Fatty acid profile in bread with flaxseed and exposed ionizing radiation with $^{60}$Co

MOURA, Neila Camargo de
SOUZA, Ivânia dos Santos
CANNIATTI-BRAZACA, Solange Guidolin
ARTHUR, Valter
DIAS, Carlos Tadeu dos S.

Resumo
A linhaça é uma planta herbácea e mais recentemente considerada como um alimento funcional. A irradiiação de alimentos é um processo que utiliza a energia de ionização emitida pelo radioisótopo $^{60}$Co para conservar os alimentos. É considerado um método de conservação eficiente pois reduz as perdas naturais causadas por processos fisiológicos (brotamento, maturação e envelhecimento), além de eliminar ou reduzir micro-organismos, parasitas e pragas, sem causar qualquer prejuízo ao alimento, tornando-os também mais seguros ao consumidor. O objetivo da pesquisa foi determinar o perfil de ácidos graxos no pão com diferentes concentrações de linhaça e exposto a diferentes doses de radiação com $^{60}$Co. O pão foi elaborado com pré-mistura e sementes marrons de linhaça, nas concentrações de 0 %, 8% e 12%. Após prontos, os produtos foram expostos à radiação de raios gama nas doses de 6,0 kGy, 8,0 kGy e 10,0 kGy. A leitura dos resultados foi feita no cromatógrafo a gás Shimadzu. As doses de radiação aplicadas provocaram diferenças entre os valores de ácidos graxos, exceto para os ácidos palmitoléico e esteárico, que não demonstraram alterações, devido ao aumento do percentual de linhaça. O ácido linoléico foi o que apresentou o maior índice, seguido de ácido linolênico.

Palavras chave: linhaça, radiação, ácido graxo, pão.

Abstract
Flaxseed is an herbaceous plant and more recently considered as a functional food. The food irradiation is a process that uses ionizing energy, with emitted gamma photons by $^{60}$Co radioisotopes on the food. It is considered an efficient conservation method because it reduces natural losses caused by physiological process (sprouting, maturation or ageing), beyond eliminate or reduce microorganism, thus making it safe to consumers. The aim of the research was to determine fatty acid profile in bread with different concentration of flaxseed and exposed to different dose of irradiation with $^{60}$Co. The bread was made with pre-mixture and flaxseed brown seeds, with 0%, 8% and 12% concentration. When ready, the products were exposed to gamma ray radiation, with 6,0 kGy, 8,0 kGy and 10,0 kGy. The reading was made in gas chromatograph Shimadzu. The dose of radiation caused differences between values of fatty acid, except to palmitoleic and stearic acids and only the palmitoleic acid did not show changes due to increase of flaxseed percentage. Linoleic acid was the one with the most content, followed by linolenic acid.

Keywords: Flaxseed; radiation; fatty acid; bread.
Resumen

La linaza es una planta herbácea y más recientemente considerado como un alimento funcional. La irradiación de alimentos es un proceso que utiliza la energía de ionización emitida por el radioisótopo 60Co para conservar los alimentos. Se considera un método eficiente para la conservación, ya que reduce las pérdidas causadas por procesos fisiológicos naturales (brotación, maduración y envejecimiento), y eliminar o reducir los microorganismos, parásitos y plagas sin causar ningún daño a los alimentos, haciéndolos más seguros también consumidor. El objetivo de la investigación fue determinar el perfil de ácidos grasos del pan con diferentes concentraciones de linaza y expuesta a diferentes dosis de radiación con 60Co. El pan se preparó con premezcla y semillas de lino marrón a concentraciones de 0%, 8% y 12%. Una vez listos, los productos a radiación gamma fueron expuestos a dosis de 6,0 kGy, 8,0 kGy y 10,0 kGy. La lectura de los resultados se hizo en cromatógrafo de gases Shimadzu. Las dosis de radiación aplicadas resultaron en diferencias entre las cantidades de ácidos grasos, excepto para el ácido palmitoleico, ácido esteárico y que no mostró cambios debido a la mayor porcentaje de linaza. El ácido linoleico fue el uno con la tasa más alta, seguido de ácido linolénico.

Palabras clave: linaza, radiación, ácidos grasos, pan.
INTRODUCTION

Nowadays linen is cultivated mainly in Canada, Argentina, USA, Russia and Ucrania (MOURA; CANNIATTI-BRAZACA; SILVA, 2009). Flaxseed world production is settled between 2.300.000 and 2.500.000 annual tons, being Canada its largest producer. Argentina is the largest producer in South America with 80 tons/year, with Brazil presenting a smaller production of 21 tons/year approximately (ALMEIDA; BOAVENTURA; GUZMAN-SILVA, 2009). In Brazil flaxseed farming is maintained by Polish and German immigrant descendants, and it is confined basically to Rio Grande do Sul State, specifically to northwest gaucho, once it requires cold weather, around 0ºC and -2ºC, to induce bloom. Its plantation occurs in the months of May and June and harvesting in the months of November, December and January. According to Vieira et al. (2012) flaxseed does not demand major farming practices, being its cultivation performed many times within a crop rotation system, with the purpose to recover exhausted soils and prevent soil wear and erosion, taking advantage of residual maize and soya fertilization.

The irradiation of food is a process that uses ionizing energy, which emits gamma photons by $^{60}$Co or $^{137}$Cs radioisotopes on the food (FARKAS; MOHÁCSI-FARKAS, 2011). This energy is called the Radiation Absorbed Dose, measured in Grays (1 Gy=1 Jkg$^{-1}$) and does not induce radioactivity in foods (SIDDHURAJU et al., 2002). This process increases stability of foods because it decreases microbiological load (FARKAS; MOHÁCSI-FARKAS, 2011).

The aim of the research was to determine fatty acid profile in bread with different concentration of triturated flaxseed and exposed to different doses of irradiation with $^{60}$Co.

LITERATURE REVIEW

Food irradiation is a treatment basic process comparable to heat pasteurisation. Irradiation is named “cold process” because processed food temperature variation is insignificant. Radiated energy is used very little by increasing the thermal energy (NABILGHOBRI, 2008). The packaged or bulk food gets controlled doses of radiation, and sanitary, phytosanitary and technical action (BRASIL, 2001).

Irradiation is used for the destruction of microorganisms and pests, pest reducing crop losses, inhibiting germination and biochemical changes of roots and prolongation of the durability of food, ensuring hygienic quality (FELLOWS, 2008).

Ionizing radiation helps to eliminate losses in post-harvest, since delays ripening of fruits and vegetables and consequently assists in transportation logistics, distribution and sales. It even replaces the use of chemicals on crops and may control infectious outbreaks in health, reducing illnesses and economic losses (OMI, 2005).

The high investment to undertake this technology, the lack of national and international promising consumer markets and culture of people averse to radiation hinder the implementation of this technique (MARQUES; COSTA, 2013).

Food with high lipid content can undergo oxidative rancidity, in addition, there is the possibility of re-infestation by insects and survival of microorganisms under the wrong dosage of radiation. Losses of vitamins and radiolysis of water are also some disadvantages. Moreover,
Fatty acid profile in bread with flaxseed and exposed ionizing radiation with $^{60}$Co

Foods unacceptable for consumption may have its bioburden eliminated, but the toxins may remain. Radiation resistance can occur by the microorganisms and some loss of nutritional food components (SILVA, 2000).

The energy of the gamma radiation is used for excitation and ionization without nuclear reaction in the matter and no radioactive residue (FARKAS, 2006). There are collisions between the radiation and product particles forming free radical ions. These free ions and the radicals, their reactions with molecules and other physical-chemical phenomena cease organic processes, causing radiolysis of water, modifies cellular DNA, destroy microorganisms, parasites and meristematic cells (HERNANDES; VITAL; SABAA-SRUR, 2003).

Ionizing radiations damages DNA, preventing reproduction, leading to death by disruption of cell membranes and inactivate enzymes. The survival of microorganisms, sensory and physicochemical changes depends on the achieved damage, irradiation time, dosage form, process temperature, oxygen, pH and chemical composition of the food (NABILGHORIIL, 2008).

Free radicals can react with other molecules altering the flavor of the food product by the production of volatile compounds and flavor as the autolysis of lipids in the presence of oxygen (oxidative rancidity) (ROSENTHAL, 2008).

**Materials and methods**

The bread was made with pre-mixture for bread and brown seeds of flaxseed. The ingredients used in the bread formulation are presented at Table 1.

**Table 1**: Ingredients used in bread preparation

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-mixture</td>
<td>5000</td>
</tr>
<tr>
<td>Yeast</td>
<td>150</td>
</tr>
<tr>
<td>Granulated Sugar</td>
<td>50</td>
</tr>
<tr>
<td>Improver</td>
<td>30</td>
</tr>
<tr>
<td>Water</td>
<td>2.600</td>
</tr>
</tbody>
</table>

The pre-mixture was composed by wheat flour enriched with iron and folic acid, salt, soy flour, emulsifier estearoil-2-lactil lactate to sodium, stabilizer polysorbate 80, flour improver, ascorbic acid, azodicarbonamide, hemicellulase enzyme, glucose oxidase, phospholipase and alpha amylase.
The dry ingredients were weighted in a Filizola© scale. In the bread with the percentage of 8% was used 400g of flaxseed and the bread with 12%, 600g of flaxseed. After being weighted, the ingredients were homogenized on a kneading machine for 10 minutes with cold water (-4°C) up to “veil point”. The bubbles were removed from the dough with a Perfecta© brand cylinder and the dough was taken to the Eco© brand volumetric divider, to standardize the size and weight of breads. The shape of the bread was made on a Lieme© brand mold. The bread was put in a roaster and carried to fermentation chamber of the Mafran© brand, in which it was maintained during 180 minutes at room temperature. The breads were baked in a Tedesco© brand turbo oven and preheated at 180°C for 15 minutes. After that, the bread was put in polypropylene packing (five units in each packing) and were heat sealed with a Matisa© brand machine to temperature in a spectrum of 200-220°C. The samples were identified with the flaxseed percentage, manufacturing date and the dose of irradiation.

The bread was sent to the Instituto de Pesquisas Energéticas e Nucleares (IPEN) (Institute of Nuclear and Energy Research), located in São Paulo, SP, Brazil, and exposed to doses of gamma ray, 6.0 kGy, 8.0 kGy and 10.0 kGy, in a multipurpose radiator, with 60Co source and rate dose 7.0 kGy/h. A control object, without irradiation, was used as comparison.

After the irradiation the samples stayed on the shelf at conditions of room humidity and temperature (26° to 34º C).

To determine fatty acid, the breads’ lipids were extracted with the Bligh e Dyer (1959) method that is based on the extraction of cold oil. After that, it was made the fatty acid esterification on oil according to Hartman and Lago (1973) method.

The reading was made in a Shimadzu gas chromatograph, model GC-14 B, with flame ionization detector, split injector, silica fused capillary column (50 m x 0,22 mm internal diameter, Shimadzu-Hicap, Australia). Chromatographic conditions were: column temperature 180º C (isothermal), carrier gas, hydrogen flow 1,05 mL/minute; detector and injector temperature of 250º C.

RESULTS AND DISCUSSION

The fatty acid profile is presented in table 2.
Table 2: Fatty acid profile (%) of bread added of flaxseed and irradiated

<table>
<thead>
<tr>
<th>Doses (kGy)</th>
<th>0% Flaxseed</th>
<th>8% Flaxseed</th>
<th>12% Flaxseed</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>22.35 ± 0.8aA</td>
<td>18.69 ± 0.2aB</td>
<td>11.71 ± 0.3abC</td>
</tr>
<tr>
<td>6.0</td>
<td>22.08 ± 0.7aA</td>
<td>16.16 ± 0.4bB</td>
<td>12.20 ± 0.5abC</td>
</tr>
<tr>
<td>8.0</td>
<td>23.81 ± 0.5aA</td>
<td>16.16 ± 0.3bB</td>
<td>13.05 ± 0.9aC</td>
</tr>
<tr>
<td>10.0</td>
<td>21.99 ± 0.6aA</td>
<td>14.86 ± 0.3cB</td>
<td>10.92 ± 0.5bC</td>
</tr>
</tbody>
</table>

Palmitoleic Acid (%)

<table>
<thead>
<tr>
<th>Doses (kGy)</th>
<th>0% Flaxseed</th>
<th>8% Flaxseed</th>
<th>12% Flaxseed</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>1.34 ± 0.7aA</td>
<td>1.21 ± 0.5aA</td>
<td>1.45 ± 0.1aA</td>
</tr>
<tr>
<td>6.0</td>
<td>1.28 ± 0.9aA</td>
<td>1.89 ± 0.3aA</td>
<td>1.03 ± 0.3aA</td>
</tr>
<tr>
<td>8.0</td>
<td>1.19 ± 0.3aA</td>
<td>1.56 ± 0.4aA</td>
<td>1.73 ± 0.3aA</td>
</tr>
<tr>
<td>10.0</td>
<td>1.32 ± 0.4aA</td>
<td>1.92 ± 0.2aA</td>
<td>1.55 ± 0.1aA</td>
</tr>
</tbody>
</table>

Stearic Acid (%)

<table>
<thead>
<tr>
<th>Doses (kGy)</th>
<th>0% Flaxseed</th>
<th>8% Flaxseed</th>
<th>12% Flaxseed</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>6.06 ± 0.4aA</td>
<td>6.04 ± 0.2aA</td>
<td>4.30 ± 0.4aB</td>
</tr>
<tr>
<td>6.0</td>
<td>6.80 ± 0.2aA</td>
<td>6.73 ± 0.4aA</td>
<td>4.32 ± 0.3aB</td>
</tr>
<tr>
<td>8.0</td>
<td>6.67 ± 0.8aA</td>
<td>6.01 ± 0.6aA</td>
<td>4.08 ± 0.4aB</td>
</tr>
<tr>
<td>10.0</td>
<td>6.56 ± 0.7aA</td>
<td>5.41 ± 0.8aA</td>
<td>3.45 ± 0.2aB</td>
</tr>
</tbody>
</table>

Oleic Acid (%)

<table>
<thead>
<tr>
<th>Doses (kGy)</th>
<th>0% Flaxseed</th>
<th>8% Flaxseed</th>
<th>12% Flaxseed</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>14.63 ± 0.2aA</td>
<td>14.46 ± 0.2aA</td>
<td>15.06 ± 0.5aA</td>
</tr>
<tr>
<td>6.0</td>
<td>12.02 ± 0.6bB</td>
<td>14.10 ± 0.6aA</td>
<td>15.14 ± 0.4aA</td>
</tr>
<tr>
<td>8.0</td>
<td>11.31 ± 0.6bB</td>
<td>14.57 ± 0.9aA</td>
<td>14.63 ± 0.4aA</td>
</tr>
<tr>
<td>10.0</td>
<td>11.78 ± 0.5bB</td>
<td>13.85 ± 0.6aA</td>
<td>14.35 ± 0.7aA</td>
</tr>
</tbody>
</table>

Linoleic Acid (%)

<table>
<thead>
<tr>
<th>Doses (kGy)</th>
<th>0% Flaxseed</th>
<th>8% Flaxseed</th>
<th>12% Flaxseed</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>51.36 ± 0.1bA</td>
<td>46.38 ± 0.6aB</td>
<td>35.10 ± 0.9aC</td>
</tr>
<tr>
<td>6.0</td>
<td>50.98 ± 0.7bA</td>
<td>43.07 ± 0.4bB</td>
<td>34.68 ± 0.5aC</td>
</tr>
<tr>
<td>8.0</td>
<td>54.86 ± 0.8aA</td>
<td>40.19 ± 0.6cB</td>
<td>35.47 ± 0.3aC</td>
</tr>
<tr>
<td>10.0</td>
<td>55.10 ± 0.7aA</td>
<td>43.56 ± 0.4bB</td>
<td>35.60 ± 0.9aC</td>
</tr>
</tbody>
</table>

Linolenic Acid (%)

<table>
<thead>
<tr>
<th>Doses (kGy)</th>
<th>0% Flaxseed</th>
<th>8% Flaxseed</th>
<th>12% Flaxseed</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>1.95 ± 0.1bC</td>
<td>14.42 ± 0.9cB</td>
<td>28.08 ± 0.6cC</td>
</tr>
<tr>
<td>6.0</td>
<td>2.31 ± 0.3abC</td>
<td>15.77 ± 0.7cB</td>
<td>29.88 ± 0.7bA</td>
</tr>
<tr>
<td>8.0</td>
<td>2.70 ± 0.2aC</td>
<td>20.30 ± 0.4aB</td>
<td>27.19 ± 0.3cA</td>
</tr>
<tr>
<td>10.0</td>
<td>2.36 ± 0.3abC</td>
<td>18.26 ± 0.9bB</td>
<td>34.88 ± 0.6aA</td>
</tr>
</tbody>
</table>

Values are the mean of three determinations (n = 3) ± SD; a Different small letter in the same column means difference dependent dose (P < 0.05) to each parameter; b Different small letter in the same column means difference dependent dose (P < 0.05) to each parameter; c Different capital letter in the same row means difference due flaxseed percentage (P < 0.05) to each parameter.
The doses of irradiation promoted differences between values of fatty acid, except to palmitoleic and stearic acids. The sample with 0% of flaxseed was also changed, oleic acid content decreasing according to the dose of irradiation increase and also increasing linoleic and linolenic acids contents. The sample of 8% of flaxseed also showed changes in linoleic and linolenic acids contents, increasing these contents according to the increase of doses. The sample of 12% of flaxseed also increased in linolenic acid content. The palmitic acid content was reduced when the 10 kGy dose was applied in the samples with flaxseed. Tipples and Norris (1965), analyzing the gamma ray effect on wheat flour lipids, observed changes in linoleic, linolenic and palmitic acids contents, where the larger changes occurred according with the doses increase. Marathe et al. (2002) and Silva et al. (2010), analyzing the effect of different doses of gamma ray on different kinds of flour, observed that irradiation did not affect the wheat flour lipids content.

In the analyzed fatty acids, only palmitoleic acid did not show changes due to increase of flaxseed percentage. However, it was detected the decrease of palmitic, linoleic and stearic acids according to flaxseed addition, in which stearic acid showed changes in samples with 12% of flaxseed. In the oleic and linolenic acids, it was possible to identify the occurrence of an increase according to the percentage of flaxseed. Mentes et al. (2008), studying flaxseed addition on breads made with wheat flour, found that polyunsaturated acids grows on bread that had flaxseed, being linolenic acid the most abundant among the fatty acids analyzed. Conforti and Davis (2006), analyzing the fatty acid profile on bread made with flaxseed flour, also observed that polyunsaturated acids increased, being oleic acid one of the highlights.

The content of flaxseed along with the dose of radiation, made noticeable the decrease of the five fatty acids analyzed, except in palmitoleic acid, when applied 10 kGy and especially when added flaxseeds is of 8%. The process of radiation causes lipid oxidation, especially on double bonds for being less stable (FARMER et al., 1942). Free radicals, formed in lipid oxidation, have a direct impact on the shelf life, causing changes in its aroma and flavor, being among the responsible factors for the fat and fatty acid profile changes (JENSEN et al., 2005).

Independent from the dose of radiation and of flaxseed content added, the amount of linoleic acid (ω -6) presented was greater when compared to linolenic acid (ω -3), with the average relationship between these fatty acids being of 1:23 in the treatment with flaxseed addition 0%; 3:1 in the treatment with flaxseed addition 8% and 1:1 in treatment with flaxseed addition 12%. The relation recommended by the World Health Organization (WHO) is the ingestion of 5:1 to 10:1 between linoleic acid (ω-6) and linolenic acid (ω-3), respectively. By that, notice that the treatment that presented this closer relationship of recommended by WHO is the flaxseed addition 0%. The most abundant fatty acid in flaxseed is linolenic acid and its content between 35 to 45, and may, in some cases, vary more or less (TOLKACHEV; ZHUChENKO, 2000). In this study, we observed that the linolenic acid content is below the quoted above. This aspect may be explained by the variety of used seeds, and the use of radiation process as well.

Among the fatty acids measured, the palmitoleic and stearic acids were the ones that showed minor contents, and the linoleic acid, followed by linolenic, presented the highest contents in the French bread. Due to the large amount of unsaturated fatty acids in bread with flaxseeds, very high doses of radiation, such as that of 10 kGy, can be harmful to both its sensory quality and stability, and should therefore be avoided.
CONCLUSIONS

The irradiation on the flaxseed addition showed changes on majority of fatty acids contents analyzed. The radiation dose of 10 kGy has significantly affected most of the content, especially unsaturated fatty acids. The fatty acids that showed changes in higher proportion were linoleic and linolenic.

Acknowledgments

To FAPESP to financial support, to Via Delícia for the seeds of flaxseeds and Cláudia Martins for the translation.
Fatty acid profile in bread with flaxseed and exposed ionizing radiation with 60 Co

REFERENCES


Fatty acid profile in bread with flaxseed and exposed ionizing radiation with $^{60}$Co


MOURA, Neila Camargo de; SOUZA, Ivânia dos Santos; CANNIATTI-BRAZACA, Solange Guidolin;
ARTHUR, Valter; DIAS, Carlos Tadeu dos S.

Fatty acid profile in bread with flaxseed and exposed ionizing radiation with $^{60}$CO


MOURA, Neila Camargo de; SOUZA, Ivânia dos Santos; CANNIATTI-BRAZACA, Solange Guidolin;
ARTHUR, Valter; DIAS, Carlos Tadeu dos S.

Fatty acid profile in bread with flaxseed and exposed ionizing radiation with $^{60}CO_2$