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THE EFFECT OF GAMMA IRRADIATION ON THE MICROBIOLOGICAL ANALYSIS ON COMMERCIAL FUNCTIONAL BRAZILIAN GREEN BANANA FLOUR.

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ABSTRACT

In Brazil, although it is qualified as a major world producers, however, the production losses are high. Nevertheless, these losses can be reduced by processing the fruit "unsuitable" for consumption into products based on green banana (pulp, rind and flour). The green banana flour shows enhanced nutrition value, with higher contents of mineral, dietary fiber, resistant starch, and total phenolics, for use in Brazilian irradiated ready - to eat foods, such as bread, macaroni, among others. Food irradiation has been identified as safe technology to reduce risk of foodborne illness as part of high-quality food production, processing, handling and preparation. Food irradiation utiliizes a source of ionizing energy that passes through food to destroy harmful bacteria and other organisms. Often referred to as "'cold pasteurization", food irradiation offers negligible loss of nutrients or sensory qualities in food as it does not substantially raise the temperature of the food during processing. The object of this work was to determine the effect of gamma irradiation on microbiological analyses of the: the number of mesophiles, *Total Coliforms at 35°C*, *Colif*orms at 45°C, *Staphylococcus aureus* and *Salmonella* spp of the green banana flour, commercially found in the Brazilian market. The microbiological analyses were carried out in conformity with the methodologies described at the Faculty of Veterinary Medicine, according to the current legislation. Irradiation was performed in a ^{60}Co Gammacell 220 (AECL) source, with dose of 3kGy at IPEN-CNEN – SP. In samples of Brazilian green banana flour, irradiated at 3 kGy, the growth of all microorganisms *(Mesophiles,Total Coliforms* at 35°C, *Coliform* at 45°C and *Staphylococcus coagulase positive*) were reduced. As a result, the application of the irradiation technique may be recommended to enhance the food safety.

1. INTRODUCTION

In Brazil, although it is qualified as a major world producers, however, the production losses are high. Nevertheless, these losses can be reduced by processing the fruit "unsuitable" for consumption into products based on green banana (pulp, rind and flour). The green banana flour shows enhanced nutrition value, with higher contents of mineral, dietary fiber, resistant starch, and total phenolics, for use in Brazilian irradiated ready to eat foods, such as bread, macaroni, among others [1] [2][3][4][5]. In general, the addition of banana starch promoted a dilution effect on protein, lipid, and ash content, while moisture content was not affected. On

the other hand, the content of resistant starch significant increasead ($p<0,05$) with an increase of banana starch [6].

The green banana fruit shows that it is rich in amylase-resistant starch, which simulates colony production of short- chain fatty acids and it is used for treating diarrheal diseases. Green banana diet improves clinical severity in childhood shigellosis and it could be a simple and useful adjunct for dietary management of this illness [7].

The substitution of wheat flour with BF(banana flour) resulted in significantly ($p < 0.05$) higher total dietary fibre (TDF), and especially insoluble dietary fibre (IDF), resistant starch (RS) and total starch contents [8].

Food irradiation has been identified as safe technology to reduce risk of foodborne illness as part of high-quality food production, processing, handling and preparation. Food irradiation utiliizes a source of ionizing energy that passes through food to destroy harmful bactéria and other organisms. Often referred to as "cold pasteurization", food irradiation offers negligible loss of nutrients or sensory qualities in food as it does not substantially raise the temperature of the food during processing [9]. Food irradiation does not replace proper food production or handling. Even with treatments that destroy 99,9% of a pathogen, some could still survive [10][11]. In this work was to determine the effect of gamma irradiation on microbiological analyses of the: number of mesophiles, *Total Coliforms at 35°C, Colif*orms at 45°C, *Staphylococcus aureus* and *Salmonella* spp of the functional brazilian green banana flour, commercially found in the Brazilian market.

2. MATERIAL AND METHODS

2.1. Material

Eight packets of green banana flour were used found in the market, in 200g pouches. Lots of the green banana flour were employed and kept at a refrigerator (4-7ºC), before and after irradiation. A portion of 25g of flour, homogenized with 225 ml of sterile peptone water (1%) in a sterile plastic bag, was analyzed for Salmonella spp; another plastic bag containing 225ml of sterile 0.1% peptone water with other homogenized 25g flour was kept for further analyses. The samples were homogenized in a stomacher for about a minute. This was considered the dilution of 10^{-1}.The enumeration of *Total Coliform at* 35°C, *Colif*orm *at* 45°C and *Staphylococcus coagulase positive* expression of the results were performed by CFU/g [12][13][14].

2.1.1. Irradiation

Irradiation was performed in a ^{60}Co Gammacell 220 (AECL) source, at a dose rate about 1.96 kGy/h with dose of 3kGy, dose uniformity factor of 1.13. Dosimetric mappping was previously performed by Fricke dosimetry.

2.1.*2* Microbiogical Analysis

Microbiological analysis of green banana flour samples were analyzed at the Faculty of Veterinary Medicine and Animal Science, before and after irradiation at IPEN-CNEN/SP. The microbiological analyses were carried out in conformity with the methodologies described at the Faculty of Veterinary Medicine, according to the current legislation, with the

following determinations: the number of aerobic mesophile ,*Total Coliforms* at 35°C, *Colif*orm at 45°C, *Staphylococcus coagulase positive* and *Salmonella spp* [12][13] [14]. The analysis of variance was applied and the mean comparisons were performed by Student test**,** considering an error of 5%.

Standard Count of microorganisms, facultative viable aerobic mesophiles and *Staphylococcus* coagulase showed to be positive. After the initial dilution (10^{-1}), the other dilutions (to 10^{-5}), in saline Peptone 0.1%, were done.

One ml of each dilution was inoculated into sterile Petri dishes and then covered with about 15 ml of Plate Count Agar (PCA), merged and maintained between 46 and 48 ° C by the method "pour plate" (plating in depth). The samples were homogenized, left to solidify at room temperature and incubated inverted at 36° C, for 48 hours. To read the mesophiles, the plates of the dilutions that had 25 to 250 *colonies were counted and the results expressed in CFU /g.*

For Staphylococcus coagulase positive reading, the plates containing between 20 and 200 colonies were selected for counting. The number of typical and atypical colonies was counted [14].

The culture medium used for detection and enumeration of Coliforms was the chromogenic medium Rapid E. coli 2 Biorad®. The Principle of the medium was based on simultaneous detection Coliforms and *Escherichia coli*. The hydrolysis of chromogenic substrates included, in the product, results in blue Coliforms colonies (β-D-glucoronidase negative / β-D-galactosidase positive) and in purple *Escherichia coli* colonies (β-D-glucoronidase positive / β-D-galactosidase positive) [12].

For *Salmonella*, the pré-enrichment took place through the incubation of sample aliquots prepared in Peptone Water, at a concentration of 1% and incubated at 36º C, for at least 16 hours and no more than 20 hours. From the pré-enrichment procedure established, 0.1 ml sample tube containing 10 ml of Rappaport Vassiliadis broth was inoculated and incubated at 41 ± 0.5 º C, in a 24-hour water bath. The selective media chosen were the XLT4 Agar Agar and Hektoen. If the incubations in the culture media reveal suspected colonies of *Salmonella*, this will be confirmed by biochemical and serological tests.

3. RESULTS AND DISCUSSION

Table 1 shows the results of the microbiological analyses of the number of *mesophilic,* total *Coliforms at 35°C, Coliforms at 45°C, Staphylococcus coagulase positive* and *Salmonella* spp of industrialized green banana flour, non-irradiated and irradiated at 3kGy. For m*esophiles, Coliformes totais, coliformes fecais and Staphylococcus coagulase positiva, a s*ignificant difference (error 5%) was observed between irradiated and non-irradiated samples. As it can be seen, gamma irradiation is effective to reduce the number of all the microorganisms of industrialized green banana flour. Salmonella spp was not found.

Table 1 Microbiological analysis of industrialized green banana flour, non-irradiated and irradiated

Microbiological Analysis	Dose	
	0kGy	3kGy
Number of aerobic mesophilic in CFU/g	$4.2X10^{6a} \pm 707$	$1.2 X 10^{4b} \pm 707$
Most probable number of Total Coliforms at 35°C in MPN/ g	$1.1 X 10^{5a \pm} 7071$	$3.1^{b} \pm 0.14$
Most probable number of Coliforms at 45°C, in MPN/ g	$1.1 X 10^{5a \pm} 7071$	$3,1^{b} \pm 0.14$
Number of Staphylococcus coagulase positive in CFU/g	$3.1 X 10^{3a} \pm 70.71$	$99.5^{b} \pm 0.70$
Salmonella spp (in 25g)	Absence	Absence

n = duplicate; Means ± Standart Desviation

a b Medium values followed by the different letters, on the same line, differ significantly from the control-sample, at 5% significance (error 5%)

In this investigation, commercial Mexican bread, made with wheat flour, was irradiated at 1,0 kGy using a Co-60 Gammabeam 651 PT irradiator facility. The total aerobic, yeast and mold counts were reduced by 96%, 25% and 75%, respectively, with the irradiation process (Agundez - Arvizu, *et al* 2006). In this study, the obtained results confirm that gamma irradiation is effective to reduce the microbial load in the green banana flour such as the Mexican bread

4. CONCLUSION

In samples of functional Brazilian green banana flour, irradiated at 3 kGy, the growth of all microorganisms *(Mesophiles, Total Coliforms* at 35°C, *Coliform* at 45°C and *Staphylococcus coagulase positive*) were reduced. As a result, the application of the irradiation technique may be recommended to enhance the food safety.

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