AN OVERVIEW OF THE STATUS OF THE SCIENCE ON DRY BEANS AND HUMAN HEALTH

The nutritional values of dry beans are many, but the relationship between consumption of dry beans and health outcomes is one that has been taken for granted for too long! The last several years have seen a resurgence of interest in unraveling those many possible health paths and a growing body of evidence is pointing to the remarkable value of beans to the maintenance, if not potential for improvement, of human wellness.

The Northarvest Bean Growers Association recently commissioned a review of the scientific literature on beans and human health by well-known nutrition researcher Dr. Maurice Bennink (http://fshn.msu.edu/directory/facultyBennink.html) of Michigan State University. Dr. Bennink and colleague Dr. Elizabeth Rondini have produced Beans and Health: A Comprehensive Review © including literature on the relationship between dry beans and health, examining studies available through early 2008. You may download a copy for your use here:

However, to assist you in "zeroing-in" on one or more areas of greatest interest to you, we have also "chunked" the report into smaller focal sections, including:

- •Bioactive Compounds in Dry Beans
- •The Relationship of Chronic Diseases to Glycemic Index and Glycemic Load
    - o Blood Glucose
    - o Insulin
    - o Body Weight
    - o Beans, Glycemic Index, and Glycemic Load
- •Low Glycemic Carbohydrates and Diabetes
- •Bean Consumption and Cardiovascular Disease (CVD)
- •Bean Consumption and Cancer
    - o Colorectal Cancer
        - ☐ Epidemiological Studies
        - ☐ Experimental Studies
    - o Breast Cancer
    - o Prostate Cancer
- •Additional Data Relating Bans to Cancer prevention
    - o Fiber and Cancer
    - o Folate and Cancer

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A Study Commissioned by the

Northarvest Bean Growers Association

Frazee, MN

INTRODUCTION

Dry beans are an economical source of concentrated vegetable protein, and are also an excellent source of fiber (both soluble and insoluble), and an excellent source of several minerals and vitamins. Despite their nutritional value, bean consumption by the average adult in the United States remains low, especially as compared with nutritional value. Numerous studies indicate that incorporating beans into the diet could aid in the prevention and/or management of chronic diseases such as diabetes, obesity, and cancer.

The leading causes of death in the USA are: (1) heart disease, (2) cancer, (3) stroke, (4) chronic lower respiratory diseases, (5) accidents, and (6) diabetes (1). It is widely accepted that environmental (i.e., non-genetic) factors, including inappropriate food choices, are major causative factors in the development of chronic diseases. Dietary guidelines to prevent and/or manage chronic diseases are made independently by professional and/or governmental agencies at national, regional, and international levels. The similarities and harmonious nature of the recommendations are quite amazing. Almost all agencies emphasize the importance of consuming legumes (pulses). Eating more beans could potentially reduce the number of premature deaths due to 4 of the top 6 leading causes of death in the USA – heart disease, cancers at certain locations in the body, stroke, and diabetes.

The purpose of this monograph is to encapsulate the recent literature on dry beans and human health. The authors begin by providing an overview of the nutrients found in dry beans, and then also address the range of bio-active compounds they are known to contain. Then, the relationship between dry bean consumption and health is reviewed, including associations with longevity, the links between chronic disease and glycemic index/load, with special attention to diabetes.

Studies of bean consumption and cardiovascular disease, as well as the association between bean use and cancer are next examined. The authors complete their review by briefly discussing research on fiber and cancer, and folate and cancer.

NUTRIENT PROFILE OF DRY BEANS

Nutrient density

Dry beans are a nutrient-dense food. Nutrient density is defined here as the amount of a nutrient in beans divided by nutrient requirement per unit of energy (i.e., 2,000 kcal). In Table 1 the right column shows the nutrient density of beans for adults. As seen in Table 1, beans are an excellent source of protein. Typically beans contain 20 - 30% protein on a dry weight basis which is greater than most plant foods. Bean protein, similar to most plant protein sources, are incomplete (i.e., one or more of the essential amino acids is(are) present in less than optimal amounts). Methionine and tryptophan are the limiting amino acids for bean protein. Consuming beans with cereal grains in proper proportions improves the nutritional value of the protein consumed and most healthy adults can consume all the amino acids they need from such a mixture. Amino acid composition is of little consequence for most North Americans unless they do not eat meat or drink milk

Dry beans are about 70% carbohydrate. Starch (43 - 45%), non-starch polysaccharides or fiber (18 - 20%), α-galactosides (starchyose, verbascose, and raffinose; 3 - 5%), and sucrose (3 - 5%) are the major types of carbohydrate. Beans are an excellent source of fiber as can be seen in Table 1. Bean fiber is roughly one-third soluble and two-thirds insoluble. Since North Americans consume less than one-half of the recommended amount of fiber, adding one serving of beans per day would increase fiber intake by 6g which is 20 - 25% of the recommended intake.

Beans are naturally low in fat with little saturated fat. The neutral lipid content is only 1.5% of the dry bean and unsaturated fatty acids make up 75% of the lipid material. As with all plant foods, beans do not

contain any cholesterol.

Beans are a significant dietary source of several essential minerals. Beans are particularly rich in magnesium, phosphorus, copper, and manganese (Table 1). Beans are a good source of potassium, a fair source of calcium and selenium, and are naturally low in sodium. Although beans contain significant amounts of iron and zinc, they do not provide as much iron and zinc as it appears in Table 1 due to low bioavailability (addressed later).

Dry beans are an excellent source of the water-soluble vitamins thiamin and folate. Adequate intakes of folate are associated with a lower risk of cardiovascular disease in some individuals (2) and with a lower risk of cancer at some sites in the body (3). One serving (½ cup) of cooked beans provides approximately 20% of the recommended daily intake (RDI) for folate (400 µg, Table 1). Beans are also a good source of riboflavin and vitamin B6.

Digestibility considerations

Both bean protein and starch are less well digested than protein and starch from cereal grains. One reason for poorer digestion is due to the physical form of the protein and starch when it enters the stomach and small intestine. Beans are generally consumed as cooked or canned beans as opposed to grinding and heat processing that occurs with cereal grains. Bean protein and starch are contained within cell walls that remain primarily intact during cooking and canning. Chewing and mixing in the stomach breaks open only a few of the cells. As a result, bean protein and starch enter the small intestine encased within fibrous cell walls. This impedes proteolysis and amylolysis. We have noted that when beans are cooked in boiling water longer than 10 min, crystalline material starts to form in the cell walls. We feel that the crystalline structures further impede the digestive process.

The digestibility of bean protein is rather low compared to animal protein and most cereal grains. True protein digestibility averages about 73%, but

digestibilities range from 65 - 85% depending upon seed coat color. Digestibility of protein from white and light colored beans is in the 80 - 85% range while protein digestibility for black, dark brown, and dark red varieties ranges from 65 - 75%. In addition to the colored seed coat pigments, some fractions of bean protein are inherently less digestible. About 61% of bean protein is in the albumin and globulin GI fractions, which are highly digestible. The globulin GII fraction (lectin fraction) comprises 10% of the bean protein and is only 60% digestible. Glutelins (18% of the protein) and protease inhibitors (0.3% of the protein) are only 40% digestible. The digestibility coefficient for the non-extractable proteins found primarily in the seed coat and cell walls (11%,) is not known but it is likely to be less than 50% based on the digestibility of protein associated with a variety of similar fiber sources. Less than optimal digestion of bean protein may be important in some countries, but it is probably of little concern for North Americans that tend to eat 1.5 - 2.5 times more protein than needed.

Bean starch is digested much slower (a positive feature) and less completely (both a negative and positive feature) than cereal starch. The slow rate of starch digestion contributes to the low glycemic index noted for beans and is a compelling reason to consume beans. The importance of low glycemic foods is discussed in a later section. Incomplete digestion of bean starch contributes to the flatulence problems associated with eating beans but it may also help protect against colon cancer.

As bean starch enters the small intestine, it is entrapped within cell walls, which partially explains the slower starch digestion. Other important aspects affecting starch digestion include extent of gelatinization and retrogradation of starch. Cooking beans in an open kettle does not provide sufficient thermal energy to cause complete gelatinization of bean starch. Canning, because of the higher temperature attained, allows nearly complete gelatinization of the starch. As would be expected, the glycemic index of canned beans is greater than

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Table 1: Nutrient compostion of dry beans

Nutrient	Unit	1 serving§ 1/2 cup	100g (DW)	Adult male DRI (RDA/AI)†	% of DRI	Adult Female DRI (RDA/AI)†	% of DRI	Nutrient‡ Density
Proximates								
Water	g	59.8						
Energy	kcal	110.3	308					
Energy	kj	461.8	1289.3					
Protein	g	7.5	20.8	56	13%	46	16%	7%
Total lipid (fat)	g	0.5	1.3					
Ash	g	1.1	3					
Carbohydrate, by difference	g	20.4	56.9	130	16%	130	16%	18%
Fiber, total dietary	g	6.2	17.3	38	16%	25	25%	6%
Sugars, total	g	0.3	0.8					
Lipids								
Fatty acids, total saturated	g	0.08	0.22					
16:00	g	0.07	0.2					
18:00	g	0.01	0.02					
Fatty acids, total monounsaturated	g	0.03	0.09					
18:01	g	0.03	0.09					
Fatty acids, total polyunsaturated	g	0.24	0.68					
18:02	g	0.1	0.28					
18:03	g	0.14	0.4					
Cholesterol	mg	0	0					
Minerals								
Calcium, Ca	mg	37.86	105.72	1000	4%	1000	4%	34%
Iron, Fe	mg	2.23	6.21	8	28%	18	12%	2%
Magnesium, Mg	mg	52.09	145.44	420	12%	320	16%	47%
Phosphorus, P	mg	118.94	332.12	700	17%	700	17%	108%
Potassium, K	mg	385.63	1076.8	4700	8%	4700	8%	350%
Sodium, Na	mg	15.64	43.67	1500	1%	1500	1%	14%
Zinc, Zn	mg	0.8	2.23	11	7%	8	10%	1%
Copper, Cu	mg	0.22	0.6	0.9	24%	0.9	24%	0%
Manganese, Mn	mg	0.4	1.11	2.3	17%	1.8	22%	0%
Selenium, Se	mcg	1.42	3.96	55	3%	55	3%	1%
Vitamins								
Vitamin C, total ascorbic acid	mg	3.2	8.93	90	4%	75	4%	3%
Thiamin	mg	0.2	0.56	1.2	17%	1.1	18%	0%
Riboflavin	mg	0.09	0.24	1.3	7%	1.1	8%	0%
Niacin	mg	0.63	1.76	16	4%	14	5%	1%
Pantothenic acid	mg	0.31	0.86	5	6%	5	6%	0%
Vitamin B-6	mg	0.12	0.34	1.3	9%	1.3	9%	0%
Folate, total	mcg	87.32	243.81	400	22%	400	22%	79%
Choline, total	mg	21.04	58.74	550	4%	425	5%	19%
Vitamin B-12	mcg	0	0	2.4	0%	2.4	0%	0%
Vitamin A, RAE	RAE	0.076	0.21	900	0%	700	0%	..
Vitamin E (alpha-tocopherol)	mg	0.29	0.8	15	2%	15	2%	0%
Vitamin K (phylloquinone)	mcg	5.98	16.71	120	5%	90	7%	5%

§Average of black, navy, pinto, kidney, great northern, and red beans. (data from www.nal.usda.gov/fnic/foodcomp/search). †DRI, Daily reference intake for adult males and females (31-50 years). ‡Expressed as amount of nutrient (g, mg, or mcg) per kcal in one serving (0.5 cup).

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the glycemic index of cooked beans. Retrograded starch is not digested within the small intestine and does not raise blood glucose. There are several factors that affect extent of starch retrogradation, but amylose content and degree of polymerization within amylose are important factors. Bean starch contains more amylose than most other sources of starch and the average degree of polymerization is 1000 – 1200, which is highly conducive to forming retrograded starch. We have utilized in vitro methods to estimate resistant starch (non-gelatinized and retrograded) in canned beans from 30 bean lines and we estimate that about 10% of bean starch in canned beans is not digestible. We predict that the amount of indigestible starch in cooked beans is 20% of the total starch content. Additional research on bean starch is warranted.

The α-galactosides (oligosaccharides) are not digested in the upper part of the small intestine due to a lack of the enzyme, α-galactosidase. The α-galactosides, resistant starch (starch not digested and absorbed in the small intestine), and fiber pass into the colon where they are fermented by colonic bacteria to produce short chain fatty acids, carbon dioxide, hydrogen, and in some individuals, methane. Soaking beans and then discarding the water reduces oligosaccharide content and may help reduce some of the abdominal discomfort associated with bean consumption. While oligosaccharides receive the blame for causing flatulence, resistant starch and fiber contribute more fermentable material (and thus more gas) than oligosaccharides. Presumably, it is the rapid fermentation of the oligosaccharides that is more bothersome to people than the slower, more consistent fermentation of the insoluble fiber. It is not known how much of the initial, rapid increase in flatulence is due to fermentation of resistant starch and soluble fiber.

Poor bioavailability of iron and zinc from plant foods is due to the presence of phytate, fiber, and phenolic compounds. Poor absorption of iron and zinc may be of little concern for North Americans (except for vegetarians), but is of particular concern in developing countries where cereal grain products and dry beans are consumed in large quantities and consumption of animal products is limited. Improving the amount of iron and zinc absorbed from beans is a major effort by "HarvestPlus", CIAT, and several Agricultural Experiment Stations.

NON-NUTRITIVE BIOACTIVE COMPOUNDS

Legumes contain a number of compounds that have potential health benefits as well as some that can reduce the bioavailability of nutrients. These compounds include saponins, phytic acid, plant sterols, phenolic compounds, enzyme inhibitors, and lectins. Much interest has been generated in examining some of these compounds with respect to chronic disease prevention.

Saponins

Saponins are amphiphilic compounds present in a wide variety of plants and herbs. Structurally, saponins in food exist as glycosides, with a hydrophobic triterpenoid or steroid (sapogenin) group linked to water-soluble sugar residues (4). The main types of steroid aglycones include the spirostan, furostan, and nautigenin derivatives whereas oleanan derivatives comprise the more common triterpenoid aglycones (5). The amount and type of sugar residues vary between saponin species, the most common being glucose, glucuronic acid, arabinose, rhamnose, xylose, and fucose attached at either the C-3 position (monodesmoside saponins) or on both the C-3 and C-22 position (bidesmoside saponins) (5). The major saponins present in Phaseolous vulgarus were identified as soyasaponin I, V, and phaseoleamide (6, 7).

Saponins are surfactants, and were initially thought to be harmful due to their strong hemolytic activity in vitro. Gestetner et al. (8) found that after feeding mice, rats, and chicks a 20% soy flour diet, neither saponins nor sapogenins were detectable in blood. Saponins were the major form present in the small intestine and sapogenins were primarily detected in the cecum and colon after hydrolysis by

microflora. Since the saponins found in dry beans are the same triterpenoid type of saponins found in soy, it is unlikely that dry bean saponins would be absorbed.

Saponins have been shown to have anticarcinogenic and antimutagenic properties in a variety of in vitro approaches (cell culture). The saponins used in these studies were from soy beans. Since dry bean saponins are similar to soy saponins, it would be expected that dry bean saponins would produce similar results. Soyasaponins reduced the growth of HCT-15 and HT-29 colon carcinoma cells and also significantly decreased TPA-associated protein kinase C activation. Because saponegins are presumably the major form of saponins present in the colon, Gurfinkel and Rao (9) looked at the effect of the chemical structure of soyasaponins on anticarcinogenic activity. Soyasaponins (I, II, III) were found to be ineffective up to 50 ppm in inhibiting cell growth, whereas soyasapogenols A and B (aglycones) effectively suppressed growth in a dose-dependent manner (6-50 ppm). These levels could easily be achieved after consumption of 10 g soy flour. Complete conversion of soyasaponins to their aglycone forms by microflora would produce approximately 25 ppm soyasapogenol A and 75 ppm soyasapogenol B (10). Only one study with saponins on carcinogenesis has been conducted in vivo. Koratkar and Rao (11) found that incorporation of soyasaponins into the diet of mice (3%) reduced the incidence of mice with ACF, and significantly decreased the number of ACF/colon and the number of AC/foci.

We did not find any research that dealt directly with dry bean saponins and health. A number of publications extol the health benefits of the steroid type of saponins. Steriod saponins are absorbable and apparently elicit numerous biological responses following systemic distribution of the saponin. However, the triterpenoid saponins are not absorbed and presumably provide benefits only in the intestine.

Phytic Acid

Phytic acid (IP6, myo-inositol hexaphosphate) is the main storage form of phosphorous in dry beans (12, 13). Different forms of phytic acid exist depending on the pH and metal ions present - phytate is the calcium salt and phytin is the calcium-magnesium salt (13). The amount of phytic acid in legumes is between 0.4% to 2.06% but can vary with genetics, environmental conditions, type of soil, and fertilizer (12-14). Consumption of foods high in phytate, mainly IP4-IP6 derivatives are known to influence zinc, calcium, and iron bioavailability by forming insoluble mineral-phytate complexes in the intestine. These effects would likely be of most concern for vegetarians and in developing countries where cereal and grain products are consumed in large quantities (15).

The suggestion that phytic acid and/or its derivatives have anti-cancer properties was first derived from epidemiological data in two Scandinavian populations (16). The incidence of colon cancer is much lower in Finland (11.9%) than in Denmark (17.9%) despite similar dietary fiber intakes (16). However, upon further analysis, the Finnish consume 20-40% more phytate than Danish populations, owing to a greater intake of rye and wheat bran products. This led to the proposal that phytic acid contributed to the lower colon cancer incidence in this group. Several experimental studies have now demonstrated that phytic acid (IP6) inhibits colon cancer development (16-19). The effect of phytic acid on colon cancer was found to be dose-dependent and adding inositol improved IP6 efficacy. IP6 is most effective if provided after carcinogen administration (i.e., during cancer promotion). IP6 was effective even when given 5 months after carcinogen administration. Phytic acid has also been demonstrated to inhibit cancer at other sites including mammary, lung, liver, skin and prostate (20-26).

The mechanisms through which IP6 and inositol derivatives inhibit tumorigenesis have not been

entirely defined. Vucenik and Shamsuddin (27) suggested that IP6 can be internalized and inhibit tumor growth by affecting cellular signaling through lower inositol derivatives. In support of this, there is some evidence that IP6 interacts with the Akt-NFκB cell survival pathway, by reducing insulin and TNF-induced Akt translocation to the cell membrane (28). Phytic acid also appears to influence the cell cycle. Treatment of cells with IP6 has been shown to cause G1 cell cycle arrest and a reduction of cells in S phase in breast (29), colon (29), and prostate cancer cell lines (30). Additionally, IP6 and inositol derivatives containing at least 3 phosphates (IP5, IP4, and IP3) can bind divalent metals, reducing iron catalyzed lipid peroxidation and high iron-induced promotion of colon tumorigenesis in rats.

Plant sterols

β-sitosterol, campesterol, and stigmasterol are the most common types of phytosterols found in food, including beans, and are structurally similar to cholesterol. The absorption of phytosterols by humans is low relative to that of cholesterol (20-50%) with only about 5% of ingested phytosterols being absorbed and the remaining 95% excreted from the colon (31). Phytosterols have been documented to decrease plasma cholesterol in humans (32) and animals (33, 34). The cholesterol reducing activity of bean sterols should help reduce cardiovascular diseases. The cholesterol lowering effect is likely due to a reduction in cholesterol solubilization into bile salt micelles resulting in a reduction in cholesterol absorption (34).

The relatively low absorption of ingested phytosterols from the intestine suggests that they can potentially affect colon carcinogenesis either directly or indirectly. Only a few animal studies, however, have been conducted to date. Phytosterols have been shown to reduce the rate of colonic epithelial proliferation and the proliferation zone in animals either induced with a carcinogen or administered 0.1-0.2% cholic acid. Addition of phytosterols (0.2-0.3%) to the diet also caused a reduction in both

preneoplastic colon lesions and colon tumorigenesis in rodents (35). Lastly, although phytosterols were not examined specifically in this study, wheat bran oil (2%) decreased colon tumor incidence, multiplicity, and tumor size and reduced tumor expression of iNOS and COX-2 in rats injected with azoxymethane (AOM) (36). The mechanisms of chemoprevention by phytosterols have been suggested to include (a) alterations in membrane phospholipid composition (37), (b) decreased formation of secondary bile acids (38), and (c) an increase in apoptosis.

Phenolic Compounds

Over the past 15 years there has been an exponential increase in publications related to plant phenolics. The great interest in this class of non-nutritive compounds is largely due to: 1) epidemiologic studies showing that plant based diets lead to less chronic diseases; 2) fruits, vegetables, and whole grains are rich in phenolic compounds; 3) many of the phenolic compounds in fruits, vegetables, and whole grains are excellent antioxidants in vitro; and 4) researchers began to suggest oxidative stress was a strong contributing factor in the development of cancer, cardiovascular diseases, and neurodegenerative diseases.

Relatively little is known about the phenolic compounds in dry beans compared to fruit, vegetables, chocolate, wine, and tea. More recently, several groups (39-41) have identified phenolic compounds in dry beans. Anthocyanins are present in black and blue-violet colored beans (39-41). A black colored Italian bean contained 170 mg of anthocyanins/kg of flour which is equivalent to 6.5 mg/serving (41). Beninger et al. (42) reported the only flavonol present in 2 lines of pinto beans was glycosides of kaempferol. The amount of kaempferol that would be consumed in one serving was 26.6 mg and 64 mg for the 2 lines. Romani et al. (40) reported the only significant flavonol in yellow, brown, and black colored beans was kaempferol glycosides, although a trace of quercetin

was detected in the brown and black variety. The amounts of kaempferol glycosides that would be consumed per serving were 23.5 and 20 mg for the 2 yellow varieties, 25.2 mg for the brown variety and 4.2 mg for the black bean. Aparicio-Fernandez et al. (41) identified glycosides of kaempferol, quercetin, and myriceten in a black bean as well as oligomers containing (epi)catechin, (epi)afzelechin and (epi)gallocatechin in the proanthocyanidin fraction (i.e., condensed tannins). No quantitative data was available from this report, but estimates based on other published studies indicate that one serving of beans would provide 300 - 1300 mg of proanthocyanidins depending on seed coat color, storage time, etc. Luthria and Pastor-Corrales (43) identified ferulic acid, p-coumaric acid, and sinapic acid in 15 varieties of raw dry beans. Caffeic acid was found only in 2 black bean varieties. The average phenolic acid that would be consumed was 11.1 mg/serving with a range of 6.8 to 17.2 mg/serving. For comparative purposes, dark blue-black berries and grapes provide 100 - 1500 mg of anthycyanins/serving. One serving of fruit provides 10 - 200 mg of phenolic acids and the amount of procyanidins from beans is equal or greater than that per serving of chocolate or green tea (44).

The studies reported above for phenolics all used raw beans which is not what we eat. Therefore, we cooked navy beans, black beans, pinto beans and small red beans and extracted the phenolic compounds. We identified protocatechuric acid, p-hydroxybenzoic acid, (+)-catechin, vanillic acid, caffeic acid, syringic acid, p-coumaric acid, ferulic acid, sinapic acid and isovanillic acid in all 4 types of beans. Ferulic acid was the predominant phenolic compound present in all beans. The flavonol, quercetin was detected in the black and red beans, while kaempherol was identified in pinto and red beans. We have not attempted to identify anthocyanins that have been identified in raw black beans and we do not have quantitative data for phenolic content at this time.

In general, polyphenolics are poorly absorbed. Of the phenolic compounds in beans, the phenolic acids would be absorbed the best and maximum plasma concentrations should occur one to three hours after beans are consumed (based on data reviewed by Manach (45)). One would not expect plasma concentrations to ever be greater than 0.5 umol/l and clearance from the blood is rapid (half-life for clearance in urine is < 2 hours). We can predict the amount of quercetin and kaempherol that would absorbed would be 1 – 2% of what is in beans (45). Blood concentrations would not exceed 0.5 umol/l. Even though the blood concentrations would be low, the clearance of quercetin (and maybe kaempherol) from the blood is slow compared to the other phenolic compounds (clearance half-life 15- 20 hr). Therefore, what little reaches the blood would be there for a longer period. Very little anthocyanins and essentially no proanthocyanidins would be absorbed (45).

Part of the poor absorption is due to the very active glucuronidation system in intestinal cells which results in the export of the glucuronidated phenolic compound back into the gut (45, 46). Even if the phenolic compound escapes glucuronidation in the enterocytes, much of the absorbed phenolic compound is glucuronidated and/or sulfated in the liver. Following glucuronidation and/or sulfation, much of the absorbed polyphenolic compound may be excreted into the bile and never appear in the blood. Rarely are glucuronidated and/or sulfated polyphenolic compounds in the blood in more than 1 umol/l concentrations when polyphenolic containing foods are consumed (45). The steady-state concentrations of polyphenolics in blood are often in the 100 - 900 nanomolar range and the concentration of unesterified polyphenolics in blood rarely exceeds picomolar concentrations.

Almost all of the research that demonstrates anti-cancer activity and biological end points that are purported to indicate a reduction in cardiovascular diseases, cancer, or untoward consequences of diabetes are done with concentrations that are 1,000 to 10,000 or more times greater than the

concentrations of unesterified polyphenolics found in humans or laboratory animals. To have physiological relevance, the experiments need to be conducted with no more than 1 or 2 μmol/l concentrations of the glucuronidated and or sulphated polyphenolic compound.

Even though polyphenolics are poorly absorbed, have low concentrations in blood, are present as conjugates with greatly reduced bioactivity, and are eliminated fairly quickly, beneficial effects have been attributed to the consumption of plant phenolics by humans (47-51) and laboratory animals (52-55). Much additional research is required to determine if the phenolic compounds found in beans are protective against chronic diseases.

The one tissue that may be exposed to the high concentrations of phenolics utilized in the in vitro studies is the gut (46, 56, 57). However, in the colon, the polyphenolics are rapidly converted into monophenolics by the colonic microflora and the biological activity of the monophenolic metabolites are unknown. Much work remains here also.

α-Amylase inhibitors

α-Amylase inhibitors are naturally present in a variety of plants, but are particularly high in common beans (58). They are large glycoprotein molecules (38-60 kDa) that are inhibitory towards mammalian α-amylases. Lajolo et al. (58) screened 150 common beans and isolated two types of inhibitors, I-1 and I-2. The α-amylase inhibitors varied to some degree in thermal stability, subunit composition, molecular weight, and ratios of I-1:I-2 between bean varieties. Inhibitor activity was not correlated with seed color. Normal cooking methods destroy most if not all of the α-amylase inhibitor activity (59). However, there are occasional public health problems resulting from consuming active α-amylase inhibitors. For example, a raw white bean preparation was sold in Japan to promote weight loss. Many people became sick and some required hospitalization due to dehydration.

Lectins (phytohemagglutins)

Lectins are large glycoprotein molecules that bind to glycoconjugates on cell membranes and can agglutinate red blood cells in vitro (60). Following ingestion, they can survive passage through the acid environment of the stomach and proteolytic activity in the duodenum. Lectins bind to epithelial cells in the small intestine thereby affecting nutrient absorption. Binding of lectins cause jejunal villi hypoplasia and crypt cell hyperplasia resulting in shorter, thicker microvilli and a reduction in brush border enzymatic activity. In the 1980s, there were several reported outbreaks of lectin poisoning in Britain as the result of consumption of incompletely cooked beans, causing severe nausea, vomiting, and diarrhea. Growth depression has also been observed in animals fed either purified lectins or raw beans. Proper cooking methods are therefore important to reduce the possibility of illness. Moist heat is more effective in eliminating lectin activity than dry heat (61).

Trypsin/chymotrypsin inhibitors

Trypsin and chymotrypsin (protease) inhibitors are present in many legumes including dry beans (62, 63). Protease inhibitors, in particular trypsin inhibitors, have generally been considered as anti-nutritional due to the long-standing observations that feeding animals raw beans causes growth depression and reduces nitrogen retention. Additionally, in rats, chickens, and growing guinea pigs, long-term feeding of raw legume flour or purified trypsin inhibitor stimulated pancreatic hypertrophy and, in rats, pancreatic adenoma development. This has raised some concern whether chronic consumption of legumes and other foods containing protease inhibitors may also produce adverse effects in humans. However, commonly employed cooking methods reduce the trypsin inhibitor activity in beans by 80-95%. Based on animal feeding studies, only 55-69% of the trypsin inhibitor activity needs to be destroyed to reduce pancreatic hypertrophy in susceptible animals and 79-87% destruction is

sufficient to allow maximum weight gain (64).

Protease inhibitors have been examined in several different model systems for the ability to suppress carcinogenesis. The most effective are those that inhibit chymotrypsin, and within this group, the Bowman-Birk inhibitor (BBI) from soybeans has been most extensively studied (reviewed in (65, 66)). Purified BBI or a concentrate enriched in BBI (BBIC) incorporated into the diet at 0.5-1% has been repeatedly shown to reduce colon carcinogenesis in both mice and rats without producing adverse effects on pancreatic lesions or body weight gain. The effects on colon carcinogenesis are most pronounced when low doses of carcinogen are administered. BBI has also been demonstrated to inhibit DMBA-induced oral carcinogenesis in hamsters, dimethylhydrazidine (DMH)-induced liver angiosarcomas and 3-methylcholanthrene (MC) induced lung tumorigenesis in mice, and in vitro, suppress radiation or chemically induced malignant transformation. More recently, the efficacy of BBIC as a potential chemopreventitive agent has been extended to phase I clinical trials in humans with some suggestion of a protective effect on benign prostate hyperplasia and oral leukoplakia (67, 68).

The exact cellular target of BBI in tissues and the mechanism of chemoprevention are not known with certainty. BBI can distribute among most tissues, except the brain, within three hours following oral ingestion. The highest concentrations are found in urine and that remaining in intestinal contents (69). Because the catalytic activity of BBI remains intact within tissue, Kennedy proposed that BBI may be inhibiting one or more enzymes involved in inducing the transformed phenotype (66, 70). In support of this, BBI has been associated with reversing the up-regulation of a proteolytic activity (Boc-Val-Pro-Arg-MCA) in the oral epithelium following carcinogen treatment and suppressing radiation-induced expression of proto-oncogenes (c-myc) in colon tissue and cancer cell lines.

We did not find any research related to the protease inhibitors in dry beans and health. The potential for dietary protease inhibitors to be beneficial to human health is controversial and the research interest in this area appears to have declined during the past 8 years.

LONGEVITY

What one eats affects disease susceptibility and survival. Comparing dietary patterns of elderly is one approach to discovering if there is a common denominator that promotes longevity. When food intake patterns of people 70 yr and older were examined in a cross-cultural study for a 5 to 7 yr period in 5 cohorts, the only statistically significant, consistent indicator of longevity was legume intake (71). For every 20g of legume consumption, there was a 7-8% reduction in mortality hazard ratio. The predominant legume consumed by the Swedish cohort was brown beans and peas and for the Greek cohorts in Australia and Greece, the predominant legumes consumed were white beans, lentils, and chickpeas. While this study doesn't provide overwhelming evidence that eating beans increases longevity, the data are intriguing!

RELATIONSHIP OF CHRONIC DISEASES TO GLYCEMIC INDEX AND GLYCEMIC LOAD

Dietary factors that promote excess glucose in the blood (hyperglycemia), excess insulin in the blood (hyperinsulinemia), and excess body fat also promote development of several chronic diseases including Type 2 diabetes, cardiovascular diseases, and cancer at several sites in the body. Hyperglycemia, hyperinsulinemia, and excess body fat are simply markers for a milieu of changes – hormones, growth factors, inflammatory products, oxidative stress, to name a few – that contribute to development of chronic diseases.

The extent to which different foods or meals raise blood glucose depends on the glycemic index of the consumed foods and the quantity of carbohydrate. The glycemic index of a food is a ratio of how much the blood glucose rises after consuming a standard amount of available carbohydrate compared to

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a standard. The glycemic load is calculated by multiplying the glycemic index of a food by the quantity of available carbohydrate eaten. The glycemic load of a meal is computed by summing the glycemic loads of all foods consumed. The following will discuss how the type of carbohydrate and the amount of the carbohydrate consumed impacts hyperglycemia, hyperinsulinemia, and body weight and thus indirectly to the development of chronic diseases.

A considerable amount of research has been devoted to studying the impact of consuming foods with a low glycemic index in contrast to high glycemic index foods. However, when one component of the diet is modified, invariably other aspects of the diet are altered also. This has led to inconsistent results. A key study (72) used meta-analysis and meta-regression to control for confounding variables that occur when prospective studies are conducted to determine the outcome of substituting low glycemic food for high glycemic foods. Since the total amount of carbohydrate consumed is important, the effect of both glycemic index and glycemic load were studied. Dietary factors considered as covariates were intakes of energy, fat, protein, and unavailable carbohydrates. The data from 45 studies were analyzed for the impact of glycemic index and glycemic load on fasting concentrations of blood glucose and insulin, blood glucose control, and body weight.

Blood glucose

Thirty-six studies provided data regarding fasting blood glucose concentrations. The studies utilized subjects that were: normal healthy, glucose intolerant, type 1 and 2 diabetics, and those at risk for heart disease. These studies reported a range in the reduction of glycemic index and glycemic load. Eating a diet with a low glycemic index significantly ($p < 0.05$) reduces fasting blood glucose in proportion to the reduction in glycemic index. Almost all low glycemic index foods are good sources of unavailable carbohydrate and therefore as the glycemic index of the diet was reduced, the

amount of unavailable carbohydrate was increased. It was determined that both a reduction in glycemic load as well as an increase in unavailable carbohydrate intake was important in causing a reduced fasting blood glucose concentration. A reduced glycemic load can be achieved by simply reducing the amount of carbohydrate containing foods that are consumed. It was further determined that a reduction in fasting blood glucose was better achieved by including low glycemic index foods and increasing unavailable carbohdrate rather than decreasing glycemic load by simply reducing the amount of available carbohydrate consumed.

Blood concentrations of glycated proteins (fructosamine and HbA1c) reflect overall control of blood glucose. Fifteen of the 36 studies provided information about fructosamine and/or HbA1c concentrations. These studies showed that overall control of blood glucose is strongly related to the glycemic index and glycemic load of the diet and the amount of unavailable carbohydrate consumed. It was suggested that optimum control of blood glucose is achieved when the diet has a glycemic index < 45, a glycemic load < 100g per day and a fiber intake of $\geq$ 25g per day.

Insulin

A reduction in insulin concentrations in fasting blood samples by switching from a high to a low glycemic index diet was achieved only when subjects had hyperinsulinemia (insulin > 100 pmol/l) initially. Non-diabetics with insulin > 100 pmol/l reduced their fasting insulin concentrations by an average of 73 pmol/l (p<0.001) when a low glycemic index diet intervention was implemented (2 studies). Likewise, fasting insulin was decreased by 73 pmol/l in overweight or obese, non-diabetics upon implementation of a low glycemic index diet only if their initial insulin was > 100 pmol/l (2 studies).

The amount of insulin required to promote glucose uptake by tissues (insulin sensitivity) is an important aspect of blood glucose control. Eighteen of the 45 studies reported measurements

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of insulin sensitivity. There was an average of 20% improvement (p< 0.004) in insulin sensitivity for the 18 studies when low glycemic index foods were substituted for high glycemic index foods. Non-diabetics (12 studies) improved (p<0.014) their insulin sensitivity by 25% and type 2 diabetics (5 studies) improved (p<0.014) their insulin sensitivity by 12%. Normal weight individuals (4 studies) did not achieve a significant improvement in insulin sensitivity while overweight and obese individuals (14 studies) had a 14% improvement (p<0.001) in insulin sensitivity. Taken together, people other than type 1 diabetics can expect an improvement in insulin sensitivity by switching from a high glycemic index diet to a low glycemic index diet.

Body Weight

There has been a steady increase in the percentage of overweight and obese individuals in North America and Western Europe. The increase in obesity is considered to be of epidemic proportions in the U.S. (73) and in most industrialized countries (73-76). For example, on a worldwide basis, more than one billion adults are overweight and more than 300 million are obese (74, 75). In the U.S. more than 60% of the adult population is overweight or obese (76). Obesity and overweight account for approximately 300,000 deaths per year in North America (77, 78) and the cost associated with excess body fat is estimated to be greater than 117 billion dollars per year (79). Most of the costs associated with excess body fat are related to type 2 diabetes, heart disease, and high blood pressure (80).

Twenty-three studies examined changes in body weight that occurred when subjects changed from a high to a low glycemic index diet. It goes without saying that a reduction body weight can occur only if there is a reduction in metabolizable energy intake. On the average, the glycemic load needed to be decreased by 17g/day before weight loss would occur. Consistent weight loss was not reported until the glycemic load was reduced by > 42g/day. If a reduction in glycemic load by substituting low glycemic index foods for high glycemic index foods resulted in less available carbohydrate (therefore a lower energy intake), weight loss occurred. The only significant factor related to weight loss was a reduced glycemic load and caloric intake; changes in fat, protein, and fiber intake that occur by substituting low glycemic index foods for high glycemic index foods could not explain the weight loss.

Clearly, if bean consumption could be increased and if there was a concomitant decrease in body weight, the public health benefit would be enormous! Since increasing bean consumption would not increase the cost of the diet, it is hard to imagine a more cost effective intervention.

Beans, glycemic index, and glycemic load

The study by Livesey et al. (72) provides very strong evidence that eating diets with a low glycemic index (< 45), a low glycemic load (<100 g equivalents per day), and more than 25g per day of unavailable carbohydrate will help normalize blood glucose, blood insulin and body weight. Controlling blood glucose, blood insulin, and body weight in turn will reduce the incidence of type 2 diabetes, cardiovascular diseases and cancer in certain parts of the body.

Beans are the perfect food to improve glycemic control. Beans have a low glycemic index, varying from 27-42% relative to glucose and 40-59% that of white bread (Table 2; (81)). Beans are also high in non-starch polysaccharides (typically 18-20%), 5% resistant starch, and 4% oligosaccharides to give a dietary fiber value of 27 - 29%. Substituting beans for foods prepared from white flour (on an equal dry weight basis) will reduce the glycemic index of the diet by about two-thirds and glycemic load by about 80%. Furthermore, consuming beans will significantly increase your intake of dietary fiber and that is particularly important for controlling blood glucose concentrations.

Table 2. Glycemic Indices of Various Beans (from (81)).

	GI White Bread *	GI Glucose *
Pinto beans	55	39
Kidney beans	42	27
Baked beans, canned	57	40
Dried beans	40	29
Black-eyed peas	59	42
Butter beans	44	31
Chick peas	47	33

* Calculated glycemic index when either white bread or glucose were used as the reference food. Expressed as a percentage of the reference food.

LOW GLYCEMIC CARBOHYDRATES AND DIABETES

Consumption of low glycemic index carbohydrates and soluble dietary fiber aids in managing some of the metabolic abnormalities associated with insulin resistance, diabetes, and hyperlipidemia.

Epidemiological evidence also suggests that long-term consumption of high glycemic index/load diets may increase the risk of developing NIDDM (82-85). Six prospective studies have reported on the relationship of glycemic index or glycemic load to risk of NIDDM. Only two studies further evaluated dietary intake among different food categories, and included an analysis on legumes (84, 86). Collectively, these studies indicate a protective role for low glycemic index diets on risk of incident NIDDM.

In the Health Professionals Study and Nurses Health Study, a 37-40% increase in diabetes was found in individuals with the highest glycemic intake compared to those having the lowest glycemic index intake after adjustment for known risk factors and cereal fiber. Foods most associated with diabetes risk included French fries, carbonated beverages, white bread, and white rice (82, 83). In a cohort from the Nurses Health study II, an increased risk of incident NIDDM was also found in young and middle aged women (24-44 years of age at baseline), when comparing highest vs lowest quintiles for glycemic index (adjusted relative risk 1.62, 95% CI 1.28-2.03) (87). Krishnan et al. (85) examined differences in glycemic indices and risk of NIDDM among a cohort of US black women. After 8 years of follow-up, they found a positive association for diabetes in women consuming higher glycemic index diets, which was surprisingly stronger in women with a BMI <25. The incidence relative risk was 1.91 (1.16-3.16, P=0.002) for women in the highest quintile (glycemic index (GI); 60.7 ± 6.8) compared to those in the lowest quintile (GI 41.9 ± 2.8). In a cohort of older Australians, Barclay et al. (84) reported a 1.75-fold increased risk of NIDDM in women < 70 years of age consuming higher GI carbohydrates, after multiple adjustments for other known risk factors (HR 1.75, 95% CI 1.05-2.92, P=0.031). Lastly, in a cohort of middle-aged Chinese women (88), individuals in the highest quintile for glycemic index and glycemic load and with a BMI > 25 had a relative risk for NIDDM of 1.30 (95% CI 1.06-1.6) and 1.52 (95% CI 1.22-1.89), respectively, after adjustment for other known

risk factors.

Two epidemiologic studies specifically related legume intake to risk of NIDDM. In a cohort of middle-aged Chinese women, Villegas et al. (86) reported a 38% reduced risk in the incidence of NIDDM for women in the highest quintile (65 g/day) of total legume intake (soybeans, peanuts, and other legumes) compared to those in the lowest quintile (12.3 g/day). This trend persisted when analyzed for "other legumes" (excluding soybeans and peanuts), with an adjusted relative risk in the highest quintile (37.1 g/day) of intake 0.76 (95% CI 0.64-0.90) compared to the lowest quintile of intake (5.6 g/day). One case-control study conducted in Europe found individuals consuming higher amounts of legumes were linked to a dietary pattern score associated with higher diabetes risk (89). The authors further acknowledged, however, that most of the legume intake in this group was attributable to a stew that also contains bacon, sausages, beef, or pork.

The ability of low GI carbohydrates to decrease risk of NIDDM may be related to lower post-prandial excursions in glucose and insulin coupled to improvements in insulin sensitivity (reviewed in (90)). High glycemic index foods are known to cause rapid elevations in blood glucose and insulin following a meal (discussed above). Chronic consumption of high glycemic index diets may in turn lead to down-regulation or desensitization of receptors for insulin, eventually contributing to insulin resistance (91). The body initially adjusts to higher circulating glucose by increasing insulin secretion from the pancreas. However, in susceptible individuals over time insulin resistance combined with exhaustion of insulin producing cells will eventually lead to type 2 diabetes (91, 92). Short-term studies in humans (93-99) indicate a role for low GI carbohydrates on improving insulin sensitivity. The accepted mechanism appears to be related to a decrease in counter-regulatory hormones (cortisol, and growth hormone) and non-esterified fatty acid release when low vs high GI foods are consumed (90). Current interest is also focusing on the role of hyperglycemia to inflammation, oxidative stress, and risk of diabetes. Increased circulating levels of the pro-inflammatory cytokines TNF α and IL-6 have been reported in insulin resistant individuals and diabetics (100-102) and serum IL-6 and C-reactive protein predict risk of diabetes in women (103). Higher plasma cytokine levels (IL-6, TNF α, and IL-18) have been reported in both normal and glucose intolerant individuals during acute hyperglycemic conditions, which were attenuated when the antioxidant, glutathione was co-administered (104). Although premature, these data suggest an intimate relationship of hyperglycemia to inflammation, and that reductions in hyperglycemia may mitigate risk of and vascular complications associated with diabetes.

It has been estimated that for every 1% reduction in HbA1c, there is a 21% reduction in risk in any end point examined for diabetes: 21% reduction for deaths, 14% for myocardial infarctions, and 37% for microvascular complications (105). Thus, the potential for low GI carbohydrates, especially beans, in the management, treatment, and delay in onset of NIDDM has profound implications for reducing morbidity and mortality associated with the disease.

BEAN CONSUMPTION AND CARDIOVASCULAR DISEASES (CVD)

Only one epidemiological study has directly examined the relationship between bean consumption and occurrence of CVD. Kabagambe et al. (106) reported that 1 serving per day of beans was associated with a 38% lower risk of myocardial infarction. More than one serving per day did not elicit a further decrease in risk for myocardial infarction. Only one study examined the relationship between legume consumption and risk of CVD. All other studies were even less precise concerning bean consumption since they examined the relationship between a "healthy eating pattern" that includes legumes and risk of CVD.

Bazzano et al. (107) reported that individuals consuming legumes at least 4 times per week had a 22% lower risk of heart disease than individuals consuming legumes less than once per week. In the epidemiological studies where legumes are consumed as part of a healthier diet plan, consistent reductions in heart disease risk have also been observed. In the Health Professionals Follow-up Study, men that adhered to a more "prudent diet" which included greater consumption of whole grains, legumes, fish, and poultry had a 30% lower risk of having heart disease. Conversely, individuals following a more "Western" diet, characterized by increased consumption of red meat, refined grains, sweets, French fries, and high fat desserts had a higher risk of heart disease (108) . Similar trends were seen in the Nurses Health Study (109). The relative risk of coronary heart disease in the 20% of women that followed the "prudent" dietary pattern more closely was 0.76 compared to 1.46 for women eating a "Western" type pattern (109). Thus, those that most consistently ate the "prudent" type of diet had one half the risk of developing heart disease compared to those that most often ate the "Western" type of diet. Lastly, a prospective study (110) utilizing the Nurses Health Study cohort found an inverse, but not significant ($p = 0.13$) trend between eating the prudent diet and a lower the risk of stroke.

Data from several human intervention trials indicate that consumption of canned (111-114) and cooked beans (111, 115-121) reduce serum cholesterol. All 11 studies found small, but statistically significant reductions in total and LDL cholesterol by eating beans. Only two studies (122, 123) did not find favorable changes in serum lipoproteins when beans were consumed. Generally, in carefully controlled clinical studies where the macronutrient intake was matched and the fiber content in the bean fed group was at least twice that of the control diet, significant reductions in both total and LDL cholesterol occurred. Changes in HDL cholesterol and triglyceride concentrations are inconsistent (111-121, 124).

Reductions in blood cholesterol due to consuming beans is modest at best (typically in the 6 - 10% range) and not likely to attract much interest by the medical profession. The study by Kabagambe et al. (106) suggests that eating beans provides protection from CVD beyond what can be explained by a small depression in blood cholesterol. It is quite possible that the wide variety of phytochemicals in beans provide protection against developing CVD. For example, publications reporting "anti-oxidant phytochemicals" protect the heart from adverse conditions in various animal models are starting to appear (125).

BEAN CONSUMPTION AND CANCER

The World Cancer Research Fund/American Institute for Cancer (3) recently published a comprehensive review that linked diet to cancer at 19 different locations in the body. Considering the etiology of cancer at many of the sites, one would expect that diet would have little impact except for excess body fat possibly increasing the risk. Beans were not considered as a separate entity, but as a group of foods labeled "pulses (legumes)". If a food, group of foods, and individual nutrients were found to be related to cancer incidence at one of the 19 sites, the relationship was classified as "decreased the risk" or "increased the risk". The strength of the evidence was classified as "convincing", "probable", "limited – suggestive", or "limited – no conclusion". To be classified as "probable", there had to be considerable data demonstrating a relationship existed.

The panel of experts did not feel that the evidence relating legume consumption to a decreased risk of developing cancer was "convincing" or even "probable" for cancer located at any of the 19 sites in the body. However, fiber containing foods were considered "probable" for reducing the risk of cancer in the colon and rectum. Since beans are rich in fiber, it can be inferred that eating beans will probably reduce one's risk of developing colon and

rectal cancer. The data relating legume consumption to a reduction of stomach and prostate cancer was considered "limited, but suggestive". It should be noted that the link between stomach cancer and legume intake is most likely based on soy and not on legumes other than soy. The study panel concluded that the data suggest that eating non-soy legumes would result in a reduction in prostate cancer (the specifics are discussed below). The panel also felt that the data relating foods rich in folate (naturally occurring or fortified) to a reduction in colon and rectal cancer was suggestive, but limited. The data relating legume consumption to cancers of the mouth, pharynx, larynx, esophagus, lung, pancreas, breast, ovary, and endometrium was too limited and no conclusion could be reached. There was no mention about dietary intake of legumes and the incidence of cancer in the nasopharynx, gallbladder, liver, cervix, kidney, bladder, or skin.

The expert panel did not include animal studies nor did they include human studies unless RR or OR with 95% CI were reported for their analyses. We feel that there are data to support that eating beans will reduce cancers of the colon, prostate, and breast, and possibly pancreas and esophageal.

The studies supporting our contention are discussed below.

BEANS and COLORECTAL CANCER

A.   Epidemiological Studies.

Despite the strong relationship of dietary habits to risk of colorectal cancer (CRC), epidemiologic studies are generally insufficient to conclude dry beans decrease CRC risk, although there is some suggestion of a protective effect. One cross-sectional study specifically related bean consumption to cancer mortality (126). Per capita data compiled from 41 countries (15 when beans were analyzed alone), revealed that countries with the greatest consumption of beans had the lowest mortality rates due to colon cancer (R=-0.68) (126). Nine case control studies have been conducted

where legume intake on CRC risk was evaluated. Five case-control studies have reported a protective effect of legume consumption on some aspect of CRC risk (127-131), three reported no association (132-134), and one reported increased risk (135). Another case-control study in Majorca reported on fiber from pulses, rather than pulse intake (136). The authors found a significant protective effect (P < 0.01) for individuals in the highest quartile of legume fiber intake, with an OR of 0.4.

In a prospective study examining dietary patterns and disease risk as part of the Adventist Health Study in the US, significant inverse associations between legume (beans, peas, lentils) consumption and colon cancer were found (137). After 6 years of follow-up, Singh and Fraser (137) reported that overall, individuals consuming legumes > 2 times/week were 47% less likely of developing colon cancer when compared to individuals consuming legumes never to < 1 time/week (RR=0.53, 95% CI 0.33-0.86). Upon further analysis, a complex relationship was detected between legume and red meat intake and body mass index (BMI). They found that individuals consuming legumes < 1 time/week and red meat ≥ 1 time/week and with a BMI ≥ 25 kg/m2 had a RR of colon cancer development of 3.19 (95% CI 1.62-6.26) compared to individuals with a BMI ≤ 25 kg/m2 and consuming legumes > 1 time/week and red meat ≤ 1 time/week. This association was stronger in men (RR = 5.10, 95% CI 1.48-17.5) than in women (RR = 2.00, 95% CI 0.78-5.11). Other large cohort studies published since this have found no association between legume intake and CRC risk (138), however two studies reported a protective effect of legumes against adenoma recurrence. Lanza et al. (139) studied changes in specific subcategories of fruit and vegetable intake to risk of adenoma recurrence as part of the Polyp Prevention Trail in the US. The authors reported a 65% reduced risk of advanced adenoma recurrence (OR=0.35, 95% CI 0.18-0.69) for subjects in the highest quartile of change in dry bean intake from baseline levels (median change in intake from baseline = 370%) compared to individuals in the

lowest quartile. There was no effect of change in bean intake on non-advanced adenoma recurrence (OR=1.01, 95% CI 0.76-1.34). In a cohort of the Nurses Health Study, women consuming four or more servings of legumes per week had a 33% lower risk of adenoma recurrence than those consuming less than one serving/week (OR=0.67, 95% CI 0.51-0.90; (140)).

B.   Experimental Studies.

Three experimental studies have been conducted specifically examining the relationship of dry bean consumption to chemically-induced colon cancer in rats. Hughes et al. (141) fed rats diets containing either pinto beans (59% wt/wt) or casein as the protein source. They found that feeding pinto beans inhibited colon tumor incidence by 52% (50% in casein-fed animals vs 24% in bean-fed animals) and significantly reduced the number of tumors that developed (1.0 ± 0.0 vs 2.5 ± 0.6 tumors/ tumor bearing animal). In a similar design, Hangen and Bennink (142) also reported a protective effect of dry beans on experimental colon cancer. They found that feeding either black beans or navy beans (75% wt/wt) inhibited colon cancer by ~57%, and similar to Hughes et al. (141), bean-fed rats also developed fewer tumors ($1.0 \pm .17$ vs 2.2 ± 1.2 tumors/tumor bearing animal). In this study, the chemoprevention of beans was associated with significantly more resistant starch reaching the colon, resulting in higher colonic acetate and butyrate production, and a decrease in body fat (142).

In the last study, Rondini and Bennink (unpublished data) corroborated chemically-induced tumor inhibition by black beans in rats. They found that black beans (74% wt/wt of diet) reduced the number of animals with colon tumors at both early (18 weeks after carcinogen administration, 8% vs 38%) and late (31 weeks post-carcinogen administration, 33% vs 75%) time points. Because the effect of beans on carcinogenesis appeared to be due to a delay in the development of tumors from normal-appearing colonic mucosa, they further

profiled gene changes in non-involved (ie, "normal appearing") distal colonic tissue from black bean- and casein-fed animals, either administered the carcinogen azoxymethane (AOM) or saline-injected controls, using microarrays. They anticipated that genes most important to black bean-induced suppression of tumorigenesis would have altered expression (increased or decreased) that paralleled tumor incidence. They identified 145 genes (90 up-regulated, 55 down-regulated) differentially expressed by beans compared to casein-fed animals. Bean-feeding induced changes in genes consistent with reduced cell proliferation and inflammation and enhanced energy metabolism. Specific molecular targets of beans that appear to corroborate reduced tumorigenesis included suppression of the pro-inflammatory gene secretory phospholipase A2 (sPLA2), the innate immune gene NP defensin 3α, as well as alterations in extracellular matrix components (collagen 1α1, fibronectin 1). These genes were induced by carcinogen (AOM) injection in both diets, but much less so in black bean-fed animals. These data provided preliminary evidence that inclusion of beans into the diet differentially affects molecular pathways in the colon important in carcinogenesis.

Breast cancer

In the Nurses Health Study, Adebamowo et al. (143) reported on intake of flavonols and flavonol-rich foods and risk of breast cancer in women who were premenopausal at baseline. A majority of breast cancer cases (89.7%) were premenopausal, 5.5% postmenopausal, and 4.8% of unknown menopausal status. They found a significant (P = 0.03) inverse association with bean and lentil intake, but not other flavonol-rich foods, and risk of breast cancer. The multivariate relative risk for highest (2 or more times/week) compared to lowest (<1 time/month) cumulative average intake was 0.76 (95% CI 0.57-1.00) and 0.67 (95% CI 0.48-0.94) when compared to baseline intake (p = 0.02). In the same cohort, however, Fung et al. (144) reported on

dietary patterns and risk of postmenopausal breast cancer and found no association (P = 0.16) between legume intake (4-6 servings/week vs < 1 serving/week) and risk of estrogen receptor negative breast cancer (multivariate RR = 0.79, 95% CI 0.51-1.22).

Three case-control studies have been conducted where bean intake was assessed. Silva et al. (145) conducted a case-control study on vegetarianism and risk of breast cancer in South Asian immigrants living in England. They reported significant, inverse associations between the highest (>107.4 g/day) and the lowest (<35 g/day) quartiles of pulse, lentil, and dhal consumption (OR=0.54, 95% CI 0.31-0.94, P = 0.007) and risk of breast cancer in middle-aged women (median age cases (51.5), controls (51.9)). A non-significant inverse association was also found for intake of non-starch polysaccharides from pulses (adjusted OR=0.66, 95% CI 0.38-1.15) when comparing highest (>2.6 g/day) to lowest (<0.9 g/day) quartile of intake. In a case-control study in Shanghai, China, Shannon et al. (146) found no association between non-soy legumes and breast cancer risk (OR=0.76, 95% CI 0.48-1.21) when comparing highest (> 3.9 servings/week) to lowest (<1.9 serving/week) quartile after adjusting for age and total energy intake. The last case-control study, conducted in Argentina, found an increased risk (OR=3.3) in individuals consuming higher intakes of pulses (147). One study reported on fiber from beans (148). Potischman (148) examined food group and micronutrient associations with risk of early-stage breast cancer in women in the US. They found an insignificant, inverse association between cases and controls for intake of fiber from beans (OR=0.88, 95% CI 0.7-1.2) when comparing the highest (>1.89g/day) to lowest quartile (<0.72 g/day) of intake. After adjustment for energy, the same trend existed, although this did not reach statistical significance.

Three ecological studies found a negative association between legume consumption and breast cancer mortality (149-151). One cross-sectional study specifically related bean consumption to breast cancer mortality (126). Per capita data from 15 countries, revealed that countries with the greatest consumption of beans had the lowest mortality rates due to breast (R=-0.70) cancer.

Prostate cancer

One case-control and one cohort study specifically identified beans and risk of prostate cancer. The case-control study reported a reduced risk for prostate cancer with increasing consumption of baked beans (152). An odds ratio of 0.844 (95% CI; 0.709 - 1.00) was determined per increase in serving of baked beans per week. The cohort study conducted in the Netherlands (153) found an inverse relationship between consumption of broad bean and risk of prostate cancer (OR 0.956 (95% CI; 0.823 - 1.11)). Both studies reported that consuming beans reduced the risk of prostate cancer, but the results from the cohort study were not statistically significant.

Four studies compared the frequency of bean and lentil consumption to risk for prostate cancer. A prospective cohort study (154) found that eating beans and lentils significantly reduced the risk of prostate cancer (OR of 0.817 (95% CI; 0.714 - 0.934)). Meta analysis of the three case-control studies (152, 155, 156) showed a statistically non-significant protective relationship between bean and lentil consumption and risk of prostate cancer OR of 0.956 (95% CI; 0.884 - 1.03). One cohort study (157) did not report quantified data but they did indicate that the association between prostate cancer mortality and bean and lentil consumption was not statistically significant. One case-control study (152) reported a non-significant increase in prostate cancer was associated with eating beans, lentils, and peas.

Kolonel et al. (158) studied dietary patterns and risk of prostate cancer in the USA and Canada. When they differentiated between soy and non-soy legumes, an inverse relationship between non-soy legume consumption and prostate cancer was determined (OR of 0.966 (95% CI; 0.941 - 0.991). Dry beans are the most commonly consumed non-

soy legume in the US and Canada, so this study suggests that eating beans helps to inhibit prostate cancer. Soy consumption was not related to prostate cancer incidence.

There were five case-control studies (152, 158-161) when the food category was broadened to include all pulses (studies cited in the above 3 paragraphs are included as part of the pulse category). These studies produced an overall OR of 0.966 (95% CI; 0.951 - 0.981) per increase in pulse serving/week. Four ecological studies reported a protective effect of legume consumption on prostate cancer risk (149, 162-164). One cross-sectional study specifically examined bean consumption and mortality rates from prostate cancer (126). Data from 15 countries revealed that countries with the greatest consumption of beans had the lowest death rates due to prostate cancer (r = -0.66).

It is important to note that the expert committee (3) concluded that the cohort study and the case-control studies showed that consuming pulses reduced the risk of prostate cancer.

ADDITIONAL DATA RELATING BEANS TO CANCER PREVENTION

Fiber and cancer

A large number of studies have examined the relationship between fiber intake and colorectal cancer with mixed results. The WCRF/AICR study panel (3) summarized 19 cohort studies. Ninety-one case-control studies have been conducted, but because of the large number of cohort studies, the case-control studies were not summarized. It was possible to conduct a meta-analysis on 8 of the cohort studies. The OR from the meta-analysis was 0.90 with a 95% CI (0.84 - 0.97) per 10g of fiber intake per day and a dose-response relationship was apparent. The expert panel concluded that foods containing fiber (naturally occurring, not added fiber) are probably protective against colon cancer based on generally consistent cohort studies, a clear dose-response, and plausible mechanisms. High

intakes of fiber have been associated with a reduced risk of esophageal cancer and cardiovascular diseases also. Pulses (legumes) and minimally processed cereals are the most concentrated sources of fiber.

Folate and cancer

Foods that are good sources of folate were identified as protecting against cancer at several sites (3). The strongest protective association for folate containing foods was noted for pancreatic cancer. The data for cancers of the colon and esophagus was strong, but not as strong as for protection against pancreatic cancer.

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