in the negative correlation between vitamin C consumption and the incidence of certain cancers. *Cancer Res.* 1994;51:1918S-1951S.


98. Salunkhe DK, Bolin HR, Reddy NR. *Storage, Processing, and Nutritional*


107. Paradis C, Castaigne F, Desrosiers T, et al. Sensory, nutrient and chlorophyll...


1994;84(1):72-76.


126. Glanz K, Rudd J, Mullis RM, Snyder P. Point of choice nutrition information,


134. Narhinen M, Nissinen A, Puska P. Changes in supermarket sales during and after


83
### APPENDIX A

**TOP CONTRIBUTORS OF BETA-CAROTENE**

<table>
<thead>
<tr>
<th>Beta-Carotene Content of Fruits &amp; Vegetables</th>
<th>100 g (µg)</th>
<th>1 Cup (µg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>sweetpotato, cooked (baked, with skin)</td>
<td>11,509</td>
<td>23,018</td>
</tr>
<tr>
<td>carrots, cooked</td>
<td>8,332</td>
<td>12,998</td>
</tr>
<tr>
<td>kale, cooked</td>
<td>8,173</td>
<td>10,625</td>
</tr>
<tr>
<td>spinach, cooked</td>
<td>6,288</td>
<td>11,318</td>
</tr>
<tr>
<td>carrots, raw</td>
<td>5,774</td>
<td>6,351</td>
</tr>
<tr>
<td>spinach, raw</td>
<td>5,626</td>
<td>1,687</td>
</tr>
<tr>
<td>collards, cooked</td>
<td>4,814</td>
<td>9,147</td>
</tr>
<tr>
<td>lettuce, green leaf, raw</td>
<td>4,443</td>
<td>2,488</td>
</tr>
<tr>
<td>mustard greens, cooked</td>
<td>3,794</td>
<td>5,312</td>
</tr>
<tr>
<td>lettuce, cos or romaine, raw</td>
<td>3,484</td>
<td>1,952</td>
</tr>
<tr>
<td>squash, winter, all varieties, cooked</td>
<td>2,793</td>
<td>5,726</td>
</tr>
<tr>
<td>peppers, sweet, red, cooked</td>
<td>2,235</td>
<td>3,017</td>
</tr>
<tr>
<td>pumpkin, cooked</td>
<td>2,096</td>
<td>5,135</td>
</tr>
<tr>
<td>melons, cantaloupe, raw</td>
<td>2,020</td>
<td>3,232</td>
</tr>
<tr>
<td>lettuce, butterhead</td>
<td>1,987</td>
<td>1,093</td>
</tr>
<tr>
<td>peppers, sweet, red, raw</td>
<td>1,624</td>
<td>2,420</td>
</tr>
<tr>
<td>broccoli, cooked</td>
<td>1,180</td>
<td>1,840</td>
</tr>
<tr>
<td>peas, edible-podded, cooked</td>
<td>629</td>
<td>1,006</td>
</tr>
<tr>
<td>mangos, raw</td>
<td>445</td>
<td>734</td>
</tr>
<tr>
<td>watermelon, raw</td>
<td>303</td>
<td>467</td>
</tr>
</tbody>
</table>

Nutrient Data Laboratory Home Page, Available at:
http://www.nal.usda.gov/fnic/foodcomp. (113)

*Frozen or canned sources not included; cooked = boiled and drained without salt unless otherwise noted.
**Based on wet weight.
APPENDIX A
TOP CONTRIBUTORS OF VITAMIN C

Vitamin C Content of Fruits & Vegetables
(in order from most to least by 100 g)  100 g (mg)  1 Cup (mg)
peppers, sweet, red, raw  190  283
peppers, sweet, red, cooked  171  232
broccoli, raw  89  78
peppers, sweet, green, raw  80  120
peppers, sweet, green, cooked  74  101
broccoli, cooked  65  101
Brussels sprouts, cooked  62  97
papayas, raw  62  87
strawberries, raw  59  98
kohlrabi, cooked  54  89
oranges, raw, all commercial varieties  53  96
orange juice, raw  50  124/8 fl oz
peas, edible-podded, cooked  48  77
cauliflower, raw  46  46
grapefruit juice, white or pink, raw  38  94
melons, cantaloupe, raw  37  59
pineapple, raw, all varieties  36  56
cranberry juice cocktail, bottled  35  90/8 fl oz
mangos, raw  28  46
cabbage, chinese (pak-choi), cooked  26  44


*Frozen or canned sources not included; cooked = boiled and drained, without salt.
**Based on wet weight.
APPENDIX B - SIGN 1

It's a fact!

1/2 Cup contains most of your daily requirement for vitamin C, even after 2 weeks in your
It's a fact! Broccoli is rich in carotenoids.

Carotenoids may lower the risk of diseases, like cataracts & macular degeneration.
It's a fact!

is a good source of a

B vitamin called folate.

Folate can ↓ the risk of heart disease & certain cancers.
APPENDIX B – CONSUMER SURVEY

1. At how many meals did you eat **vegetables** last week?
   
   
   0 – 3  4 – 7  8 – 11  12 – 15  16 – 19  20 – 21

2. Approximately, at how many meals did you eat **broccoli** last week?
   
   
   0   1   2   3   4   5+

3. Did you notice the nutrition message by the broccoli display?  ____ Yes  ____ No
   (If your answer is “no” then STOP and skip to question # 7.)

4. Did you read it?  ____ Yes  ____ No
   (If your answer is “yes” then go to # 5. If your answer is “no” then go to # 6.)

5. What did the message say? ____________________________________________
   ____________________________________________
   ____________________________________________
   ____________________________________________

6. Did it affect your decision to purchase broccoli today?  ____ Yes  ____ No

7. Are you:  ____ Male  or  ____ Female

8. Please circle your age range?
   
   18-19  20-29  30-39  40-49  50-59  60-69  70-79  80+

9. What is your ethnicity?
   
   ____ Caucasian  ____ Korean
   ____ Chinese  ____ Samoan/Polynesian
   ____ Filipino  ____ Southeast Asian
   ____ Hawaiian/part Hawaiian  ____ Spanish/Puerto Rican
   ____ Japanese  ____ Other:  _____________________________

10. What is the highest level of education you have completed?
    
    ____ Grade school  ____ Vocational training  ____ Other:  __________
    ____ Junior high school/middle school  ____ College (4 year)
    ____ High school  ____ Graduate school or professional school

11. Are you the primary food shopper for your family/household?  ____ Yes  ____ No

12. What is the average amount of money you spend on groceries per week for your
    family/household?
    $50 or less  $51 - $100  $101 - $150  $151-$200  $201 or more

13. How many people are in your family/household?  __________

*Thank you for participating!*
Dear participant,

I am a graduate student at the University of Hawaii in the Nutrition department and doing a project that involves nutrition education in the supermarket. The purpose of this project is to see if various nutritional messages displayed next to the broccoli will affect shoppers' decisions to purchase broccoli. The information collected will be used to help nutritionists design materials that will promote the consumption of fruits and vegetables.

All of the information that I have collected from you is completely anonymous and you will not be identifiable in any way. The information will only be used by my supervising professors and myself.

If you have any questions about participating in this study, please contact me, Ana Valiere, at 778-9736, or my supervising professors – Dr. Dian Dooley at 956-7021 and Dr. Alvin Huang at 956-3840. If you cannot obtain satisfactory answers to your questions or have comments or complaints about your treatment in this study, contact: Committee on Human Studies, University of Hawai‘i, 2540 Maile Way, Honolulu, Hawai‘i 96822; phone (808) 956-5007.

Thank you for participating!