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# THERMAL STRESS ANALYSIS OF DIETARY FATS

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***Abstract***

*The aim of this paper is the assessment of thermal stress effects on dietary fats. For this purpose, oils and fats from those sold on the Romanian market, intended for home use and in public food establishments were selected (sunflower oil, palm oil, lard and vegetable fat). These oils and fats are some of the most used ones in cooking. Dietary fats were subjected to heat stress by gradually heating to different temperature levels as well as repeated exposure to high temperatures. Subsequently, the influence of temperature on the characteristic parameters of food fats was analysed: free acidity, indices of: acidity, iodine, peroxide, saponification, refraction, water content and volatile substances, unsaponifiable matter. The results of the present study provide useful information for choosing a dietary fat suitable for frying food, in order to minimize the undesirable effects for the health of the consumer and to obtain a quality fried product.*

***Key words***: *animal fat, dietary fats, oil, Romanian market, thermal stress.*

## INTRODUCTION

Dietary fats are part of the macronutrients category, which, along with proteins and carbohydrates, are important nutrients that the body needs to function normally (Banu et al., 2002; Alais et al., 2020; Oteng & Kersten, 2020). In the daily diet, the intake of fats is ensured by the consumption of fats of animal and vegetable origin, as such (butter, lard, oils) or contained in food (meat and meat products, dairy products, fish, avocados, olives, oleaginous seeds) (Lee, 2017).

The classification of fats is based on several criteria: origin and properties, consistency at $20^0$C, type of fatty acids that predominate in their composition. Another criterion for classifying dietary fats is the water content (Banu et al., 2002; Alais et al., 2020). Thus, they can be divided into three types: vegetable and animal oils (maximum 0.2% water content), animal fats (maximum 8% water), butter and margarine (maximum 16% water) (Malesza et al., 2021).

In the context of the globally accelerated increase of edible oils prices, the trend in choosing them for consumption, especially for the thermal preparation of food by frying, is directed towards the purchase of products at affordable prices, without emphasizing quality. For the same reason, when frying, oil is used more times than recommended, which is not beneficial, neither for health nor for the quality of the fried products (Radzikowska et al., 2019; Santos et al., 2018; Ding et al., 2022).

During the frying process, chemical reactions that occur cause irreversible changes in the properties of the oily product, during the entire thermal process (Choe & Min, 2007; Bhat et al., 2022; Choe & Min, 2007; Matthäus, 2010). These reactions are determined by the biochemical composition of the oil and also by the physico-chemical stress factors (temperature, light, presence of oxygen in the air) (Hassanzadazar et al., 2018).

Thermal stability evaluation is an important tool to determine the optimal utilization of the oil process in order to maximize profitability and satisfy consumer preferences and consumer safety requirements (Zhuang et al., 2022; Negishi et al., 2003).

The frequent consumption of foods in the preparation of which heated oil was used may trigger a postprandial pro-inflammatory status (Malesza et al., 2021; Lee, 2017).
At the same time, the consumption of products thermally prepared in burnt oil should be avoided, as there is a risk of atherosclerosis and rheumatoid arthritis (Radzikowska et al., 2019). This oil is more dangerous because it doesn't necessarily have a burning taste, so it's not easy to identify (Ioannou et al., 2023).
Compounds formed during frying can contribute to the imbalance of the microbiome, that can lead to inflammatory bowel disorders (Narula et al., 2021). At the same time, lower microbiome diversity as a result of fried food consumption is associated with lower immunity and digestive problems (Wastyk et al., 2021).

## MATERIALS AND METHODS

The edible fats subjected to heat stress (oils and fats) were selected from those sold on the Romanian market, intended for domestic use and in public catering establishments: oils obtained from oilseeds (sunflower, rapeseed), oil fruits (palm), vegetable fat (margarine) and animal fat (lard).
The ingredients, as well as the recommendations regarding how to use each dietary fat can be found in Table 1 and are those written on the label/packaging of each product.

Table 1. Characteristics of the analysed dietary fats

| Product name | Ingredients | Recommendations |
|---|---|---|
| Refined sunflower oil "Vita D'or" | refined sunflower oil rich in monounsaturated fatty acids (oleic acid) | Recommended for frying |
| "Yunus" palm oil | refined vegetable oil (palm oil and its fractions), antioxidant (butylhydroxytoluene (BHT) - E321), antifoam (dimethylpolysiloxane - E900a) | Recommended for frying |
| "Angst" lard | lard, unsalted sea salt | - |
| Spreadable vegetable fat 72%, "Vita D'or" | vegetable fats (palm, coconut), water, rapeseed oil, emulsifiers: lecithin, mono- and diglycerides of fatty acids 0.2% salt, flavour, acidity corrector: citric acid, vitamin A, vitamin D, dye: carotenes | Product for baking and cooking |

**Samples preparation**
The thermal stress was applied in a temperature gradient regime (100⁰C, 200⁰C and 240⁰C, 30 minutes for each temperature) (program 1), respectively constant temperature (cycles of 30 minutes at 175⁰C) (program 2). The work scheme is presented in Figure 1.

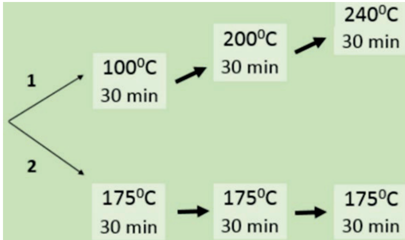


Figure 1. Scheme of thermal stress applied to food fats

**Analysis methods used for sample characterization**
Both the raw (before thermal stress) and the heat-treated samples were analysed according to the standardized analysis methods mentioned in Table 2.

Table 2. Standardized methods used for physico-chemical characterization of samples

| No. | Analysed parameter | Unit | Standardized method |
|---|---|---|---|
| 1. | Free acidity | % oleic acid | SR EN ISO 660-2020 |
| 2. | Acidity index | mg KOH / g fat | |
| 3. | Iodine index | g I / 100 g | SR EN ISO 3961:2013 |
| 4. | Peroxide index | meq $O_2$ / kg | SR EN ISO 3960-2017 |
| 5. | Saponification index | mg KOH / g fat | SR EN ISO 3657:2020 |
| 6. | Unsaponifiable | % | SR EN ISO 18609:2002 |
| 7. | Refractive index | - | SR EN ISO 6320:2017 |
| 8. | Water and volatile substances | % | SR EN ISO 662:2002 |

**Free acidity** is the percentage of free fatty acids from the total fatty acids contained in the fat sample, which is determined by dissolving a quantity of the sample in an alcohol-ether mixture, followed by titration with KOH solution in the presence of phenolphthalein.
The **acidity index** depends on the amount of free fatty acids resulting from basic hydrolysis and autoxidation of fats. Therefore, this index allows to appreciate the degree of fats freshness. Thus, the

smaller it is, the fresher the fat is, which means that the rancidity process has not started.
The **iodine index** indicates the unsaturation degree of the fat, varies with storage time and represents the amount of iodine, expressed in grams, which is added to the double bonds of unsaturated fatty acids in 100 g of triacylglycerol and titrated with sodium thiosulphate solution, in the presence of starch as an indicator.
The **peroxide index** is a parameter that reflects the content of oxygen in the form of peroxide (hydroperoxide) in a fat, indicating the degree of fat freshness. The peroxides formed from the oxidation of polyunsaturated fatty acids from fats in the presence of atmospheric oxygen react with potassium iodide, releasing iodine which is titrated with sodium thiosulphate in the presence of starch.
The **saponification index** represents the quantity of potassium hydroxide in mg, required for the saponification, respectively the transformation into soap, of one gram of oil. This index characterizes the molecular weight of the fatty acids that make up the oils.
**Unsaponifiables** are the organic substances in oils and fats that do not react with alkalis and therefore do not form soap. This category includes higher aliphatic alcohols, pigments, aromatic and volatile compounds. They are soluble in common solvents.
The **refractive index** is also a parameter used in assessing the quality of food fats and represents the ratio of the sine of the angle of incidence to the sine of the angle of refraction. Its determination was made using an Abbe refractometer. For oils, the determinations were made at $20^0$C, and for fats which are solid at room temperature, at $40^0$C.
The determination of the **water content and volatile substances** was carried out by oven drying at 105°C until constant mass, followed by cooling the sample and weighing to the nearest 0.001 g.

## RESULTS AND DISCUSSIONS

Dietary fats were analysed before being subjected to thermal treatments, the analysis methods being those presented in the previous Table. Each sample being analysed in triplicate. The results obtained fall within the limits provided in the quality standards (Table 3).

Table 3. Physico-chemical characterization of food fat samples, before the application of thermal stress

| S | Free acidity, % oleic acid | Acidity index, mg KOH/g fat | Iodine index, g I/100 g | Peroxide index, meq O2/kg | Refractive index | Water and volatile substances, % | Saponification index, mg KOH / g fat | Unsaponifiable, % |
|---|---|---|---|---|---|---|---|---|
| 1 | 0.21 | 0.5 | 128 | 2.12 | 1.47 | 0.02 | 189 | 0.98 |
| 2 | 0.12 | 0.3 | 51 | 2.85 | 1.46 | 0.02 | 198 | 1.15 |
| 3 | 0.5 | 3.4 | 65.2 | 6.67 | 1.47 | 9.4 | 194 | 0.77 |
| 4 | 0.45 | 4.95 | 34 | 2.45 | 1.45 | 15.9 | 200 | 1.88 |

*S-Sample

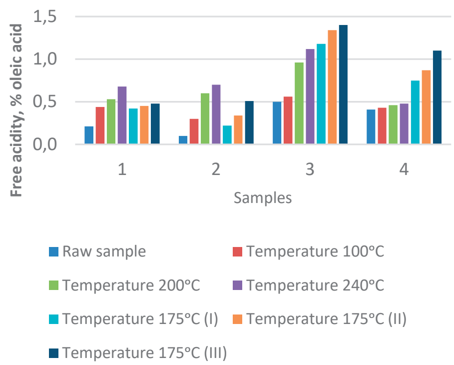


Figure 2. Comparison of free acidity values for both heat stress programs

According to the graph shown in Figure 2, the increase in free acidity values of all samples with temperature increase shows an accumulation of free fatty acids, which is in accordance with the fact that fat degradation is driven by increasing temperature.
As can be seen in Figure 2, sample 1 has a better stability during program 2 compared to program 1, while in sample 3 there is an almost threefold increase in the amount of free fatty acids.

Table 4. Acidity index values (mg KOH/g fat) of the analysed samples

| S | Raw sample | Temperature | | | | | |
|---|---|---|---|---|---|---|---|
| | | $100^0$C | $200^0$C | $240^0$C | $175^0$C (I) | $175^0$C (II) | $175^0$C (III) |
| 1 | 0.5 | 0.56 | 0.62 | 1.35 | 0.58 | 0.6 | 0.62 |
| 2 | 0.3 | 0.98 | 1.62 | 2.56 | 0.45 | 0.7 | 0.83 |
| 3 | 2.4 | 2.6 | 2.8 | 3.1 | 2.7 | 3.1 | 4.5 |
| 4 | 4.8 | 4.9 | 5.2 | 6.3 | 5.1 | 6.4 | 6.8 |

*S-Sample

The increase in the acidity indices values (Table 4) is the consequence of the accumulation of free fatty acids at each temperature step, which results in fats degradation.

In the graphic representation from Figure 3, the degree of unsaturation is reflected by the obtained values. Thus, the samples with the highest content of saturated fatty acids are the margarine samples (4), the palm oil (2) and the lard sample (3) and, finally, the refined oil (1).

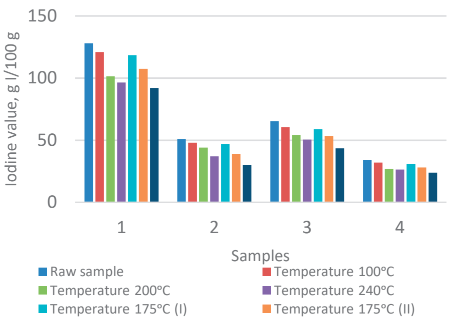


Figure 3. Comparison of iodine index values for both heat stress programs

For all samples, the degree of unsaturation decreases with increasing temperature, as a result of fat degradation reactions.

According to Figure 3, it can be concluded that the variation of the unsaturation degree of the four samples is the same, regardless of the applied program.

Regarding the variation of the peroxide indices (Figure 4), the obtained values show that, at temperatures higher than 200$^0$C, fat degradation occurs with the formation of considerable amounts of free radicals and, implicitly, peroxides, best example being sample 4, where the values of the peroxide increased by about 100%.

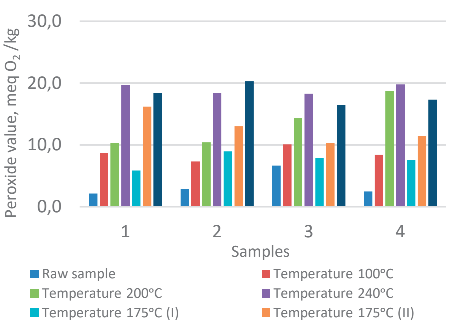


Figure 4. Comparison of peroxide index values for both heat stress programs

These values continue to increase at 240$^0$C, which shows that the thermal degradation increases as the temperature goes high. At the end of both programs, comparable peroxide index results are obtained for all samples, meaning that they have the same level of degradation.

The formation of free fatty acids causes an increase in the amount of saponifiable substances, as can be seen in Figure 5.

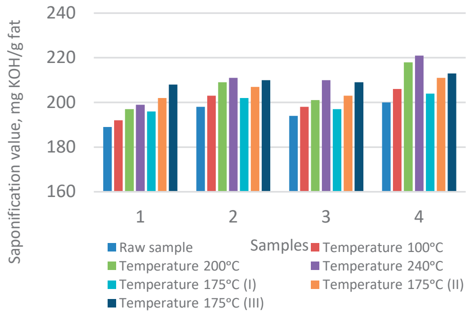


Figure 5. Comparison of saponification index values for both heat stress programs

The increase in saponification index occurs with increasing temperature and keeps the same trend for all studied dietary fats.

Regarding the influence of the temperature programs, the saponification index values are comparable for all samples, so the optimal program cannot be chosen.

The thermal degradation of food fats also results in the formation of unsaponifiable matter, thus causing their quantity to increase as the temperature increases. According to the graph in Figure 6, a significant difference can be observed in the variation of the amounts of unsaponifiables for sample 2. In the case of program 2, the amount of unsaponifiable matter increases with increasing temperature, the variation being the same for all samples. The lowest amount of unsaponifiables was determined for the lard sample, while vegetable fat had the highest values.

Thermal stress influences the values of the refractive indices of fats, as a result of the increase in the concentration of fatty acids resulting from thermal degradation. Thus, there is an increase in the value of the refractive index as the products resulting from the autoxidation reaction accumulate.

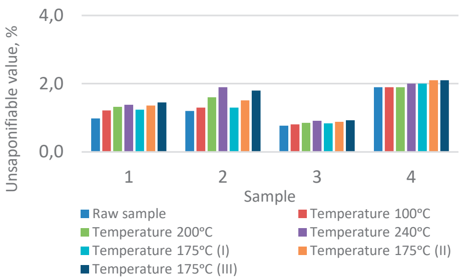


Figure 6. Comparison of unsaponifiables values for both heat stress programs

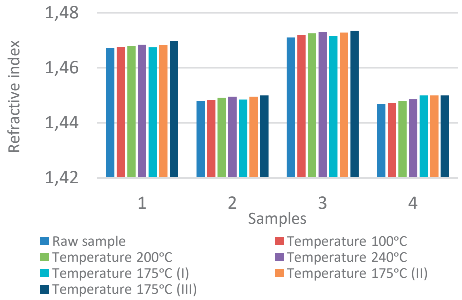


Figure 7. Comparison of refractive index values for both heat stress programs

As can be seen in Figure 7, in the case of the studied samples, the refractive index values are directly proportional to the increase in temperature, in both thermal stress regimes. Furthermore, the variations in values for both heat stress programs are comparable, the difference between the final value of each program and the initial value being around 0.002.

During heat treatment, evaporation of water from fats, as well as volatile substances, takes place. The decrease in the values of this parameter is more pronounced in the case of program 1 (Table 5).

Table 5. Comparison of water content values for both heat stress programs

| S | Raw sample | Temperature | | | | | |
|---|---|---|---|---|---|---|---|
| | | 100°C | 200°C | 240°C | 175°C (I) | 175°C (II) | 175°C (III) |
| 1 | 0.018 | 0.015 | 0.013 | 0.008 | 0.016 | 0.013 | 0.010 |
| 2 | 0.019 | 0.016 | 0.014 | 0.009 | 0.017 | 0.015 | 0.012 |
| 3 | 9.400 | 8.800 | 7.900 | 7.300 | 8.700 | 8.100 | 7.500 |
| 4 | 0.450 | 0.120 | 0.070 | 0.020 | 0.090 | 0.070 | 0.060 |

*S-Sample

## CONCLUSIONS

Analysing the results obtained following the application of the two temperature programs on food fats, the following can be concluded:

- sample 1 (Refined sunflower oil "Vita D'or") has a better stability during program 2 compared to program 1, while in sample 3 ("Angst" lard) there is an almost threefold increase in the amount of free fatty acids;
- the samples with the highest content of saturated fatty acids are the margarine samples (4), the palm oil (2) and the lard sample (3) and, finally, the refined oil (1);
- referring to the variation of the peroxide indices, the obtained values show that, at temperatures higher than $200^0$C, fat degradation occurs with the formation of considerable amounts of free radicals and, implicitly, peroxides, best example being sample 4, where the values of the peroxide increased by about 100%;
- regarding the influence of the temperature programs, the saponification index values are comparable for all samples, so the optimal program cannot be chosen;
- the lowest amount of unsaponifiables was determined for the lard sample, while vegetable fat had the highest values;
- the variations of refractive index values for both heat stress programs are comparable, the difference between the final value of each program and the initial value being around 0.002;
- the decrease in the values of water content and volatile substances is more pronounced in the case of program 1.

The final conclusion is that, for all food fats analysed from the point of view of thermal stability, heating to $100^0$C is safe, and for repeated heating to $175^0$C it is recommended to do it twice at most, to avoid their sharp degradation and, implicitly, the occurrence of negative effects on the health and quality of products prepared by frying.

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