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Making Sense of Olive Oil: Simple Experiments To Connect Sensory Observations with the Underlying Chemistry

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Supporting Information

ABSTRACT: In the last decade, our understanding of the chemistry of olive oil has dramatically improved. Here, the essential chemistry of olive oil and its important minor constituents is described and related to the typical sensory categories used to rate and experience oils: color, aroma, bitterness, and pungency. We also describe experiments to explore some of the characteristics of olive oil related to its status as a new health food. Simple qualitative experiments on olive oil can be done in conjunction with tastings of the oil. First, we establish the relationship between the color of an object and the absorption of light by its molecular constituents using gummy candies and laser pointers. Then, the color of the various oils can be measured quantitatively using an iPhone app (Irodori). Illuminating the oil with a green laser produces a startling red fluorescence in the presence of the natural chlorophyll in some olive oils. Relatively straightforward colorimetric assays can reveal the presence of unsaturated fatty acids, healthy antioxidants such as phenols (unique to olive oils), contaminating peroxides, and the level of free fatty acid that is a telltale sign of poorly treated or stale oils. A final comparison of the sensory observations from tasting with the chemical and spectroscopic analysis provides an introduction to the science behind food standards and the sensitivity of our own sensory apparatus.

KEYWORDS: *High School/Introductory Chemistry, First Year Undergraduate/General, Second-Year Undergraduate, Interdisciplinary/Multidisciplinary, Laboratory Instruction, Public Understanding/Outreach, Agricultural Chemistry, Fluorescence Spectroscopy, Food Science*

INTRODUCTION

Olive oil is unique among oils in that it is a product of a fruit, rather than a seed. As such, it has a different constitution, particularly with respect to the flavor profile, but also in the presence of a range of antioxidants and pigments. It also is central to Mediterranean cuisine and culture, and is integral to classical and modern literature. The rise of olive oil as the perceived "healthy oil"[1−3] and the new international standards[4−6] make it a subject of intense current interest. As such, the olive offered a perfect theme about which to teach basic ideas of chemical structure and to connect natural biomolecules to the development of the characteristic color, taste, and smell of the oils.

This work began as a summer workshop in a small town, Yeni Foça, on the western coast of Turkey where the goal was to develop and implement a curriculum that highlights the olive and olive oil. Working collaboratively with scholars from the humanities and social sciences from Turkey and the United States, we created a workshop on olives, titled *Zeytin*—the Turkish word for olive. Our experiments were done in laboratories that included olive groves, some with trees such as the one shown in Figure 1 below that is more than 1000 years old, and olive presses, including one at Klozomenai, which dates back to 600 BC.[7−9] Most of these experiments were performed at two sites in Yeni Foça, the Taşköy Olive Press, and the Eray Restaurant.

The audience for Zeytin, which continues under the aegis of the nonprofit agency Ege'de Atölye (Aegean Workshop), is predominantly Turkish high school and university students or recent graduates curious about the liberal arts approach to higher education. Some are interested in learning more about the region or about the olive industry. Several of the

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Figure 1. An ancient olive tree near Yeni Foça, Turkey said to be 1500 years old.

participants have been sons and daughters of olive grove owners or the grove owners themselves. Because education in Turkey is tracked after high school, most participants had little more than a high school background in chemistry. Some participants had been trained in the sciences, but in particular areas that were not relevant to the topics at hand and no one was a current science major.

Over the last three years, we have also presented workshops and seminars in the United States on olive oil chemistry, both to the New England Association of Science Teachers at Keene State College, Amherst College and Colby College. Most recently, we did an abbreviated version of the workshop at the First International Olive Oil Competition in New York City, to an audience of olive growers, testers, gourmet chefs, and a variety of other professionals who were not trained in science. The success of these workshops suggests that these labs appeal not only to high school and first year science students, but also to a general audience.

An earlier article on olive oil in this *Journal*[10] focused mainly on the triglyceride and fatty acid content, and included several interesting experiments on oleic acid (named for the olive). In the intervening two decades, the prominence of olive oil in global food culture,[11] and the regulation of its production and marketing has changed dramatically. Additional chemical components in olive oil have been discovered, and olive oil has been recognized as an important research topic.[12] It seemed appropriate to revisit this unique edible substance, which may be the most healthy oil for consumption.[13]

This discussion of the chemistry of olive oil has been organized largely in accordance with the tasting categories used in common tasting protocols (we have found that this discussion can be greatly enhanced by a tasting of several olive oils). There are four broad categories in which the sensory experiences are placed: color, aroma, bitterness, and something ambiguously called pungency (a feeling in the back of the throat that may induce coughing). To the extent possible, compounds associated with our sensations of olive oil have been demonstrated with qualitative experiments. In addition, simpler versions of some of the tests associated with olive oil standards (which may deviate from the sensory tests) are demonstrated. **Detailed instructions about each of these experiments are included in the** Supporting Information.

The results of these experiments can be compared with those from the tasting of the various oils. Did the compound that was most green have more chlorophyll? Did the most bitter tasting oil contain more or less phenolic antioxidants?

OLIVE OIL PRODUCTION: STANDARDS FOR OLIVE OIL PRODUCTION AND MARKETING

A series of limits, based on both sensory and chemical tests, have been developed to determine the purity of oil and the absence of adulterants.[5] The introduction of European Union standards began in the early 1990s,[4] and the clear and consistent labeling of oils was the first step in a growing trend to provide the consumer with more information. The United States[6] and Turkey[14] have also begun to regulate which oils can bear the coveted label of virgin, extra virgin, or cold pressed. Strong and clear standards are useful in that they provide the consumer with the best possible oil, diminish the opportunity for fraud, and reward producers who are vigilant in the production of their olive oil.[15−17]

The major grades of edible olive oil are extra virgin, virgin, olive-pomace, and refined olive oils. Extra virgin and virgin olive oils are both the result of a purely mechanical pressing process, without the addition of solvents. Sometimes the phrase “cold-pressed” will be added, as the temperature of the press can be manipulated to affect the yield (more oil is yielded at higher temperatures, accompanied by a loss of quality). The difference between extra virgin and virgin oils is the level of allowed free fatty acids (less than 0.8% for extra virgin versus 2.0% for virgin) and the absence of “sensory defects” in extra virgin olive oil. Olive pomace oil—oil expressed by adding solvents to the pressed solids (pomace) to extract additional oil—cannot currently be marketed as “olive oil”. An additional class of oil with a free fatty acid content of over 3.3% is called “lampante” (literally, for burning in lamps).

Production of Olive Oil

The production of olive oil sounds deceptively simple: crush the olives and extract the oil. The yield and flavor of olive oil depend on the variety and location of trees and on when and how the olive is picked. As olives mature, they progress from green, through a mixed color called veraison, to black. Green olives produce a lower yield of oil that has the most grassy and bitter flavor, and that has the highest antioxidant concentration and shelf life. Veraison olives give perhaps the optimum processing (high yield, high antioxidant concentration and shelf life, and a mixture of fruity and bitter flavors). Black olives give a high yield of “sweet” oil that is lower in antioxidants and shelf life but that is not very bitter.[18]

Shortly after harvest (less than 1 day for the highest quality of oil; ideally less than 8 h), the olives are crushed or ground to produce a paste. This paste undergoes a unique process called malaxation, in which the paste is thoroughly mixed at a controlled temperature (around 30 °C) for approximately 1 h. During this process, there is both a physical consolidation of the oil and also a maturation of flavor components. Flavor changes can include the development of desirable aromas and the destruction of undesirable flavors such as intense bitterness. After the malaxation comes the separation. The traditional process is a two-stage, three phase separation—first the combined liquids from the solid paste, then the oil from the aqueous layer. The solid, called olive pomace, can be extracted further with solvents to produce olive-pomace oil, burned, or discarded. We encountered one farmer who discovered that wild pigs enjoyed eating the olive pomace. The aqueous layer is called olive mill waste (OMW) or "black water." Introduction of centrifugal separation has allowed a one step or two-phase separation—the oil is separated directly from the olive paste, producing oil and a mixture of olive paste and OMW. Varying amounts of water are added to the mixture in the separation, but ideally this is minimized to reduce the volume of wastewater produced. While OMW is somewhat toxic and poses an expensive disposal problem, it is the subject of intense research to discover economically feasible uses.[12]

The oil produced can be filtered to reduce particulates or sold as is. While the technology has improved over the past few millennia, the fundamental process has remained unchanged. Archeologists at Klazomenai (Urla, Turkey) have discovered the remnants of an olive press with a clever phase separation system dating back to 600 BCE.[7−9,19] The reconstruction is shown in Figure 2.

Figure 2. Detail of the reconstructed olive press at Klazomenai, Urla, Turkey.

■ EXPERIMENT 1: PRODUCING YOUR OWN OLIVE OIL

A crude demonstration (or hands-on experiment) of the production of olive oil can be accomplished by simply crushing a few olives and draining off the liquid for examination. We have found that two good olive choices are fresh olives, which give a larger aqueous phase, or dark olives such as Kalamata olives, which lead to a colorless oil phase and a dark aqueous phase. Drawing the mixture into a pipet accentuates the separation and permits the two phases to be easily visualized. Having students realize that the oil layer is on the top and the aqueous layer is on the bottom allows for a lively conversation about how the lower density of olive oil (or any oil) causes it to float on water.

Sensory Analysis of Olive Oil: What Chemicals Give Rise to the Distinctive Aroma?

The aroma of olives and oil is chemically complex, distinct from other oils, and used by connoisseurs to distinguish highest quality olive oils. This aroma is affected by changes in the treatment of the olives (which unlike seeds will continue to ripen or decay after harvest), the conditions of the pressing and separation, and storage conditions of the oil. Even the irrigation of trees has an effect on aroma and bitterness.[20,21]

Figure 3 shows a few of the compounds with 5 and 6 carbons that account for many of the hundreds of major volatile

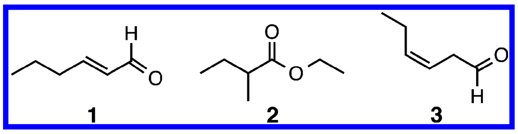

Figure 3. Some aroma compounds from olive oil: **1**, (*E*)-2-hexenal (green apple-like and astringent); **2**, ethyl 2-methylbutanoate (fruity); **3**, (*Z*)-3-hexenal (grassy).

compounds reported to be responsible for the aroma of olive oils.[22] Hexanal, *E*-2-hexenal, hexan-1-ol, and 3-methylbutan-1-ol are found in most virgin olive oils. Compounds that give olive oil its fruitiness include ethyl 2-methylpropanoate, ethyl 2-methylbutanoate, ethyl 3-methylbutanoate, and ethyl cyclohexanecarboxylate. The aldehydes, especially (*Z*)-3-hexenal, give the oils a grassy note, while 4-methoxy-2-methyl-2-butanethiol gives a black currant aroma.[23]

Of course, simple concentrations of aroma compounds are not indicators of their sensory impact; Reiners and Grosch divided the concentration by the threshold detection limit to get an odor activity value that was useful in identifying the most important compounds.[23]

Some aroma compounds are produced during the ripening process (very little olive aroma is associated with unripe olives), but much of the flavor is produced during the processing from linoleic and α-linolenic acids through a sequence of enzymatic reactions known as the lipoxygenase pathway, which is stimulated when olives are crushed during the process of oil extraction.[24] The crush releases enzymes such as lipoxygenases, hydroperoxide lyases, alcohol dehydrogenases, and acyl transferases that produce major fragrance components during malaxation.[25] It takes judgment and experience by the olive press manager to choose the proper conditions for malaxation.

Bitterness

Bitterness, like sweetness, is a sensation mediated by transmembrane G-proteins similar to rhodopsin.[26] Polyphenols in olive oil are the most likely source of this bitter taste.[27] Specifically, the main contributors to the bitterness sensation in olives are derivatives of the secoiridoids oleuropein (**4**) and ligstroside (**5**) (Figure 4). As oleuropein and ligstroside are quite water-soluble, their aglycones (compounds **6** and **7**, produced by the hydrolysis of the glycosidic linkage) are more likely to be the related compounds in the oil, and are considerably less bitter than the parent glycosides. The concentration of these bitter compounds is highest in green olives, and diminishes as the olives mature.[28]

Polyphenolic and flavonoid components are relatively easily oxidized, increasing their water-solubility and thus reducing their concentration in the oil (they end up in the aqueous waste water). This oxidation can be reduced by doing stages of the production (for example, malaxation) under a nitrogen or reduced oxygen atmosphere.[29,30]

Interestingly, some of these compounds (particularly oleuropein aglycone) have been shown to be quite active against HER2-specific breast cancer cells in vitro.[31] Other health claims range from skin protection, through cardioprotective actions like antiatherogenic and blood pressure

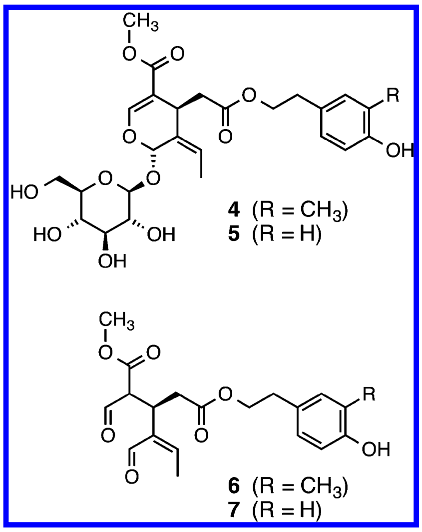

Figure 4. Bitter compounds found in olives and olive oil.

reduction to neuroprotective action, among others.[32] Oleuropein is also found in olive leaves, which are used for making tea.

A type of olive (*hurma*) that grows in older groves near Izmir, Turkey, has virtually no bitterness on harvest. The oil percentage of hurma olives is approximately 25.4%, and is of very good quality. Hurma olives are fermented on the tree with the help of the fungus *Phomo oleae*, which is local to this region.[33] Its geographic preference is unexplained, but the olives are used both for oil and for eating, as they require no curing.

Pungency

Pungency, an irritation in the back of the throat that often causes a cough, is poorly understood at a mechanistic level. Compounds must pass through the epithelium of mucus cells to reach shallow nerves in the back of the throat.[34] The compound responsible for this property, oleocanthal, shown in Figure 5, has a biological activity similar to that of the non-steroidal anti-inflammatory drug ibuprofen.[35,36]

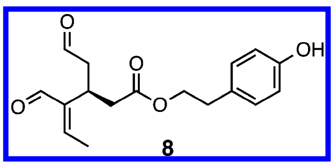

Figure 5. Oleocanthal is an anti-inflammatory compound naturally present in pungent olive oil.

The desirability of pungency is hotly debated in olive oil circles. While there is no simple qualitative test specifically for oleocanthal, we should note that a rapid NMR method for quantifying oleocanthal, suitable for advanced student laboratories, was recently published.[37]

EXPERIMENT 2: OLIVE OIL TASTING

A preliminary olive oil tasting helps set the stage for the experiments that follow. Expert tasters place samples of oil in small blue glass cups to mask the color, but we have had good luck with small plastic cups for less critical tasters. Our typical tasting begins by noticing the color of the oil followed by smelling the oil after gently warming it using the heat of your hand as shown in Figure 6. Tasting a small sample involves thoroughly coating the tongue, followed by an intake of breath over the oil in the mouth. Loud slurping sounds will accompany a proper tasting. While comparing several oils, students should observe and record the color, aroma, taste (bitterness), and pungency. Oils are included from a variety of different grades and varietals.

Figure 6. Tasting of the olive oil at the olive oil press Taş Koy.

Molecular models for olive oil components: triglycerides, free fatty acids, hydroxytyrosinol, and oleocanthal are displayed and used in the discussion. Students' questions of the meaning of virgin and extra-virgin oils, which refer to the level of free fatty acids (FFA), can be answered quickly when models of chemical structures of triglycerides are in hand.

Color and Spectroscopic Properties of Olive Oils

Olive oil is usually yellow, due largely to lutein at approximately 10−20 mg/kg. In addition, β-carotene is present at 1−5 mg/kg, lending an orange hue.[38,39] Figure 7 shows the major biomolecules that contribute to the color in olive oils. Other carotenoids present in olive oil but at lower concentration include α-carotene, phytofluene, ξ-carotene, auroxanthin,

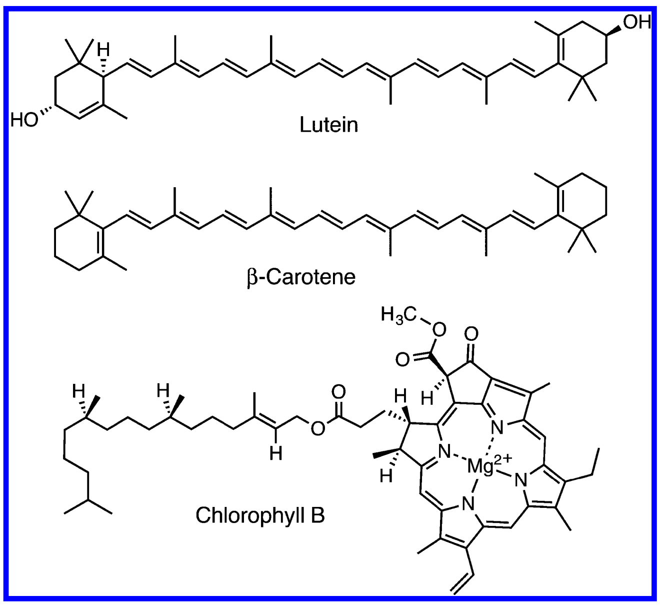

Figure 7. Common pigments associated with olive oil.

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luteoxanthin, violaxanthin, neoxanthin, and neochrome.[39] Depending on the ripeness of the olives, chlorophyll A and B may also be present, at 1−5 mg/kg. Other green colors may come from pheophytin A and B.[38] Both chlorophyll and pheophytins are fluorescent.[40]

Because green olive oil is associated with green, "fresh" olives, some may prefer a green olive oil to a yellow one. Given the role of chlorophyll as a photosensitizer (converting triplet to singlet oxygen in the presence of light), it poses an extra risk to the quality of olive oil on storage, especially after opening.

■ EXPERIMENT 3: THE RELATIONSHIP BETWEEN ABSORBED LIGHT AND OBSERVED COLOR

To understand the relationship between the presence of a particular molecule that absorbs light and the color of the substance that contains it, students performed an experiment with colored gummy candies and colored hand-held laser pointers according to a modified experiment previously reported.[41] Here, students discover as is shown in Figure 8 that the light from the green laser is absorbed by the red candy but transmitted by the green candy. Similarly, light from a red laser is transmitted by red candy and absorbed by the green candy. Students can measure how many candies of each color it takes to completely absorb the laser light. This experiment vividly underscores the relationship between the color of objects and the particular wavelengths of light that are absorbed and transmitted.

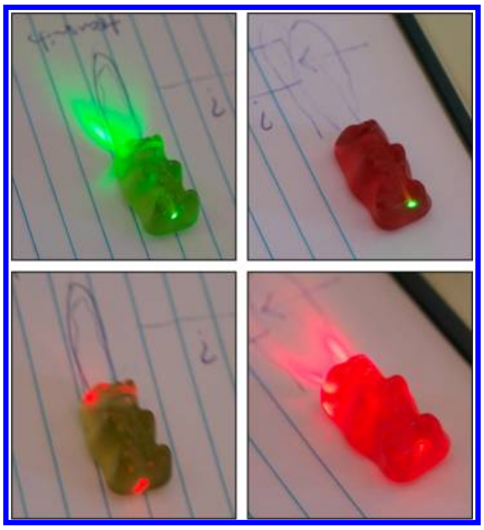


Figure 8. Demonstration of the relationship between the color of an object and the transmission of light. Green candies in column 1 transmit green light and absorb red. Red candies of column 2 absorb green light and transmit red. Fruit gum candy bears are shown.

■ EXPERIMENT 4: MEASURING THE VARIANCE IN THE COLOR OF OLIVE OILS

The color of olive oil can relate to the maturity of the olives used to make it: more green indicates more chlorophyll hence younger olives, more yellow indicates more lutein hence riper olives. Students can download and use an application for the iPhone, Irodori,[42] to measure the Red/Green/Blue (RGB) contributions to the color of the olive oils. A photo is taken with the iPhone, and the program breaks it down into color swatches. The user selects the most characteristic swatch for analysis and the RGB contribution (transmitted light) for that swatch is reported. To do this experiment reproducibly, students will need to account carefully for all of the elements of good spectroscopy: the need for uniform illumination, comparable path length, and good background control.

The best photos were taken under identical illumination conditions, with the same volume of solution, and using a white piece of paper as a background.

Students report the normalized RGB content for each of the oils by dividing the RGB for each oil swatch by the RGB of the background. As suggested by one reviewer, it is also possible to use the gummy candies (used in experiment 3) as samples for this exercise.

■ EXPERIMENT 5: USING FLUORESCENCE TO DETECT CHLOROPHYLL

As mentioned above, some olive oils may contain chlorophyll. These oils will appear greener than those that do not contain chlorophyll. While chlorophyll by itself does not add to the flavor profile of the oil, it is correlated with the oil being from a younger olive. These oils often contain more antioxidants and have a greener olive flavor.

Fluorescence can be used to identify the presence of certain materials. Background material for talking with students about fluorescence can be found in a previous article in this *Journal*.[43] Chlorophyll absorbs light in the blue region of the electromagnetic spectrum and fluoresces red. A green laser pointer provides enough energy to excite the chlorophyll nicely.[44] As shown in Figure 9, two oils that appear very similar in color can be distinguished by the fluorescence of chlorophyll in the sample on the right. A thorough study of olive oil fluorescence is available.[45]

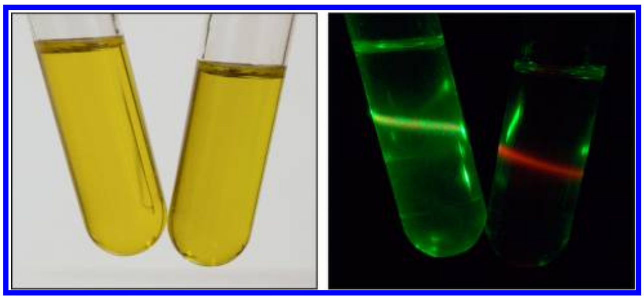


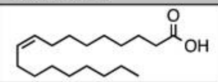
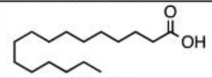
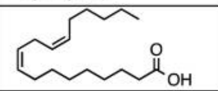
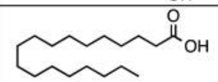
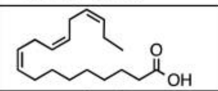
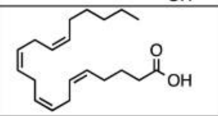
Figure 9. In ambient light (left panel), the slightly greener cast of the oil on the right is barely perceptible. However, when the two olive oils are illuminated with a green laser pointer (right panel), the red fluorescence of the oil on the right shows the presence of chlorophyll. The green beam in the oil on the left is due to scattering of the laser light; the absence of red fluorescence shows that chlorophyll is not present.

Most green food dyes are not fluorescent. Food color could be added to an inexpensive oil to make it appear as though it was a green olive oil. This deception can easily be detected by shining a green laser pointer through the olive oil. Even if it is green oil, if it does not fluoresce, it does not contain chlorophyll.

Qualitative Chemical Analysis of Olive Oil: Fatty Acid Chemistry

The most prevalent compounds in olive oil are the triglycerides, which give rise to the main physical properties of the product. While the dominant triglyceride is triolein (with three oleic acid moieties), a number of other fatty acids are present. Table 1

Table 1. Common Fatty Acids Related To the Standards for Evaluating Olive Oil

Shorthand/ Formula	Structure	IUPAC Name	Common Name	Melting Point (°C)	Approx. Percent in Olive Oil
18:1 (Δ^{9}) $C_{18}H_{34}O_2$		(*Z*) 9-Octadecenoic Acid	Oleic Acid	13	70
16:0 $C_{16}H_{32}O_2$		Hexadecanoic Acid	Palmitic Acid	63.1	10
18:2 ($\Delta^{9,12}$) $C_{18}H_{32}O_2$		(*9Z, 12Z*) 9,12-Octadecadienoic Acid	Linoleic Acid	5	10
18:0 $C_{18}H_{36}O_2$		Octadecanoic Acid	Stearic Acid	69.6	2
18:3 ($\Delta^{9,12,15}$) $C_{18}H_{30}O_2$		(*9Z, 12Z, 15Z*) 9, 12, 15-Octadecatrienoic Acid	α-Linolenic Acid	-11	1
20:4 ($\Delta^{5,8,11,14}$) $C_{20}H_{32}O_2$		(*5Z, 8Z, 11Z, 14Z*) 5, 8, 11, 14-Eicosatetraenoic Acid	Arachidonic Acid	-49.5	0

shows a subset of these with typical amounts, although there is considerable variation in samples. The abundance of oleic acid aids the production of oleoylethanolamide (OEA), which prolongs a sense of fullness between meals.[46] The absence of arachidonic acid and low levels of linolenic and stearic acids in authentic olive oil allow these compounds to serve as a signal for adulteration with other vegetable oils.

EXPERIMENT 6: FREE FATTY ACID CONTENT

Under proper harvest conditions, very little hydrolysis of triglycerides is seen. There are a number of bad harvest practices that lead to hydrolysis, however, including improper storage (too long, too warm or under too much mechanical pressure) and the presence of pests or mold. Since these bad practices lead to bad tasting oil (even though one can seldom taste the free fatty acids themselves), one of the primary international standards for olive oil is the level of free fatty acid that arises from hydrolysis of the triglycerides in the olives prior to processing

The industry standard analysis for free fatty acids (FFA) is a titration with standard NaOH. This uses a hazardous mixed solvent of equal parts ethanol and diethyl ether. The search for alternatives is exemplified by a recently published NMR method for FFA determination.[47] We required a faster and simpler qualitative use, and designed a reagent to give a simple yes or no answer as to whether the oil met a certain standard. Briefly, the reagent uses a fixed amount of standard NaOH and contains a pH indicator. If the level of FFA in the oil to be tasted exceeds the threshold amount, it consumes the base in the reagent and the indicator changes from blood red to yellow. If the level of FFA in the oil is below the threshold, the addition of oil does not consume the base, and no color change occurs leaving the reagent blood red. While this does not give the actual percent FFA, it very quickly shows whether or not the oil is above or below the threshold level.

Running the test simply requires mixing exactly equal volumes of olive oil and reagent in a small test tube or vial as shown in Figure 10. The color develops very quickly, making

Figure 10. Three Alizarin Yellow tests on an oil with 2% free oleic acid. From left to right, the reagent (base plus indicator) was chosen to test for 0.8%, 1.6%, and 3.0% oleic acid. This low quality oil fails the first two tests, and only passes the third, as would be expected.

this test much faster than a titration. When the aqueous layer remains red, the oil meets or exceeds the standard chosen. If instead, the aqueous layer turns yellow, the oil has failed and the FFA is higher than the cutoff. The emulsion that forms, especially with higher concentrations of oleic acid, did not affect the evaluation of the reagent color in the bottom aqueous phase.

EXPERIMENT 7: DEMONSTRATING UNSATURATION

Iodine can be used to demonstrate unsaturation that is characteristic of olive oil and many other vegetable oils. Using tincture of iodine purchased from a local drug store, students added two drops of tincture of iodine solution to the oils, and mixed well, noting the color immediately and at the end of the period. The distinctive purple color of the iodine disappears in the presence of a double bond, and is replaced by a brown to orange charge transfer complex.[48] The color of the iodine-containing samples can be compared with iodine mixtures with mineral oil as a negative control (no double bonds; stays purple) and canola oil as a positive control (polyunsaturated; turns orange). This particular assay evolves over time, with a first orange product that occurs immediately changing to a decolored solution at the end. The very slow reaction illustrates why despite naming the typical measurement of fatty acid unsaturation "iodine number" (the number of grams of iodine that would be consumed per 100 g of fat or oil), the actual reagent used to measure this value is the much more rapidly reacting I−Cl.[49]

EXPERIMENT 8: PEROXIDE CONTENT

Most peroxide content in olive oil is organic hydroperoxides (an example is shown in Figure 11), which can originate due to mistreatment/spoilage of olives, during the crush and malaxation steps or during storage if exposed to oxygen for long periods. Peroxide formation is accelerated dramatically by light exposure, especially in the presence of chlorophyll pigments,[50] but can also form in stored oil exposed to oxygen. While the peroxides themselves do not contribute to the taste of olive oil, they can lead to flavor changes over time. Therefore, high quality olive oils should have a peroxide value of less than 20 mequiv/kg. In this assay, we used peroxide strips that detect organic peroxides at concentrations well below that value. Organic hydroperoxides form one of the major challenges to antioxidants in olive oil.

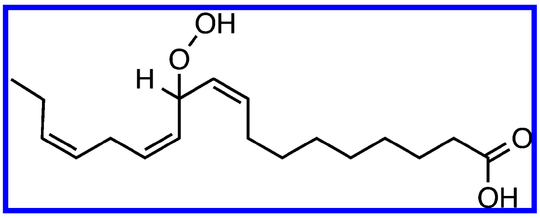


Figure 11. One of several possible hydroperoxides formed from α-linolenic acid.

EXPERIMENT 9: ANTIOXIDANT CONTENT

Polyphenolics and Vitamin E in olive oil are proven antioxidants.[51] To demonstrate the presence of antioxidants, we turned to classic blueprint chemistry. We asked students to add olive oil to a mixture of aqueous iron(III) chloride and potassium ferricyanide solution. The characteristic Prussian Blue color that develops in the presence of reducing agents can be used in a qualitative test for phenols.

The chemistry of antioxidant assays[52] can make an interesting discussion in a more advanced course. Not only are there several possible mechanisms for the redox chemistry, but assays (including ours) use a nonbiological oxidant, necessitating the development of an explicit connection to establish that health or other effects correlate with measured antioxidant capacity. In the case of olive oil, one well-established role for antioxidants is the preservation of fresh olive oil after it has been exposed to air. On the other hand, the health effects of these compounds are so broad that they are likely to have mechanisms beyond simple antioxidant capacity.

SUMMARY

Olive oil is a complex, interesting, and economically important material found in most households. Using olive oil as the subject of chemical analysis helps to motivate the understanding of basic chemical properties such as acidity, chemical structure, and oxidation and reduction in first and second semester chemistry classes. At the same time, the growing status of olive oil as "the healthy oil"[53] makes it of general interest to a broader audience that includes all health conscious consumers. We have found that these nonscientists can be induced to learn chemistry when they understand that it will help them make wise food choices on their way to better health. Courses taught to nonmajors might include these experiments in a lab or demonstration format.

ASSOCIATED CONTENT

Supporting Information

Instructor's notes 1−9 and worksheets 1−9. This material is available via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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