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SUBJECT OF PETITION:	Authorization of a health claim for monounsaturated fatty acids from olive oil and coronary heart disease
SUBMITTED TO:	Office of Nutritional Products, Labeling and Dietary Supplements (HFS-800) Center for Food Safety and Applied Nutrition Food and Drug Administration Harvey W. Wiley Federal Building 5100 Paint Branch Parkway College Park, MD 20740

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Health Claim Petition: Monounsaturated Fatty Acids from Olive Oil and Coronary Heart Disease

Volume 1 of 3

The North American Olive Oil Association

August 28, 2003

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I. INTRODUCTION

The undersigned, the North American Olive Oil Association (NAOOA), submits this petition pursuant to section 403(r)(4) of the Federal Food, Drug, and Cosmetic Act with respect to the ability of monounsaturated fatty acids (MUFAs) from olive oil and certain olive oil-containing products to reduce the risk of coronary heart disease (CHD). The claim would apply to eligible products that contain at least 3.4 grams of olive oil per reference amount customarily consumed (RACC).

The NAOOA is committed to supplying consumers with quality products in a fair and competitive environment; to fostering a clear understanding of the different grades of olive oil; and to expounding the benefits of olive oil in nutrition, health, and the culinary arts. We believe a health claim linking the MUFAs in olive oil with reduced risk of CHD has important public health implications. An in depth assessment of the literature demonstrates that there is significant scientific agreement that MUFA-containing moderate fat diets that are low in saturated fat and cholesterol result in favorable serum lipid profiles. Olive oil is one of the richest sources of MUFAs (primarily oleic acid) among edible oils sold in the United States, and its effect on serum lipids has been more extensively studied than any other source. In addition, olive oil contains a variety of non-glyceride components that likely contribute to its cardioprotective properties. Finally, olive oil is versatile and well accepted by American consumers.

FDA has long recognized the potential role of MUFAs in managing the risk of CHD. The agency made the following observations in authorizing the current health claim on "Dietary Saturated Fat and Cholesterol and Heart Disease" (58 FR 2739, 2740, January 6, 1993):

> FDA is aware of the recent and ongoing research efforts on the possible beneficial role of *cis*-forms of MUFA's in helping Americans to find a practical means of reducing saturated fat intake without changing total dietary fat intakes...FDA, however, considers this issue outside the scope of this rule. In the proposed rule, the agency noted that, because of the extremely large volume of scientific research on lipids and cardiovascular disease and because of the extremely limited time constraints of the 1990 amendments, it had limited its science review to an evaluation of the relationship of saturated fat and cholesterol intakes to risk of CHD. Therefore, in both the proposed and final rules, FDA has limited the health claim to saturated fats and cholesterol.
>
> FDA notes that the rapidly expanding science base may now, or in the future, be adequate to support that *cis*-monounsaturated fatty acids have a beneficial role in reducing blood total and LDL-cholesterol levels. However, because the question of whether this nutrient/disease relationship is appropriate for a health claim is outside the scope of this rulemaking, the question should be the subject of a petition for a health claim in accordance with the provisions of the final rule on general requirements for health claims published elsewhere in this issue of the **Federal Register**.

Considerable information on the benefits of diets rich in MUFAs has accumulated since FDA published these comments. Numerous studies have shown that dietary MUFAs lower total cholesterol (T-C) and low density lipoprotein cholesterol (LDL-C) when substituted for saturated fat, and newer data suggest they also have an independent effect (Yu *et.al.*, 1995; Clarke *et.al.*, 1997). In addition, it has been shown that MUFAs do not lower plasma high density lipoprotein cholesterol (HDL-C) or raise triglycerides (TG) as high carbohydrate (CHO) diets often do, which prompted the Food and Nutrition Board (2002) to conclude, "These data indicate that in weight-stable individuals, a high monounsaturated fatty acid-low saturated fatty acid diet results

in a more favorable metabolic profile with respect to total cholesterol, HDL cholesterol, and triacylglycerol concentrations." This conclusion is also reflected by the latest dietary recommendations in the Dietary Guidelines for Americans (U.S. Department of Agriculture, U.S. Department of Health and Human Services, 2000), the American Heart Association (Krauss, 2000) and the National Heart, Lung and Blood Institute (NHLBI) (National Cholesterol Education Program, 2001).

The NAOOA believes there is now ample evidence to support FDA's speculation that, "*cis*-monounsaturated fatty acids have a beneficial role in reducing blood total and LDL-cholesterol levels," and we are pleased to respond to the agency's "invitation" to submit this petition.

The model language for the proposed claim is, "Monounsaturated fats from 13.5 grams per day of olive oil (one tablespoon) may reduce your risk of heart disease when included in a moderate-fat diet low in saturated fat and cholesterol." The disclosure statement, "See nutrition information for fat and saturated fat content," would also be used when appropriate.

II. REGULATORY RATIONALE

The proposed claim is based on the approach FDA used to authorize health claims for soluble fiber from certain foods and risk of CHD (21 CFR § 101.81). Specifically, we propose that MUFAs be designated as a marker for the cholesterol-lowering potential of foods containing them, and that individual sources of these fatty acids (e.g. olive oil) be evaluated on a case-by-case basis to determine their eligibility for the claim.

The agency explained this approach in the final rule for the health claim on oats and CHD (62 FR 3584, 3590, January 23, 1997):

> In the proposal, the food substances that were the subject of the claim were oat bran and rolled oats and the products that contain them. The agency stated that the β-glucan soluble fiber content of these products is an appropriate marker for identifying the cholesterol-reducing potential of these products (61 FR 296 at 308) and established levels for β-glucan in foods that would qualify for the claim.
>
> Based on its review of the comments, however, the agency has concluded that β-glucan is the primary component of whole oats that is responsible for the effect that consuming these foods has on the risk of CHD. Therefore, the agency has concluded that the substance-disease relationship that is appropriately the subject of a claim is that between β-glucan soluble fiber from whole oats and CHD.

The agency further explained that compounds other than β-glucan, such as tocotrienols, might contribute to the cardioprotective effect of oat-containing products, and that β-glucan is not the appropriate object of the claim:

> The agency has carefully reviewed the comments and evidence submitted on the issue of the significance of the β-glucan in the oat products and is persuaded that β-glucan soluble fiber is the primary, but not the only, component in whole oats that affects serum lipids. ß-glucan thus plays a significant role in the relationship between whole grain oats and the risk of CHD…
>
> FDA, therefore, concludes that it is appropriate to change the food substance that is the subject of this authorization for claims from oat bran and rolled oats to β-glucan soluble fiber from whole oats.

The NAOOA believes this approach is directly applicable to MUFAs from olive oil. A review of the literature provided later in this document suggests that the hypocholesterolemic effect of olive oil is due primarily to its high MUFA content. However, olive oil is unique from other common edible oils because it is obtained by pressing washed, crushed olives, rather than by

solvent extraction. As a result, olive oil contains numerous components (e.g. polyphenols, sterol esters) other than MUFAs that likely contribute to its cardioprotective properties. It is therefore appropriate to designate MUFAs as a marker for eligibility for the claim, but to evaluate specific foods on a case-by-case basis.

This reasoning is analogous to why FDA did not authorize a health claim for β-glucan from non-oat sources such as barley. The agency acknowledged that while there is evidence that a variety of food sources containing this substance would lower blood cholesterol, it lacked criteria for differentiating among such sources (62 FR 3584 at 3587). The agency also noted that it had not reviewed the, "totality of evidence on these other sources of the fiber" but structured the regulation in such as was as to accommodate such sources in the future,

> Nonetheless, the agency recognizes that it is likely that consumption of other sources of β-glucan soluble fiber in addition to those that are the subject of this rulemaking will affect blood cholesterol levels. For this reason, and for reasons described elsewhere in this document in response to related comments about other soluble fibers, FDA is adopting a final rule that is structured so that it can be amended to establish a framework that will accommodate claims for other sources and types of soluble fibers and the risk of CHD.

In fact, the agency subsequently extended eligibility of the claim to additional sources of β-glucan (i.e. the soluble fraction of α-amylase hydrolyzed oat bran or whole oat flour) (67 FR 61773, October 2, 2002) and to psyllium (an additional form of soluble fiber) (63 FR 8103, February 18, 1998). The same approach can be used to modify the proposed claim to include other sources of MUFA if the agency is provided with sufficient data to demonstrate that they also reduce the risk of CHD.

In summary, the NAOOA believes that the approach FDA has taken for the authorization of health claims under 21 CFR § 101.81 is equally applicable to MUFAs from olive oil, and we strongly recommend that it be used to authorize the proposed claim described in this petition.

Attached hereto, and constituting a part of this petition, are the following:

III. PRELIMINARY REQUIREMENTS

Petitions for health claims pertaining to a component of a food to be consumed at other than decreased dietary levels are required by 21 CFR § 101.70 to demonstrate that certain preliminary requirements are met: that the object of the proposed claim conforms to the definition of a "substance" in § 101.14(a)(2); that the substance is eligible for a health claim according to § 101.14(b), which specifies it must be "associated with a disease or public health-related condition for which the general U.S. population, or an identified U.S. population subgroup…is at risk…"; that it contributes, "taste, aroma, or nutritive value, or any other technical effect listed in 21 CFR § 170.3(o); and that the substance is safe and lawful at the level necessary to justify the claim under the food safety provisions of the Federal Food, Drug, and Cosmetic Act.

As noted above, the "substance" of the proposed claim is MUFAs from olive oil. Olive oil is proposed as a dietary source of MUFAs, but is not, in and of itself, the object of the claim. The NAOOA believes that MUFAs comply with all preliminary requirements for health claims, as discussed below:

A. MUFAs are a substance under 21 CFR § 101.14 (a)(2)

The definition of a "substance" under 21 CFR § 101.14 (a)(2) is "...a specific food or component of food, regardless of whether the food is in conventional food form or a dietary supplement that includes vitamins, minerals, herbs, or other similar nutritional substances." MUFAs (including oleic acid (18:1ω9) and other *cis*-MUFAs) are components of food. These compounds are also important nutritional substances that provide energy to the body and are key components in membrane structural lipids – especially nervous tissue myelin (Food and Nutrition Board, 2002). Furthermore, the NHLBI (National Cholesterol Education Program, 2001) recommends that up to 20% of total calories come from dietary sources of MUFAs because of their beneficial effects on blood lipids. We believe these properties quality MUFAs as a substance under 21 CFR § 101.14 (a)(2).

B. Heart disease is a major public health concern in the United States

Cardiovascular disease (CVD) is the leading cause of mortality in the U.S. and accounted for 39.4% of all deaths during 2000 (American Heart Association, 2002). CVD was listed as a primary or contributing cause of death on approximately 1,415,000 death certificates in 2000, and the total direct and indirect cost of CVD in this country was estimated to be $351.8 billion. Fifty-four percent of all CVD deaths are due to CHD. In addition, FDA has authorized several CHD-related health claims since enactment of the Nutrition Labeling and Education Act (NLEA): dietary saturated fat and cholesterol and risk of coronary heart disease (21 CFR § 101.75); fruits, vegetables, and grain products that contain fiber, particularly soluble fiber, and risk of coronary heart disease (21 CFR § 101.77); soluble fiber from certain foods and risk of coronary heart disease (21 CFR § 101.81); soy protein and risk of coronary heart disease (21

CFR § 101.82); and an interim final rule for plant sterol/stanol esters and risk of coronary heart disease (21 CFR § 101.83).

C. MUFAs contribute nutritive value to the diet and functionality to food products

The definition of "nutritive value" under 21 CFR § 101.14 (a)(3) is "a value in sustaining human existence by such processes as promoting growth, replacing loss of essential nutrients, or providing energy." As noted above, MUFAs provide nutritive value to the diet by serving as a source of energy. Energy is an essential component of the diet and Estimated Energy Requirements (EERs) for different age-gender segments of the population have recently been established (Food and Nutrition Board, 2002). MUFAs in the context of the proposed claim are most appropriately considered in a nutritional sense as components of TGs. However, MUFAs may also be used to provide functionality to food products in the free form or as components of mono-, di- or triglycerides. For example, oleic acid is authorized as a direct food additive under 21 CFR § 172.860 for use as a lubricant, binder and a defoaming agent, technical effects listed in 21 CFR § 170.3 (o)(14), (18) and (29).

D. MUFAs are safe and lawful

MUFAs are ubiquitous, natural components of the food supply. As noted in paragraph A above, the most recent dietary recommendations from the NHLBI (National Cholesterol Education Program, 2001) recommend that these fatty acids supply up to 20% of total energy in the diet. In addition, the most prevalent form of MUFA (oleic acid) is specifically authorized for the addition to food under 21 CFR §§ 182.90, 172.860, 182.70, 172.210, 175.105, 176.180, 177.1200 and 177.2600.

IV. SUMMARY OF SCIENTIFIC DATA SUPPORTING THE CLAIM

A. Introduction

A comprehensive review of the published observational studies, controlled feeding trials, meta-analyses and third-party dietary recommendations (e.g. AHA revised dietary guidelines, the National Cholesterol Education Program, Adult Treatment Panel III guidelines) demonstrate that the cardioprotective effects of diets rich in MUFAs have been well accepted by the nutrition and public health communities. MUFAs exert much of their protective effect by replacing cholesterol-raising saturated fatty acids (SFAs) in the diet, but they also function through an independent effect on blood lipids. The cardioprotective properties of olive oil have been more thoroughly studied than any other source of MUFAs. Although the mechanisms by which this food exerts its cardioprotective effect are not completely understood, it is likely that its high MUFA content is the primary factor. Nevertheless, olive oil contains numerous additional components that may also be cardioprotective. In addition, olive oil is chemically stable and does not increase the susceptibility of serum lipoproteins to the formation of atherogenic oxidation products as polyunsaturated fatty acids (PUFAs) do. The combination of these beneficial effects of olive oil make it uniquely qualified to bear the proposed claim. The NAOOA strongly believes that the scientific evidence discussed below demonstrates that there is significant scientific agreement to support the proposed claim.

The NAOOA is aware that FDA is reluctant to authorize a health claim on the grounds that a food or substance in a food displaces a dietary constituent that is associated with increased risk of a disease (e.g. SFAs). Although there is evidence that MUFAs exert an independent effect on blood T-C and LDL-C, the majority of their benefit is likely due to the fact that they can displace

saturated and *trans* fatty acids (TFAs) in the diet. Despite the agency's reluctance, we believe it is entirely appropriate for this mechanism to be given credence in consideration of the proposed claim.

Section 403(r)(3)(A) of the Federal Food, Drug, and Cosmetic Act states that a health claim shall be authorized if, "the Secretary determines that such information will assist consumers in maintaining healthy dietary practices." The intent of this legislation is to improve public health. Congress did not put limits on the type of biological mechanism required to accomplish this goal. As will be discussed in this petition, the totality of scientific evidence clearly demonstrates that MUFAs from olive oil may reduce the risk of CHD when consumed as part of a moderate-fat diet low in saturated fat and cholesterol. We believe this statement is true at face value, and the fact that a portion of the beneficial effect comes from the displacement of SFAs and/or TFAs (and a reduced dependence on carbohydrates for this purpose) is of no practical significance. In fact, to deny providing the public with this information in the form of a health claim would be contrary to both the legislative intent of the NLEA and FDA's public health mandate.

Nevertheless, the NAOOA appreciates that the agency does not want to be put in a position that forces it to authorize a health claim for any substance that can displace a negative dietary component. We believe, however, that MUFAs are virtually unique in this regard. Specifically, in order to prompt a meaningful biological effect on CHD, a substantial amount of the beneficial dietary component(s) would be required to displace the negative one(s). Therefore, it must be nutritionally appropriate for considerable amounts of the beneficial component(s) to be consumed routinely as part of a balanced diet. The only candidates for the replacement of SFAs

and/or TFAs (sources of dietary energy) are MUFAs, PUFAs, carbohydrates (including polysaccharides and simple sugars), protein and alcohol. We believe that MUFAs are the only macronutrient that clearly qualifies for this purpose. As noted above, the NHLBI (National Cholesterol Education Program, 2001) recommends that up to 20% of total calories be provided by MUFAs – more than two times the recommendation for PUFAs. With respect to CHD, dietary carbohydrates (including simple sugars) tend to reduce serum HDL-C and increase TG (National Cholesterol Education Program, 2001; Food and Nutrition Board, 2002). On the other hand, moderate-fat diets in which MUFAs partially replace carbohydrates (as the proposed claim states), result in a more favorable blood lipid profile. The recommended intake of PUFAs is limited to 10% of total energy because greater amounts may increase the susceptibility of LDL to the formation of atherogenic oxidation products (National Cholesterol Education Program, 2001; O'Byrne *et.al.*. 1998). The safety of prolonged consumption of large amounts of protein has not been adequately studied (Foster *et.al.*, 2003; Samaha *et.al.*, 2003), and the use of alcohol as a dietary replacement is obviously inappropriate. We therefore believe that MUFAs are the optimal macronutrient for dietary displacement, and that FDA can easily define criteria that would permit the rational application of this mechanism to the authorization of health claims.

In summary, we believe that the proposed claim will benefit public health by a combination of the inherent cholesterol-lowering properties of MUFAs as well as by enabling consumers to obtain a more favorable blood lipid profile by using MUFAs as a partial alternative to carbohydrates to reduce consumption of SFAs and/or TFAs. MUFAs are uniquely suited for this purpose, and may be the only dietary component for which such a mechanism can be justified. Finally, the overriding imperative for authorization of the proposed claim should be benefit to

the population, and not whether there is an inherent biochemical mechanism. We therefore strongly urge FDA to consider the ability of MUFAs to replace SFAs, TFAs and carbohydrates as an appropriate mechanism, at least in part, for authorization of the proposed claim.

B. Regulatory precedent

FDA has accepted the validity of serum lipids as a biomarker for CHD and has used this rationale for the approval of several other CHD-related health claims. The preamble to the Interim Final Rule for the health claim on plant sterol/stanol esters and CHD (65 FR 54686, 54690, September 8, 2000) states,

> ... the agency based its evaluation of the relationship between consumption of plant sterol/stanol esters and the risk of CHD primarily on changes in blood total and LDL-C cholesterol resulting from dietary intervention with plant sterol/stanol ester-containing products. A secondary consideration was that beneficial changes in total and LDL-C cholesterol should not be accompanied by potentially adverse changes in HDL-C cholesterol. This focus is consistent with that used by the agency in deciding on the dietary saturated fat and cholesterol and CHD health claim, §101.75 (56 FR 60727 and 58 FR 2739); the fiber-containing fruits, vegetables, and grain products and CHD claim, §101.77 (56 FR 60582 and 58 FR 2552); the soluble fiber from certain foods and CHD claim, §101.81 (61 FR 296, 62 FR 3584, 62 FR 28234, and 63 FR 8119) and the soy protein and CHD claim §101.82 (63FR 62977 and 64 FR 57700).

In addition, the agency has recognized that health claims can be authorized without a detailed understanding of the mechanism involved. Specifically, the Final Rule authorizing a health claim for soy protein and coronary heart disease (64 FR 57700, 57709, October 26, 1999) states,

> Other comments reviewed various possible mechanisms for the cholesterol-lowering effects of soy protein and some argued that until the mechanism of action of soy protein is clearly established, no health claim should be authorized. **FDA notes, however, that such knowledge is not necessarily required for authorization of a health claim.** (emphasis added)

Furthermore, the agency's document, "Guidance for Industry – Significant Scientific Agreement in the Review of Health Claims for Conventional Foods and Dietary Supplements" (page 9, December 22, 1999) states,

> Measurement issues generally focus on substances in food, but the same principles apply when the substance of interest is itself a food. While a single food can be the subject of a health claim, existing experience is that the subject is more likely to be a group of foods, such as fruits, vegetables, and grains, which have been associated with a reduced risk of heart disease and of cancer. This identification, and consequently measurement, of a food group is, in turn, most likely to occur because it is not possible to identify and, therefore, measure a particular component of these foods that is responsible for the benefit.

The precedent established by the agency in adopting these positions is applicable to the proposed claim. In addition, as noted earlier in this petition, the NAOOA believes the approach FDA applied in the authorization of a health claim for soluble fiber from oat bran, rolled oats and whole oat flour (21 CFR §101.81) is directly applicable to olive oil as a source of MUFAs (see 62 FR 3584 at 3586). With respect to that claim, soluble fiber (i.e. β-glucan), serves as a marker for the cardioprotective properties of oat products that contain it, but additional factors (e.g. tocotrienols) may also contribute to this effect. In addition, other sources of β-glucan (e.g. barley) may not have similar effects on CHD due to myriad factors including the impact of other constituents, physical form or processing.

With respect to the current petition, MUFAs serve as a marker for the cardioprotective properties of olive oil, but additional factors specific to each dietary source may enhance or detract from their protective properties. Therefore, the eligibility of other MUFA-containing oils must be considered on a case-by-case basis. A comprehensive analysis of the properties of olive oil that uniquely qualify it as a source of MUFAs for the proposed claim, and a discussion of the

available literature on the ability of MUFAs from olive oil to reduce the risk of CHD, is provided below.

C. The role of olive oil in reducing the risk of CHD

There are several compelling reasons why olive oil should be specified as a source of MUFAs for the proposed claim. This food is a rich source of MUFAs compared to other edible oils available in the marketplace; olive oil is well accepted by U.S. consumers for use in cooking and at the table; the cardioprotective properties of olive oil have been more thoroughly documented than for any other food; and unlike most other edible oils, olive oil contains numerous non-glyecride compounds that may contribute to its cardioprotective properties.

1. Olive oil is a rich source of MUFAs

Olive oil has long been recognized as one of the richest sources of MUFAs available. Table 1 (see next page) provides the concentration of MUFAs of common fats and oils used in the United States.

Olive oil is composed of more than 70% MUFAs – predominantly oleic acid (18:0). Indeed, the name "oleic acid" is derived from the word "olive" because olive oil is the most concentrated natural source of this fatty acid. Olive oil provides more than three times the amount of MUFAs as soybean oil (the most widely used edible oil in the United States) and nearly 25% more than canola oil which is also recognized as a rich source of MUFAs. The only oils that have a higher concentration of oleic acid than olive oil are high oleic sunflower and safflower oils. These

products have been specifically developed to maximize their content of this fatty acid. As noted below, olive oil also contains polyphenols and other potentially cardioprotective substances that are not prominent in these products.

Table 1
Monounsaturated Fatty Acid Content of Edible Oils

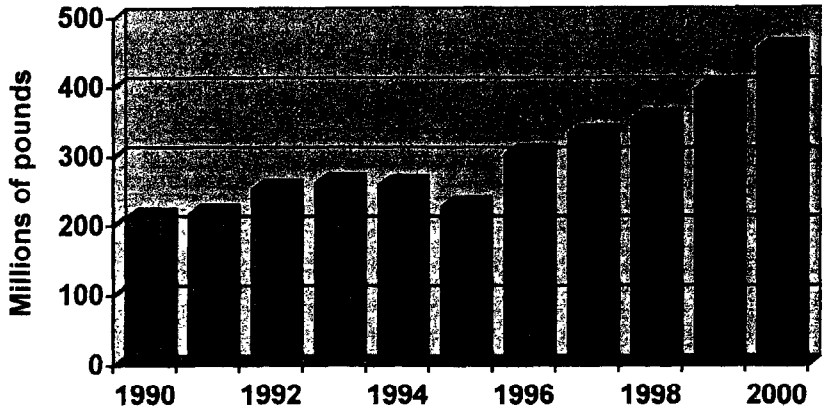
Product	**MUFA Content (g/100g)***
Butter	23.4
Canola oil	58.9
Coconut oil	5.8
Corn oil	24.2
Cottonseed oil	17.8
Lard	41.5
Olive oil	***72.5***
Palm oil	37.0
Palm kernel oil	11.4
Peanut oil	46.2
Safflower oil (high oleic)	74.6
Sesame oil	39.7
Soybean oil	23.3
Sunflower oil (high oleic)	83.5

*Source: USDA National Nutrient Database for Standard Reference, Release 15

2. Olive oil is well accepted by U.S. consumers

Figure 1 shows that the use of olive oil in the U.S. (based on government disappearance data) has increased steadily during the past decade.

Figure 1
Disappearance of Olive Oil in the United States



Source: USDA, Oil Crops Situation and Outlook Yearbook, October 2002

This steady increase in the use of olive oil is also reflected by sales data. Olive oil was purchased by 31.5% of U.S. households during 2000 and accounted for 32.3% of the $1.2 billion cooking oil market – more than any other oil[1].

Quantitative consumer research regarding the increasing popularity of olive oil has not been published, but its unique flavor compared to other oils, and an increasing recognition of its health benefits as part of the Mediterranean diet are likely factors. The NAOOA believes that the availability of an FDA-authorized health claim for olive oil would stimulate additional use of this healthful food.

3. The cardioprotective properties of olive oil have been documented more than any other source of MUFAs.

[1] Data from the North American Olive Oil Association: http://www aboutoliveoil.org/aboutoliveoil/index.asp

A comprehensive search of the literature related to MUFAs and CHD identified 77 dietary intervention studies published since 1989. The data in Table 2 provide a categorization of this research according to the source of MUFA studied.

Table 2
Categorization of Dietary Intervention Studies with Respect to Source of MUFAs

Source of MUFAs	**Number of studies**
Olive oil	35
Multiple sources including olive oil	17
Canola/rapeseed oil	8
High oleic sunflower oil	5
High oleic safflower oil	1
Multiple sources not including olive oil	3
Not specified	2
Meta-analyses	6
TOTAL	77

Approximately two-thirds of the studies identified in this area used olive oil (alone or in combination with other oils) as a source of MUFAs – far greater than for any other single fat or oil. These studies are summarized in Appendix B. Studies pertaining to other sources of MUFAs that were identified are: eight studies pertaining to canola oil (Truswell *et.al.*, 1992; Valsta *et.al.*, 1992; de Lorgeril *et.al.*, 1994; Gustafsson *et.al.*, 1994; Howard *et.al.*, 1995; Sundram *et.al.*, 1995; Jenkins *et.al.*, 1997; Hodson *et.al.*, 2001), five studies using high-oleic sunflower oil (Wardlaw *et.al.*,1990; Clevidence *et.al.*, 1997; Gumbiner *et.al.*, 1998; Ashton *et.al.*, 2001; Vessby *et.al.*, 2001), one study with high-oleic safflower oil (Grundy, 1986), three studies using blends of MUFA sources that did not include olive oil (Grundy *et.al.*, 1988; Luscombe *et.al.*, 1999; Parker *et.al.*, 2002) and two studies where the source of MUFAs was not identified (Ginsberg *et.al.*, 1990; Muller *et.al.*, 2003).

The NAOOA believes that olive oil should be specified as a source of MUFAs for the proposed claim based on the preponderance of evidence available to assess its cardioprotective properties. If additional information is provided to the agency with respect to other sources of MUFAs, the regulation authorizing the health claim can easily be amended.

4. Olive oil contains substances other than MUFAs that likely contribute to its cardioprotective properties

The effect of minor constituents in olive oil on CHD and other chronic diseases has received considerable attention. Olive oil is unique among common edible oils because it is obtained by physically pressing washed, crushed olives rather than extracting the lipid component with organic solvents. This technique allows numerous minor constituents in olives to be transferred to the oil (Owen *et.al.*, 2000).

The non-glyceride components of olive oil include hydrocarbons, non-glyceride esters, tocopherols, alkanols, flavonoids, anthocyanins, hydroxy- and dihydroxyterpenic acids, sterols, phenolic constituents and phospholipids (Visioli and Galli, 1998). The phenolic compounds in olive oil have received the most attention because they are absorbed by humans in a dose-dependent fashion (Visioli *et.al.*, 2000a), are potent antioxidants and may have additional protective effects (Visioli *et.al.*, 2000). The phenolic compounds present in olive oil include:

- Caffeic acid
- Hydroxytyrosol
- Hydroxytyrosol esters
- Ligstroside
- Oleuropeine
- Synapic acid
- Syringic acid

- Tyrosol
- Vanillic acid

Several recent papers (Visioli and Galli, 1998; Visioli *et.al.*, 2000; Owen, 2000; Visioli *et.al.*, 2002) have reviewed the mechanisms by which olive oil phenolic compounds are thought to be cardioprotective. Such compounds have been shown to inhibit LDL-C oxidation *in vitro* and *in vivo* (Visioli *et.al.*, 2002). In addition, a recent study (Oubiña *et.al.*, 2001) with 14 post-menopausal women fed extra virgin olive oil or high-oleic acid sunflower oil found that serum peroxides and thromboxane B_2 levels in stimulated platelet-rich plasma were significantly higher after a sunflower oil (i.e. PUFA) diet than after an olive oil diet.

Other documented biological activities of olive oil phenolic compounds include: inhibition of apoprotein derivatization; inhibition of platelet aggregation; scavenging of superoxide and other reactive oxygen species; inhibition of peroxynitrite-induced tyrosine nitration; scavenging of hypochlorous acid; increased nitric oxide production by LPS-challenged macrophages; inhibition of neutrophil respiratory burst; inhibition of bacterial growth and activity; cytostasis; hypotensive action; decreased isoprostane excretion in humans and in sidestream smoke-exposed rats; and increased plasma antioxidant capacity (Tonga *et.al.*, 2003; Visioli *et.al.*, 2002).

In summary, olive oil is uniquely qualified for designation as a source of MUFAs with respect to the proposed claim because it is a rich source of oleic acid, contains a wide range of other compounds which have been shown to have potentially cardioprotective properties *in vitro* and *in vivo*, is well-accepted by consumers and has been more extensively studied than other edible oils.

D. The role of MUFAs in CHD risk management

1. Observational studies

Epidemiologic studies have shown that diets containing MUFAs and other unsaturated fatty acids are associated with reduced risk of CHD compared to diets higher in SFA or TFA. Early work by Keys *et.al.* (1986) reported a significant inverse association (r = -0.42) between MUFA intake and the 15-year death rate among 11,579 members of the Seven Countries Study cohort. More recently, Ascherio (2002) reviewed the prospective cohort studies on dietary fat and CHD and made the following conclusions with respect to MUFAs,

> Oleic acid is the major monounsaturated fat in the diet and is found in animal fats, dairy foods, meat, vegetable oils (notable olive and canola [rapeseed] oils), and nuts. Replacement of saturated fat with monounsaturated fat is associated with a 30% reduction in CHD, 3 times more than that associated with replacement of saturated fat with carbohydrate.

Seven observational studies published since 1989 were identified that are germane to the proposed claim. These studies will not be reviewed in detail because the agency places much greater weight on the results of randomized intervention studies in considering whether to authorize health claims, but the results of these studies are briefly noted below.

Trevisan *et.al.* (1990) found that consumption of olive oil (and other vegetable oils) was associated with lower serum cholesterol and systolic blood pressure in a cohort of 4,903 Italian men and women aged 20 to 59 years.

Posner *et.al.* (1991) found that dietary energy from total fat and MUFAs was positively associated with CHD in a younger cohort (aged 45-55 years) of the Framingham population.

There was no association between dietary lipids and CHD in the older cohort (56-65 years). The authors noted that a partial explanation for this finding may be that the MUFAs consumed by this cohort are largely derived from animal products that are also rich sources of SFAs.

Similar results were obtained by Esrey *et.al.* (1996) from the Lipid Research Clinics Prevalence Follow-Up Study cohort. Dietary intake from 4,546 subjects was analyzed with respect to CHD incidence during a 12-year follow-up period. There was no association between dietary lipids and CHD among older participants (60-79 years), but a positive association between total energy, SFAs and MUFAs after adjusting for age, gender, energy intake, serum lipids and other CHD risk factors. The authors noted that uncontrolled variables such as fruit and vegetable intake may have confounded the results. In addition, TFA intake was not accounted for in this study.

Hu *et.al.* (1999) reported that MUFAs and PUFAs were inversely associated with CHD among 80,082 members of the Nurses' Health Study cohort during a 14-year follow-up period. The data showed that CHD risk increased when carbohydrates were substituted for MUFAs or PUFAs with the greatest effect due to PUFAs. Intake of SFAs and TFAs were positively associated with CHD among these subjects. The size and established nature of this cohort adds to its credibility compared to the smaller studies discussed above.

A small prospective cohort study with 330 elderly Australian subjects followed for approximately five years reported a 17% reduction on overall mortality associated with

consumption of a Mediterranean-type diet (Kouris-Blazos *et.al.*, 1999). MUFA intake was also significantly associated with reduced all-cause mortality.

A recent case-control study (Fernández-Jarne *et.al.*, 2002) reported significantly reduced risk of a first heart attack among subjects in the upper quintile of energy-adjusted olive oil consumption among 171 heart attack patients compared to 171 hospital-matched controls.

In summary, observational studies in this area are consistent with the hypothesis that dietary MUFAs and other sources of unsaturated fatty acids are inversely associated with CHD risk. However, such studies are of limited use in confirming this hypothesis due to the limited quality of some studies, the fact that fatty acid composition of diets is more difficult to accurately assess than the intake of individual foods, the possibility of uncontrolled confounding variables and the inherent inability of epidemiologic studies to prove causality.

2. Intervention studies

A review of the criteria FDA has used to identify germane studies for consideration of CHD-related health claims was described in the Interim Final Rule for the Sterol/Stanol ester health claim (65 FR 54686 at 54691). These criteria include:

> ...(1) Present data and adequate descriptions of the study design and methods; (2) be available in English; (3) include estimates of, or enough information to estimate, intakes of plant sterols or stanols and their esters; (4) include direct measurement of blood total cholesterol and other blood lipids related to CHD; and (5) be conducted in persons who represent the general U.S. population. In the case of criterion (5), these persons can be considered to be adults with blood total cholesterol levels less than 300 mg/ dL, as explained below.

This document also provided additional information on the criteria FDA uses to assess the strength of individual studies in establishing a link between dietary components and serum lipid biomarkers (65 FR 54686 at 54692):

> The general study design characteristics for which the agency looked included selection criteria for subjects, appropriateness of controls, randomization of subjects, blinding, statistical power of the studies, presence of recall bias and interviewer bias, attrition rates (including reasons for attrition), potential for misclassification of individuals with regard to dietary intakes, recognition and control of confounding factors (for example monitoring body weight and control for weight loss), and appropriateness of statistical tests and comparisons. The agency considered whether the intervention studies that it evaluated had been of long enough duration, greater than or equal to 3 weeks duration, to ensure reasonable stabilization of blood lipids.

The minimum level of reduction in T-C or LDL-C that the agency considers necessary for the authorization of a health claim has not been rigidity defined. However, a decrease in total cholesterol of 4.4 percent (10.0 milligrams mg/dL) and in LDL-C of 4.9 percent (7.8 mg/dL) was regarded as significant in authorizing a health claim for oats and coronary heart disease (62 FR 3584 at 3586), and similar levels were used to justify authorization of the health claim for soy protein and CHD (64 FR 57700 at 57708).

The role of MUFAs in the management of CHD risk was reviewed by the Food and Nutrition Board of the National Academy of Sciences (1989) in its landmark publication, *Diet and Health. Implications for Reducing Chronic Disease Risk (Diet and Health)*. This report summarized early work by Keys and Hegsted that showed MUFA lowers T-C when substituted for SFAs in the diet, but that the isocaloric replacement of MUFAs for dietary carbohydrate had no discernable effect. However, the report also noted that later work by investigators including Grundy, and Mensink and Katan found, "... that high-MUFA diets did not lower HDL-C

concentrations as did replacement of SFAs by carbohydrates" (FNB, 1989). The report concluded,

> The effects of MUFAs on serum lipoprotein levels are important, because olive oil or other oils rich in these fatty acids could be used, along with or as an alternative to carbohydrates, as replacement of SFAs in diets designed to lower serum cholesterol, and particularly, LDL-C levels. If the early and recent findings regarding the effects of MUFAs on LDL-C and HDL-C levels are consistently confirmed, the use of oils rich in these fatty acids makes possible the design of diets that would lower LDL-C levels, maintain HDL-C levels, and be more palatable than very-low-fat diets.

Ample evidence has been published since the *Diet and Health* report to confirm that dietary MUFAs lower T-C and LDL-C when fed at the expense of dietary SFAs, and that these fats do not have deleterious effects on serum lipids as carbohydrates do. In addition, newer data suggest that MUFAs may be more desirable than PUFAs as a replacement for SFAs because they have comparable effects on CHD biomarker risk profiles but do not increase the susceptibility of serum lipids to oxidation (Hargrove *et.al.*, 2001). The NAOOA believes that olive oil has the potential to make important public health contributions with respect to each of these three dietary strategies (i.e. as a direct replacement for dietary SFA and as alternatives to dietary PUFAs and carbohydrates for replacing SFAs). We have therefore grouped the intervention studies that will be discussed below into to these categories.

All randomized, controlled studies designed to assess the effect of olive oil on CHD published since the *Diet and Health* report that provided olive oil as a distinct source of MUFAs, had an intervention period of at least three weeks and were conducted in healthy individuals are discussed in detail below. These studies meet FDA's criteria for consideration in the

authorization of health claims. Six studies that used subjects with diabetes mellitus are also discussed. These studies are included because the management of CHD-risk is especially important in this population, and olive oil may have beneficial effects on glucose and insulin control in addition to its cardioprotective properties. Furthermore, there were an estimated 16.7 million type-2 diabetics in the U.S. in 2001 (Mokdad, 2003), and the incidence of this condition is predicted to increase dramatically in parallel with escalating obesity rates. We therefore believe studies using non-insulin dependent diabetes mellitus (NIDDM) subjects have special relevance from a public health perspective, and should be considered with respect to the proposed claim.

A tabular summary of all studies involving olive oil, regardless of FDA's quality criteria, is provided in Appendix B. These studies include intervention trials published since 1989 that used olive oil as a source of MUFAs either exclusively or in conjunction with other foods. In addition, studies conducted using non-healthy subjects (e.g. severe hypercholesterolemia, end stage renal disease, myocardial infarction patients) and studies with short intervention periods or other design limitations are included in Appendix B to provide the agency with a thorough representation of the available literature pertaining to olive oil. In addition, taken collectively, these studies provide additional, albeit suggestive, evidence that olive oil reduces the risk of CHD.

a. Olive oil vs. saturated fatty acids

Ng *et.al.* (1992) compared the effects of a diet rich in SFA with a diet rich in MUFA on serum lipids and lipoproteins in 33 normocholesterolemic men and women. All subjects were fed a

control diet for four weeks in which coconut oil was the sole cooking fat used. The participants were then randomized to diets in which coconut oil was replaced with either olive oil or palm oil using a crossover design protocol with six weeks per dietary intervention. There was no washout period used between the test diets. Cooking fats provided 23% of daily energy, and approximately two-thirds of the total daily fat intake. Fatty acid profiles of the diets differed in that intakes of SFA were highest during the coconut phase (approximate levels as a percent of energy: SFA 26%, MUFA 4%, PUFA 1%), while intakes of MUFA were highest during the olive oil phase (SFA 8.5%, MUFA 22%, PUFA 3%). The palm oil diet contained intermediate levels of both SFA and MUFA (SFA 16%, MUFA 14%, PUFA 4%). Subjects were provided with their major meal (lunch) at the research kitchen, and with test oils for cooking at home. All subjects kept daily dietary records including a cooking oil logbook. Unused cooking oil was returned at the end of each successive phase. Two research dietitians, who met with subjects at the research center at least twice, and at the subject's homes at least once during each intervention phase, monitored compliance. Based on the record of attendance for lunch served at the research center (averaging 78%), and on diet records from home, compliance was estimated to be close to 90%. Several changes in serum lipid and lipoprotein levels were evident between the olive oil and coconut oil phases. Serum T-C was significantly lower after the olive oil phase compared to the coconut oil phase, by 15.8% in men (197mg/dL vs. 234mg/dL) and 17.2% in women (188mg/dL vs. 227mg/dL). Serum LDL-C was also significantly reduced by olive oil consumption, by 18.8% in males (165mg/dL vs. 134mg/dL) and 19.6% in females (158 mg/dL vs. 127mg/dL). Serum TG levels were 11.9% lower in men, and 15.6% lower in women consuming the olive oil diet. HDL-C was 7.5% lower in men consuming the olive oil diet compared to the coconut oil diet, but did not differ significantly between dietary phases in

women. There were no significant differences in any lipid or lipoprotein parameters measured between the olive oil and palm oil phases. This study demonstrated that substitution of SFA with MUFA results in a more favorable lipoprotein profile. The authors state that replacement of lauric and myristic acid with palmitic acid plus oleic acid also appears to have a beneficial impact on an important index of thrombogenesis (that of the thromboxane/prostacyclin ratio in plasma).

Mata et.al. (1992) investigated the effects on plasma lipoproteins of replacing dietary SFA with MUFA from olive oil. Following a four week run-in period during which time energy requirements were assessed by 24-hr recall and a food frequency questionnaire, 21 healthy, normolipidemic women first received a moderately high fat diet (35%) rich in saturated fat (SFA19%, MUFA14%) for four weeks, followed by a diet rich in MUFA (SFA 11%, MUFA 22%) for six weeks, in two consecutive phases. Meals were identical between the study periods, with only the type of oil used in the preparation of the meals differing between study periods. Intakes of cholesterol, fiber, carbohydrate, PUFA, and protein were held constant between dietary interventions. Subjects were teachers living at a boarding school where all meals were prepared in the facility's kitchen and consumed in the school's dining hall. One of the investigators was present twice weekly during preparation of meals, and a detailed record was kept of the food and oil used in meal preparation. Compliance was monitored using a daily questionnaire, and by measurement of fatty acids in the cholesterol-ester fraction in plasma. Analysis of meal homogenates indicated that actual intakes of MUFA and other dietary components, as a percentage of energy, were highly similar to calculated values. Plasma T-C decreased by 8.9% (5.27mmol/L to 4.80 mmol/L) in subjects after consumption of the high-

MUFA diet, compared with the high-SFA diet. This change was accounted for largely by an 18.6% (3.44mmol/L to 2.80mmol/L) decrease in LDL-C. Both HDL-C and TG increased significantly during the MUFA phase of the trial. Plasma HDL-C increased by 5.6% (1.42mmol/L to 1.50mmol/L), and TG increased by 10.5% (0.95mmol/L to 1.05mmol/L). The authors conclude that in this female population a diet rich in MUFA produced a lipoprotein profile consistent with decreased atherogenic risk. The authors further state that MUFAs could be the fat of choice to substitute for the excess of SFA in the Western diet.

Kris-Etherton (1993) used a randomized, controlled, double-blind, crossover design to study the effects of whole food diets on plasma lipids among 39 healthy, normocholesterolemic (mean T-C: 120-205 mg/dL), non-obese (mean body weight: 70 kg, BMI 23) young (mean age: 26 years) men. The study was designed to test the effect of individual saturated fatty acids (as well as MUFAs and PUFAs) from whole food sources including cocoa butter, dairy butter, olive oil and soybean oil. Twelve subjects served as controls and consumed their habitual diet (34% energy from fat, 11.1% SFA, 12.3% MUFA, 7.2% linoleic acid), 48% CHO, 16% protein, 2% alcohol and 318 mg cholesterol. The remaining 18 subjects were randomized to one of four experimental diets using a Latin Square design. All diets had similar amounts of total fat (37%), CHO (51%,), protein (13%), cholesterol (360 mg) and dietary fiber (18g). The fatty acid distribution of the diets were: Olive oil (OO): 6.0% SFA (0.01% 12:0, 0.11% 14:0, 4.5% 16:0, 1.4% 18:0), 27.2% MUFA, 2.3% 18:2.; cocoa butter (CB): 20.9% SFA (0.01% 12:0, 0.18% 14:0, 9.3% 16:0, 11.4% 18:0), 13.2% MUFA, 2.1% 18:2; soybean oil (SO): 6.3% SFA (0.01% 12:0, 0.11% 14:0, 4.5% 16:0, 1.7% 18:0), 10.1% MUFA, 17.8% 18:2; and butter (B) 21.0% SFA (08% 12:0, 3.5% 14: 0, 9.3% 16:0, 4.5% 18:0), 10.1% MUFA, 1.7% 18:2. The investigators

furnished all food. Breakfast and dinner (which provided 90% of the test fat) was consumed at the research center under supervision. Compliance was assessed by dietitians who recorded food and beverage consumption twice each day. In addition, fecal fatty acids were determined after each test diet and reflected the unique profile of the experimental diet. Energy intake was adjusted to maintain constant body weight throughout the study. The results comparing diets with OO vs. those with saturated fat (B and CB) are discussed below. The results comparing the diets containing OO and SO (a source of PUFA) are discussed in the next section of this document. As expected, the OO diet compared to the B and CB diets resulted in significant decreases in T-C (24 and 13, respectively), LDL-C (21 and 11 mg/dL) and the LDL/HDL ratio (0.5 and 0.3). There was no significant change in TG or HDL-C. The SO diets were also hypocholesterolemic, and exhibited a greater decrease in T-C and LDL-C vs. B and CB than were seen with OO. However, there was no difference in the LDL/HDL ratio after feeding the SO and OO diets. These data clearly demonstrate that OO reduces T-C and LDL-C when fed as a substitute for sources of dietary SFA.

Lichtenstein *et.al.* (1993) studied the effects of a diet rich in MUFA from olive oil on plasma lipids and lipoproteins in 15 healthy, modestly hypercholesterolemic middle-aged and elderly men and women, compared to a baseline diet similar to the typical American diet (i.e. rich in SFA). Subjects consumed a moderate fat diet (35% of energy) for 32 days, followed by each of three test diets in random order. Each test diet was based on the NCEP Step II diet, and provided a total of 30% fat, in identical foods, with the exception that 2/3 of the daily total fat intake was derived from one of three test oils – canola, corn, or olive. Both subjects and investigators were blinded to subject randomization throughout the study. All food and beverages during the trial periods were provided by the investigators or packaged for take-out. Each phase lasted 32 days,

with a one to two week washout period in between. The olive oil diet provided 17% of energy from MUFA, but only 6.9% energy from SFA, while the baseline diet provided approximately equal levels of MUFA and SFA (MUFA 12.2%, SFA 12.9%). Plasma fatty acid profiles were analyzed in part to assess compliance, and were found to be characteristic of the fatty acid content of the diet. Compared to the baseline diet, plasma T-C decreased by 7.2% in subjects during the MUFA phase (221mg/dL to 205mg/dL), due primarily to a 13.2% decrease in plasma LDL-C (152mg/dL to 132 mg/dL). Consumption of a high-MUFA diet did not significantly change HDL-C or TG levels compared to the baseline diet. The authors conclude that replacing saturated fat with MUFA as part of the current NCEP Step 2 recommendations in middle-aged and elderly modestly hypercholesterolemic subjects results in reductions in LDL-C.

Seppänen-Laakso *et.al.* (1993) utilized a randomized, parallel feeding study to determine the effect of substituting margarine on bread with water-oil emulsions of OO or rapeseed oil (RO) on serum lipids and fatty acid distribution in among 57 middle aged men (30) and women (27) in Finland. The subjects were healthy with normal to moderately elevated serum T-C concentrations (5.0 – 8.5 mmol/L) and TGs less than 3.5 mmol/L. All subjects consumed at least three slices of bread with margarine daily. The subjects were assigned to either a control group (n=11), an OO group (n=23) or a RO group (n=23) alphabetically according to last name at the beginning of the study. There were no significant differences in gender distribution or age among the groups. The control group continued to consume their habitual diet throughout the experiment. The OO and RO groups replaced margarine with water-oil emulsions of the two test fats, respectively for six weeks followed by resumption of the baseline diet for an additional six weeks. No other dietary changes were made. There were no significant differences among the

diets in energy, protein, carbohydrate, alcohol, cholesterol or dietary fiber. The MUFA content of the OO diet was significantly higher than the baseline or control diets (16.8 % of energy vs. 13.1 and 13.2, respectively), and lower in PUFA (6.0, 7.1 and 5.5) but there were no significant differences in SFA (13.6, 15.2 and 15.5),. The RO diet contained significantly more PUFA than the baseline or control diets (9.4 % of energy vs. 6.8 and 5.5, respectively), but the MUFA (15.7, 14.0 and 13.2) and SFA (14.9, 15.8 and 15.5) concentrations were statistically similar. The subjects maintained 3-day dietary diaries before and towards the end of the substitution period, and consumption of the oil emulsions was monitored at three and six weeks. Dietary compliance was evident from changes in the concentration of serum fatty acids, which mirrored the fatty acid distribution on the test fats. The OO diet resulted in a significant decrease in serum LDL-C compared to baseline at three weeks (4.25 mmol/L vs. 3.93) but the effect was not apparent at the end of the intervention period (4.08 mmol/L). Nevertheless, subjects in this group experienced a significant decrease in LDL-C (3.93 mmol/L) at the end of the study (after an additional six weeks of resuming the habitual diet). There were no significant changes in serum T-C at three or six weeks, but the 12-week value was significantly lower than baseline (6.28 vs. 5.93 mmol/L, respectively). HDL-C did not change with OO feeding throughout the experiment. There were no changes in T-C or LDL-C in response to RO feeding during the substitution phase, but there was a significant decrease in these values compared to baseline at the end of the 12-week study (T-C = 6.07 mmol/L vs. 5.72; LDL-C = 4.13 vs. 3.82). HDL-C concentrations increased in response to RO feeding at three weeks compared to baseline (1.33 mmol/L vs. 1.41, respectively), but the effect was no longer evident at the end or the six-week substitution period (1.37 mmol/l). There were no significant changes in body weight during the experiment. It is unclear why the effects of OO and RO on serum lipids that were evident after 3 weeks of

intervention did not persist after an additional three weeks of feeding the test fats. Nor is it clear why changes in T-C and LDL-C were apparent at the end of the experiment after the habitual diets were resumed for six weeks. The relatively subtle changes in dietary fatty acids may partially explain the results. In addition, although the lipid profiles of the control group did not change significantly during the 12-week experiment, there was a downward trend. It is possible a similar trend contributed to the significant differences seen at the end of the study in the intervention groups. Despite the somewhat confusing results, this study shows that OO can have positive effects on LDL-C when fed as a replacement for margarine in free-living subjects. This change occurred despite a non-significant change in the SFA content of the diet.

Choudhury *et.al.* (1995) compared the effects of feeding a diet rich in MUFA with a diet rich in SFA for 30 days on plasma lipoprotein profiles. Twenty-one healthy young men and women recruited from the university community participated in a randomized crossover trial, with no washout period. During the pre-experimental period, subjects completed dietary histories to estimate 'usual' dietary intakes, and were given individual dietary counselling sessions in which they were instructed to replace most of their usual dietary fat with the test oils provided (either palm oil or olive oil). Subjects were told to exclude butter, margarine, cooking oils, nuts, visible fat on meat, cream, chocolate, baked goods, eggs, avocados, olives, and poultry skin, and to restrict occasions of eating out and take-away foods to not more than once a week. Sheets of printed information were used to reinforce dietary protocols. Skim milk, reduced-fat milks, grain and cereal products, fruits and vegetables, jams, and soft drinks were all allowed. Subjects received test oils weekly, individualized as approximately 50% of their usual dietary fat intake. All meals were prepared by participants at home. Food intakes were measured by assessment of

daily food records completed throughout the study. Compliance was monitored by weekly phone contacts by a dietitian, and by review of food diaries and of household daily records of test oil use when subjects came to collect their test oils. Energy intakes remained relatively constant on the test diets compared to baseline, although fat intakes fell slightly (palm oil 30% of energy, olive oil 31%, baseline 34%). Body weights did not change significantly during the trial. All subjects were reportedly able to incorporate the test fats into their daily menu, which was confirmed by measurement of individual fatty acids in different plasma lipid fractions and platelet membrane lipids. MUFA intake was determined to be 19% of energy during the olive oil phase, 12% during the palm oil phase, and 13% at baseline. Corresponding values for SFA were 8.5, 14 and 15%, respectively. Linoleic acid intake was significantly greater in the palm oil group compared to the olive oils group (4.3 % of energy vs. 3.4%, respectively). In both dietary groups, there was a decline in plasma T-C, TG, and HDL-C compared to baseline. Plasma T-C was reduced by 16.3% (5.53 mmol/L to 4.65mmol/L) and plasma TG fell by 19.5% (1.18 mmol/L to 0.95 mmol/L) in the olive oil diet group from baseline. Plasma HDL-C inexplicably fell 37.5% in subjects consuming the olive oil rich diet, but also fell by 22.9% in subjects during the palm oil phase. Plasma LDL-C decreased by 6% during the olive oil phase, although this effect was not significant. Both palm oil and oleic acid rich diets effectively lowered plasma T-C compared to a typical Western diet (the baseline diet). The authors concluded that in young adults of normal weight and plasma cholesterol concentrations, exchange of oleic for palmitic acid at 17% of dietary energy in diets low in cholesterol resulted in identical plasma T-C and LDL-C concentrations. The lack of an effect of olive oil compared to palm oil may partially be explained by the difference in linoleic acid, but is still surprising. As the authors note, palmitic acid may be less hypercholesterolemic than lauric and myristic acids. These results may have

limited practical application in the U.S. because palm oil is used infrequently[2], and the leading sources of SFA (e.g. dairy products, red meat) are rich in lauric and myristic acids. Nevertheless, this study provides strong evidence that OO can have a favorable affect of serum lipids when compared to a baseline diet that is significantly higher in palmitic as well as lauric and myristic acids as was the case in this study.

Nicolaïew *et.al.* (1998) studied the effect of feeding diets rich in MUFA from extra virgin olive oil or high-oleic sunflower oil on serum lipids using a randomized, crossover design. Ten lean (mean BMI = 22.4), normocholesterolemic men (mean T-C = 4.66 mmol/L) aged 25-40 years were randomized to one of the test diets for three weeks before being switched to the alternate diet after a one-week washout period. The baseline (i.e. usual diet) was consumed during this period. The test diets contained 35% of energy from fat (10.5% SFA; 21% MUFA and 3.5% PUFA). The composition of the test diets was identical except for the source of MUFA. Changes in serum lipids were compared to the baseline diet which had a mean fatty acid composition of 16% of energy from SFA, 15% MUFA and 5% PUFA. The diets consisted of common foods including fruits, vegetables, starches, meats, fish and dairy products. The quantities of dairy products, low-fat meat, and fish were indicated precisely. The subjects were not permitted to consume alcohol. Compliance was determined by analysis of the fatty acid composition of plasma TGs and phospholipids, which showed an increase in oleic acid concentration following both experimental diets. The OO diet resulted in a decrease in serum T-C of 5.2% (5.00 to 4.74 mmol/L) and in LDL-C of 9.2% (3.37 to 3.06 mmol/L), compared to the usual diet but the changes were not statistically significant. There was also no significant difference in these biomarkers after the SO diet. HDL-C and TGs were unchanged by the OO or

[2] USDA, Oil Crops Situation and Outlook Yearbook, October 2002

SO diets at the end of the intervention period compared to the baseline diet. Changes in body weight were not reported. The primary purpose of this study was to examine the effect of non-lipid constituents of OO on LDL-C oxidation rather than its affect on serum lipids. Consequently, the study used a small number of normocholesterolemic subjects, which probably explains why the changes in serum lipids did not reach statistical significance. Nevertheless, this study provides suggestive evidence that OO imparts favorable changes to CHD biomarkers when it replaces dietary SFA.

Kris-Etherton *et.al.* (1999) compared the lipid and lipoprotein profiles of 22 healthy men and women after consumption of an Average American Diet (AAD) or three intervention diets high in MUFA for four weeks in a double-blinded, randomized, cross-over trial. MUFAs in the intervention diets were provided by olive oil (OO), peanut oil (PO), or peanuts and peanut butter (PPB). The investigators supplied all foods during the trial periods, and subjects were not allowed to consume non-study foods or beverages. Subjects ate breakfast and dinner on weekdays at the metabolic diet study center, and consumed pre-packaged meals for lunch and on weekends. Compliance was monitored by body weight measurements and a dietary assessment questionnaire administered daily. Subjects were allowed a four to 11 day break between study periods in order to enhance compliance. While fat intake remained fairly constant across study phases, at 34 to 36% of energy, dietary intakes of MUFA were higher during the MUFA intervention periods, accounting for 17 to 21% of energy, versus only 11% of energy during the AAD phase. Most of this difference resulted from a decrease in SFA intake during the MUFA interventions to 7 to 8% of energy from 16%, although there was some variation in PUFA intakes (PUFA: OO 6% of energy, PO 9%, PPB 10%). All three MUFA intervention diets

resulted in similar, significantly lower serum T-C, LDL-C, and TG compared to the AAD, without adversely affecting HDL-C levels. Compared to the AAD, subjects on the OO diet had 11.5% lower T-C (4.79 mmol/L vs. 5.41 mmol/L) and 15.3% lower LDL-C (2.98 mmol/L vs. 3.52 mmol/L). TGs were 13.5% lower in subjects on the OO diet (1.15mmol/L vs. 1.33 mmol/L). HDL-C levels were almost identical in subjects on the OO diet compared to the AAD group (1.28 mmol/L vs. 1.29 mmol/L, respectively). This study also included a fifth dietary group based on the AHA Step II diet that was low in fat (25% of energy). Compared to this group, the OO diet group had a 22.3% lower TG (1.15 mmol/L vs. 1.48 mmol/L), but did not differ significantly in T-C, LDL-C or HDL-C levels[3]. The authors concluded that "a high-MUFA, cholesterol-lowering diet is superior to a low-fat diet such as the Step II diet" especially since, unlike the Step II diet, the high-MUFA diet lowers TG and does not decrease HDL-C.

Williams et al (1999) investigated the effects of a diet rich in MUFA with the typical diet of the United Kingdom (high in SFA) on blood lipids and lipoproteins. In a randomized, single blinded, cross-over trial, 23 young men with a family history of CHD, and 30 healthy middle-aged men consumed diets containing identical foods differing only in fat type, for a period of 8 weeks, with a 4 to 6 week wash-out period between trials. Foods provided by the investigators accounted for approximately two-thirds of individuals total daily fat intakes, and were in the form of spreads and cooking fats made from olive oil[4] for use in meal preparation, biscuits, puddings, and pre-made meals. Commercially available low-fat meals, potatoes, stir-fry vegetables, and rice were also provided to offer more choices, and subjects were able to add the

[3] The superscript for the T-C value of the Step II diet in Table 2 should be "b" rather than "a" as printed in the paper (personal communication with the senior author).
[4] The paper did not report the MUFA source used to make the fats used for the experimental diets, but personal communication with the authors revealed that it was olive oil.

cooking fats and spreads to these. The remaining one-third of fat in the diet came from milk, cream, ice cream, and yogurts, and from meals consumed during the two 'free-meal' periods each week. Free-meal periods were included to aid compliance, and subjects were guided in the amounts and types of foods that were allowable. In a further effort to optimize compliance, foods were also provided for subjects' partners for use during food preparation. Dietary intakes were assessed from diet diary records completed during weeks 3 and 7 of each arm of the study, and covering four diet days and one free day. Dietary compliance was self-assessed after 4 and 8 weeks on each arm of the study, using a questionnaire that incorporated a linear-analogue scale. Dietary intakes of MUFA were significantly higher during the MUFA phase of the trial compared to the SFA phase in both young men (18.3% of energy versus 13.8%, respectively) and middle-aged men (17.2% vs 13.2%). This difference occurred largely at the expense of total SFA, as PUFA intake did not differ significantly across diets in either age group studied. Within age groups, total energy intake derived from fat (38%), and total daily energy intake, did not differ significantly between diet phases. Middle-aged men showed an 11% decrease in LDL-C from baseline on the MUFA diet, with no change in response to the high-SFA control diet. Young men, however, showed a decrease in LDL-C of 7.8% on the MUFA diet with a 6.2% increase on the SFA diet. When data were pooled for all subjects, T-C was significantly lower, by 10% (5.0 mmol/L vs. 4.5mmol/L) at the end of the MUFA period compared to the end of the SFA period. LDL-C was also significantly lower after consumption of the MUFA rich diet, falling by 13.9% (from 3.6 mmol/L to 3.1mmol/L). There were no differences in HDL-C or TG between different diets or from baseline to the end of either diet phase. A significant but small weight gain of approximately 1 kg was experienced by subjects in both groups compared to baseline. The authors conclude that "a high-MUFA diet offers a practical means of achieving

target SFA intakes and of reducing plasma total and LDL-cholesterol concentrations, and may be more acceptable to some consumers than advice to reduce fat intakes to levels less than 33% of dietary energy".

Fuentes *et.al.* (2001) measured the effects of a diet rich in SFA on plasma lipids and lipoproteins compared with a diet rich in MUFA or the NCEP-1 diet (low fat, low SFA). During a 28-day baseline period, 22 healthy, hypercholesterolemic men were fed a high-fat diet rich in SFA (38% of energy as fat, SFA 20%, MUFA 12%). Subjects were then randomly assigned in a crossover design to two diets for another two, 28-day periods without washout: the NCEP-1 diet (28% total fat), and a diet high in MUFA (a typical Mediterranean diet enriched with olive oil). The high-MUFA diet was also high in fat (38% of energy) but this was primarily in the form of MUFA (22%), with relatively low intakes of SFA (10%). All three diets provided 6% of energy as PUFA. Compliance was determined by measuring fatty acid enrichment of plasma lipoproteins at the end of each phase for comparison with dietary fatty acid profiles. Participants also kept food diaries where they recorded any significant dietary events. Plasma T-C was significantly lower, by 7.6%, in subjects after the high-MUFA diet phase than at the end of the high-SFA phase (6.0mmol/L vs. 6.5mmol/L). Plasma LDL-C was also significantly lower after the high-MUFA compared to the high-SFA phase, by 13.5% (3.8mmol/L vs. 4.4mmol/L). Plasma TG and HDL-C levels did not vary significantly between the high-SFA phase and the high-MUFA phase. The decrease in plasma LDL-C, coupled with the lack of change in HDL-C, contributed to the significant 8% decline in LDL-C/HDL-C ratio observed after the MUFA period compared to the SFA period (3.24 vs. 3.52). In this study, replacement of SFA with MUFA also resulted in improved endothelial function. The authors concluded that it may be possible to correct early

expression of the atherosclerotic disease process by prescribing a lipid-lowering diet, such as a diet rich in MUFA.

In summary, the scientific literature published since 1989 confirms the conclusions of the *Diet and Health* report that MUFA-containing diets lower serum T-C and LDL-C when fed as a replacement for SFAs. All but one of the studies reviewed in this section showed that olive oil significantly reduced serum T-C and/or LDL-C compared to a baseline diet higher in SFAs. The only exception (Nicolaïu *et.al.*, 1998) reported reductions in these lipids after feeding an olive oil-containing diet but the results were not statistically significant. However, the study used a small sample size because its primary objective was to examine the effect of olive oil on LDL oxidation.

Two studies (Ny *et.al.*, 1992; Choudhy *et.al.*, 1995) reported that both olive oil and palm oil reduced T-C and LDL-C compared to baseline diets with relatively high concentrations of lauric and/or myristic acid. These results are consistent with the recent meta-analysis of Mensink *et.al.* (2003) that showed lauric and myristic acids increase T-C significantly more than palmitic acid when fed as an isocaloric replacement of for carbohydrate. A similar pattern was seen with respect to LDL-C, but the difference was statistically significant only for lauric acid. (Interestingly, all three of these fatty acids increased HDL-C as well as T-C and LDL-C so that their affect on the T-C:HDL-C ratio was small.)

In addition, a meta-analysis of olive oil studies was conducted at the request of the NAOOA to objectively assess the effect of olive oil consumption on serum lipids[5] (this report is provided in Appendix C). This analysis included human intervention studies published in English that used randomized, controlled parallel or crossover designs. Studies were required to report data (including appropriate variance estimates) in sufficient detail to allow the calculation of mean treatment minus control effects. In addition, an intervention period of three weeks or longer was required and only studies with healthy participants (including subjects with moderate hypercholesterolemia) were considered.

Five studies (providing six treatments) that compared the effect of olive oil with a higher saturated fat diet met the inclusion criteria for the meta-analysis (Choudhury *et.al.*,1995; Kris-Etherton *et.al.*, 1993; Mensink and Katan, 1989; Ng *et.al.*, 1992; Williams *et.al.*, 1999). The results of this analysis are presented in Table 3. The olive oil intervention groups experienced a significant reduction in T-C (-8.8%) and in LDL-C (-11.3%) compared to diets higher in SFA. There were no changes in HDL-C or TG.

Table 3
Overall Effect of Olive Oil as Compared to Saturated Fat Diets

Variable	Pooled Effect Size (mg/dl)	95% Confidence Interval (CI)	Percent Change: Olive oil vs. Saturated Fat
Total Cholesterol	-17.7	-10.0 -25.3	-8.8
LDL-Cholesterol	-15.5	-8.64, -22.3	-11.3
HDL-Cholesterol	-1.97	0.23, -4.17	-4.4
Triglycerides	-3.22	7.15, -13.5	-3.1

*$p<0.05$

[5] Fulgoni, V. "Effects of Olive Oil on Blood Lipids: Results of a Meta Analysis of Human Clinical Trials – Final Report. Nutrition Impact, LLC." August 8, 2003.

Taken together, the studies discussed in this section provide strong support for the well-accepted premise that diets containing olive oil exerts a hypocholesterolemic effect when used to replace sources of SFAs (e.g. butter) in the American Diet. This conclusion is confirmed by the meta-analysis of olive oil studies provided in Appendix C.

b. Olive oil vs. PUFA

Mensink and Katan (1989) compared the effects of a MUFA-rich diet to a PUFA-rich diet on serum lipids using a randomized parallel intervention protocol. Fifty-eight healthy adult men and women paired by sex and oral contraceptive use consumed a control diet high in SFA (19.3% of total energy) for 17 days and were then randomly assigned to a MUFA-rich diet enriched with olive oil or a PUFA-rich diet enriched with sunflower oil for the next 36 days. A total of 6.5% of the SFA content of the control diet was replaced with MUFAs and PUFAs in the MUFA and PUFA-rich diets, respectively. Both of the experimental diets were also supplemented with a margarine high in linoleic acid. Subjects consumed 35.7g/d MUFA on the control diet at baseline (11.5% MUFA, 4.6% PUFA, 19.3% SFA), 47g/d in the MUFA-rich group (15.1% MUFA, 10.8% PUFA, 12.9% SFA) and 34g/d on the high-PUFA diet (7.9% MUFA, 12.7% PUFA, 12.6% SFA). All subjects were provided with food for consumption at home and were allowed to choose a limited number of fat-free and cholesterol-free items that provided 9-10% of total daily energy intake. Dietary records were kept of all chosen foodstuffs and adherence to the diets was confirmed by fatty acid enrichment of the serum cholesterol esters. Consumption of the MUFA-rich diet resulted in a significant 14.1% decrease in T-C and a 17.9% decrease in LDL-C and no change in HDL-C and TG compared to baseline. The reductions in T-C and LDL-C were significantly greater in the high-MUFA group compared to

the PUFA-rich group. The authors concluded that a mixed diet rich in MUFA was as effective as a diet rich in PUFA in lowering LDL-C.

Connor *et.al.* (1993) used a randomized, double-blind, placebo-controlled crossover design to study the effect of supplementing the diet of 16 subjects with NIDDM with 15 g per day of OO or fish oil for six months. The average age of the subjects was 58.7 years (46-72) and they were overweight or obese (average BMI = 30.6; range = 26.3-35.5). A baseline diet containing 30% of energy as fat, 55% carbohydrate, 15% protein and 300 mg cholesterol was provided for three months. The SFA content of the diet was not reported. The subjects were then randomized to the OO (placebo) or fish oil supplement group (experimental group) and continued to consume the baseline diet. The fish oil preparation (Promega ®) provided 4.1 g of eicosapentaenoic acid (EPA) and 1.9 g docosahexaenoic acid (DHA) per day. The subjects were seen on a monthly basis to receive the supplements at which time body weight was measured, dietary histories were obtained and blood samples for serum lipids, fasting glucose and fatty acid composition were obtained. There were no differences between T-C (228 mg/dL for OO vs. 225 for fish oil), or HDL-C (39 mg/dL vs. 38) after supplementation. The OO placebo resulted in significantly lower LDL-C than the fish oil supplement (117 mg/dL vs. 145). However, VLDL-C and TG were lower in the experimental group compared to the OO controls. There were no differences in parameters related to diabetic control (e.g. plasma glucose, concentration, glycosylated hemoglobin, C-peptide) between the two groups. There were no changes in body weight during the experiment. This study was designed primarily to assess the safety of fish oil supplementation among NIDDM patients but provides additional data to compare the effects of OO and PUFA on serum lipids.

A study described in the previous section of this document (Lichtenstein *et.al.*, 1993) that showed feeding OO or canola oil lowers serum T-C and LDL-C compared to a diet high in SFAs also studied the effect of corn oil on these parameters. The OO and corn oil diets contained similar amounts of carbohydrate, fat and SFA (6.9% of energy) while the OO diet was higher in MUFAs (16.99% of energy vs. 8.98) and lower in PUFAs (3.85% of energy vs. 11.21) than the corn oil regimen. All three vegetable oils lowered T-C and LDL-C compared to a baseline diet with 35% energy from fat and 12.9% energy from SFA. OO lowered T-C significantly less than corn oil or canola oil (7, 13 and 12%, respectively), compared to baseline, but did not lower HDL-C as was the case with both canola oil and corn oil. All three oils had similar affects on LDL-C and there was no significant difference in the ratio of total to HDL-C. A comparison of serum lipids at the end of the treatment periods for the three oils showed that T-C was slightly (but significantly) higher after feeding OO compared to corn or canola oils (205, 194 and 194 mg/dL, respectively), but there were no differences in LDL-C, HDL-C, TG, or the T-C/HDL-C ratio. The authors concluded, "Although differential effects were seen after the consumption of the three different oil-enriched diets in some plasma lipid measures, none of these oils had a significant advantage in terms of altering the overall lipoprotein profile."

Kris-Etherton *et.al.* (1993) compared the effect of diets containing OO and Soybean oil (SO) with diets higher in SFA from butter (B) or cocoa butter (CB). The protocol and diet composition for this randomized, double-blind, controlled, crossover study was provided in the previous section of this document. The OO diet compared to the B and CB diets resulted in significant decreases in T-C (24 and 13, respectively), LDL-C (21 and 11 mg/dL) and the LDL/HDL ratio (0.5 and 0.3). However, analogous data for SO showed that the effect on T-C

(37 and 26 mg/dL) and LDL-C (31 and 20 mg/dL) were significantly greater than for OO. Nevertheless, because OO tended to increase HDL-C compared to SO (2 mg/dL), there was no difference in the LDL/HDL ratio after feeding either source of fat. The authors conclude that SO is significantly more hypocholesterolemic than OO when fed to young, healthy normocholesterolemic males and speculate that the effect is largely due to differences in the linoleic acid content of the two fats. Nevertheless, this study shows that overall risk of CHD, as measured by the LDL/HDL ratio, is equivalent between diets containing equal amounts of OO and SO.

Nydahl *et.al.*. (1994) used a randomized, controlled, crossover design to compare the effects of a diet enriched with MUFAs to one enriched with PUFAs on serum lipids. Twenty-six men and women with clinical hyperlipidemia (T-C>6.5 mmol/L, TG>2.0 mmol/L) including 7 patients with CHD, participated in two consecutive 3.5 week treatment periods with no washout between treatments. Each experimental diet supplied 30% fat as total energy (8% SFA), and contained olive oil (MUFA diet) or corn oil (PUFA diet) as the main source of fat. Participants consumed 40.5g/d MUFA and 21.6g/d PUFA on the MUFA-rich (15% energy as MUFA, 4% PUFA) and PUFA-rich diets (8% MUFA, 11% PUFA), respectively. The SFA content of the two diets was identical (8% of energy). Participants were free-living throughout the study. Subjects collected prepared food items from a metabolic ward three times a week and were given specific instructions on how the food should be stored and consumed. Participants recorded foods they were not able to eat and returned uneaten foods. The fatty acid composition of the plasma cholesterol esters at the end of each dietary treatment period confirmed compliance with the diets. Compared to baseline levels, the MUFA diet resulted in a significant 17% reduction in T-

C, and 19% reduction in LDL-C. HDL-C levels decreased 10% from baseline in the MUFA group, and there were no significant changes in TG. There were no significant differences in serum lipids between the MUFA and PUFA groups. The authors concluded that the addition of MUFA or PUFA to a low SFA diet is equally effective in reducing T-C and LDL-C.

Pedersen *et.al.*, (2000) compared the effects of different diets rich in OO, rapeseed oil and sunflower oil on plasma lipids. Eighteen lean (BMI = 23; range = 18-27), healthy subjects aged 20-28 years (mean 24 years) participated in a randomized, double-blind crossover experiment with three week treatment periods separated by washout periods of 5-12 weeks. Three diets containing OO, sunflower oil (SO) or rapeseed oil (RO) contained the same amount of total fat (35% of energy), carbohydrates (52-53 % of energy) and cholesterol (257-259 mg per 10 MJ). The total SFA content of the diets was similar (9-11% of energy), but the OO diet contained slightly more palmitic acid (19% of total fat) compared to the SO (16%) or RO (15%) diets. Total MUFA content of the OO, SO and RO diets was 21, 9 and 18% of energy, respectively while analogous data for PUFAs were 3, 15 and 7% of total energy. Subjects received all foods from the laboratory and constant weight was maintained by adjusting the diet as necessary. Lunches during the week were consumed under supervision, and other meals were provided as a package with preparation guidelines. Subjects were instructed not to change their habitual diets during the washout periods, but composition of these diets was not provided. There were no significant differences in fasting plasma TG, T-C, HDL-C or the ratios of LDL-C/HDL-C or T-C/HDL-C between the OO and SO diets, however the latter two ratios were lower after the RO diet. Subjects fed the OO diet had significantly higher LDL-C (2.16 mmol/dL) than when they were fed the SO diet (1.89 mmol/L). VLDL-C was also significantly different between the two

diets (OO = 0.33 mmol/L vs. SO = 0.27 mmol/L). In summary, this study found small, but statistically significant differences in LDL-C and VLDL-C between subjects fed OO compared to a diet high in PUFA from SO, but the ratios of T-C and LDL-C to HDL-C were unchanged. These data suggest that the effect of OO and SO on individual lipoprotein fractions in lean, normo-cholesterolemic subjects is somewhat different, but their impact on CHD risk as judged by overall lipid profiles is comparable.

An extension of the previous study (Pedersen *et.al.*, 2000) by Nielsen et.al. (2002) compared the effects of different diets rich in olive oil, rapeseed oil and sunflower oil on plasma lipids in a very similar population. Eighteen healthy adult males participated in a double-blinded randomized crossover study consisting of three periods of three weeks with strict dietary control and 4-22 weeks wash-out periods in between. The experimental diets provided 30% of total energy as fat and contained 50g per 10 MJ (2380 kcal) of olive oil (OO), rapeseed oil (RO) or sunflower-seed oil (SO) corresponding to 19% of total energy. The OO was rich in MUFA and contained 58% MUFA and 8% PUFA. The SO contained 26% MUFA and 40% PUFA. The OO and SO were identical in SFA content. The fatty acid composition of the RO was intermediate between the OO and SO and provided 49%, 22% and 28% of total fat as MUFA, PUFA and SFA, respectively. Subjects were fed lunch on weekdays and other meals for the day as well as weekend meals were pre-packaged for consumption at home. Good subject compliance was confirmed from food records and plasma analysis of TG and cholesterol ester fatty acids. Consumption of the MUFA-rich OO supplement resulted in higher fasting plasma concentrations of T-C, LDL-C and VLDL-C compared to the SO diet rich in PUFA (12.4%, 14.6% and, 17.9%, respectively). Levels of T-C, LDL-C, TG and VLDL-C were also significantly higher in the OO

group compared to RO. There were no differences in HDL-C between the dietary groups. The OO diet had favorable effects on postprandial lipoprotein oxidation characteristics. The propagation rate during *in vitro* oxidation of VLDL particles was significantly lower after three weeks on the OO diet (4.78 nmol/mg/min) compared to the SO diet (10.06 nmol/mg/min). In addition, the lag time for VLDL oxidation was significantly longer for the OO diet (190.0/min) compared to the SO diet (147.3). The authors concluded that the high MUFA OO diet resulted in higher blood lipids compared to RO and SO but reduced susceptibility of lipoproteins to oxidation. This study does not support the notion that OO exerts similar effects on serum lipids compared to PUFAs in young, lean male subjects. The relatively small sample size (n = 18) and the fact that most of the subjects were normocholesterolemic (average T-C = 4.71 mmol/L; range 2.46 – 6.01) may have influenced the results, but the study was well designed and appeared to be well executed.

Kratz *et.al.* (2003) used a randomized, parallel design to study the effect of feeding a MUFA-rich diet high in OO compared to a MUFA diet rich in alpha-linolenic acid from rapeseed oil (RO) and a PUFA-rich diet from sunflower oil (SO) on plasma Apolipoprotein A-IV. Forty-eight participants (23 men) finished the study. The subjects were lean (mean BMI = 23) with a mean T-C of 4.95 mmol/L. All subjects were instructed to consume a baseline diet containing 38% energy as fat (19% SFA, 11.3% MUFA, and 5.6% PUFA) for two weeks prior to the treatment phase of the study. Total fat content of the experimental diets was similar (38.2 – 38.7% of energy). The OO, SO and RO diets contained 10.7, 10.0, 9.2% and of energy from SFAs, respectively. Analogous data for MUFAs were 23.2, 8.7 and 19.1% of energy and 3.4, 18.4 and 9.0% of energy for PUFAs. Experimental diets were fed for four weeks and energy intake was

adjusted to maintain stable body weight throughout the study. As expected, Apo A-IV fell among all subjects while consuming the high SFA baseline diet (from 97.2 to 83.6 mg/L). Concentrations of this apolipoprotein increased similarly for all three experimental diets (OO = 89.8 mg/L, SO = 99.5 mg/L, RO = 101.3 mg/L). The purpose of this study was to examine the effect of replacement of dietary SFAs with MUFAs and PUFAs on plasma Apolipoprotein A-IV (Apo A-IV) and consequently data on T-C, LDL-C and HDL-C were not provided. The authors conclude, "...diets rich in unsaturated fatty acids, independent of the degree of unsaturation, gender, and Apo A-IV genotype, increase plasma Apo A-IV concentrations compared with a baseline diet rich in SFA in healthy men and women."

In summary, the studies discussed in this section provide evidence that the substitution of olive oil or PUFAs for dietary SFAs has comparable affects on serum T-C, LDL-C and HDL-C. Three of the studies were entirely consistent with this hypothesis (Mensink and Katan, 1989; Connor *et.al.*, 1993; Nydahl *et.al.*, 1994). Three studies reported differences between olive oil and PUFA-containing diets in T-C and/or LDL-C (Lichtenstein *et.al.*, 1993; Kris-Etherton *et.al.*, 1995; Pedersen *et.al.*, 2000), but the ratio of T-C/HDL-C or LDL/HDL was unchanged because olive oil diets tended to increase HDL-C compared to the PUFA-containing diets. One study (Kratz *et.al.*, 2003) did not provide complete data on serum lipids, but showed that diets rich in unsaturated fatty acids had beneficial affects on Apolipoprotein A-IV as PUFA. Only one study (Nielsen *et.al.*, 2002) failed to provide evidence that olive and PUFAs have similar affects on serum lipids, but did show that olive oil diets resulted in lipoproteins that were less susceptible to oxidation compared to those arising from PUFA-containing diets.

These results are also consistent with a meta-analysis (Gardner and Kraemer, 1995) that found no difference in the hypocholesterolemic effect of MUFAs and PUFAs when fed in diets with similar concentrations of total fat, SFA, fiber and cholesterol (see discussion of this paper below). Truswell and Choudhury (1998) criticized this meta-analysis because its rigorous inclusion criteria limited the number of studies analyzed. However, this approach was necessary to ensure that the data being combined resulted in a valid comparison between the effects of MUFAs and PUFAs on blood lipids.

The meta-analysis conducted at the request of the NAOOA (see Appendix C) for studies with a direct comparison between olive oil and PUFA-containing diets is also consistent with these conclusions. Six studies met the inclusion criteria for this analysis (Lichtenstein *et.al.*, 1993; Mensink and Katan, 1989; Nielsen *et.al.*, 2002; Nydahl *et.al.*, 1994; Sirtori *et.al.*, 1992). The results, which are presented in Table 4, show that there is no statistically significant difference in T-C, LDL-C or HDL-C between the olive oil and PUFA-containing treatments. Plasma TGs were significantly higher after the olive oil diets. This unexpected result did not appear to be due to total carbohydrate because the total fat content of the control and experimental diets were similar. It is possible that the composition of the carbohydrate (e.g. simple sugars) was responsible for the change in TGs. The practical significance of this effect is difficult to ascertain because the pooled baseline TG concentration was relatively low (106 mg/dL) and olive oil feeding resulted in insignificant changes in TGs compared to other dietary fats in all but one of the studies included in the meta-analysis (Mensink and Katan, 1989).

Table 4
Overall Effects of Olive Oil as Compared to Polyunsaturated Fat Diets

Variable	Pooled Effect Size (mg/dl)	95% CI	Percent Change: Olive oil vs. PUFA
Total Cholesterol	7.22	14.9, -0.48	3.8
LDL-Cholesterol	4.93	11.8, -1.92	3.9
HDL-Cholesterol	0.49	2.84, -1.85	1.1
Triglycerides	12.5*	23.6, 1.30	11.8

*$p<0.05$

Finally, although the studies discussed above suggest that PUFAs and MUFAs have similar effects on serum lipids when fed as replacements for SFAs, the former may increase the susceptibility of serum lipoproteins to oxidation while MUFAs are much less prone to do so (O'Byrne *et.al.*. 1998). Several recent papers (Hargrove *et.al.*, 2001; Kratz *et.al.*, 2002; Strychar *et.al.*, 2003) as well as the study by Nielsen *et.al.* (2002) discussed in this section have shown that diets containing olive oil result in serum lipoproteins that are more resistant to oxidation than diets rich in PUFA.

c. Olive Oil vs. Carbohydrates

Baggio *et.al.*. (1988) compared the effects of a diet enriched with MUFA with a standard low-fat diet on serum lipids in 11 moderately hypercholesterolemic (T-C: 5.9mmol/L, TG:0.96 mmol/L) men admitted to a metabolic ward. The study used a crossover design consisting of two three-week feeding interventions, but the order of treatments was not randomized. For the first three weeks, subjects consumed diet I which was a low-fat, CHO-rich diet (28% energy as fat, 56% CHO) containing 43g/d of MUFA (12.6% of energy). During the second three week intervention, subjects received diet II which was a high MUFA diet (38% fat, 46% CHO)

obtained by substituting most fats from diet I with olive oil and removing some of the CHO in order to keep the diets isocaloric. Subjects consumed 82.8g/day of MUFA (25% of energy) during the second three-week intervention period. The low-fat and OO-rich diets had the same P:S ratio (0.35 and 0.36, respectively), and comparable levels of PUFA (4.1 and 3.5% of energy) and SFA (11.7 and 9.7% of energy, respectively). Dietary compliance was assessed by the evaluation of the fatty acid profile of erythrocyte membranes before and after the olive oil-rich diet. The high MUFA diet resulted in a significant 9.5% decrease in T-C, 12.2% decrease in LDL-C and 25.5% decrease in TG compared to the low-fat CHO-rich group. HDL-C levels did not differ between the two groups. The authors concluded that olive oil could be used for the control of plasma and LDL-C cholesterol as a valid alternative to CHO.

Garg *et.al.*. (1988), compared the effects of a high CHO and high MUFA diet on plasma lipids in 10 moderately hypercholesterolemic (T-C>200 mg/dL or TG>200 mg/dL) men with NIDDM. All subjects were on insulin therapy and none had a history of ketosis. A randomized, crossover design was employed. After consuming a baseline American Diabetes Association (ADA)-recommended diet (30% energy from fat, 50% CHO) during a seven day run-in period, subjects were given diets consisting of a high MUFA diet in which olive oil replaced CHO (50% energy as fat, 35% CHO) and a low fat high CHO diet (25% fat, 60% CHO). Subjects consumed the diets in random order during two 28-day hospitalizations in a metabolic ward separated by a 6-22 day washout period during which patients were out of the hospital. Subjects consumed the ADA baseline diet during the 6-22 day washout period. Olive oil was the main source of fat in the high MUFA diet, whereas a mixture of corn oil and palm oil was used for fat in the high CHO diet. Both experimental diets were isocaloric and provided similar amounts of SFA and PUFA.

Consumption of MUFA increased from 36.5g/d (14% of energy) at baseline to 88.3g/d (33% of energy) in the high MUFA group and decreased to 24g/d (9% energy) in the high CHO group. The high MUFA diet resulted in a significant 12.9% reduction in T-C compared to baseline. As compared with the high CHO diet, the high MUFA diet resulted in a significant 25% and 34% reduction in TG and VLDL-C respectively and a 13% increase in HDL-C. As expected, the levels of LDL-C did not differ significantly in patients on the two test diets. The authors concluded that partial replacement of complex carbohydrates with MUFA in the diets of patients with NIDDM does not increase the level of LDL-C and may improve the levels of TG and HDL-C. The applicability of this study to the healthy U.S. population is compromised by the fact that although the patients were classified as NIDDM, they were receiving daily injections of human insulin. In addition, some of the patients had a history of CHD although none were taking lipid-lowering medication.

Garg *et.al..* (1992) used a randomized, crossover design to compare the effects of High CHO and Low CHO/High MUFA diets on plasma lipids in eight adult men with mild NIDDM. All subjects had stable body weights (mean wt = 89 kg, BM I = 30) and glycemic control before entering the study (fasting glucose <10mM) and had not received drug treatment or insulin therapy for the previous four months. Subjects consumed a baseline diet according to ADA recommendations (55% energy as CHO, 27% fat) for five days and were then randomly assigned to experimental diets for 21 days with a 6.5 day washout between interventions. The experimental diets consisted of a low CHO/high MUFA diet (35% CHO, 50% fat) that provided 87g/day MUFA (32% of energy) mainly from olive oil and a high CHO diet (60% CHO, 25% fat) containing 32.4g/day MUFA (12% of energy). Patients were allowed to consume plain

coffee or tea in restricted amounts. All experimental diets were provided during two 21-day hospitalizations in the research center. The low CHO/high MUFA diet resulted in an 11.8% increase in HDL-C, a 21.5% decrease in TG and a 21.5% decrease in VLDL-C compared with the high CHO diet. There were no significant differences in T-C or LDL-C between the high CHO and low CHO/high MUFA groups. The authors concluded that, in patients with mild NIDDM, high CHO diets do not improve glycemic control and they raise plasma TG and VLDL-C levels while reducing HDL-C levels.

Rasmussen *et al.* (1993) used a randomized crossover study to compare the effects of a high MUFA diet with a high CHO diet on blood lipid levels in 15 free-living adult men and women with NIDDM. Half of the subjects received additional oral antidiabetic drugs while the other half was treated with diet alone. After consuming their habitual diets during a 2-week run-in period, participants were randomly allocated to a 3-week treatment with a high-CHO or high-MUFA diet with a 3 week washout period between treatments. The composition of the baseline diets (40% energy from fat, 40% CHO) was calculated according to 4-day diet records maintained by the subjects during the run-in period. The high-MUFA diet (50% fat, 30% CHO) was based on the patient's food records and was supplemented with MUFAs in exchange for CHOs (bread, potatoes, rice). Olive oil was the main source of fat in the high MUFA diet and it was supplied in special rolls and meat dishes prepared and supplied frozen for subjects to consume at home. Subjects in the high MUFA group were also allowed daily consumption of 10-20 g of almonds or nuts daily but were not allowed butter, margarine or avocados. The high-CHO diet was enriched mostly by bread, potatoes, and rice. Subjects consumed 71 g/d (30% of energy) and 26 g/d (11% energy) of MUFA on the high-MUFA and high-CHO diets

respectively. The PUFA content was similar in all diets. Participants kept weighed food records during baseline and the last week of each diet intervention (3 working days and 1 weekend day) to estimate energy intake and diet composition. There were no significant differences between diet groups in TG, TC, LDL-C, HDL-C and LDL/HDL. Compared to the high CHO diet, the high MUFA group had favourable effects on blood pressure and blood glucose responses. The authors concluded that a diet rich in MUFA has beneficial effects on blood pressure and glucose metabolism with no adverse effects on blood lipids in subjects with NIDDM.

Garg *et.al.*. (1994) studied the effects of a high MUFA diet on plasma lipids in 42 adult men and women with NIDDM (fasting glucose: 101-199 mg/dL, fasting TG: 54-440mg/dL) in a randomized, crossover design study. All patients were receiving glipizide therapy. Subjects were randomized to a high MUFA diet (45% energy as fat, 40% CHO) containing olive oil as the main source of fat or a high CHO diet (30% fat, 55% CHO) for six weeks with a seven day washout between interventions. The high MUFA and high CHO diets provided 70g/day (25% of energy) and 28g/day (10% of energy) of MUFA respectively and equal amounts of SFA and PUFA. All patients ate at least one meal at the metabolic unit on weekdays and were supplied with the rest of their food in packages to be consumed at home. Study compliance was monitored by interviews with dietitians and returned portions of unconsumed food. The high MUFA diet resulted in a significant 35% decrease in TG and 19% decrease in VLDL-C compared to the high CHO group. Serum levels of T-C, LDL-C and HDL-C did not differ between the two dietary groups. This study suggests that a high MUFA diet results in favorable VLDL-C and TG levels in patients with NIDDM compared to a high CHO diet.

Lopez-Segura *et.al.* (1996) determined the effect of a MUFA-rich diet in comparison to a low-fat diet on blood lipids using a randomized, crossover design. Twenty-one healthy, normocholesterolemic (T-C <220mg/dL) male students (mean age, 23.4 ± 5.6 yrs) consumed a low fat NCEP-1 diet (30% energy from fat, 54.5% CHO) during an initial 24-day baseline period. After this phase, subjects were randomized to one of two dietary treatments consisting of a high fat high-MUFA diet containing a lower content of CHO (39% energy from fat, 44% CHO), or the high-MUFA diet supplemented with cholesterol for two 24-day periods with no washout between treatments. Afterward, all subjects consumed the NCEP-1 diet supplemented with cholesterol for a period of 24 days. Consumption of MUFA increased from 38.5g/d (13.5% energy) at baseline to 72g/d (24% of energy) in the high MUFA diets based on substitution of olive oil for cookies, bread and marmalade found in the NCEP-1 diet. The content of SFA and PUFA was similar in the NCEP-1 and high MUFA diets. All meals were cooked and consumed in the school canteen. Good compliance was suggested in this study based on enrichment of oleic acid in the cholesterol ester fraction of LDL during the MUFA diets. Consumption of both of the experimental diets rich in MUFAs did not result in any significant changes in T-C, LDL-C, TG, HDL-C or VLDL-C compared with the low fat, CHO-rich NCEP-1 diets. Plasma PAI-1 activity and serum insulin were significantly decreased in the high MUFA groups compared with the CHO-rich NECP-1 diets. The authors concluded that reduction of plasma PAI-1 activity and insulin levels may be a phenomenon involved in the lower incidence of CHD in populations that have a high oleic acid consumption.

Morgan *et.al.* (1997) compared the effects of a very-low fat diet with a low-fat diet supplemented with MUFA on serum lipid levels using a randomized, crossover design. Twenty-

four healthy men and women with hypercholesterolemia (T-C >6mmol/L) consumed and recorded their usual self-selected diets for a two week run-in period and were then randomly assigned to a low-fat MUFA rich diet and a very low fat, CHO-rich diet for a period of three weeks each with no washout between treatments. The low-fat, MUFA-rich diet contained olive oil and an olive oil-based margarine and provided 25.6% percent of energy as fat and 49% as CHO. The very low fat, CHO-rich diet contained no added fat, included a carbohydrate supplement drink, and provided 10% of energy as fat and 64% as CHO. The SFA content of the experimental diets was comparable and was significantly lower than baseline. The baseline diet provided 36.5% of energy as fat, and 43% CHO. All subjects were provided with preweighed daily packages of very lean beef (raw or precooked) and sample menus and recipes for meat dishes, desserts, and cakes. Subjects were allowed to eat non-fat dairy products, egg whites, and vegetables and fruits except avocado, soy beans, and olives and they were encouraged to eat grains and cereal products. Foods with added fat, any meat (other than what was provided), fish, egg yolk, nuts and other fats and oils were excluded from the diet. Subjects provided seven-day weighed food records each week of the study and met weekly with a dietitian to ensure dietary compliance. Subjects consumed 34.7g/d (15% of total energy), 34.8g/d (15.5% energy) and 8.3g/d (3.8% energy) MUFA on the baseline, high-MUFA, and very low fat high-CHO diets, respectively. Compared to baseline, the high MUFA diet resulted in a significant 11.4% reduction in T-C, 12.5% reduction in LDL-C, 12.5% reduction in HDL-C, and no change in TG and the LDL-C/HDL-C ratio. Concentrations of T-C, LDL-C and HDL-C fell significantly during the very low fat, high CHO diet as well compared to baseline. There was however, a significantly greater drop in HDL-C, a larger increase in TG and a significant increase in the LDL-C/HDL-C ratio on the very low fat, high CHO diet compared to the low fat, MUFA-rich

group. The authors concluded that a low-fat diet enriched with olive oil provides advantages over a very-low fat diet in the control of serum lipoproteins among persons with hypercholesterolemia.

Thomsen *et.al.* (1999) studied the effect of high-MUFA vs. high-carbohydrate diets on CHD risk factors in the children of NIDDM patients. Sixteen subjects (mean age = 35 years) with at least one relative with type 2 diabetes participated in the study. The subjects were slightly overweight (mean BMI = 25.8) but were normolipidemic and had normal oral glucose tolerance tests. The subjects were randomized to either a high MUFA or high carbohydrate diet after consuming their habitual diet for a four-week run-in period. The test diets were fed for four weeks at which time the subjects were switched to the other diet after a four-week washout period consuming the habitual diet. The baseline diet contained 35.8% energy from fat and 13.6% energy from SFA. The carbohydrate diet provided the following percentages of energy: fat, 27.8; SFA, 8.8; MUFA, 8.1; PUFA, 6.7; and carbohydrate 52.5. Analogous values for the MUFA diet were: fat, 42.2; SFA, 9.4; MUFA, 23.9; PUFA, 5.5; and carbohydrate, 41.0. The SFA and PUFA content of the two diets were not significantly different. There were no differences in T-C, LDL-C, or TG at the end of the experimental periods, however HDL-C and Apo A-I concentrations were significantly higher after the MUFA diet. The authors conclude that high MUFA and high carbohydrate diets have essentially similar effects on serum lipids in people at high risk for type 2 diabetes, however HDL-C and Apo A-I were higher on the high MUFA diet.

Rodriguez-Villar *et.al.*. (2000) compared the effects of a high MUFA, olive oil-rich diet to a high CHO diet on blood lipids in 12 patients with NIDDM. The subjects were not using

hypolipidemic medication and were receiving treatment with diet or oral hypoglycemic agents. After consuming their usual low-fat, high CHO diet during the pre-inclusion period, patients were randomly prescribed a low-fat, high CHO-diet (29% energy as fat, 54% CHO) or a high-fat, high MUFA diet (40% fat, 43% CHO) supplemented with olive oil in a random crossover design consisting of 6 week interventions with no washout between treatments. Initially and weekly during each diet, patients met with a dietitian and completed three-day food records (including 1 weekend day). Patients were trained to follow recommended diets, which differed only in fat and complex CHO content from their usual diets. The test diets were limited in red meat, eggs and whole-fat dairy products and the consumption of vegetable products and fish was emphasized. Consumption of cereal products, legumes, and fruits was increased in the high-MUFA group and the use of olive oil was restricted in the CHO diet. The high MUFA diet provided 55g/d of MUFA corresponding to 25% of total energy and the high-CHO diet contained 25g MUFA, corresponding to ≈ 12% of daily energy intake. Dietary compliance was determined to be excellent throughout the study based on a deviation of less than 15% between actual intake from food records and prescribed nutrient intake. Consumption of the high-MUFA diet did not result in significant changes in T-C, TG, LDL-C or HDL-C compared to the high-CHO diet. The authors concluded that a diet high in MUFA containing olive oil is a good alternative diet to the traditional low-fat diet for patients with type 2-diabetes mellitus.

Strychar et.al. (2003) investigated the effects of a high-MUFA diet compared to a diet rich in carbohydrates (CHO) on plasma lipoprotein profiles in 26 type I diabetics who were otherwise healthy. Subjects underwent 2 months of intensive insulin therapy to optimize glycemic control and normalize lipoprotein levels, and were then randomized to one of two dietary interventions

using a crossover design. Isoenergetic dietary prescriptions were based on the food choice system adapted from the Canadian Diabetes Association Good Health Eating Guide, and subjects were given information sheets that included details of their diet prescription, including total daily number of food choices, meal plan, and sample menus. The two diets were similar, except that during the high-MUFA phase subject's diets contained fewer starch choices and more fat choices (e.g. olive oil based salad dressings). Intakes were measured using 5-day dietary records completed during and at the end of each phase. Compliance was monitored by telephone calls four days per week, in which subjects reported glycemic readings, and were asked whether they were following the diet, and whether they were consuming olive oil during the high-MUFA phase. Only seven subjects completed both dietary study periods. In these subjects, fat intakes were 30% of energy in the baseline group (11% SFA, 13% MUFA, 6% PUFA), 37% of energy in the high-MUFA group (10% SFA, 21% MUFA, 6% PUFA) and 27% of energy during the high-CHO phase (9% SFA, 12% MUFA, 6% PUFA). Plasma T-C, LDL-C and HDL-C did not differ significantly between diet groups at the end of the trial. Plasma TGs were 18% lower during the high-MUFA phase compared to the Hi-CHO phase (1.17mmol/L vs. 1.42mmol/L). Many of the subjects preferred the Hi-MUFA diet, and the authors concluded that MUFA may be an alternative strategy to improve overall dietary adherence rates in diabetes.

In summary, the olive oil dietary intervention studies published since 1989 confirm the preliminary conclusions of the *Diet and Health* report that MUFAs do not cause adverse effects on plasma HDL-C or TGs that often result from low-fat, high-carbohydrate diets. Seven of these studies found that olive oil-containing diets resulted in improved concentrations of T-C, LDL-C, HDL-C, or TGs compared to low-fat, high-carbohydrate diets (Battio *et.al.*, 1998; Garg *et.al.*,

1988, 1992, 1994; Morgan *et.al.*, 1997;Thomsen *et.al.*, 1999; Strychar *et.al.*, 2003). One additional study (Kris-Etherton *et.al.*, 1999) (see the MUFA vs. saturated fat section of this document) found that an olive oil diet resulted in lower concentrations of T-C and TGs than an NCEP Step II diet with no difference in LDL-C or HDL-C. The three remaining studies discussed in this section (Rasmussen *et.al.*, 1993; Lopez-Segura *et.al.*, 1996; Rodriguez-Villar *et.al.*, 2000) and one study discussed in the saturated fat section (Fuentes *et.al.*, 2001) found no differences in blood lipids between diets rich in olive oil and those high in carbohydrate. These three studies support the notion that olive oil is equivalent (but not superior) to a high-carbohydrate diet.

This conclusion was confirmed by the meta-analysis conducted for the NAOOA on studies that compared olive oil with low-fat, high-carbohydrate diets (see Appendix C). Eleven studies met the inclusion criteria for this analysis (Fuentes *et.al.*, 2001; Garg *et.al.*, 1988, 1992, 1994; Kris-Etherton, *et.al.*, 1999; Lopez-Segura *et.al.*, 1996; Morgan *et.al.*, 1997, Rasmussen *et.al.*, 1993; Rodriguez-Vilar *et.al.*, 2000; Strychar *et.al.*, 2003; Thomsen *et.al.*, 1999). The results of this analysis (presented in Table 5) show that there were no differences in T-C, or LDL-C between the OO and high-carbohydrate diets, but olive oil resulted in significantly higher HDL-C and lower TG.

Table 5
Overall Effect of Olive Oil as Compared to Low-Fat Diets

Variable	Pooled Effect Size (mg/dl)	95% CI	Percent Change: Olive oil vs. Low-Fat Diets
Total Cholesterol	-0.63	6.60, -7.86	-0.3
LDL-Cholesterol	0	6.56, -6.56	0.0
HDL-Cholesterol	3.01	5.36, 0.67	7.2
Triglycerides	-20.0	-7.53 -32.4	-12.2

*$p<0.05$

Finally, both the FNB (2002) Macronutrient report and the NHLBI (National Cholesterol Education Program, 2001) ATP III report came to the same conclusion that moderate-fat diets rich in MUFAs do not lower HDL-C or raise TGs as diets high in carbohydrates often do. An additional advantage of using MUFAs as an alternative to carbohydrates as a replacement for dietary SFAs is that moderate fat MUFA-containing diets may favorably affect plasma insulin and glucose. Three of the studies conducted in type-2 diabetics found that serum glucose or insulin concentrations responded favorably to high MUFA diets (Garg *et.al.*, 1988, 1994; Rasmussen *et.al.*, 1993) while two studies found no effect (Garg *et.al.*, 1994; Rodriquez-Villar *et.al.*, 2000).

3. Meta-Analyses

Six meta-analyses have been published on the effect of dietary MUFAs on blood lipids. These analyses provide evidence that MUFAs have an *independent* effect on serum lipids in addition to their role in the displacement of dietary SFAs.

Grundy and Vega (1988) performed a meta-analysis of liquid diet feeding trials to evaluate how individuals vary in their responses in plasma concentrations of T-C and LDL-C to the substitution of saturated fatty acids for unsaturated fatty acids. All studies included employed liquid-formula diets in which the only variable was the type of fat. Two studies were included in this meta-analysis that compared a MUFA rich dietary formulation (in both cases high-oleic safflower oil was the sole fat source) with SFA rich formulations. The first study was carried out in 17 subjects and utilized palm oil as the fat source in the high-SFA group. The second study was carried out in seven patients, with coconut oil as the sole source of fat during the high-SFA

phase. Both studies were conducted using a randomized, cross-over design. Test subjects in these studies generally had high-normal concentrations of plasma triglycerides, but highly varied plasma T-C concentrations. In the first study, plasma T-C levels were an average of 37 mg/dL lower during the MUFA phase of the trial, compared to the palm oil phase. Subjects' individual responses ranged from a 1 mg/dL to 79 mg/dL decline in T-C. Similarly, mean plasma LDL-C was 30mg/dL lower in subjects after the high-MUFA phase compared to the high-SFA phase. Individuals ranged in their LDL-C response to replacement of SFA with MUFA from a 5 mg/dL to a 68 mg/dL decrease. In the second study, mean plasma T-C was 34 mg/dL lower after the MUFA phase than after the coconut oil phase, representing individual subjects declines ranging from 8 mg/dL to 53 mg/dL. Plasma LDL-C was also 32 mg/dL lower after the MUFA phase than after the coconut oil phase in this study. A 5 mg/dL to 61 mg/dL range of reduction in plasma LDL-C was experienced by individuals when SFA in coconut oil were substituted by MUFA in this trial. The authors state that individual responses to the high-MUFA diets are similar in their degree of variability to those seen when high-linoleic safflower oil (rich in PUFA) was the test oil. The authors conclude that data from this meta-analysis imply that the composition of the diet (i.e. a diet rich in SFA rather than MUFA or PUFA) "may be more important in causation of primary hypercholesterolemia than has generally been realized".

Mensink and Katan (1992) performed a meta-analysis of 27 well-controlled dietary intervention studies published between 1970 and 1991 to calculate the effect of changes in carbohydrate and fatty acid intake on serum lipid and lipoprotein levels. The studies included were all original articles that met the following criteria: 1) food intake and composition was highly regulated (i.e. metabolic-ward conditions), with cholesterol levels that did not vary between groups, and with

dietary fatty acids being the single variable; 2) the designs that compared experimental diets only to baseline were excluded (specifically randomized crossover trials, parallel trials, and Latin square design were acceptable); 3) feeding periods had to be of sufficient length to stabilize lipid responses (i.e. >14d); and 4) subjects in studies had to be relatively healthy (i.e. not suffering from gross disturbances of lipid metabolism). Diets enriched in very long chain PUFA, trans isomers of unsaturated fatty acids, or stearic acid were excluded. Data from studies included were used to estimate multiple regression equations for the mean changes in serum lipids and lipoproteins. Isocaloric replacement of 1% of daily dietary energy intake as carbohydrates with MUFA (or 6g of carbohydrate with 2.7g of MUFA) is predicted by the regression coefficient for MUFA to result in a 0.009mmol/L increase in HDL-C. The regression coefficient for the change in serum LDL-C predicted to result from the replacement of 1% of dietary energy from carbohydrate with MUFA is –0.006mmol/L. Although this value is negative, it was not significantly different from zero. For the predicted change in T-C, per percent of energy, the regression coefficient for MUFA (specifically oleic acid) was found to be –0.007mmol/L. While the predicted correlation coefficient for a change in serum TG was –0.022 when carbohydrates are substituted by MUFA, this value did not differ significantly from zero. The replacement of 10% of energy from SFA by MUFA is predicted to result in a 0.33mmol/L decrease in LDL-C, and a small (0.03mmol/L) decrease in HDL-C. The authors conclude that "according to the present analysis, replacement of saturated by unsaturated fatty acids produces a more favourable lipoprotein profile than does replacement by carbohydrates," so long as body weights, and other related factors, remain equal.

Hegsted et.al. (1993) conducted a meta-analysis of the combined published data on the effects of dietary fatty acids (including MUFA) and cholesterol on serum cholesterol and lipoprotein cholesterol evaluated in groups of human subjects. Studies included in this analysis were of two types. The first type, metabolic studies, are those done under carefully controlled conditions in which food was prepared and fed to subjects, although often not under metabolic-ward conditions. The second type, field trials, are trials in which the diet was modified by instruction or a combination of instruction and provision of some foods. Only data obtained with diets composed of ordinary foods, excluding those obtained with liquid-formula diets, were used. Data on hydrogenated, isomerised, or transesterified fats, *trans* fatty acids, fish oils, and studies of weight-reducing diets were excluded. In addition, trials with very short experimental periods (7-10 days) and those with inadequate descriptions of the fatty acid content of the dietary regimen were excluded. Regression analysis was used to model changes in serum lipids in mmol/L in response to changes in dietary fatty acids as a percentage of energy, including MUFA. The predictability of the regression equation for changes in serum T-C is not improved when changes in MUFA are considered, indicating that the regression coefficient for MUFA is not significant, although it is negative (-0.00318). The regression coefficient for MUFA in the equation to predict changes in HDL-C was positive, but also not significant. Regression equations for LDL-C did not take into account changes in MUFA level. The authors conclude that from this analysis, no effect of MUFA can be demonstrated on serum cholesterol.

Gardner and Kraemer (1995) conducted a meta-analysis to examine whether MUFA or PUFA have a differential effect on serum lipid levels. Studies were included if they were independent and randomized, and contained at least two intervention groups that were similar in all respects

(total fat, SFA, fiber, dietary cholesterol) except for levels of MUFA and PUFA. Fourteen studies met these stringent criteria. All foods were provided by the researchers in 10 of these studies, three provided principal fat sources and one provided dietary advice by registered dietitians. For LDL-C, eleven of fourteen studies found no difference in the effect size between diets rich in MUFA and diets rich in PUFA, while 1 found a significant positive effect size (i.e. LDL-C levels were higher on the MUFA diet) and two found a significant negative effect size (i.e. LDL-C levels were lower on the MUFA diet). Virtually identical results, of a highly similar magnitude, were seen for T-C (data not provided by the authors). For HDL-C, 10 of the 14 effect sizes were not statistically significantly different from zero. Two studies showed a significant positive effect size, and two showed a significant negative effect size. Nine of the fourteen studies showed effect sizes that were not significantly different between PUFA and MUFA for TG. The remaining five studies all had significant positive effect sizes (i.e. where TG levels were lower on the high-PUFA diets). When studies in which high SFA diets were contrasted with high MUFA diets, six of seven studies found that MUFA reduced LDL-C, while the effect sizes of the seventh study were borderline significant. The individual study effect sizes for T-C were virtually identical to those for LDL-C (data not provided). For HDL-C, five of seven studies showed effect sizes that did not differ significantly from zero. The remaining two studies that found significant negative effect sizes of MUFA compared to SFA on HDL-C were small (<0.5SD). None of the high-MUFA vs. high-SFA effect sizes for TG were significantly different from zero. The authors conclude that there is no significant difference in serum T-C, LDL-C or HDL-C levels between diets relatively high in MUFA versus PUFA when fat intake is derived primarily from common plant and vegetable oils.

Yu et.al. (1995) conducted a meta-analysis of 18 studies in order to develop predictive equations for the relationship between the change in plasma lipid and lipoprotein levels and the change in intakes of dietary fatty acids, including relative intakes of MUFA (which were derived from oleic acid, olive oil, canola oil, and high-oleic safflower oil). The authors analyzed well-controlled studies performed in a total of 682 normocholesterolemic healthy men and women using multiple regression analysis. According to the regression equations developed in this meta-analysis, MUFA decreased total cholesterol in both men and women, although this effect was not statistically significant (regression coefficients: –0.0137 in males, and –0.0197 in females). MUFA also decreased LDL-C, and this effect was significant in males (regression coefficients: 0.0181 in males, $p<0.01$, and –0.0253 in females). Conversely, MUFA increased HDL-C, an effect that was also significant only in males (regression coefficients: -0.0091 in males, $p<0.01$, and –0.0080 in females). The authors conclude that the present study shows that "MUFA significantly decrease serum total cholesterol, and LDL-C and increase HDL-C concentrations". Based on results from the predictive equations generated in this study, the authors speculate that the effect of MUFA on serum cholesterol and LDL-C is dependent on the amount of SFA (specifically 12:0, 14:0, and 16:0). The authors explain that "when hypercholesterolemic SFA in the diet are low, the independent cholesterol-lowering effect of MUFA is observed. When 12:0-16:0 SFA are high, the cholesterol-lowering effect of MUFA is obscured, and MUFA appear to have a neutral effect".

Clarke et.al. (1997) performed a meta-analysis of 72 metabolic ward studies of solid food diets in healthy volunteers to determine the quantitative importance of dietary fatty acids to blood concentrations of cholesterol. Only studies that ensured compliance (metabolic ward), with

dietary interventions persisting for at least 2 weeks, and sufficient dietary information to contribute to the analysis, were considered. Multivariate regression analysis produced regression coefficients for the effects of isocaloric substitutions of dietary fatty acids for complex carbohydrates on blood cholesterol concentrations. Every 1% exchange of carbohydrate with MUFA, was predicted to result in a 0.005mmol/L increase in T-C, a 0.008mmol/L decrease in LDL-C, and a 0.006mmol/L increase in HDL-C. However, only the effect of MUFA on HDL-C was significant, indicating that changes in MUFA have little effect on T-C and LDL-C when substituted for carbohydrates. Changes in SFA intakes were the strongest predictor of blood cholesterol levels. Replacement of 5% of calories as SFA (e.g. 12% of dietary energy reduced to 7%) with MUFA was predicted to result in a 0.24mmol/L reduction in T-C. The authors conclude that the reduction in blood cholesterol shown by their review with isocaloric replacement of saturated by unsaturated fats appears within just a few weeks and is greater than is sometimes appreciated.

In conclusion, the meta-analyses discussed above confirm that dietary MUFAs lower serum T-C and LDL-C when fed as a replacement for dietary SFAs. However, these data also suggest that MUFAs modify serum lipids by an *independent* mechanism. The meta-analysis by Yu *et.al.* (1995) included 18 feeding studies where the amount of individual saturated fatty acids in the diets were published. Regression analysis of these data showed that adding MUFAs to the diet (while holding other fatty acids constant) resulted in a decrease in both T-C and LDL-C according to the following equations: ΔT-C(mmol/L) = 0.0522Δ12:0-16:0 – 0.0008Δ18:0 – 0.0124ΔMUFA – 0.0248ΔPUFA and ΔLDL-C(mmol/L) = 0.0378Δ12:0-16:0 + 0.0018Δ18:0 – 0.0178ΔMUFA – 0.0248ΔPUFA. Based on this analysis, T-C would fall by 0.0124 mmol/L

(0.48 mg/dL) and LDL-C would decrease by 0.0178 mmol/L (0.69 mg/dL) for each 1% increase in energy from MUFAs while holding all other dietary fatty acids constant. This small, but significant change occurs independent of the displacement of dietary SFA. The authors suggest that the independent hypocholesterolemic effect of MUFAs is dependent on the SFA content of the diet, and that the effect was not seen in the analysis by Hegsted (1965) because it was masked by a higher content of dietary lauric, myristic and palmitic acids (12.38% of energy combined) compared to their analysis (9.17 % of energy). The cholesterol content of the diet may also influence the effect of MUFAs on serum lipids.

The meta-analyses reviewed above also show that the effect of MUFAs on serum HDL-C and TGs is *independent* of the saturated fat content of the diet. For example the analysis of Mensink and Katan (1992) of 27 studies predicted that serum HDL-C would increase by 0.34 mg/dL ($p<0.01$) and that serum TG would decrease by 1.99 mg/dL ($p<0.001$) for each 1% of energy in which MUFAs replaced carbohydrate. In addition, the epidemiologic data from the Women's Health Study (Hu *et.al.*, 1999) show that CHD incidence is reduced when MUFA replaces carbohydrate regardless of SFA intake. These data demonstrate that the cardioprotective properties of dietary MUFAs are not due exclusively to the replacement of dietary SFA.

4. Statements and recommendations from governmental and professional organizations

The beneficial role of unsaturated fatty acids, including MUFAs, on management of CHD risk has been acknowledged in the most recent dietary recommendations from governmental and non-

governmental organizations. A brief summary of these public health recommendations is provided below.

a. The Dietary Guidelines for Americans

The Dietary Guidelines for Americans (United States Department of Agriculture and United States Department of Health and Human Services, 2000) represent official government policy with respect to nutrition. The most recent edition of the 'Guidelines states,

> **Unsaturated fats (oils) do not raise blood cholesterol.** (emphasis added) Unsaturated fats occur in vegetable oils, most nuts, olives, avocados, and fatty fish like salmon. Unsaturated oils include both *monounsaturated fats* and *polyunsaturated fats*. Olive, canola, sunflower and peanut oils are some of the oils high in monounsaturated fats. Vegetable oils such as soybean oil, corn oil, and cottonseed oil and many kinds of nuts are good sources of polyunsaturated fats. Some fish such as salmon, tuna, and mackerel, contain omega-3 fatty acids that are being studied to determine if they offer protection against heart disease. Use moderate amounts of food high in unsaturated fats, taking care to avoid excess calories.

b. Healthy People 2010

Healthy People 2010 (United States Department of Health and Human Services, 2000) outlines specific health objective for the nation. This document acknowledges that unsaturated fatty acids (including MUFAs) "can help lower health risks".

> The major vegetable sources of monounsaturated fatty acids include nuts, avocados, olive oil, canola oil, and high-oleic forms of safflower and sunflower seed oil. The major sources of polyunsaturated fatty acids are vegetable oils, including soybean oil, corn oil, and high-linoleic forms of safflower and sunflower seed oil and a few nuts, such as walnuts. **Substituting monounsaturated and polyunsaturated fatty acids for saturated fatty acids can help lower health risks.** (emphasis added).

c. Adult Treatment Panel III (ATP III)

The NHLBI of the National Institutes of Health (NIH) recently published evidence-based guidelines on the management of CHD risk (National Cholesterol Education Program, 2001). These guidelines concluded, "Monounsaturated fatty acids lower LDL cholesterol relative to saturated fatty acids (A2, B2)[6]. Monounsaturated fatty acids do not lower HDL cholesterol nor raise triglycerides (A2, B2)." In addition, olive oil is featured in two sample menus for "Traditional American Cuisine" provided in this report.

d. Dietary Reference Intakes for Macronutrients

The Food and Nutrition Board (2002) of the National Academy of Sciences recently issued comprehensive dietary recommendations for macronutrient intake in the United States and Canada. This report established an "Acceptable Macronutrient Distribution Range" (AMDR) for dietary fat of 20 to 35% of total calories for the adult population. The upper range of the AMDR was set higher than the 30% of total calories previously recommended by other organizations in order to accommodate the fact that moderate fat diets, rich in unsaturated fatty acids, are cardioprotective. With respect to MUFAs, this report concludes,

> These data indicate that in weight-stable individuals, a high monounsaturated fatty acid-low saturated fatty acid diet results in a more favorable metabolic profile with respect to total cholesterol, HDL cholesterol, and triacylglycerol concentrations. Figure 11-4 shows that with increased monounsaturated fatty acid intake, there is a favorable reduction in the total cholesterol:HDL cholesterol ratio. Furthermore, a meta-analysis of feeding studies estimated that the regression coefficients for the effects of monounsaturated fatty acids on LDL and HDL cholesterol concentrations were – 0.008 and +0.006, respectively, suggesting a slight positive benefit (Clarke et. al., 1997).

[6] These designations specify the type and strength of evidence used to assess the literature. A = major randomized controlled clinical trials (RCTs), B = smaller RTCs and meta-analyses of other clinical trials; 1 = very strong evidence, 2 = moderately strong evidence.

This report also summarized the available experimental data (see Figure 2 below) to show that HDL-C increases and TGs decrease as percent of energy from total fat increases (i.e. decreased dietary carbohydrate) when intake of SFAs are low.

Figure 2
Relationship between Percent of Total Fat intake and Change in Triacylglycerol (TAG) and HDL Cholesterol Concentrations

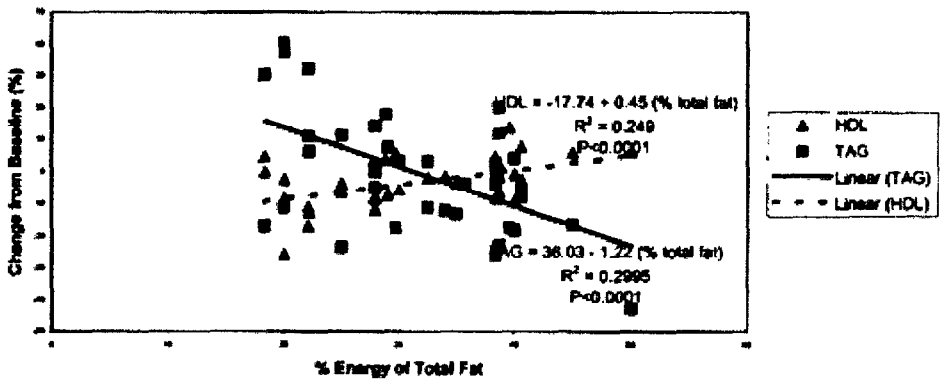


FIGURE 11-1 Relationship between percent of total fat intake and change in triacylglycerol (TAG) (▬) and HDL cholesterol (—) concentrations. Regression equations for percent change in serum TAG and HDL cholesterol predicted by percent total fat in the experimental diets of controlled-feeding studies comparing low-fat, high-carbohydrate diets to high-fat diets. Weighted least-squares regression analyses were performed using the mixed procedure to test for differences in lipid concentrations (SAS Statistical package, version 8.00, SAS Institute, Inc., 1999). Percent of energy from total fat varied from 18.3% to 50%. All diets were low in saturated fat (less than 10% energy). Using these equations, for every 5% decrease in total fat, HDL cholesterol would decrease by 2.2% and triacylglycerol would increase by 6%. SOURCE: Berry et al., 1992; Curb et al., 2000; Garg et al., 1988, 1992a, 1994; Ginsberg et al., 1990; Grundy, 1986; Grundy et al., 1988; Jansen et al., 1998; Kris-Etherton et al., 1999; Lefevre et al., unpublished; Lopez-Segura et al., 1996; Mensink and Katan, 1987; Nelson et al., 1995; Parillo et al., 1992; Pelkman et al., 2001; Perez-Jimenez et al., 1995, 1999, 2001.

Source: FNB Macronutrient Report, 2002

In conclusion, the most knowledgeable and respected public health organizations in the U.S. have concluded that MUFAs can help reduce the risk of CHD when consumed as part of an energy appropriate diet moderate in total fat and low in SFAs. MUFAs lower T-C and LDL-C when fed as a replacement for SFA, and also exert a smaller, independent effect (Food and Nutrition Board, 2002). In addition, MUFAs do not decrease HDL-C, or increase TGs as high-

carbohydrate diets often do. This observation was used by the NHLBI to conclude that MUFAs can contribute up to 20% of total calories in a heart-healthy diet (National Cholesterol Education Program, 2001). Collectively, current dietary guidelines and recommendations provide strong support for the premise that MUFAs from olive oil can help reduce the risk of CHD when consumed as part of a moderate-fat diet low in saturated fat and cholesterol.

E. Significant Scientific Agreement

The NAOOA strongly believes that the totality of publicly available scientific evidence shows that there is significant scientific agreement among experts qualified to evaluate such data that monounsaturated fatty acids from olive oil can reduce the risk of CHD when consumed as part of a moderate-fat diet low in saturated fat and cholesterol. This conclusion is based on the following observations:

- A large predominance of literature that shows olive oil reduces blood concentrations of T-C and LDL-C when fed as a replacement for dietary SFAs
- A large predominance of literature that shows olive oil does not increase TG or lower HDL-C as high-carbohydrate diets do
- A large predominance of literature that shows olive oil does not increase susceptibility of lipoprotein fractions to oxidative changes as PUFAs do
- Universal acknowledgement from governmental and public health organizations that MUFAs can reduce the risk of CHD when fed as part of an energy-appropriate moderate-fat diet low in saturated fat in which MUFAs serve as a partial alternative to carbohydrates as a replacement of SFAs
- The cardioprotective effects of olive oil have been more thoroughly documented than any other source of MUFA
- Olive oil contains numerous substances in addition to MUFAs that may contribute to its cardioprotective properties

The NAOOA respectfully requests that FDA conclude that the significant scientific agreement standard in this area has been met, and move swiftly to authorize the proposed claim.

V. MINIMUM EFFECTIVE DOSE

The meta-analysis of human olive oil intervention studies conducted for the NAOOA (see Appendix C) can be used to determine the minimum daily effective "dose" of MUFAs from olive oil that would be required to achieve a reduction in blood T-C and LDL-C similar to that elicited by substances already authorized by FDA for CHD-related health claims. The pooled data for studies that compared olive oil with diets higher in SFAs were used for this calculation. The average amount of MUFA from olive oil used for dietary treatments in these studies was 8.5 percent of energy. Based on the standard 2,000-calorie diet used for nutrition labeling, this amount of MUFA provides 170 Kcal (2,000 Kcal x 0.085 = 170 Kcal from MUFAs). Olive oil contains 73.7% MUFAs so the amount required to provide 170 Kcal MUFA is 25.6 grams ((170 Kcal MUFA ÷ 9 Kcal per gram) ÷ 0.737 = 25.6 g olive oil). With respect to blood T-C, the magnitude of reduction between the olive oil and SFA diets was 8.83% (T-C decreased 17.7 mg/dl from a baseline concentration of 200.5 mg/dl). Therefore, the percentage change in T-C per gram of olive oil was -0.34 (-8.83 ÷ 25.6 g = -0.34). Finally, the amount of olive oil necessary to prompt a 5 percent reduction is T-C would be 14.7 grams (5 ÷ 0.34 = 14.7 g olive oil) containing 10.6 grams MUFA. Similar calculations for LDL-C, which was reduced by 11.3% by the olive oil interventions, (-15.5 mg/dl from a baseline concentration of 137 mg/dl) reveals that 11.4 grams of olive oil (containing 8.4 grams MUFA) would result in a 5 percent reduction in LDL-C. Therefore, the proposed minimum daily dose of MUFAs is 10 grams (the average of the amount required to reduce T-C and LDL-C by 5%) or approximately 13.5 grams

of olive oil. This value is comparable to the RACC for olive oil of (one tablespoon). Therefore, the proposed minimum effective daily dose of olive oil to be fed as part of a low-saturated fat diet is 13.5 grams or one tablespoon.

The proposed minimum effective dose of 13.5 grams olive oil per day is intentionally conservative. A five percent reduction in T-C and LDL-C was used to calculate this dose despite the fact that reductions of serum T-C and LDL-C as low as 4% have been used to authorize health claims. FDA stated in the preamble to the final rule for the psyllium health claim (63 FR 8103 at 8109)

> Similarly, there is no basis to require that the qualifying criteria for a substance associated with risk of CHD be based on the amount of that substance to elicit a 5 percent reduction in blood total- and LDL-cholesterol levels. The data on psyllium seed husk suggests that the magnitude of the effect on blood lipids for intakes of about 10 g/d of psyllium seed husk ranges from 4 to 6 percent for blood total-cholesterol and about 4 to 8 percent for LDL-cholesterol levels in conjunction with diets low in saturated fat and cholesterol (Ref. 7). Although modest in size, these are clinically significant reductions in blood lipids and translate to a reduced risk of CHD for individuals with hypercholesterolemia...

In addition, the meta-analysis provided in Appendix C included two studies (Choudhury *et.al.*, 1995; Ng *et.al.*, 1992) that compared olive oil to palm oil. These studies reported a significant hypocholesterolemic effect for olive oil and palm oil compared to baseline, but no significant difference between the two treatments. Although the applicability of these data to the American diet (where palm oil intake is very low) is questionable, they were retained in the meta-analysis to ensure a conservative estimate of the effect of olive oil on CHD risk.

The effective daily dose of 13.5 g olive oil per day is contingent on it being consumed as part of a diet lower saturated fat because the studies used to calculate this dose included olive oil in such diets. This fact is implicit in the proposed claim because it specifies that olive oil be consumed "as part of a diet low in saturated fat and cholesterol." The current intake of saturated fat in the U.S. typically exceeds dietary recommendations. Data from the 1994-96 and 1998 Continuing Survey of Food Intake by Individuals (CSFII) (see appendix D) show that the current average intake of saturated fat among U.S. adults at least 20 years of age is 11.0 percent of energy, and that only 11.3% of this population consumed less than seven percent of energy as saturated fat as recommended by the NHLBI (National Cholesterol Education Program, 2001). As a result, a large majority of consumers who adhere to the proposed claim will automatically reduce dietary saturated fat.

VI. NATURE OF THE FOOD ELIGIBLE TO BEAR THE CLAIM

The NAOOA requests that products that contain at least 3.4 grams of olive oil per RACC and are low in saturated fat (21 CFR § 101.62(c)(2)), contain no more than one gram of *trans* fatty acids per RACC, and contain no more than 20 mg cholesterol per RACC be eligible to bear the proposed claim. Products that are essentially pure olive oil would not be required to be low in saturated fat and would be exempted from the total fat disqualifier level, the 50-gram criterion of the saturated fat disqualifier level and the 10% DV nutrient contribution requirement. All olive oil-containing products other than those that are essentially pure olive oil would be subject to the general health claim requirements in 21 CFR § 101.14.

A. Minimum amount of olive oil per RACC

As noted above (see section V) 13.5 grams per day of olive oil is sufficient to lower T-C and LDL-C by five percent when consumed as part of a diet low in saturated fat. The NAOOA proposes that a minimum of 3.4 g olive oil per RACC be required for a food to bear the claim. This amount is based on the premise that consumers should have the flexibility to consume the minimum effective dose by eating up to four servings of olive oil-containing foods per day (13.5 g ÷ 4 servings/d = 3.4 g/serving).

FDA has traditionally considered that a typical daily food consumption pattern is composed of three meals and a snack per day (58 FR 2302, 2379, January 6, 1993). This dietary pattern was used to define the minimum content criterion for three CHD-related health claims: soy protein (64 FR 57700 at 57713); ß-glucan soluble fiber from whole oats (62 FR 3584 at 3592); and soluble fiber from psyllium seed husks (63 FR 8103 at 8109). The NAOOA believes the same consumption pattern should be applied to the proposed claim for the reasons provided below.

Many consumers may choose to incorporate olive oil into their diets throughout the day by using the essentially pure product in cooking, at the table as an alternative to butter or other spreads, as a salad dressing or other direct use. However, the availability of a wide variety of olive oil-containing prepared foods with reasonable amounts of total fat would provide additional opportunities for consumers to obtain the benefit of this high-MUFA food.

As noted above, FDA typically uses four servings per day to determine the minimum amount of a substance that must be contained in a food product to be eligible for health claims. An

exception to this approach was the claim for sterol-stanol esters and CHD. In this instance the agency chose to use two servings per day because there are few plant sterol-containing foods available in the marketplace so that it would be difficult for individuals to consume four servings per day. FDA was also reluctant to recommend more frequent consumption of such foods because they are necessarily high in fat (FR 65 54686 at 54707).

The NAOOA does not believe this situation exists for the proposed claim. A list of olive oil-containing foods that meet the general criteria for health claims (i.e. the disqualifier levels and the 10% DV minimum nutrient content criterion) that were consumed by participants in the 1994-96 and 1998 CSFII surveys is provided in Appendix E. This list shows that a wide variety of olive oil-containing foods were consumed. Many of these foods are home-prepared dishes and few commercially available products are currently available[7]. However, one of Congress' original objectives for the NLEA was to provide incentives for food manufacturers to develop products that help consumers eat healthier diets. In the case of the proposed claim, such products would likely be patterned after recipes currently used by consumers. By using four servings per day to determine the minimum olive oil content, products in the range of 3.4 – 6.75 grams per RACC could be eligible to bear the claim. Many of these foods are vegetable, pasta, rice and fish-based dishes that contain moderate amounts of total fat, and could frequently be consumed as part of a diet consistent with the Dietary Guidelines for Americans. A higher minimum requirement for olive oil would not be in the best interest of consumers, or the food industry, because it would necessitate that more fat be added to certain claim-eligible products, make it more difficult to achieve the low-saturated fat requirement and would add to the cost of such

[7] Spaghetti sauce appears to be the only packaged food eligible to bear the claim.

products. We therefore, strongly recommend that 3.4 grams olive oil per RACC be established as the minimum amount necessary to bear the claim.

B. Total fat content

As noted previously, leading public health authorities have altered traditional recommendations that Americans consume a low-fat, high-carbohydrate diet as a means to reduce the risk of CHD. For example, the fifth edition of the Dietary Guidelines for Americans (U.S. Department of Agriculture, U.S. Department of Health and Human Services, 2000) modified the previous "fat" guideline from, "Choose a diet low in fat, saturated fat, and cholesterol," to, "Choose a diet that is low in saturated fat and cholesterol and **moderate in total fat"**(emphasis added). In addition, the latest report of the National Cholesterol Education Program (ATP III) increased the upper recommended level of total dietary fat from 30 to 35% of calories in order to accommodate up to 20% of calories from MUFAs and up to 10% of from PUFAs (National Cholesterol Education Program, 2001). Similarly, the FNB (2002) has established an AMDR for total fat of 20-35% of energy for the same reason.

The majority of currently authorized CHD-related health claims (21 CFR §§ 101.75, 101.77, 101.81) require that foods meet the "low-fat" definition (21 CFR §101.62 (b)(2)) in order to be eligible to bear a claim. However, FDA has recognized the need for several important exceptions to this policy. For example, the agency initially proposed that foods eligible to make the soy-CHD claim be required to be low in fat, but eliminated this requirement because total fat intake is not directly related to CHD, and because the inherent fat content of soybeans would have prevented many products made from whole beans from making the claim (64 FR 57700 at

57717). In addition, the agency chose not to impose a low-fat criterion on products eligible to make the sterol/stanol ester health claim because fat is the only vehicle capable of delivering these cardioprotective substances which were deemed to have important public health significance (65 FR 54686 at 54708). FDA also noted that this policy was consistent with the fifth edition of the Dietary Guidelines for Americans, which recommends "moderate" rather than low" fat diets. More recently, the agency sanctioned a qualified health claim for nut-containing products that are low in saturated fat and cholesterol, but not necessarily low in total fat[8].

Olive oil consumption is not associated with higher overall fat intake in the U.S. population. Data from the 1994-96 and 1998 CSFII were analyzed to determine the dietary intake patterns of olive oil consumers vs. non-consumers (see Appendix D). Subjects aged 20 years or more (n=9,221) who were not pregnant or lactating were included in the analysis. These subjects were categorized as essentially non-olive oil consumers (0-1.9 g olive oil per day from any source), light olive oil consumers (2-4.9 g olive oil per day) and heavy olive oil consumers (5 g olive oil per day or more). The intake of energy, total fat and fatty acids according to these olive oil consumption categories are presented in Table 6 (see next page).

The data in Table 6 show that energy and total fat intake did not differ significantly between olive oil consumption categories. Light olive oil-consuming subjects consumed significantly *less* total fat (expressed percent of energy) than non-consumers and similar amounts compared to subjects in the high olive oil group. Subjects in the light olive oil group also consumed significantly less saturated fat than non-consumers when expressed as g/d and percent of energy.

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[8] Letter to Mr. D.J. Soetaert from Dr. Christine L. Taylor dated July 14, 2003 (Docket No. 02P-0505) http://www.cfsan.fda.gov/~dms/qhcnuts2.html

Table 6
Energy, Fat and Fatty Acid Intake of U.S. Adults According to Olive Oil Intake

Dietary Factor	**Non consumers (0-1.9 g/d) n = 8,804**	**Light consumers (2.0-4.9 g/d) n = 180**	**Heavy consumers (5+ g/d) n = 237**
OO (g/d)	0.1	3.2	11.7
Energy (Kcal)	1,988	1,928	2,076
Total Fat (g)	74.4	67.7	76.8
Total Fat (% of energy)	33.3[a]	30.7[b]	33.3[ab]
Percent of subjects with <35% Kcal from Total Fat	58.6[a]	71.0[b]	59.3[ab]
Saturated Fat (g)	25.0[a]	21.7[b]	23.3[ab]
Saturated Fat (% of energy)	11.1[a]	9.8[b]	10.0[ab]
Percent of subjects with <7% Kcal from Saturated Fat	10.8[a]	19.6[b]	22.1[b]
Percent of subjects with <10% Kcal from Saturated Fat	38.1[a]	55.1[b]	51.8[b]
Monounsaturated Fat (g)	28.5[a]	25.9[a]	32.5[b]
Polyunsaturated Fat (g)	15.1	14.5	15.2

Values not sharing an alphabetic superscript differ significantly ($p<0.05$) by a priori Bonferonni contrast.

There was also a tendency for heavy olive oil consumers to take in less saturated fat than non-consumers (10.0 vs. 11.1 g/d), but the difference was not statistically significant. Both olive oil consumption groups were more likely to meet current dietary recommendations for saturated fat (i.e. <7 or 10% of energy) than non-olive oil consumers. High olive oil subjects consumed significantly more MUFAs than the other categories but there was no difference in the intake of PUFAs.

The NAOOA believe this analysis of U.S. dietary intake patterns provides additional evidence that foods eligible to bear the proposed claim need not be low in total fat. These data clearly show that olive oil consumption is not associated with increased intake of total fat or saturated

fat in the U.S. population. Data from the heavy olive oil consumption category are particularly noteworthy because the mean olive oil intake in this group (11.7 g/d) approximates the daily dose specified in the proposed claim.

In summary, the NAOOA believes there is now compelling evidence that foods need not be low in total fat in order to reduce the risk of CHD as long as they are low in saturated fat. FDA has acknowledged this fact in previous rulemakings. In addition, dietary analysis show that olive oil consumption is not associated with a significant increase in total fat (and a *beneficial* association with saturated fat). We, therefore, request that a low-fat criterion not be imposed on foods eligible to bear the proposed claim.

C. Saturated/*trans* fat content

The NAOOA believes that all olive oil-containing products, with the exception of products that are essentially pure olive oil, should be required to be low in saturated fat as specified in 21 CFR § 101.62(c)(2). We also believe that such products should contain no more than one gram of *trans* fatty acids (TFA) per RACC as described in the agency's recent final rule (68 FR 41434, July 11, 2003). If FDA establishes a nutrient content claim for low-*trans* fatty acids in the future, we believe that it should be added to the criteria for the proposed claim. This criterion would help ensure that consumers who choose products bearing the proposed claim reduce the SFA content of their diets as they transition to MUFA-containing foods.

D. Cholesterol content

All of the CHD-related health claims that have been authorized to date require that eligible foods be low in cholesterol as defined by 21 CFR § 101.62 (d). Like all plant-based foods, olive oil does not contain cholesterol. The NAOOA believes that olive oil-containing formulated products should be required to contain ≤ 20 mg cholesterol per RACC (or per 50 g if the RACC is 30 g or less or two tablespoons or less) according to 21 CFR § 101.62(d)(2)(ii)(A) in order to be eligible to make the proposed claim.

E. Exemptions

As noted above, products that are essentially pure olive oil would need to be exempt from the total fat disqualifier level, the 50-gram criterion of the saturated fat disqualifier level and the 10% DV nutrient contribution requirement in order to qualify for the proposed claim. The NAOOA believes that the latest scientific information clearly justifies these exemptions. In addition, we believe that such exemptions are imperative from a public health perspective because pure olive oil is the form most frequently used by consumers. Furthermore, if the exemptions are not granted, it would be virtually impossible to explain why olive oil-containing products would be permitted to bear the claim but pure olive oil was not.

1. Total fat/saturated fat disqualifier levels

As noted in paragraph "B" above, nutrition science and public health policy has evolved to recognize the benefits of moderate-fat diets that are low in saturated fat as alternatives to more traditional low-fat, high-carbohydrate diets. In addition, the analysis of the totality of available scientific evidence presented in this document clearly demonstrates that MUFAs from olive oil

have beneficial effects on CHD risk factors when consumed as part of a moderate-fat diet that is low-saturated fat. Despite their health benefits, products that are essentially pure olive oil exceed the disqualifier level for total fat and saturated fat (due to the 50-gram criterion) as defined in 21 CFR § 101.14(a)(4). Therefore, the NAOOA respectfully requests an exemption from these requirements for the following reasons:

Consumption of pure olive oil is not associated with increased intake of total fat. Data from the 1994-96 and 1998 Continuing Survey of Food Intake by Individuals (CSFII) were analyzed to determine the dietary intake patterns of consumers of pure olive oil vs. non-consumers. The analysis was similar to that discussed above (see p 78). Subjects aged 20 years or more (n=9,221) who were not pregnant or lactating were included in the analysis. These subjects were segregated according to whether they had reported consumption of pure olive oil (USDA foodcode = 82104000) at least once during the two-day survey. The intake of energy and total fat for this population categorized by consumption of pure olive oil is presented in Table 7.

Table 7
Intake of Energy and Total Fat Among Pure Olive Oil Consumers and Non-Consumers

Dietary Factor	**Non-consumers n = 9,118**	**Consumers n = 103**
OO (g/d)	0.0	10.7
Energy (Kcal)	1,991	1,908
Total Fat (g)	74.4	72.6
Total Fat (% of energy)	33.1	33.9
Percent of subjects with <30% energy from Total Fat	33.3	32.4
Percent of subjects with <35% of energy from Total Fat	58.8	61.3

There were no significant differences in energy or total fat intake between the subjects who had consumed pure olive oil during the two days of the survey compared to those who had not. These data show that olive oil consumption is not associated with the intake of total fat in the U.S. population.

FDA has established an important precedent for granting exemptions from the total fat disqualifier level when appropriate to do so. Such an exemption was granted for products making the sterol/stanol ester health claim. The agency cited four criteria it considered in making this decision (65 FR 54686 at 54709). These criteria were: whether the disease in question is of public health significance; whether the absence of an exemption from the disqualifier level would severely limit the number of foods that would qualify to bear the claim; whether there is evidence that the population to which the health claim is targeted is not at risk for the disease; and whether there are other public health reasons for granting the exemption.

FDA concluded that sterol/stanol ester-containing foods should be granted the requested exemption because CHD is a significant public health concern, because lack of an exemption would severely limit the foods that would qualify for the claim and because sterol/stanol ester-containing products have a significant potential to benefit public health by virtue of the fact that they can lower serum T-C and LDL-C without adversely affecting HDL-C. The agency also justified the exemption by concluding that, "...current scientific evidence does not indicate that diets high in unsaturated fat are associated with CHD...", and cited the 2000 Dietary Guidelines for Americans which states, "Choose a diet that is low in saturated fat and cholesterol and **moderate in total fat**" (emphasis added).

More recently, FDA sanctioned the use of a qualified health claim for whole or chopped nuts that exceed the total fat disqualifier level. The agency concluded that, "...an appropriately qualified claim about consumption of most nuts would assist consumers in maintaining healthy dietary practices, provided that the label bears a disclosure statement that compiles with 21 CFR § 101.13(h) (i.e. "See nutrition information for fat content.")[9]."

The NAOOA believes that all of the criteria used by FDA to justify the total fat disqualifier level exemption for sterol/stanol ester-containing foods and whole or chopped nuts also apply to the proposed claim for MUFAs from olive oil, and we respectfully request that the exemption be granted.

FDA has not established a precedent for providing an exemption to the saturated fat disqualifier level, but we sincerely believe such an exemption is warranted for products that are essentially pure olive oil. As explained below, such products contain only 1.8 g of SFA per serving, the saturated fat in olive oil is *always* accompanied by more than six times that amount of unsaturated fat, olive oil users do not consume more SFA than non-olive oil users and are more likely to meet dietary recommendations for saturated fat intake; and the scientific literature clearly shows that olive oil consumption results in reduced risk of CHD when consumed as part of a low-saturated fat diet.

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[9] Letter to Mr. D.J. Soetaert from Dr. Christine L. Taylor dated July 14, 2003 (Docket No. 02P-0505) http://www.cfsan.fda.gov/~dms/qhcnuts2.html

a. Saturated fat content per serving

As noted previously, olive oil has a saturated fat content of 1.8 g per RACC. This value compares favorably with that of other "healthy" foods including avocado (3.6 g), cooked chicken breast (2.1 g) low-fat cottage cheese (1.4 g), cooked coho salmon (1.7 g), cooked rainbow trout (1.8 g) and plain low-fat yogurt (2.5 g).

The only reason that olive oil exceeds the saturated fat disqualifier level is because it must qualify on both a serving size basis and (because it has a small RACC) a 50-gram basis. We believe this requirement is inappropriate. As discussed above, scientific studies demonstrate that olive oil provides a health benefit despite its saturated fat content. Moreover, we believe it is unfair that the foods listed above with similar SFA concentrations as olive oil do not exceed the saturated fat disqualifier level while olive oil does. In addition, the average SFA content of the nuts recently sanctioned by FDA for a qualified health claim (almonds, hazelnuts, peanuts, pecans, pistachio nuts and walnuts) is very similar to that of olive oil (1.6 g per RACC vs. 1.8, respectively). The NAOOA believes that these comparisons alone justify exempting olive oil from the saturated fat disqualifier level.

b. Olive oil has a very favorable ratio of unsaturated to saturated fatty acids

The fatty acid distribution of olive oil is shown in the Table 8. These data show that the ratio of unsaturated fatty acids (MUFAs + PUFAs) to SFAs is 6.2.

Table 8
Fatty Acid Distribution of Olive Oil

Fatty Acid Group	Concentration in Olive Oil (%)
Saturated	13.5
Monounsaturated	73.9
Polyunsaturated	10.0

Source: USDA National Nutrient Database for Standard Reference. Release 16

Products that are essentially pure olive oil are the only foods for which an exemption from the saturated fat disqualifier level is being proposed. This narrow application ensures that the saturated fat content of exempted products will *always* be accompanied by an abundance of unsaturated fat.

c. Olive oil consumers do not consume more saturated fat than non-olive oil consumers

Analysis of the CSFII database for the intake of saturated fat among users and non-users of pure olive oil is presented in Table 9.

Table 9
Saturated Fat Intake Among Pure Olive Oil Consumers and Non-Consumers

Dietary Factor	Non-consumers (Mean Olive Oil intake = 0.0 g/d) n = 9,118	Consumers (Mean Olive Oil intake = 10.7 g/d) n = 103
Energy (Kcal)	1,991	1,908
Saturated Fat (g)	24.9	21.0*
Saturated Fat (% of energy)	11.0	9.7**
Percent of subjects with <7% energy from Saturated Fat	11.3	17.3
Percent of subjects with <10% of energy from Saturated Fat	38.6	54.7*

*$p<0.05$, **$p<0.01$ by t-test for means or by Chi square test for percentages

Subjects whose diets included pure olive oil consumed significantly less saturated fat per day than their non-olive oil-consuming counterparts. In addition, olive oil-consumers were significantly more likely to achieve the benchmark of no more than 10% of calories from saturated fat. These data clearly show that olive oil is not associated with increased intake of SFA in the U.S. and may, in fact, help consumers meet dietary recommendations for macronutrients.

d. Olive oil is hypocholesterolemic despite the saturated fat it contains

Public health should be the overriding determinant on whether olive oil is entitled to bear the proposed claim. An overwhelming predominance of the scientific literature (including a meta-analysis of olive oil studies) provides compelling evidence that olive oil lowers T-C and LDL-C when included in a moderate-fat diet that is low in saturated fat. Furthermore, these data show that olive oil does not cause unfavorable effects on HDL-C or TGs. The NAOOA believes it would be contrary to the science, and to the interests of public health, to prevent olive oil from bearing the proposed claim because it contains a small (1.8 g) amount of saturated fat per RACC.

Products which make the proposed claim that exceed the disqualifier levels would be required to bear the disclosure statement, "See nutrition information for fat and saturated fat content" as specified in 21 CFR § 101.13(h). This statement would alert consumers to the fact that such foods contain these components and would also call attention to additional nutrition information that can help them make informed dietary choices.

In summary, the NAOOA urgently recommends that FDA grant exemptions from the total fat disqualifier level and the 50-gram criterion of the saturated fat disqualifier level for products that are essentially pure olive oil. These exemptions are consistent with the latest dietary recommendations and are justified by more than 40 dietary intervention studies that show pure olive oil has beneficial effects on CHD biomarkers. In addition, olive oil contains only a small amount of SFA per serving, has a very favorable unsaturated to saturated fatty acid ratio and does not contribute to SFA intake in the American population. The NAOOA believes that these justifications are compelling, and uniquely qualify products that are essentially pure olive oil for the requested exemptions.

2. 10% DV nutrient contribution requirement

Foods must contain at least 10% DV of protein, dietary fiber, calcium, iron, vitamin A or vitamin C per RACC in order to bear a health claim unless otherwise exempt by regulation (21 CFR § 101.14 (e)(6)). The agency explained the rationale for this requirement in the preamble to its final rule on the general principles concerning approval of health claims (58 FR 2478, 2521, January 6, 1993), which states, " Thus, FDA finds merit in the suggestion that foods bearing health claims should be those consistent with dietary guidelines, and that the value of health claims should not be trivialized or compromised by their use on foods of little or no nutritional value."

Since the initial rulemaking for health claims, FDA has proposed to exempt certain fruits and vegetables as well as many enriched grain products from the 10% DV nutrient contribution requirement (60 FR 66206, 66214, December 21, 1995). The agency's proposal states,

"Moreover, diets high in fruits, vegetables and grain products have been associated with various specific health benefits, including lower occurrence of coronary heart disease...and therefore, are exactly the types of foods that should be included in the diet to reduce the risk of specific diet-related diseases." FDA further stated that it would consider providing additional exemptions from the 10% DV requirement if it were provided with sound justification to do so. Indeed, the agency granted such a request for salad dressings to bear the sterol/stanol ester claim. An important consideration in this decision was that although salad dressings are low-nutrient dense foods, "...they are often consumed with foods rich in nutrients and fiber. Salads, for example, are usually rich in vegetables that provide important nutrients at significant levels, e.g., tomatoes – vitamins A and C; carrots – vitamin A; spinach – vitamin A and calcium." (see 65 FR 54686 at 54711).

The NAOOA believes that similar rational can be used to exempt olive oil from the 10% minimum nutrient contribution requirement. Analysis of data from the CSFII database as described above show that individuals who consumed pure olive oil on at least one day of the two-day survey consumed a significantly higher percentage of the RDA for dietary fiber (65.4%), vitamin E (67.8%) and vitamin C (160.4%) than subjects who did not (54.6%, 54.6 and 115.3, respectively). This increase in nutrient density was probably due to the fact that olive oil-consumers included significantly more fruits and vegetables in their diet compared to non-consumers as shown by the data in Table 10.

Table 10
Food Guide Pyramid Servings for Adult (≥20 Years)
Consumers and Non-consumers of Pure Olive Oil

Pyramid Food Group Servings	**Non-consumers (Mean Olive Oil intake = 0.0 g/d) n = 9,118**	**Consumers (Mean Olive Oil intake = 10.7 g/d) n = 103**
Grain group	6.7	6.6
Vegetable group	3.4	4.5**
Fruit group	1.5	2.3**
Dairy group	1.3	1.2
Meat group	5.3	4.6
Added sugar (% Kcal)	14.7	10.1**

** $p<0.01$ by t-test for means

In addition to a higher fruit and vegetable intake, survey participants who consumed pure olive oil consumed less added sugar had a higher Health Eating Index (HEI) score than non-consumers (68.6 vs. 63.9, respectively; $p<0.05$).

In conclusion, the scientific evidence presented in this petition shows that olive oil can reduce the risk of CHD when consumed as part of a moderate-fat diet that is low saturated fat. Furthermore, like salad dressing, olive oil is typically consumed with foods that are rich sources of the nutrients specified in 21 CFR § 101.14 (e)(6). Olive oil is frequently used in combination with vinegar as a dressing for salads, as a component of tomato sauce used with pasta and as an alternative to butter or margarine for breads. Furthermore, olive oil is ideal for stir-frying vegetables or sautéing fish and other nutrient-dense foods. Olive oil clearly does not fit the profile of jellybeans and other foods that would "trivialize the value of health claims" and we respectfully request that products that are essentially pure olive oil be exempted from this requirement.

VII. LABELING REQUIREMENTS

Foods eligible to bear the proposed claim would be required to declare the grams of MUFAs and PUFAs per serving in the Nutrition Facts panel as stipulated in 21 CFR § 101.9(c)(2)(iii)-(iv).

VIII. DIETARY CONSIDERATIONS

Petitions for new health claims are required to assess, "the potential effect of the use of the proposed claim on food consumption," (21 CFR § 101.70(f)). Analysis of the CSFII database for dietary patterns among consumers of olive oil from all dietary sources (i.e. pure olive oil as well as olive oil in prepared foods) was used to make this determination. The results show that olive oil consumption is associated with better dietary quality and unlikely to contribute to higher energy intake or incidence of obesity. Additional details of this analysis are provided below.

A. Dietary pattern

As noted earlier, data from the 1994-96 and 1998 CSFII were analyzed to determine the dietary intake patterns of olive oil consumers vs. non-consumers. Subjects aged 20 years or more (n=9,221) who were not pregnant or lactating were included in the analysis. These subjects were categorized as essentially non-olive oil consumers (0-1.9 g olive oil per day from any source), light olive oil-consumers (2-4.9 g OO per day) and heavy olive oil consumers (5 g olive oil per day or more).

The percentage of consumers who met current dietary benchmarks according to olive oil consumption category is presented in Table 11.

Table 11
Percentage of Adults (≥20 years) Meeting Dietary Benchmarks
by Olive Oil Consumption Category

Nutrient	**Non consumers (Mean Olive Oil intake = 0.10 g/d) n = 8,804**	**Light consumers (Mean Olive Oil intake = 3.2 g/d) n = 180**	**Heavy consumers (Mean Olive Oil intake = 11.7 g/d) n = 237**
Protein	66.0%	70.7%	74.4%
Carbohydrate	89.1	90.5	93.6
Total Dietary Fiber	6.7[a]	17.9[b]	12.6[ab]
Vitamin A	44.7[a]	71.3[b]	59.7[b]
Vitamin E	8.0[a]	8.6[ab]	16.7[b]
Vitamin C	43.8[a]	69.0[b]	57.2[b]
Thiamin	69.0	75.6	76.5
Riboflavin	77.1	82.8	78.9
Niacin	76.7[a]	84.2[ab]	85.7[b]
Vitamin B-6	59.9	68.1	66.5
Folate	13.9	16.5	19.7
Vitamin B-12	72.0	69.4	65.5
Calcium	18.2[a]	9.5[b]	20.2[a]
Phosphorus	85.6	87.1	91.0
Magnesium	17.0	16.9	23.8
Iron	65.0	74.0	69.7
Zinc	55.1	60.1	55.8
Copper	67.1[a]	78.7[b]	81.6[b]
Selenium	88.0[a]	88.4[ab]	94.7[b]

Values not sharing an alphabetic superscript differ significantly ($p<0.05$) by a priori Bonferonni contrast using SUDAAN

The data in Table 11 show that light olive oil consumers are significantly more likely to receive 100% RDA of dietary fiber, vitamin A, vitamin C and copper compared to non-olive oil consumers. Furthermore, heavy olive oil consumers were more likely to meet this nutrition benchmark for vitamin A, vitamin E, vitamin C, niacin, copper and selenium. The only significantly negative association was that light olive oil users (but not heavy consumers) were less likely to consume the RDA for calcium than non-consumers. As noted in Table 13 below,

this observation is likely due to the fact that olive oil consumers tend to be older than non-consumers and may be drinking less milk.

Olive oil contains approximately 5% DV of vitamin E per RACC, which may partially explain why heavy consumers were more likely to receive 100% RDA of this nutrient. However, olive oil does not contain appreciable amounts of other micronutrients. Therefore, it is apparent that olive oil users are consuming more foods that contain these nutrients than their non-olive oil-consuming counterparts. The data in Tables 12 and 13 provide additional insights into this observation.

Table 12
Mean Healthy Eating Index Scores for Adults (≥20 Years) by Olive Oil Consumption Category

HEI Component	**Non consumers (Mean Olive Oil intake = 0.10 g/d) n = 8,804**	**Light consumers (Mean Olive Oil intake = 3.2 g/d) n = 180**	**Heavy consumers (Mean Olive Oil intake = 11.7 g/d) n = 237**
Healthy Eating Index	62.6[a]	68.9[b]	67.1[b]
Grains Score	6.4	6.6	6.6
Vegetable Score	6.3[a]	7.5[b]	7.4[b]
Fruits Score	3.6[a]	5.6[b]	4.7[b]
Milk Score	5.0	4.7	5.3
Meat Score	6.7	6.4	6.4
Total Fat Score	6.7[a]	7.6[b]	6.7[a]
Saturated Fat Score	6.6[a]	7.6[b]	7.3[b]
Cholesterol Score	7.7	7.9	8.1
Sodium Score	6.2	6.5	6.3
Variety Score	7.4[a]	8.5[b]	8.3[b]

Values not sharing an alphabetic superscript differ significantly ($p<0.05$) by a priori Bonferonni contrast using SUDAAN

The data in Table 12 show that olive oil consumers have a significantly higher Healthy Eating Index (HEI) score compared to their non-olive oil-eating counterparts. In particular, olive oil

consumers ate significantly more fruits and vegetables and incorporated more variety into their diets than non-olive oil consumers. As discussed earlier in the section on disqualifier level exemptions, olive oil consumers were also more likely to meet SFA recommendations. These data are very consisted with adherence to consumption patterns recommended by the Food Guide Pyramid as presented in Table 13.

Table 13
Percent of Adults (≥20 Years) Who Meet the Food Guide Pyramid Serving Recommendations by Olive Oil Consumption Group

Pyramid Food Group	**Non consumers (Mean Olive Oil intake = 0.10 g/d) n = 8,804**	**Light consumers (Mean Olive Oil intake = 3.2 g/d) n = 180**	**Heavy consumers (Mean Olive Oil intake = 11.7 g/d) n = 237**
Grain group	35.1	40.2	44.4
Vegetable group	41.6[a]	63.2[b]	61.1[b]
Fruit group	21.6[a]	43.9[b]	32.1[b]
Dairy group	16.6[a]	10.2[b]	21.5[a]
Meat group	40.2	36.9	35.8
Discretionary fat (<30% of energy)	75.6[a]	85.0[b]	73.8[a]
Added sugar (<10% of energy)	33.4[a]	50.1[b]	54.5[b]

Values not sharing an alphabetic superscript differ significantly ($p<0.05$) by a priori Bonferonni contrast using SUDAAN

Olive oil consumers were significantly more likely to eat the recommended number of servings of fruits and vegetables than their non-olive oil-consuming counterparts. Consistent with the data in Table 11 on calcium, light olive oil consumers were less likely to receive the recommended number of servings of dairy, but this observation did not extend to the heavy olive oil consumers. Interestingly, both olive oil consumption groups were significantly more likely to limit added sugar intake to less than 10% of energy than the non-olive oil-consuming group.

In summary, analysis of the 1994-96 and 1998 CSFII databases clearly show that olive oil consumers selected a more nutrient dense diet that included more fruits and vegetables than subjects who consumed only a trivial amount of olive oil. These data do not support the contention that including 13.5 g/d of olive oil would result in unfavorable dietary changes.

B. Dietary energy and obesity

CSFII data for Body Mass Index (BMI) and energy intake by olive oil consumption groups is presented in Table 14.

Table 14
Body Mass Index and Energy Intake of Adults (≥20 Years) by Olive Oil Consumption Group

Parameter	**Non consumers (Mean Olive Oil intake = 0.10 g/d) n = 8,804**	**Light consumers (Mean Olive Oil intake = 3.2 g/d) n = 180**	**Heavy consumers (Mean Olive Oil intake = 11.7 g/d) n = 237**
BMI (kg/m^2)	26.1	25.9	25.2
Energy (Kcal)	1,988	1,928	2,076
Energy (%Estimate Energy Requirement)	78.4	78.5	80.9
Energy (% of subjects receiving ≥100% EER)	19.8	16.0	17.7

Values not sharing an alphabetic superscript differ significantly ($p<0.05$) by a priori Bonferonni contrast using SUDAAN

There were no significant differences in Body Mass Index (BMI) or energy intake among the three olive oil consumption groups. There was a tendency for lower energy consumption among the light olive oil users and for a higher calorie intake among heavy olive oil consumers, but these differences were not statistically significant. These data do not suggest that olive oil intake (from a combination of the oil itself and olive oil-containing foods) is associated with excess energy intake or weight gain.

Analogous data for subjects who consumed any amount of pure olive oil (USDA foodcode 82104000) compared to non-olive oil consumers is presented in Table 15.

Table 15
Body Mass Index and Energy Intake of Adults (≥20 Years) Who Consumed Pure Olive Oil Compared to Non-Olive Oil Consumers

Parameter	**Non consumers (Mean Olive Oil intake = 0.0 g/d) n = 9,118**	**Consumers (Mean Olive Oil intake = 10.7 g/d) n = 103**
BMI (kg/m^2)	26.1	24.9*
Energy (Kcal)	1,991	1,908
Energy (%EER)	78.5	76.5
Energy (% of subjects receiving ≥100% EER)	19.7	14/8

*$p<0.05$ by Chi square test

Subjects who consumed any amount of pure olive oil had a significantly *lower* BMI than their non-olive oil-consuming counterparts. In addition, although there was a trend toward lower energy intake among the olive oil consumers, the difference was not statistically significant. These data augment the results for olive oil consumption from all dietary sources presented in Table 14, and suggest that olive oil itself is not associated with increased obesity or energy intake among American consumers.

Willett and Leibel (2002) recently reviewed the literature and concluded that dietary fat is not a major determinant of body fat. This conclusion is based on several observations: Within geographic areas of similar economic development, regional intake of fat and prevalence of obesity are not positively correlated. In addition, short-term clinical trials have shown that

subjects randomized to receive a lower percentage of calories from fat tend to have a modest reduction in body weight compared to individuals given higher amounts of fat. However, longer-term studies (one year or longer) have shown that fat consumption within the range of 18 – 40% of energy has little or no effect on body fatness. The authors also note that obesity in the U.S. has increased dramatically at a time when fat intake (expressed as a percent of energy) has decreased substantially.

Observational data on the association between olive oil and indicators of obesity are limited, but tend to support the conclusions expressed by Willett and Leibel (2002). González *et.al.* (2000) found an inverse association between olive oil consumption and BMI among 25,814 Spanish women when expressed as grams per day ($p<0.0001$), but not as percent of energy ($p = 0.2402$). Olive oil was not related to BMI among 15,635 men in the same population. Ferro-Luzzi *et.al.* (2002) published a review paper that concluded increasing obesity rates in Greece are associated with high fat intakes, but the paper was strongly criticized by Trichopoulos (2002) for being "unscientific" and failing to adequately address the effect of physical inactivity on obesity. Garaulet *et.al.* (2001) found that central obesity was inversely correlated with the concentration of MUFAs in adipose tissue from 84 obese subjects, and Fernández-Jarne *et.al.* (2002) found no difference in BMI between 171 CHD patients and an equal number of healthy controls despite a significant difference in olive oil consumption.

Recent experimental data show that dietary fat need not be restricted in order to achieve and maintain significant weight reduction for up to 1.5 years. McManus *et al.* (2001) reported that subjects fed a moderate-fat, weight-reduction diet containing olive oil and other sources of

unsaturated fatty acids were significantly more likely to remain compliant, and lost more weight after 18 months than subjects fed a low-fat, high-carbohydrate diet. This increased adherence to a weight loss regimen was attributed largely to increased palatability of the foods in the moderate-fat diet.

In addition, two randomized intervention studies designed to test the safety and efficacy of carbohydrate-restricted diets (Foster *et.al.*, 2003; Samaha *et.al.*, 2003) found that subjects who were instructed to limit intake of carbohydrates but permitted *ad libitum* consumption of fat and protein, lost more weight than subjects instructed to consume a low-fat, high-carbohydrate diet.

The reason why olive oil does not appear to contribute to excess weight gain is not clearly understood. However, Piers *et.al.* (2002) reported increased postprandial fat oxidation rates after a high olive oil meal than after a high SFA (cream) meal in humans, which suggests there may be a metabolic effect. In addition, olive oil may be more satiating than other dietary fats. A recent small study (Wansink and Linder, 2003) showed that restaurant patrons consumed 23% less bread when it was served with olive oil than when it was served with butter.

In conclusion, a considerable body of data suggest that daily incorporation of 13.5 grams of olive oil into a low-saturated fat diet is unlikely to contribute to weight gain or obesity. Data from the 1994-96 and 1998 CSFII surveys show that olive oil consumers (either as the pure oil or from the pure oil and that derived from foods) do not consume more calories than subjects whose diets did not contain olive oil. In addition, there were no differences in BMI between olive oil consumption groups including all foods, and a significantly lower BMI among consumers of

pure olive oil vs. non-consumers. Furthermore, the observational data suggest that dietary MUFAs (and olive oil in particular) are not associated with BMI, and may be inversely associated in some populations. The experimental data clearly show that moderate- or high-fat diets can result in greater weight loss than low-fat, high-carbohydrate regimens. Finally, as the CSFII data show, olive oil is eaten with a variety of foods. This usage pattern suggests that consumers encouraged to include 13.5 grams of olive oil per day into a moderate-fat diet that is low in SFA and cholesterol are likely to substitute it for other dietary fats that are higher in SFAs as is intended by the claim. The NAOOA, therefore, believes that the proposed claim is unlikely to contribute to excess energy intake or obesity.

IX. PROPOSED MODEL HEALTH CLAIM

The following model statements are recommended for the proposed claim:

> "Monounsaturated fats from 13.5 grams per day of olive oil (one tablespoon) may reduce your risk of heart disease when included in a moderate-fat diet low in saturated fat and cholesterol."

> "Moderate-fat diets low in saturated fat and cholesterol that include monounsaturated fats from olive oil (13.5 grams, or one tablespoon) per day may reduce your risk of heart disease."

The disclaimer statement, "See nutrition information for fat and saturated fat content," would appear as part of the claim without any intervening material and in the same type size and boldness for products that are essentially pure olive oil.

The NAOOA believes that these model statements accurately reflect the totality of available science that demonstrates MUFAs from olive oil reduce serum T-C and LDL-C when consumed as part of a moderate-fat diet low in SFAs. The proposed model statements specify the amount of olive oil necessary to achieve the beneficial effect, and clearly state that MUFAs must be included in a diet low in saturated fat. This wording is intended to convey the importance of limiting dietary SFAs, and the fact that olive oil should be "included" in the diet rather than added to it.

X. DETERMINATION OF COMPLIANCE

The proposed claim requires that products contain at least 3.4 grams of olive oil per RACC in order to qualify. In cases where the claim appears on products that are essentially pure olive oil, compliance will be obvious because the RACC for such products is 30 g, and the weight of any non-olive oil ingredients are insignificant. However, for products that are composed primarily of non-olive oil ingredients, compliance with the claim will not be obvious. Furthermore, there are no AOAC approved methods that would allow FDA to determine the olive oil content of such products analytically.

The NAOOA proposes that information supplied by the manufacturer be used to establish compliance when the claim is used on formulated foods that are not essentially pure olive oil.

This approach is similar to that used to determine the eligibility of products to bear the health claim for soy protein and CHD (21 CFR § 101.82 (c)(2)(ii)(B)). This provision requires manufacturers to maintain records such as, "... recipes or formulations, purchase orders for ingredients...", or any other information that reasonably substantiates the claim. In addition, NAOOA proposes that manufacturers choosing to make the claim be required to maintain records sufficient to substantiate the claim for as long as the products are marketed, and to provide these records, on written request, to FDA.

XI. REQUEST FOR INTERIM FINAL RULE

FDA has the authority under Section 403(r)(7) of the Federal Food, Drug, and Cosmetic Act to issue an interim final rule for a health claim if such action is necessary for public health. The NAOOA believes that the three criteria specified by the act that enable FDA to take this approach have been satisfied:

A. "Enable consumers to develop and maintain healthy dietary practices."

The scientific evidence demonstrates that consuming MUFAs from olive oil is a healthy dietary practice. As noted earlier, a meta-analysis of human intervention studies shows that the inclusion of 13.5 grams of olive oil per day in a diet that is low in saturated fat and cholesterol would reduce blood T-C and LDL-C by approximately 5%. This reduction equates to a decrease in CHD incidence of approximately 10%. In addition, such a practice is consistent with recommendations of the NHLBI (National Cholesterol Education Program, 2001), the American Heart Association (Krauss *et.al.*, 2000) and the Dietary Guidelines for Americans (U.S. Department of Agriculture, U.S. Department of Health and Human Services, 2000). The

proposed claim would provide an important new opportunity to educate consumers and would serve as an ongoing reminder about this healthy practice.

B. "Enable consumers to be informed promptly and effectively of important new knowledge regarding nutritional and health benefits of food."

Packages of olive oil have not been permitted to provide information on its cardioprotective benefits, which has restricted the dissemination of this information and failed to provide an incentive for the food industry to develop and market additional olive oil-containing products that are low saturated fat and cholesterol. As a result, consumers are generally focused on reducing their intake of total fat rather than substituting MUFAs for SFAs. Twenty-four percent of a sample of 1,001 supermarket shoppers reported that they were "eating less fats/oils," in a recent survey (Food Marketing Institute, 2003), but not a single consumer mentioned reducing their intake SFAs or increasing that of unsaturated fats. Publication of an interim final rule would reinforce this message. In addition, publication of an interim final rule would provide an immediate incentive for food manufacturers to develop additional olive oil-containing foods that are low in SFA and cholesterol.

C. "Ensure that scientifically sound nutritional and health information is provided to consumers as soon as possible"

An interim final rule would shorten the length of time necessary to provide information on the cardioprotective properties of olive oil in labeling by approximately one year. A 10% reduction in CHD incidence could result in a yearly savings of 51,000 lives, prevent 340,000

hospitalizations, and reduce health care costs by $19 billion based on statistics compiled by the American Heart Association (2002).

In conclusion, the NAOOA believes that all three conditions for an interim final rule have been met. FDA took this approach for the sterol/stanol esters health claim (65 FR 54686 at 54713), and we believe the public health rationale to do the same for MUFAs from olive oil is even more compelling.

XII. ENVIRONMENTAL IMPACT ASSESSMENT

The NAOOA chooses to avail itself of the categorical exclusion with respect to an environmental impact assessment provided by 21 CFR § 25.32(p). Accordingly, an environmental impact assessment is not required for this submission.

XIII. CONCLUSION

The NAOOA respectfully requests that FDA issue an interim final rule authorizing the use of the proposed claim as quickly as possible. The totality of evidence presented in this petition clearly demonstrates that including MUFAs from 13.5 grams per day of olive oil in a moderate-fat diet that is low in saturated fat and cholesterol would reduce the risk of CHD. In addition, authorization of the proposed claim would help educate consumers about the importance of using MUFAs to displace SFAs in the diet, and provide an incentive for the food industry to develop more olive oil/MUFA-containing foods that are low in SFA and cholesterol.

XIV. PROPOSED REGULATORY TEXT

§ 101._____ Health claims: monounsaturated fatty acids from certain foods and risk of coronary heart disease (CHD).

(a) *Relationship between diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from certain foods and risk of CHD.* (1) Cardiovascular disease means diseases of the heart and circulatory system. Coronary heart disease (CHD) is one of the most common and serious forms of cardiovascular disease and refers to diseases of the heart muscle and supporting blood vessels. High blood total cholesterol and low density lipoprotein (LDL) cholesterol levels are associated with increased risk of developing coronary heart disease. High CHD rates occur among people with total cholesterol levels of 240 milligrams per deciliter (mg/dL) (6.21 millimole per liter (mmol/l)) or above and with LDL cholesterol levels of 160 mg/dL (4.13 mmol/l) or above. Borderline high-risk blood cholesterol levels range from 200 to 239 mg/dL (5.17 to 6.18 mmol/l) for total cholesterol, and from 130 to 159 mg/dL (3.36 to 4.11 mmol/l) for LDL cholesterol. The scientific evidence establishes that diets high in saturated fat and cholesterol are associated with increased levels of blood total- and LDL-cholesterol and, thus, with increased risk of CHD.

(2) Populations with a low incidence of CHD tend to have relatively low blood total cholesterol and LDL-cholesterol levels. These populations also tend to have dietary patterns that are moderate in total fat composed predominantly of unsaturated (i.e. monounsaturated and polyunsaturated) fatty acids from certain foods, such as olive oil, and also are relatively high in plant foods that contain dietary fiber and other potentially cardioprotective components.

(3) Scientific evidence demonstrates that diets that include monounsaturated fatty acids and are low in saturated fat and cholesterol may reduce the risk of CHD.

(b) *Significance of the relationship between moderate-fat diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from certain foods and the risk of CHD.* (1) CHD is a major public health concern in the United States. It accounts for more deaths than any other disease or group of diseases. Early management of risk factors for CHD is a major public heath goal that can assist in reducing risk of CHD. High blood total and LDL cholesterol are major modifiable risk factors in the development of CHD.

(2) Intakes of saturated fat exceed recommended levels in the diets of many people in the United States. One of the major public health recommendations relative to CHD risk is to consume less than 7 percent of calories from saturated fat and 25-35 percent of calories from total fat with up to 20 percent of calories from monounsaturated fatty acids. Recommended daily cholesterol intakes are 300 milligrams (mg) or less per day. Scientific evidence demonstrates that diets low in saturated fat and cholesterol are associated with lower blood total- and LDL-cholesterol levels. Monounsaturated fatty acids from certain foods, when included in a low saturated fat and cholesterol diet, also helps to lower blood total- and LDL-cholesterol levels.

(c) *Requirements* – (1) General. All requirements set forth in § 101.14 shall be met, except as set forth in this paragraph. The label and labeling of foods containing the claim shall declare the content of polyunsaturated fatty acids and monounsaturated fatty acids per serving as set forth in § 101.9(c)(2)(iii)-(iv).

(2) *Specific requirements – (i) Nature of the claim.* A health claim associating moderate-fat diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from certain foods with reduced risk of heart disease may be made on the label or labeling of a food described in paragraph (c)(2)(ii) of this section, provided that:

(A) The claim states that diets that are moderate in total fat and low in saturated fat and cholesterol and that include monounsaturated fatty acids from certain foods "may" or "might" reduce the risk of heart disease.

(B) In specifying the disease, the claim uses the terms "heart disease" or "coronary heart disease";

(C) In specifying the substance, the claim uses the term "monounsaturated fatty acids" or "monounsaturated fats" qualified by the name of the eligible source of monounsaturated fatty acids provided in paragraph (c)(2)(ii) of this section. Additionally, the claim may use the name of the food product that contains the monounsaturated fatty acids;

(D) In specifying the fat component, the claim uses the terms "saturated fat" and "cholesterol";

(E) The claim does not attribute any degree of risk reduction for CHD to moderate-fat diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from the eligible food sources from paragraph (c)(2)(ii) of this section; and

(F) The claim does not imply that consumption of moderate-fat diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from the eligible food sources from paragraph (c)(2)(ii) of this section is the only recognized means of achieving a reduced risk of CHD;

(G) The claim specifies that a daily dietary intake of 10 grams of monounsaturated fatty acids is necessary to reduce the risk of coronary heart disease and/or specifies the amount of one or more food sources from paragraph (c)(2)(ii) of this section that contains 10 grams of monounsaturated fatty acids, expressed in grams and/or units of common household measure;

(ii) *Nature of the substance* – *Cis*-monounsaturated fatty acids as defined in § 101.9(c)(2)(iii), from the food sources listed below:

(1) Olive oil

(2) [Reserved]

(iii) *Nature of the food eligible to bear the claim.* (A) The food product shall contain a minimum of 3.4 grams of olive oil, which contains 2.5 grams of monounsaturated fatty acids as described in paragraph (c)(2)(ii), per reference amount customarily consumed (RACC), as determined by reference to § 101.12. FDA will assess whether the required amount of olive oil is present for products other than those that are essentially pure olive oil as specified in paragraphs (c)(2)(iii)(B), (D) and (F) based on information identified and supplied by manufacturers, such as recipes or formulations, purchase orders for ingredients, or any other information that reasonably substantiates the amount of olive oil in the product. Manufacturers must maintain records sufficient to substantiate the required amount of olive oil for as long as the products are marketed, and must provide these records, on written request, to appropriate regulatory officials;

(B) The food shall meet the nutrient content requirements in § 101.62 for "low saturated fat" food, except for foods that are essentially pure olive oil (including pure olive oil and pure olive oil with a maximum of 1% additional ingredients that include flavors, colors and/or preservatives) are not required to meet this requirement;

(C) The food contains 20 mg or less of cholesterol per RACC and per 50 g if the RACC is 30 g or less or 2 tablespoons or less (for dehydrated foods that must be reconstituted before typical consumption with water or a diluent containing an insignificant amount, as defined in § 101.9(f)(1), of all nutrients per RACC, the per 50-g criterion refers to the "as prepared" form);

(D) The food must meet the limit for total fat and saturated fat in § 101.14 (a)(4), except for foods that are essentially pure olive oil (including pure olive oil and pure olive oil with a maximum of 1% additional ingredients that include flavors, colors and/or preservatives) provided that label of the food bears a disclosure statement that complies with § 101.13(h);

(E) The food must contain one gram of less of *trans* fat as defined by § 101.9(2)(ii);

(F) The food must meet the minimum nutrient contribution requirement in §101.14 (e)(6) except that foods that are essentially pure olive oil (including pure olive oil and pure olive oil with a maximum of 1% additional ingredients that include flavors, colors and/or preservatives) are not required to meet this requirement; and

(G) The exemptions established by subparagraphs (B), (D) and (F) of this paragraph apply only to olive oil within the scope of those subparagraphs. Other foods that include olive oil as an ingredient are not exempted from any provisions of § 101.14(a)(4) and (e)(6).

(d) *Optional information.* (1) The claim may state that the development of heart disease depends on many factors and may identify one or more of the following risk factors for heart disease about which there is general scientific agreement: A family history of CHD; elevated blood total and LDL cholesterol; excess body weight; high blood pressure; cigarette smoking; diabetes; and physical inactivity. The claim may also provide additional information about the benefits of exercise and management of body weight to help lower the risk of heart disease.

(2) The claim may state that the relationship between intake of moderate-fat diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from the eligible food source from paragraph (c)(2)(ii) of this section and reduced risks of heart disease is through the intermediate link of "blood cholesterol" or "blood total- and LDL-cholesterol;"

(3) The claim may include information from paragraphs (a) and (b) of this section, which summarize the relationship between moderate-fat diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from certain foods and coronary heart disease and the significance of the relationship;

(4) The claim may state that a moderate-fat diet low in saturated fat and cholesterol that includes monounsaturated fatty acids from the eligible food source from paragraph (c)(2)(ii) is consistent with "Nutrition and Your Health: Dietary Guidelines for American," U.S. Department of Agriculture (USDA) and Department of Health and Human Services (DHHS), Government Printing Office (GPO);

(5) The claim may state that individuals with elevated blood total- and LDL-cholesterol should consult their physicians for medical advice and treatment. If the claim defines high or normal blood total- and LDL-cholesterol levels, then the claim shall state that individuals with high blood cholesterol should consult their physicians for medical advice and treatment;

(6) The claim may include information about the number of people in the United States who have heart disease. The sources of this information shall be identified, and it shall be current information from the National Center for Health Statistics, the National Institutes of Health, or "Nutrition and Your Health: Dietary Guidelines for Americans," U.S. Department of Agriculture (USDA) and Department of Health and Human Services (DHHS), Government Printing Office (GPO).

(e) *Model health claims.* The following model health claims may be used in food labeling to describe the relationship between moderate-fat diets that are low in saturated fat and cholesterol and that include monounsaturated fatty acids from certain foods and reduced risk of heart disease:

(1) Monounsaturated fats from [gram weight of the monounsaturated fatty acid source from paragraph (c)(2)(ii) of this section that provides 10 grams of monounsaturated fatty acids] grams per day of [monounsaturated fatty acid source from paragraph (c)(2)(ii) of this section and, if desired, the name of the food product] may reduce your risk of heart disease when included in a moderate-fat diet low in saturated fat and cholesterol.

(2) "Moderate-fat diets low in saturated fat and cholesterol that include [gram weight of the monounsaturated fatty acid source from paragraph (c)(2)(ii) of this section that provides 10 grams of monounsaturated fatty acids] grams from [name of monounsaturated fatty acid source from paragraph (c)(2)(ii) of this section and, if desired, the name of the food product] per day may reduce the risk of heart disease."

XV. CERTIFICATION

I hereby certify that, to the best of my knowledge, this petition is a representative and balanced submission that includes unfavorable information as well as favorable information known to me to be pertinent to the evaluation of the proposed health claim.

Respectfully submitted,

THE NORTH AMERICAN OLIVE OIL ASSOCIATION

By Bob Bauer /GT

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