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An Intervention Study Using Irradiated Ethnic Ready to Eat Foods in Immunocompromised Patients

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Abstract: Food poisoning derived from contaminated foods can have serious health consequences, particularly for immuno-compromised patients as well as specific target groups where fresh food supply might be limited such as the army, people who live in remote areas and those exposed to natural disasters. Food processing through ionizing radiation as a food safety measure can be applied to the targeted foods either at a medium or high radiation dose, either alone or in combination with other techniques. Radiation sterilization of ethnic ready to eat foods has been considered as safe. Products which have high sensory qualities, hygienic and nutritionally sound may play a major role in accelerating recovery process through maintaining the dietary diversity of such population groups. In this study, the ethnic safe ready to eat (RTE) foods prepared were those from animal origins, i.e., fish, red meat and poultry meat, respectively while the formula, recipe and design product type was maintained based on WHO Recommended Dietary Allowance (RDA) for immuno-compromised patients. Irradiation was carried out at a ⁶⁰Co irradiation facility and the process was maintained under cryogenic condition to protect the essential dietary nutrients against free radical attack. To reduce the undesirable oxidative changes in the food matrix containing fats, the processing was also done on non transparency packaged frozen products after evacuation to eliminate atmospheric oxygen. Irradiated food samples were then stored at room temperature (28-30°C). The foods were administered to immunocompromised residents of the National Narcotic Board (NNB) in Bogor over a 21 day period. The 45 participants were randomly allocated into 3 groups with n = 15 per group. Group I (control) consumed regular foods as prepared by NNB, group II consumed unirradiated ready to eat foods and group III consumed radiation sterilization ready to eat foods namely fish *pepes*, *rendang* beef, *semur* and processed chicken. Blood samples were taken from all participants, both pre and post intervention and tested for albumin and lymphocyte proliferation, while BMI was also assessed. Radiation sterilization ready to eat foods were rated positively in sensory evaluation tests. No difference in effects of the intervention on Body Mass Index (BMI) measurements was found, but there was some indication that the intervention using irradiated RTE foods increased albumin levels and maintained lymphocyte count. Data were analyzed and calculated statistically using software and tested for its significance.

Key words: Ethnic ready to eat (RTE) foods, immuno-compromised patients, intervention studies, recommended dietary allowance, ionizing radiation

INTRODUCTION

Irradiation offers a potential benefit to enhance microbiological safety of food and of maintain nutritional and sensory quality while extending shelf-life. The revised regulation of food irradiation in Indonesia is already in place. The regulation, No. 701/MENKES/PER/VIII/2009, has been stipulated under the decree of Ministry of Health of the Republic of Indonesia on 28 August 2009 and replaces both PERMENKES No.826/MENKES/PER/XII/1987 and PERMENKES RI No.152/MENKES/SK/II/1995. Annex 1 of the new regulation partly

includes the application of medium dose applied for microbiological decontamination of meat and meat products and the high dose for sterilization and extend storage life of the products at ambient temperature purposes of ready to eat meals as well as shelf-stable foods (Anonymous, 2009). The benefits of irradiation as sanitary and phytosanitary treatments of many types of food products is increasing each year and it is applied by food industries for commercial use.

Food irradiation is a non thermal process, so the treated food is close to the natural state both in appearance and

taste, unlike other technical processes involving heat treatment which may lead to some unacceptable changes in food. Various positive impacts can be achieved after the process, resulting in improvement of microbiological and parasitological-toxicological safety of foods without impairing their overall quality including the sensory attributes (Placek *et al.*, 2004; Anwar *et al.*, 2008) before and after storage (Anonymous, 2010a). Little data were available on the effect of irradiation on minimally processed food and composite food as well as prepared meals for specific target groups including immunocompromised patients such as those with Human Immunodeficiency Virus/Acquired Immuno-deficiency Syndrome (HIV/AIDS). An intensive series of *in vitro* and *in vivo* studies, as a part of risk assessment monitoring programme, showed that a high irradiation dose at 45 kGy of irradiated ethnic ready to eat (RTE) foods demonstrated that such foods were safe and wholesome (Irawati *et al.*, 2010, 2011; Irawati and Sani, 2012). Because of the benefits of the process, high dose irradiation can also potentially facilitate the availability of food for immunocompromised hospital patients where the food is safe to eat and of acceptable nutritional and organoleptic quality (Anonymous, 2010b).

Conventional methods applied to sterilize the food for patients are mostly autoclaving, micro waving and ultra violet irradiation, but these introduce some adverse effects. Other thermal sterilization methods in order to prepare a clean diet can be acceptable though some detrimental effects on certain quality parameters still occur. The existing technology has in fact shifted the food criteria from sterile diets to low microbe diets (Narvais *et al.*, 2003). RTE foods can be pasteurized or sterilized by irradiation in the final package and then reheated by microwave cooking prior to serving and any nutritional losses are small. Nevertheless the loss of micro and macro nutrients can be suppressed by selecting appropriate irradiation conditions and proper packaging material used in this purpose.

A coordinated research project (CRP) on "Development of Safe, Shelf-Stable and Ready-to-Eat Food through Radiation Processing" was implemented by the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture in 1996-2007 to evaluate the role of irradiation for such food. Such irradiation was shown to offer promise as a sanitary treatment to ensure microbiological safety and shelf-life extension of several types of food products, including chilled ready-to-eat meat products (Irawati *et al.*, 2009). The following CRP on Development of Irradiated Foods for Immuno-compromised Patients and other Potential Target Groups started in 2010 and was initiated to prepare a variety of foods for immuno-compromised patients as well as for other specific target groups, e.g., post natural disaster. Type of foods were sterile meals and ethnic ready-to-eat-food of intermediate moisture content including products derived from animal origins such as

fish *pepes*, *rendang* meat and processed chicken (Anonymous, 2003). Moist foods, including ready to eat meals packed in laminate pouch under vacuum condition should be irradiated at cryogenic temperature (Anonymous, 1995; Irawati *et al.*, 2005, 2007) in particularly for high radiation dose in order to inactivate free radical movement and radical reactions (Diehl, 1990). A previous study on the intervention of ethnic safe shelf-stable foods using gamma rays to HIV positive respondents showed a preliminary result that the respondents' acceptance of such products depended on their clinical and nutritional status, education and psychological well-being (Simanungkalit *et al.*, 2013).

The objective of this research was to study the impact of intervention with radiation sterilized RTE meals in immunocompromised patients on nutritional status. It was hypothesized that the application of radiation technology in complete set RTE meals would be acceptable for immunocompromised patients and improve their nutritional status, aid their recovery process from illness and improve quality of life. This would be achieved through the ability to consume higher quality and a greater variety of foods. The major goal of medical nutrition therapy is to optimize nutritional status, immunity, prevent loss of weight and lean body mass.

MATERIALS AND METHODS

Materials: Radiation sterilization of (RTE) foods was performed based on three types of animal origins, including fish base (gold fish and anchovy *pepes*), red meat base (spicy *rendang* beef and semur beef) and poultry base (chicken meat). These foods were prepared in order to study the effects of sterile foods on nutritional status of NNB respondents, considered as immuno-compromised patients. Some unirradiated foods, as prepared regularly by NNB kitchen, were used in the control group, as were unirradiated ethnic RTE. Middle Chain Triglycerides (MCT) as an energy source, rich with lauric acid, is considered as a natural food supplement containing high saturated fatty acids (92%) which is an antioxidant agent (Kono *et al.*, 2000). MCT was also given to all NNB residents 3 times a day at 10 ml after meals within 21 day-food intervention.

Methods: Food testing of some irradiated RTE foods i.e., fish *pepes*, anchovy *pepes*, *rendang* beef and *semur* beef, was conducted prior to the intervention study. The test was done by 40 NNB residents as volunteers to conduct sensory evaluation of radiation sterilized ethnic RTE foods based on general appearance, texture, flavour, taste and odour according to a 5 point Hedonic scale (Anonymous, 2003).

The intervention study of radiation sterilization RTE foods on Hepatitis C residents was carried out using methods developed during preliminary research (Simanungkalit *et al.*, 2013). All participants received information regarding the purpose of intervention study and nutrition

education prior to the study. The selected residents, 45 people who inclusion criteria, were randomized into 3 intervention groups; each group consisted of 15 people and received a different type of food. Group 1 was consumed unirradiated regular foods prepared from NNB's kitchen; Group II consumed some unirradiated foods prepared by BATAN and Group III consumed some main meals irradiated foods prepared by BATAN. Pre and post tests conducted included Body Mass Index, calculated from body weight and height were measured using a calibrated weighing scale and microtoise, respectively. This measurement may indicate nutritional status and determine the potential risk of degenerative disease of the respondent. Blood was collected by an accredited clinical laboratory in Bogor for further analysis of albumin and lymphocyte count.

Standard methodologies and techniques applied for quality assessments: Blood samples were collected from individual residents for pre and post test analysis (Burtis and Ashwood, 1994; Ravel, 1995) including albumin (spectrophotometry) and lymphocytes counts (laser light scattering). The assessments of the blood were conducted by a Reputable Private Clinical Laboratory Services in Bogor.

RESULTS AND DISCUSSION

People living with HIV and AIDS are immunocompromised and at greater risk for developing life threatening complications from a food borne illness. Many of these illnesses are characterized by fever, diarrhea and weight loss. It is important to prevent food borne illness because it can lead to opportunistic infections (Simanungkalit *et al.*, 2013). For most healthy people food borne illness, is not life threatening. This is not the case for those with compromised immune systems.

It is published elsewhere irradiation greatly reduces bacteria, insects and parasites that contaminate food or cause spoilage and deterioration. Irradiation can drastically reduce the presence of these disease-causing agents, providing a much broader margin of safety. Used in combination with other food safety measures, it can drastically reduce the risk of illness for consumers (Anonymous, 2002) and hospitals sometimes use irradiation to sterilize food for immunocompromised patients (Bruyn, 2001).

Sensory evaluations of irradiated RTE foods prior to intervention studies conducted by volunteer residents at NNB: HIV/AIDS and its complications, drug therapies and/or malignancies can result in malnutrition (Anonymous, 2002) and also might change their appetite. Therefore it may seem difficult sometimes to follow good nutrition recommendations due to health complications. An extra sensory evaluation was carried out during the trial to find out which foods taste good and

which are rejected. Food testing was done to figure out the sensory response and acceptance of NNB resident to the irradiated foods in general. Results of food testing based on sensory attributes showed that almost all residents accepted the irradiated foods with high acceptability values (Fig. 1 to 4).

Intervention studies of radiation sterilization RTE foods on immunocompromised patients to improve their nutritional status:

Nutrition is the most important food component for patients who are immunocompromised, because lack of nutrition in the diets may lead to reduced immune system. Of all the body's systems, the immune system responds most sensitively to subtle changes in nutrition status. Impaired immunity open the way for infectious. Decreased nutritional status will proportionally decrease albumin level and lymphocyte count. Low levels of albumin are associated with several other diseases, including Hodgkin's disease and HIV, the precursor to AIDS. Low albumin levels are also a predictor of mortality. People with albumin levels below 3.5 g/dl are approximately twenty times more likely to die from all causes than those with albumin levels of 5.0 g/dl. Some researchers reported that reduction of body mass cells and albumin serum might increase the morbidity. Lymphocytes play a major role to increase body immunity (Mc. Corkindale *et al.*, 1990; Don and Kaysen, 2004).

Patient who are immunocompromised such as those with HIV/AIDS influence nutritional status by having increased resting energy expenditure, decreased nutritional intake, anorexia and malabsorption. Prevention of malnutrition is one of the keys to extending the lifespan of persons living with HIV/AIDS. Ongoing nutritional assessment and monitoring is necessary for the person living with HIV/AIDS, especially if oral feedings do not meet nutritional needs. If oral feedings do not meet nutritional needs, then a different approach must be initiated. There are many ways to deal with these problems and improve quality of life. In this study it was hypothesized that immunocompromised respondents treated with irradiated RTE foods might improve their nutritional status and immune system function.

The results of comparative studies on BMI of pre- and post-treatment in group I, II, III is presented in Table 1. It is shown in the Table that mean values of pre treatment BMI group I was 22.3 ± 3.9 while the mean post treatment BMI was 22.6 ± 3.1 kg/m², group II mean pre treatment BMI was 22.2 ± 2.2 and post treatment 22.9 ± 2.2 and group III mean pre treatment BMI was 23.5 ± 2.8 and post treatment 23.7 ± 2.7 , respectively.

The post treatment BMI was not significantly different to the pretreatment BMI ($p > 0.05$) in group I, II or III. Overall result showed that BMI from Group I, II and III were still in the normal range (18-25 kg/m²) both during pre and post tests. The majority of the patients who developed a

secondary infection, however, lost weight despite the use of supplements (Mangili *et al.*, 2006; Scalfi *et al.*, 1990; Fleck, 1989). This study showed that BMI was still in normal limit.

Table 2 shows that the mean values of pre treatment and post treatment serum albumin levels in group I were 4.58 ± 0.24 and post treatment 4.66 ± 0.36 mg/L, respectively. In group II the mean values of pretreatment and post-treatment serum albumin levels were 4.52 ± 0.28 and 4.52 ± 0.26 mg/L ($p > 0.05$), respectively. Table 2 indicates that there was no any significant different in albumin content at pre and post treatments in group I nor II. Albumin content in this group was still in normal condition.

However, in group III post-treatment serum albumin level was significantly higher than the pretreatment level ($p < 0.05$). This study has shown that serum albumin levels increase significant in response to post treatment RTE foods, compare with albumin level post treatment in the groups consuming non irradiated foods which did not change significantly ($p > 0.05$).

Albumin is an important protein that is found in most animal tissues. It is the fact that albumin levels are the most important indicator of health status. Albumin serum is one important parameter in measuring morbidity and mortality of patient during hospitalization. Statistical reports demonstrated that albumin levels correlate closely with nutritional status and immune system (Mc. Corkindale *et al.*, 1990; Don and Kaysen, 2004).

If bacteria, viruses, fungi entering the body simply did their damage, that would be a serious problem. In order to fight off invading bacteria, viruses, fungi and other invaders, the body sends the immune system into action. The immune system utilizes many protein-based substances to fight off the invasion. The immune system

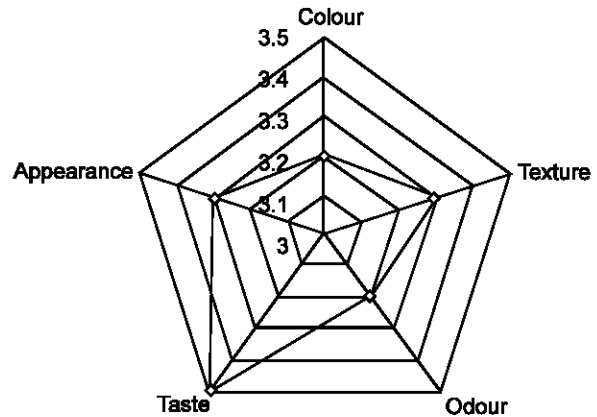


Fig. 1: Sensory evaluation of radiation sterilization of fish pepes as conducted by residents as volunteers at NNB in Bogor

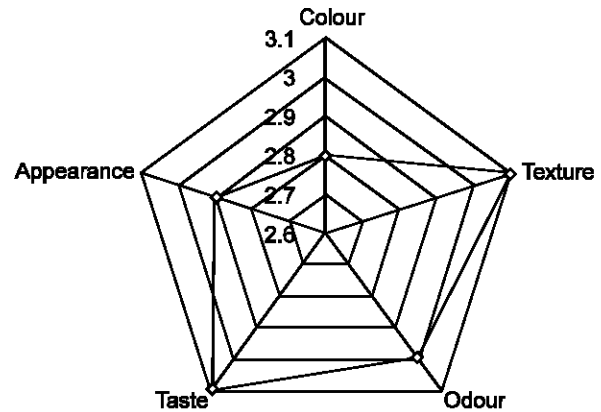


Fig. 2: Sensory evaluation of radiation sterilization of anchovy pepes as conducted by residents as volunteers at NNB in Bogor

Table 1: Comparison between pre- and post-treatment BMI in group I, II, III

Parameter	Values		
BMI (kg/m ²)	Pretreatment	Post-treatment	p-value
Group I	22.3±3.9	22.6±3.1	0.294
Group II	22.2±2.2	22.9±2.2	0.080
Group III	23.5±2.8	23.7±2.7	0.090

Table 2: Comparison between pre- and post-treatment serum albumin in group I, II, III

Parameter	Values		
Serum albumin (g/L)	Pretreatment	Post-treatment	p-value
Group I	4.58±0.24	4.66±0.36	0.314
Group II	4.52±0.27	4.53±0.25	0.907
Group III	4.51±0.19	4.58±0.12	0.047

Table 3: Comparison between pre- and post-treatment lymphocyte count in group I, II, III

Parameter	Values		
Lymphocyte (cells/mL)	Pretreatment	Post-treatment	p-value
Group I	30.93±7.20	28.14±8.30	0.374
Group II	33.22±6.60	29.58±8.72	0.029
Group III	36.02±12.10	33.04±4.30	0.449

rapidly produces more protein-based substances to defend the body. Since there can only be so many proteins in bodily fluids at one time, if the antibodies and other proteins associated with infection or injury rise, then the albumin must fall (Fleck, 1989). Remaining healthy is the only way to keep the antibodies and other proteins at normal levels, leaving "room" for plenty of albumin. When the concentration of immune system proteins goes up, the concentration of other proteins must fall (Scalfi *et al.*, 1990; Fleck, 1989). One of the proteins that decreases when the immune system lower is albumin. Albumin levels drop when the immune system engages in a battle with invading bacteria, viruses, fungi and other germs. It also falls when the body is forced to deal with an onslaught of toxins and other dangerous substances that we inhale, drink, or eat, or that get into the body through the skin, respiratory system, or other ports of entry. The fact that the albumin is good, a powerful defense, or otherwise it would die.

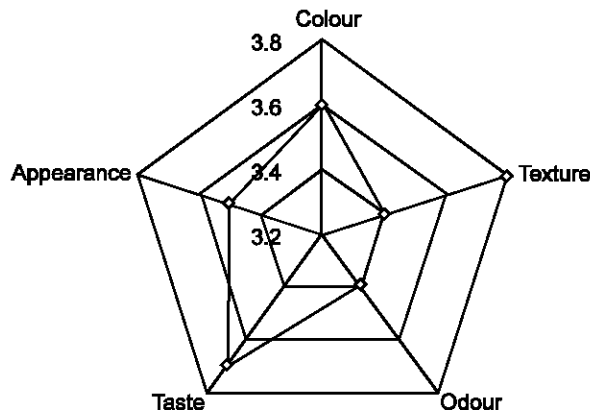


Fig. 3: Sensory evaluation of radiation sterilization of spicy *rendang* beef as conducted by residents as volunteers at NNB in Bogor

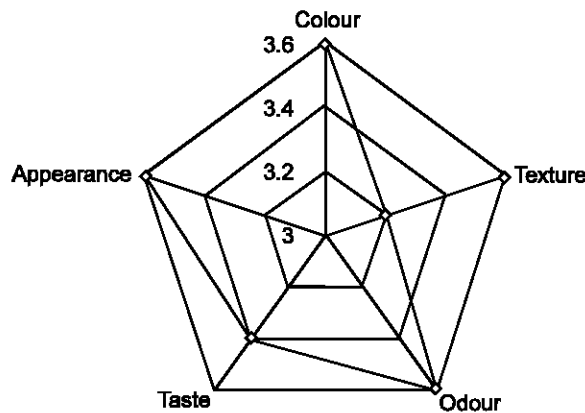


Fig. 4: Sensory evaluation of radiation sterilization of *semur* beef as conducted by residents as volunteers at NNB in Bogor

But the unintended consequence, the shortfall in albumin, is harmful in the long run. Inflammation, inadequate protein and caloric intake which cause anorexia, mal absorption of nutrient, increased resting energy metabolism all occur in immunocompromised patients with HIV/AIDS. Both inflammation and malnutrition reduce albumin concentration by decreasing its rate of synthesis, while inflammation alone is associated with a greater Fractional Catabolic Rate (FCR) and, when extreme, increased transfer of albumin out of the vascular compartment. A vicious cascade of events ensues in which inflammation induces anorexia and reduces the effective use of dietary protein and energy intake and augments catabolism of the key somatic protein, albumin (Mc. Corkindale *et al.*, 1990; Don and Kaysen, 2004).

It is well known that irradiation food sterilization at the dose of 45 kGy could eliminate all the contaminant substances including spore form pathogenic bacteria, mold and yeast (Ravel, 1995) and might result an

increase antioxidant level (Irawati *et al.*, 2011) and the nutritive substances will be more easily digested (Irawati and Sani, 2012). High quality, hygienic and wholesome of various irradiated ethnic ready to eat foods can serve as accelerating recovery process through safe foods intake diet of the immunocompromised patients (Anonymous, 2003).

The results of comparative studies on lymphocyte count of pre- and post-treatment in group I, II, III is presented in Table 3. The mean value of lymphocyte count in group I was 30.93 ± 7.2 cells/mL pre treatment and 28.14 ± 8.3 cells/mL post treatment. The post treatment lymphocyte cell count was not significantly lower than the pretreatment count ($p > 0.05$). Table 3 shows that both at pre and post tests the lymphocytes contents reduced significantly in group II who consumed non irradiated food ($p < 0.05$), but it was not significantly reduced in group III who consumed irradiated RTE food ($p > 0.05$). Lymphocytes respond to infection by rapidly dividing and produce large protein known as antibodies. Patients with immunocompromise the lymphocyte count is low. Albumin level and lymphocyte count has also been shown to be a good index of immune system. The major goals of medical nutrition therapy in immunocompromised patients is to increase immune system. In this study concluded that intervention residents treated with irradiated RTE foods increases serum albumin level and maintains lymphocyte level.

Conclusions and recommendation: It can be concluded from study that radiation sterilization of some ethnic ready to eat foods derived from animal origin were acceptable for immunocompromised people. An intervention study of the irradiated foods on immunocompromised patients with HIV/AIDS demonstrated that consumption of such foods increases albumin status and maintained lymphocyte numbers, suggesting these foods can improve nutrition status and maintain the immune system of immunocompromised patients.

It can be suggested that type and composition of diets should be first well defined and characterized according to RDA before preparation for conducting the test. Some researchers informed that persons live with HIV/AIDS must not consume certain herbal such as Echinacea, mistletoe herbs and woody nightshade stem. Clinical study on this subject is complex due to unstable emotion and psychological effect of the people with HIV/AIDS who joined the group, but most residents at NNB were very cooperative during the observation and interview.

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