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Applications of Differential Scanning Calorimetry (DSC) in Oils and Fats Research. A Review

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Abstract

This review is designed to be a comprehensive review in a new way to help you to understand the principle and theory of Thermal Analysis with special emphasis on Differential Scanning Calorimetry (DSC) as a new fast-growing and important technique used for authentication, characterization and detecting adulterations of oils and fats. DSC is a powerful instrument that measures the energy absorbed or released as a function of time or a controlled temperature profile. The sensor of the DSC is the heat flux plate which is designed to give superior performance and rugged reliability. The heat flux plate is capable of measuring small energy changes over the entire temperature range. Examples of measurements with DSC are Oxidative Stability, Melting Enthalpy, Glass Transition, Heat of Crystallization, Purity Determination and Heat Capacity. DSC can be used as a rapid method for assessment of oxidative stability, prediction of shelf life and evaluation of the quality of edible oils during refining. DSC holds a potential to be used as the reliable and reproducible technique for the detection of adulteration of animal body fat added in ghee individually and in combination of vegetable oil. DSC method is faster, require less sample size and no chemicals or solvents compared to other conventional, modern oxidative stability methods and conventional shelf life estimation.

Keywords: *Oils, Fats, Thermal Analysis, DSC*

Introduction

Thermal analysis is one of the oldest analytical techniques. Throughout history, people have used simple heat tests to determine whether materials were genuine or fake. The year 1887 is looked upon as the dawn of present-day thermal analysis. It was then that Henri Le Chatelier, the famous French scientist, carried out his first thermometric measurements on clays. Just a few years later in 1899, the British scientist Roberts-Austen performed the first differential temperature measurements and so initiated the development of DTA. Commercial instruments did not however appear until the 1960s. Since then, thermal analysis has undergone fifty years of intense development. As a result, the use of thermal analysis has expanded into many new research and application fields in different industries. In many cases, the thermal analysis techniques employed to analyze new materials require specific accessories and measurement parameters as well as special sample preparation [1].

Differential scanning calorimetry (DSC) is the most widely used of the thermal techniques available to the analyst and provides a fast and easy to use method of obtaining a wealth of information about a material, whatever the end use envisaged. It has found use in many wide ranging applications including polymers and plastics, foods and pharmaceuticals, glasses and ceramics, proteins and life science materials; in fact virtually any material, allowing the analyst to quickly measure the basic properties of the material. Many of the application areas are

dealt with in greater depth within the chapters of this book, and the principles involved extend to many other materials that may not be mentioned specifically. It is in fact a fascinating technique and the purpose of this introduction is to provide an insight into this method of measurement, to provide the necessary practical guidance a new user will need to go about making measurements, and to give understanding about the information that can be obtained and how to interpret the data [2].

The differential scanning calorimeter (DSC) is a fundamental tool in thermal analysis. It can be used in many industries – from pharmaceuticals and polymers, to nanomaterials and food products. The information these instruments generate is used to understand amorphous and crystalline behavior, polymorph and eutectic transitions, curing and degree of cure, and many other material properties used to design, manufacture, and test products. The biggest advantage of DSC is the ease and speed with which it can be used to see transitions in materials. If you work with polymeric materials of any type, the glass transition is important to understanding your material. In liquid crystals, metals, pharmaceuticals, and pure organics, you can see phase changes or polymorphs and study the degree of purity in materials. If you are processing or distilling materials, knowledge of a material's heat capacity and heat content change (called enthalpy) can be used to estimate how efficiently your process is operating. For these reasons, DSC is the most common thermal analysis technique and is found in many analytical, process control, quality assurance, and R&D

laboratories [3].

In Edible oils research, Oxidative Stability, or the Oxidative Induction Time (OIT), test is often studied in both DSC and TGA. This is normally done by heating a material to a set temperature under an inert gas and switching to air or oxygen once it has equilibrated. The time needed for the material to begin to burn is then recorded. Normally, in a power compensated DSC, a flow-thru cover is used to remove the smoke from the DSC as quickly as possible.

High Pressure (HP) DSC is used for several reasons: first, an oxidative stability test may take too long at atmospheric pressures to be convenient. An example would be looking at an antioxidant package in motor oil. Secondly, some reactions from water or methanol as a byproduct, leading to foaming in the sample. Higher pressure suppresses this. Thirdly, some reaction kinetics are affected by pressure and running the reaction under controlled pressure is needed to study this effect. Finally, transitions, like the Tg and boiling point, are responsive to pressure and running DSC under pressure allows you to study that process. For boiling points, pressure DSC also allows you to calculate the vapor pressure of the sample.

Kinetic studies on the DSC can be done using scanning methods, where the sample is either heated through a temperature ramp, or isothermally, where the sample is held at a set temperature. In the latter case, the ramp rate to that temperature should be as fast as possible to minimize the effect of the ramp. Data from these methods can be exported to TIBCO Spotfire®, Excel® or another program for analysis or run through several commercially available programs. The advantage of using DSC for kinetic studies is it tends to be faster and more straightforward than other methods.

Choosing the thermal analysis technique

The thermal analysis technique that can be used to measure a particular property depends on the effect or property you want to measure. The following table gives an overview of the best (■) and alternative (■) techniques [1].

Table 1: *Overview of the Best (■) and Alternative (■) Thermal Analysis Techniques*

	DSC	TGA	TMA	DMA
Physical properties				
Specific heat capacity	■	■	–	–
Expansion coefficient	–	–	■	■
Young's modulus	–	–	■	■
Physical transitions				
Melting and crystallization	■	–	■	■
Evaporation, sublimation, drying	■	■	–	–
Glass transition, softening	■	–	■	■
Polymorphism (solid-solid transitions)	■	–	■	–
Liquid crystals	■	–	–	–
Purity analysis	■	■	–	–
Chemical properties				
Decomposition, degradation, pyrolysis, oxidation, stability	■	■	–	–
Composition, content (moisture, fillers, ash)	■	■	–	–
Kinetics, reaction enthalpies	■	■	■	■
Crosslinking, vulcanization (process parameters)	■	–	■	■

Principle of Thermal Analysis

The definition of Thermal Analysis: "A group of techniques in which a property of the sample is monitored against time or temperature while the temperature of the sample, in a specified atmosphere, is programmed" [4].

Figure 1 shows the configuration of Thermal Analysis instrument includes the followings:

• Detection Unit: Furnace, sample and reference holder, and sensor, heat and cool the sample in the furnace, and detects the sample temperature and property.

• Temperature Control Unit: Controls the furnace temperature.

• Data Recording Unit: Records the signals of sensor and sample temperature, and analyzes them.

Temperature control, data recording and analysis are all computer-controlled. The combination of the furnace and sensor enables the various types of the measurement techniques. This computer can be connected to the several instruments which has the other types of measurement techniques, enables the simultaneous measurement and analysis.

Table 2. shows the measurement techniques for each property in Thermal Analysis (TA). Each technique is called DTA, DSC, TG, TMA and DMA.
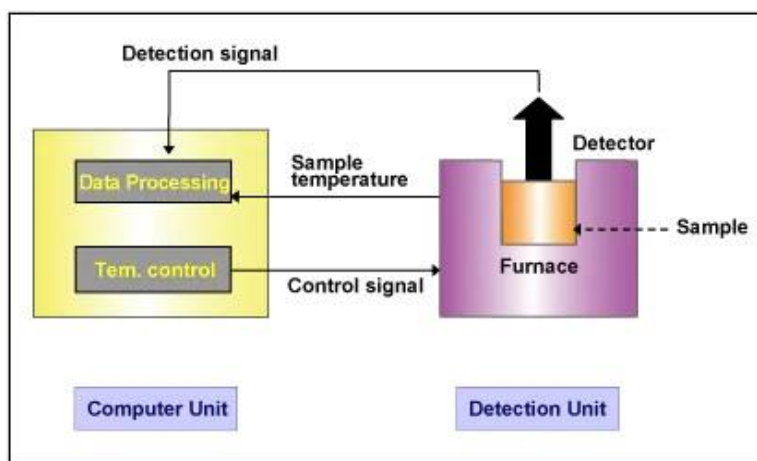

Figure 1: Block diagram of Thermal Analysis instrument

Table 2: The measurement techniques for each property in Thermal Analysis

Abbrev.	TA Measurement Technique	Property	Unit
DTA	Differential Thermal Analysis	Difference temperature	°C or μV*
DSC	Differential Scanning Calorimetry	Enthalpy	W = J/sec
TG	Thermogravimerty	Mass	gram
TMA	Thermomechanical Analysis	Deformation	meter
DMA	Dynamic Mechanical Analysis	Elasticity	$Pa = N/m^2$

*Thermocouple electromotive force.

Table 3: The effects and properties found in various materials are measured in each TA technique.

Phenomenon/Property	Technique			
	DSC	TG	TMA	DMA
Melting	◯	—	Δ	Δ
Glass transition	◯	—	◯	◯
Crystallization	◯	—	Δ	◯
Reaction (Curing / Polymerization)	◯	Δ	◯	◯
Sublimation / Evaporation / Dehydration	Δ	◯	—	—
Thermal decomposition	Δ	◯	—	—
Thermal expansion / Thermal shrinkage	—	—	◯	—
Thermal history	◯	—	◯	◯
Specific heat capacity	◯	—	—	—

◯ : Measurement Object

Δ : Some compatible

— : Not measured

Table 3. shows which effects and properties found in various materials are measured in each technique.

The main application areas of DSC concern the determination of the melting, the glass transition, the crystallization, the chemical reactions, the thermal history, and the specific heat capacity. Though the sublimation, the evaporation, and the thermal decomposition can also be measured, this technique is not usually used because of the mass change by the decomposition and the corrosion of the sensor by the decomposed gas.

This is definition of DSC by JIS (Japanese Industrial Standard). DSC is the generic term for the following two measurement methods:

• Heat Flux DSC

A technique in which the temperature of the sample unit, formed by a sample and reference material, is varied in a specified program, and the temperature difference between the sample and the reference material is measured as a function of temperature.

• Power Compensation DSC

A technique in which difference of thermal energy that is applied to the sample and the reference material per unit of time is measured as a function of the temperature to equalize their temperature, while temperature of the sample unit, formed by the sample and reference material, is varied in a specified program.

Differential Scanning Calorimetry (DSC)

Description of DSC

DSC is a commercially available instrument which has two (2) types: Heat Flux Type and Power Compensation Type. Figures 2, 4 show the block and schematic diagrams of Heat Flux DSC as an example, whereas, Figure 3 shows block diagram of DSC furnace. Heat Flux DSC comprises the sample and reference holder, the heat resistor, the heat sink, and the heater. Heat of heater is supplied into the sample and the reference through heat sink and heat resistor. Heat flow is proportional to the heat difference of heat sink and holders. Heat sink has the enough heat capacity compared to the sample. In case the sample occurs endothermic or exothermic phenomena such as transition and reaction, this endothermic or exothermic phenomena is compensated by heat sink. Thus the temperature difference between the sample and the reference is kept constant. The difference the amount of heat supplied to the sample and the reference is proportional to the temperature difference of both holders. By calibrating the standard material, the unknown sample quantitative measurement is achievable.
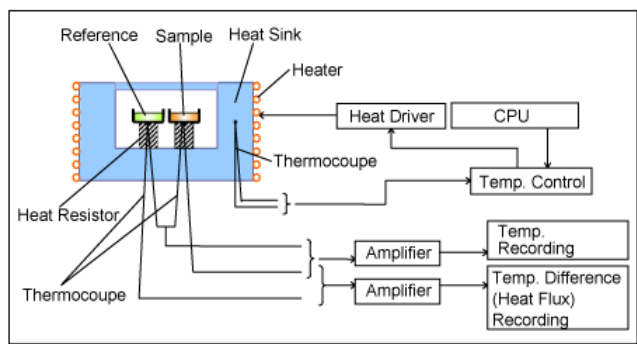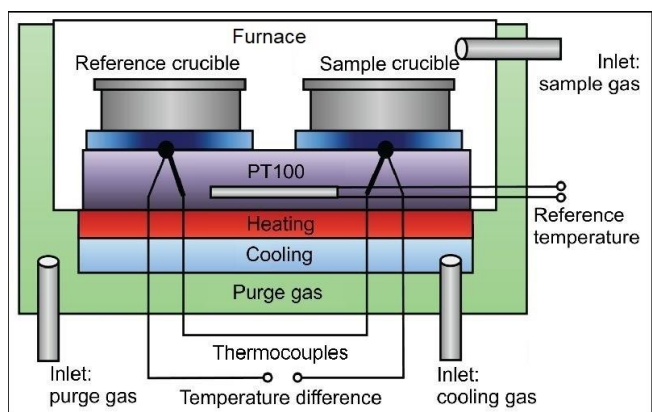
Figure 2: *Block Diagram of Heat Flux DSC*

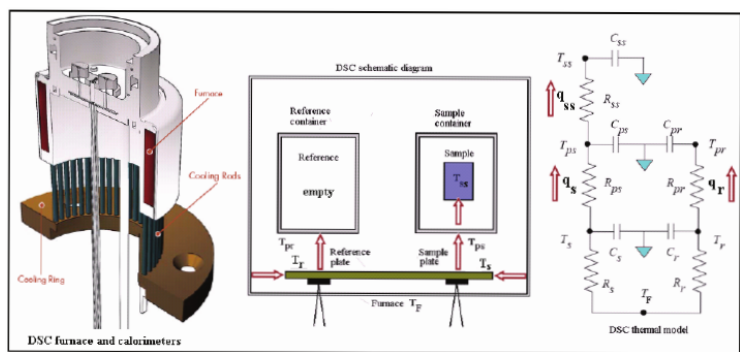**Figure 3:** *Block Diagram of DSC Furnace*

Figure 4: *Schematic Diagram of Heat Flux DSC*

DSC Measuring Principles

Figure 5 shows DSC Measuring Principles as following:

a) Power compensation DSC: The electric heating power required to compensate the difference in heat flow is measured

b) Heat flux DSC: The temperature difference caused by the difference in the heat flow of the sample and reference is measured.

* Aluminium pans are the most commonly used DSC pans.

* Important to ensure that the contact between the sample and the pan is very good.

* Large contact area is desirable, in order to have uniform heat transfer.

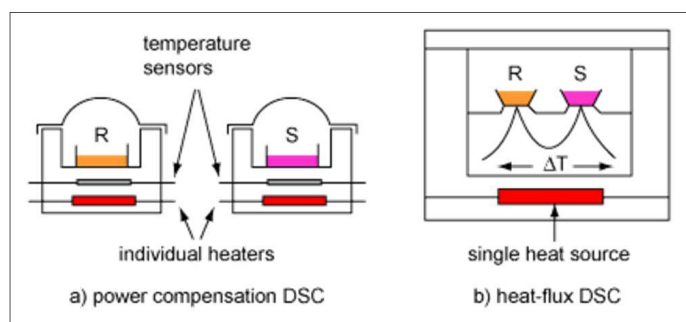* Usually, the reference is an empty pan covered with a sealed lid.

Figure 5: *DSC Measuring Principles*

Endothermic and Exothermic Heat Flow

Figure 6 shows DSC Thermogram as following:

• Endothermic event: heat (energy) absorbed by sample

• Exothermic event: heat (energy) released by sample

• Glass transition: Endothermic

• Melting: Endothermic

• Crystalisation: Exothermic

• Vaporization: Endothermic

• Chemical reactions: Exothermic

• Oxidation: Exothermic

• Degradation: Exothermic

Principles of DSC and types of measurements made

A definition of DSC

A DSC analyzer measures the energy changes that occur as a sample is heated, cooled or held isothermally, together with the temperature at which these changes occur. The energy changes enable the user to find and measure the transitions that occur in the sample quantitatively, and to note the temperature where they occur, and so to characterize a material for melting processes, measurement of glass transitions and a range of more complex events. One of the big advantages of DSC is that samples are very easily encapsulated, usually with little or no preparation, ready to be placed in the DSC, so that measurements can be quickly and easily made.

Heat flow measurements

The main property that is measured by DSC is heat flow, the flow of energy into or out of the sample as a function of temperature or time, and usually shown in units of mW on the y-axis. Since a mW is a mJ/s this is literally the flow of energy in unit time. The actual value of heat flow measured depends upon the effect of the reference and is not absolute. What matters is that a stable instrumental response or baseline is produced against which any changes can be measured. The starting point of the curve on the y-axis may be chosen as one of the starting parameters, and it should be set at or close to
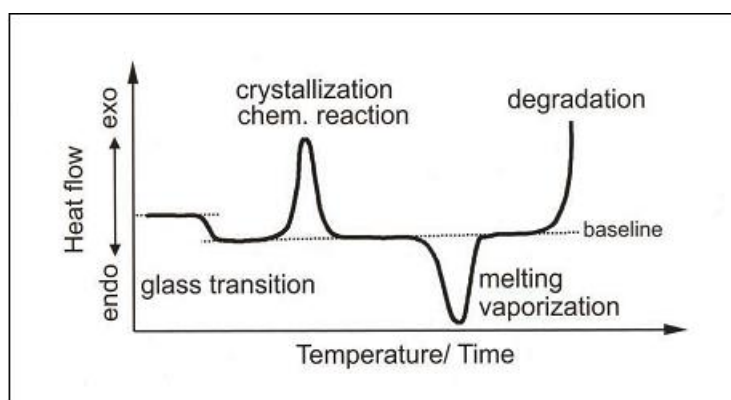

Figure 6: *DSC Thermogram*

zero. Two different conventions exist for the display of the heat flow curve: one shows endotherms in the downward direction, the other upward. Traditionally, with heat flux systems endotherms are shown as going down, since endothermic transitions result in a negative temperature differential, whilst with power compensation systems they are shown as going up since with this principle endothermic transitions result in an increase in power supplied to the sample. The value of measuring energy flow is that it enables the analyst to identify the range of different transitions that may occur in the sample as it is heated or cooled.

Specific heat (Cp)

The specific heat (heat capacity, Cp) of a material can be determined quantitatively using DSC and is designated Cp since values are obtained at constant pressure. Traditionally, this is done by subtracting a baseline from the heat flow curve in the manner described below, but values may also be obtained using modulated temperature technique. The subtracted curve referenced against a standard gives a quantitative value of Cp. The accuracy that can be obtained depends upon the instrument and method in use.

In practice the traditional standard test method provides a fairly rapid method for determination of Cp and many manufacturers provide software specifically designed to comply with this. Three runs are required, each consisting of an isothermal period, temperature ramp and final isotherm. This method is applied identically to the succeeding runs:

1. First run: a baseline with uncrimped empty pans placed in the furnace.

2. Second run: as above but adding a reference (typically sapphire) to the sample pan.

3. Third run: replace the reference with your sample.

The three curves are brought up on the screen, isothermals matched, data subtracted and referenced against the standard. Most software packages will do this automatically, and if the differing weight and heat capacity of sample pans are taken into account then the baseline and reference runs may be used for subsequent samples, provided the DSC is stable. In fact, because the procedure is based on a subtraction technique between measurements made at different times, any drift will cause error. The DSC must be very stable and in practice it is best not to use an instrument at the extremes of its temperature range where stability may be compromised. The standard most often used is sapphire, and the mass used should be similar to the sample; in any event the sample should not be a great deal larger or errors will be increased. This method relies on the measurement of the heat flow of the sample compared to that of an empty pan. Whilst there may be a number of factors which dictate the scan rate of choice it should be noted that faster scan rates result in increased values of heat flow giving increased accuracy of measurement, and this also minimizes the time of the run and potential drift of the analyser. It has been reported that fast scan rates used by fast scan DSC can give extremely accurate data [2].

A similar principle is employed in stepwise heating methods where the temperature may be raised by only a fraction of a degree between a series of isotherms. This is reported to give a very accurate value for Cp because of the series of short temperature intervals.

Specific heat data can be of value in its own right since this information is required by chemists and chemical engineers when scaling up reactions or production processes, it provides information for mathematical models, and is required for accurate kinetic and other advanced calculations. It can also help with curve interpretation since the slope of the curve is fixed and absolute, and small exothermic or endothermic events identified. Overall, it gives more information than the heat flow trace because values are absolute, but it does take more time, something often in short supply in industry [2].

Enthalpy

The enthalpy of a material is energy required to heat the material to a given temperature and is obtained by integrating the heat capacity curve. Again many software packages provide for the integration of the Cp curve to provide an enthalpy curve. Enthalpy curves are sometimes used for calculations, for example when calculating fictive temperature, and can help in understanding why transitions have the shape they do. In the cases where amorphous and crystalline polymer materials exhibit significantly different enthalpies, the measurement of enthalpy can allow an estimate of crystallinity over a range of temperatures as the polymer is heated [2].

Derivative curves

Derivative curves are easily obtained from the heat flow curve via a mathematical algorithm and aid with interpretation of the data. Typically they can help define calculation limits, and can aid with the resolution of data, particularly where overlapping peaks are concerned. The first derivative curve is useful for examining stepwise transitions such as the glass transition. The second derivative of a peak is more easily interpreted than the first derivative. In this case the data are inverted, but any shoulders in the original data will resolve into separate peaks in the second derivative curve. It is particularly useful for examining melting processes to help identify shoulders in the peak shape due to multiple events.

Applications of DSC in Oils and Fats Research

Two kinds of fats were investigated in this study: palm oil and cocoa butter. Cocoa butter is one of the most precious and useful vegetable fat obtained from the cocoa beans. Palm oil is an edible vegetable oil high in saturated fats and free of trans fats. A lot of methods for determining oxidative stability are known. Differential Scanning Calorimetry is in the group of thermoanalytical methods and allows the determination of oxidation parameters without the need for chemicals substances. With the use of DSC, polythermal and isothermal research can be carried out. The aim of this research was to analyse the oxidative stability of cocoa butter and palm oil. The kinetics parameters: activation energy (Ea), preexponential

factor Z were investigated with used differential scanning calorimetry (DSC). The initial temperature (onset) and maximum temperature (final) of oxidation process, and melting characteristic for both lipids were determined by DSC. The kinetics parameters, temperatures (onset and maximum) were investigated at oxygen atmosphere. The melting characteristic was investigated at nitrogen atmosphere. Calibration was done with indium standards. The averages from measurements of Ton and Tmax for each lipid at a given temperature were determined as the intersection of the extrapolated baseline and the tangent line (leading edge) of the recorded exotherm. The range of temperatures during determine melting characteristic were –80 to 80°C. If the heating rate of the system was constant for the test conditions, then the temperatures obtained: Ton and Tmax were characteristic of the system and could be used as parameters differentiating the resistance (stability) of fats and oils to thermal decomposition. Palm oil was characterized by a lower activation energy at onset temperature than cocoa butter, it may be due to the fact that palm oil was unrefined [5].

The oxidative stability of vegetable oils mainly depends on their fatty acid composition, their degree of unsaturation, and the presence of compounds with antioxidant activity. A research article reports on the effects of the process of roasting oil seeds, prior to pressing them, on the basic characteristics of the oils produced and their oxidative stability. The differential scanning calorimetry (DSC) technique was used to study the process of oxidation of the oil samples in an oxygen-flow cell. Chromatographic analysis revealed that roasting the seeds increased the levels of chlorophyll and β-carotene in all the cold-pressed oils. Similar results were observed for the oil's antioxidant activity, measured by the scavenging 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical method. Results also indicated that roasting seeds prior to pressing them for oil had a positive effect on the oil's stability, as determined by the DSC method. This manifested in both the extension of oxidation induction time and the final oxidation time. Most researchers publishing in the subject of DSC oils oxidation consider only induction time parameter. However, significant differences were also observed in the part of the DSC curve after the induction time [6].

27 market and edible cold-pressed oils from 10 different oilseeds were analyzed. Oxidative stability and chemical composition of oils were evaluated. The oils were investigated for their primary quality, fatty acid composition, total phenolic content and antioxidant activity. Rancimat and pressure differential scanning calorimetry (PDSC) were used to assess oils oxidative stability. Principal component analysis (PCA) was conducted to determinate impact of selected chemical characteristics on tested oils' oxidative stability in accelerated modes. According to the statistical analyses, oils were divided into 4 groups, which depend on the method of oxidative stability evaluation did not differ [7].

With the use of DSC method, the assessment of oxidative stability of sunflower and soybean oils, and also efficiencies of used herbal plant extracts were performed. SFO containing a slightly higher level of unsaturated fatty acids compared to SBO appeared to be less stable. TON values were higher for soybean oil samples without the addition of herbal plant extracts and also when they were enriched with marjoram, thyme, and oregano extracts in comparison with sunflower oil samples. The calculated activation energy values for the oxidation reaction of tested oil samples showed that the effectiveness of used herbal plant extracts varied with the type of herbal plant extract added to oil samples and with their concentration. Oregano extract at concentration of 0.07% was effective for protection of SFO against oxidation and thyme extract at concentration of 0.07% improved the oxidative stability of SBO. Therefore, these herbal plant extracts can be recommended as a potent source of natural antioxidants replacing synthetic antioxidants for protection of edible oils against oxidation [8].

Differential scanning calorimetry (DSC) was for the first time applied to evaluate cooling and heating curves and related thermal properties of olive oil during refining process steps starting from two series of samples at different initial peroxide values (low, 15 meq O2/kg and high, 30 meq O2/kg). Thermal properties were statistically correlated by means of principal component analysis (PCA) to those substance classes (products deriving from oxidation, polymerization and hydrolysis of triacylglycerols (TAG)) that could exert adverse effects on consumer health and a negative contribution to the shelf-life of the refined oil. A concomitant significant increase of K270 and polymerized TAG, as a general decrease of oxidation level was exhibited by oils during bleaching. Crystallization significantly shifted towards lower temperatures as well as onset temperature of heating, enlarging transition range, starting from bleaching and at both oxidation levels. PCA was performed to tentatively discriminate among samples according to different refining steps and/or initial level of oxidation correlating thermal properties and chemical results. The first principal component clearly clustered samples according to different refining steps and this differentiation was possible on the basis of the relation of thermal parameters with polymerized TAG conjugated dienes and trienes (K232 and K270, respectively). This preliminary goal should lead to deepen the study of DSC application in the evaluation of the quality of olive and other vegetable oils during refining [9].

DSC was used to detect adulteration of pure ghee with caprine body fat when added singly (at 5, 10 and 15%) and in combination with groundnut oil (GNO) (at 5, 10 and 15%). Samples were analyzed for transition behavior in terms of crystallizing and melting curves. When compared to pure ghee, adulterated ghee samples showed a shift in the midrange temperature of thermal curves, indicating the presence of foreign fats. The results revealed that the detection of adulteration was possible at the lowest level of the study (5%), irrespective of the nature of the adulterants. Differential scanning calorimetry is a fingerprinting technique that can be efficiently used for the detection of adulteration of milk fat by cheaper oils and fats, based on the rise in midrange temperature of crystallisation and melting peaks. It can be concluded that CBF when added individually and in combination with GNO in PCG and PBG

could be detected even at the lowest level of study (5%). Also, the change in shape of the melting curve in adulterated samples further provided a clear indication of adulteration. Thus, DSC holds a potential to be used as the reliable and reproducible technique for the detection of adulteration of animal body fat added in ghee individually and in combination of vegetable oil. However, further strengthening of the technique is required in field conditions [10].

The use of differential scanning calorimetric heating thermograms was investigated to detect the presence of lard, beef tallow, and chicken fat as contaminants in sunflower oil. Sunflower oil samples spiked separately with lard, beef tallow, and chicken fat in levels ranging from 1 to 20% (w/w) were analyzed using high performance liquid chromatography to get triacyl-glycerol profiles and differential scanning calorimetric to obtain their heating profiles. The results showed that below 20% (w/w) level of contamination, both lard and beef tallow in sunflower oil can be detected using characteristic contaminant peaks appearing in the higher temperature region (0–50°C) of differential scanning calorimetric curve. However, chicken fat contamination in sunflower oil did not show any characteristic peaks in this region, but caused changes only in the exiting thermal transitions in the low temperature region of the differential scanning calorimetric curve. Based on the characteristic differences in the peak size, shape, and position, it was also possible to make a distinction between lard and beef tallow contaminations in sunflower oil. The DSC melting curve region of SFO from 0 to 50°C was found to be sensitive to the TAG compositional changes caused by different animal fats. While the changes caused by BT in thermal profiles could even be seen at a 2% level, the changes due to LD could be seen only after an 8% level. Based on the characteristic shape, size, and position of the contaminant peaks, it was possible to distinguish lard contamination from other animal fat contaminations in SFO [11].

The applicability of differential scanning calorimetry (DSC) for the detection of water content in butter was evaluated. High correlation coefficients were found between the water content and the enthalpies of the ice melting/water crystallization. The correlation equations were adopted to calculate the water content for seven tested kinds of butter, and the results were compared with the values, obtained by using the reference method. The difference between the water content determined by the reference method and by DSC ranged between 0.2 and 2.6% for the measured enthalpy of ice melting, and between 1 and 5.6% for the enthalpy of water crystallization. In relation to the data obtained, it can be concluded that the parameter of ice melting enthalpy can be used in the identification of adulterations or confirmation of butter authenticity [12].

CONCLUSIONS

Advances of DSC technique have led to new applications for authentication, characterization and detecting adulteration of oils and fats. DSC can be used as a rapid method for assessment of oxidative stability, prediction of shelf life of oils and fats and evaluation of the quality of edible oils during refining. The DSC method is faster, require less sample size and no chemicals or solvents compared to other conventional and modern oxidative stability methods and conventional shelf life estimation.

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