DNA COMET ASSAY AS A RAPID DETECTION METHOD OF IRRADIATED BOVINE MEAT BY ELECTRON BEAM

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Introduction: The presence in food of pathogenic microorganisms, such as *Salmonella* species, *Escherichia coli* O157:H7, *Listeria Monocytogenes* or *Yersinia enterolitica*, is a problem of growing concern to public health authorities all over the world. Thus, irradiation of certain prepackaged meat products such as ground beef, minced meat, and hamburgers may help in controlling meatborne pathogens and parasites. Pathogenic microorganisms and parasites in meat products, which are commonly consumed raw, are of particular importance. Up to now, only electron-beam accelerators and gamma-ray cells have been used for commercial applications. At the international conference on "The Acceptance, Control of, and Trade in Irradiated Food", it was recommended that governments should encourage research into detection methods (Anon, 1989). Already five international standards are available to food control agencies. A number of physical, chemical, and biological techniques of detection of irradiated foods have been discussed in the literature. A rapid and inexpensive screening test employing DNA Comet Assay to identify radiation treatment of food has been described by Cerda et al. (1997). This method is restricted to foods that have not been subjected to heat or other treatments, which also induce DNA fragmentation. Advantages are its simplicity, low cost and speed of measurement. This method was proposed to the European Committee for Standardization (CEN) as a screening protocol (presumptive) and not as a proof (definitive). The DNA comet assay have been yielded good results with chicken, pork, fish meat, exotic meat, hamburgers, fruits and cereals. In this work we studied a DNA fragmentation of bovine meat irradiated by electron beam. **Experimental:** Bovine meat was purchased in local shops in São Paulo. Irradiation was performed with electron beam of accelerator facility of Radiation Dynamics Inc., USA (E= 1,5 MeV, I=25mA). The irradiation doses were 3,5; 4,5; 5,5; and 7,0 kGy at chilled conditions, and 3,5; 4,5; 6,0; 7,0 and 8,0 kGy at frozen conditions. The thickness of meat was less than 0,5 cm. Briefly, meat samples were crushed with a mortar and pestle and was transferred to 1ml ice-cold PBS. This suspension was stirred for 5 minutes and filtered. 100µl cell suspension was mixed with 500µl of low-melting agarose (0,8% in PBS). 100µl of this mixture was spread on pre-coated slides. The caste slide were immersed in lysis buffer (0,045M TBE, pH 8.4, containing 2,5% SDS) for 15 minutes. Electrophoresis was carried out using the same TBE buffer, but devoid of SDS, at a potential of 2V/cm for 2 minutes. Silver staining was carried out for 20 minutes following fixing. Duplicate measurements for each sample were carried out and 100 cells were counted for each dose level. The migration patterns of DNA was evaluated with a standard microscope. **Results and Discussion:** In the work with Comet Assay, increasing DNA degradation was characterized by different migration patterns of DNA. Some differences were observed between chilled and frozen conditions. The storage time influenced the DNA degradation. **Conclusions:** It was concluded that the comet assay could be used for detection of processing of irradiated bovine meat by electron beam. Also, this method could be used as a freshness indicator. **Acknowledgments**: The authors are grateful to CAPES and IPEN/CNEN-SP.