Biocatalysis and Agricultural Biotechnology 10 (2017) 30–37

Contents lists available at ScienceDirect

Biocatalysis and Agricultural Biotechnology

journal homepage: www.elsevier.com/locate/bab

Effect of gamma irradiation on the nutritional and antinutritional qualities of *Vigna aconitifolia* (Jacq.) Marechal: An underutilized food legume

Pious Soris Tresina[a], Koilpitchai Paulpriya[a], Veerabahu Ramasamy Mohan[a,*], Solomon Jeeva[b]

[a] *Ethnopharmacology Unit, Research Department of Botany, V.O.Chidambaram College, Tuticorin, Tamil Nadu, India*
[b] *Department of Botany and Research Centre, Scott Christian College (Autonomous), Nagercoil, Tamil Nadu 629003, India*

ARTICLE INFO

Keywords:
Gamma irradiation
Vigna aconitifolia
Vitamins
Amino acids
PDCAAS
IVPD

ABSTRACT

Effect of gamma irradiation on the nutritionally potent underutilized wild legume *Vigna aconitifolia* at various doses (2, 5, 10, 15 and 25 kGy) were assessed for its nutritional and antinutritional factors. Gamma irradiation significantly enhanced the crude protein content at all doses, while crude lipid, crude fibre and ash resulted in a significant dose-dependent decline. Raw seeds are rich in potassium, phosphorus, magnesium, manganese and vitamins (niacin and ascorbic acid); significant diminution was reported in irradiated seeds. The essential amino acids of raw and gamma irradiated seeds were comparable with the FAO/WHO recommended pattern. A significant dose -dependent increase in IVPD on irradiation was observed. High amount of saturated fatty acids decreased after irradiation. However, linoleic acid, palmitoleic acid and eicosenoic acid were increased after irradiation at 25 kGy. Irradiating the seeds with gamma rays significantly curtailed the levels of the toxic non-proteinaceous amino acid, L-DOPA, hydrogen cyanide, trypsin inhibitors, oligosaccharides and phytohaemagglutinins. The aromatic compound, phenols, the water soluble polyphenols and tannins showed a dose-dependent significant increase. The overall findings are the indications to improvise the nutritional traits of the gamma irradiated underutilized tribal pulse, *V. aconitifolia* which could be a good source of protein for human consumption.

1. Introduction

Protein energy malnutrition is a widespread problem throughout the world and has both health and economic consequences. It is the most common deficiency disease especially in developing countries (FAO/WHO, 2001). Due to an inadequate supply of proteins of animal origin, nutritionalists, researchers and government organizations worldwide are searching for reliable, cheap and high quality proteins of plant origin. The need to provide inexpensive plant-based protein supplements has led to the examination of underutilized dicotyledonous seeds for human and livestock consumption (Adebowale and Lawal, 2004; Bhat and Sridhar, 2008).

Legumes represent an important component of human diet in several areas of the world, especially in the developing countries, where they complement the lack of proteins from cereals, roots and tubers. Among European countries, a higher legume consumption is observed around Mediterranean, with daily consumptions between 8 and 23 g/capita (DAFNE, 2009). Legumes are low in fat, and rich in proteins, complex hydrocarbons, and minerals (Geil and Anderson, 1994) and exhibit lower glycaemic index compared to other starchy foods. In addition, legumes contain a rich variety of phytochemicals, including phytosterols, natural antioxidants and bioactive carbohydrates (Amarowicz and Pegg, 2008; Rochfort and Panozzo, 2007), which if consumed in sufficient quantities may help to reduce tumour risk (Mathers, 2002). Epidemiological and intervention studies indicated that legume consumption is inversely associated with the risk of coronary heart disease (Bazzano et al., 2001), Type II diabetes mellitus (Villegas et al., 2008) and obesity (Rizkalla et al., 2002), and results in lower LDL cholesterol and higher HDL cholesterol (Anderson and Major, 2002; Bazzano et al., 2008). For these reasons, legumes are considered an ideal complement to cereals in vegetarian diets and they gain increasing attention as functional food items. Within the context of the adoption of a healthier diet, it is recommended that legume consumption should increase in the western diet (Leterme, 2002; Bell and Sears, 2003; Kalogeropoulos et al., 2010).

Nonetheless, the utilization of these underutilized legumes is limited due to the presence of certain antinutritional compounds. The removal of the undesirable components from the dry legume seed is essential for improving their nutritional qualities and for effectively utilizing them to their full potential as food. To achieve this, several

* Corresponding author.
E-mail addresses: tresina1987@gmail.com (P.S. Tresina), priyaabraham88voc@gmail.com (K. Paulpriya), vrmohanvoc@gmail.com (V.R. Mohan), solomonjeeva@gmail.com (S. Jeeva).

http://dx.doi.org/10.1016/j.bcab.2017.02.002
Received 8 September 2016; Accepted 3 February 2017
Available online 08 February 2017

processing techniques such as germination, soaking and cooking and dry heat treatment have been used (Kakati et al., 2010; Khandelwal et al., 2010; Janardhanan et al., 2003; Bhat et al., 2007; Vadivel and Pugalenthi, 2008).

Irradiation treatment as a method of preservation to enhance the shelf-life or to improve the hygienic qualities of raw and processed foods and agricultural commodities has been well established worldwide. Radiation processing has proved to be an effective means of disinfestation and decontamination of food and agricultural products (Anonymous, 1991; Loaharanu, 1994). Radiation treatment itself or in combination with other processing methods has been shown to reduce or eliminate some of the anti-nutrients in cereals and legumes (Farag, 1989; Sattar et al., 1990; Siddhuraju et al., 2002a; Bhat et al., 2008; Tresina and Mohan, 2011) security. A joint FAO/IAEA/WHO study group reviewed the toxicological, nutritional and radiation-induced chemical and physical aspects of irradiated foods above 10 kGy and concluded that application of ionizing radiation at 10 kGy or higher doses will be safe and nutritionally adequate (WHO, 1999).

Literatures on the nutritional and antinutritional properties of *Vigna aconitifolia* seeds are available. Siddhuraju et al. (1994) reported the proximate composition, minerals, seed protein fractions, amino acids, fatty acids and antinutritional factors of *Vigna aconitifolia*. The total and resistant starch (RS), dietary fibre (DF) and soluble sugars including oligosaccharides of *V. aconitifolia* were determined by Bravo et al. (1999). Effect of soaking and heat processing on the levels of antinutrients and digestible proteins of *V. aconitifolia* were reported by Vijayakumari et al. (1998). There are insufficient reports about possible effects of gamma irradiation on nutritional value of *V. aconitifolia*. Consequently, the present investigation was commenced to explore the impact of gamma irradiation on the nutritional and antinutritional factors of the underutilized legume, *V. aconitifolia*.

2. Materials and methods

2.1. Collection of seeds

The mature seed materials of *Vigna aconitifolia* (Jacq.) Marechal were collected from Sivagiri Hills, Tamil Nadu. Soon after the collection, the seeds were sun dried for 2–3 days and were surface cleaned with muslin cloth and physically damaged, immature and insect infested seeds were eliminated.

2.2. Irradiation

Seed samples (each-50 g) packed in polyethylene pouches were irradiated at different doses of gamma irradiation (2, 5, 10, 15 and 25 kGy) at room temperature (25 ± 1 °C) using a Cobalt -60 Gamma cell 5000 unit at Radiological Safety Division, Indira Gandhi Center for Atomic Research, Kalpakam, Tamil Nadu. Seed samples packed similarly without irradiation served as control. The seed samples were powdered and stored in screw capped bottles for further usage.

2.3. Analyses of proximate composition

The moisture content (%) was determined by drying 50 transversely cut seed in an oven at 80 °C for 24 h and is expressed on a percentage basis. The air-dried samples were powdered separately in a Wiley mill (Scientific Equipment, Delhi, India) to 60-mesh size and stored in screw capped bottles at room temperature for further analysis.

The nitrogen content was estimated by the micro-Kjeldahl method (Humphries, 1956) and the crude protein content was calculated (N x 6.25). Crude lipid content was determined using Soxhlet apparatus (AOAC, 2005). The ash content was determined by heating 2 g of the dried sample in a silica dish at 600 °C for 6hr (AOAC, 2005). Total dietary fibre (TDF) was estimated by the non-enzymatic-gravimetric method (Li and Cardozo, 1994).

The nitrogen free extract (NFE) was obtained by difference (Muller and Tobin, 1980). The energy value of the seed (kJ) was estimated by multiplying the percentages of crude protein, crude lipid and NFE by the factors 16.7, 37.7 and 16.7, respectively (Siddhuraju et al., 1996).

2.4. Minerals and vitamins analyses

Five hundred mg of the ground legume seed was digested with a mixture of 10 ml concentrated nitric acid, 4 ml of 60% perchloric acid and 1 ml of concentrated sulphuric acid. After cooling, the digest was diluted with 50 ml of deionised distilled water, filtered with Whatman No. 42 filter paper and the filtrates were made up to 100 ml in a glass volumetric flask with deionised distilled water. All the minerals except phosphorus were analysed from a triple acid-digested sample by an atomic absorption spectrophotometer – ECIL (Electronic Corporation of India Ltd., India) (Issac and Johnson, 1975). The phosphorus content was determined colorimetrically (Dickman and Bray, 1940).

Ascorbic acid and niacin contents were extracted and estimated as per the method given by Sadasivam and Manickam (1996).

2.5. Amino acid analyses

The total seed protein was extracted by a modified method of Basha et al. (1976). The extracted proteins were purified by precipitation with cold 20% trichloroacetic acid (TCA). A protein sample of 30 mg was hydrolysed by 6N HCL (5 ml) in an evacuated sealed tube, which was kept in an air oven maintained at 110 °C for 24 h. The sealed tube was broken and the acid removed completely by repeated flash evaporation after the addition of de-ionized water. Dilution was effected by means of citrate buffer pH 2.2 to such an extent that the solution contained 0.5 mg protein ml^{-1}. The solution was passed through a millipore filter (0.45 μM) and derivitized with O-phthaldialdehyde by using an automated pre-column (OPA). Aminoacids were analysed by a reverse – phase HPLC (Method L 7400, HITACHI, Japan) fitted with a denali C_{18} 5 μm column (4.6×150 mm). The flow rate was 1 ml/min with fluorescence detector. The cystine content of protein sample was obtained separately by the Liddell and Saville (1959) method. For the determination of tryptophan content of proteins, aliquots containing known amounts of proteins were dispersed into glass ampoules together with 1 ml 5 M NaOH. The ampoules were flame sealed and incubated at 110 °C for 18 h. The tryptophan contents of the alkaline hydrolysates were determined colorimetrically using the method of Spies and Chamber (1949) as modified by Rama Rao et al. (1974). The contents of the different amino acids were expressed as g100g-1 proteins and were compared with FAO/WHO (1991) reference pattern. The essential amino acid score was calculated as follows:

$$\text{Essential amino acid score} = \frac{\text{grams essential amino acid in 100g of total protein}}{\text{grams of essential amino acid in 100g of FAO/WHO (1991) reference pattern}} \times 100$$

2.6. Determination of in vitro protein digestibility (IVPD)

In vitro protein digestibility (IVPD) of unprocessed and processed seed samples was determined using the multi-enzyme techniques (Hsu et al., 1977).

The protein digestibility corrected amino acid score (PDCAAS) of EAA was calculated based on EAA requirements for adults (FAO/WHO, 1991):