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Original Article

Effect of Different Thermal Techniques on Anti-Nutritional Compunds with Special Refefence to Hydrogen Cyanide in *Linum usitatissimum*

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ABSTRACT

Flax is one of the world's primordial crops with proceedings of human cultivation extending back to thousands of years. **Objective:** To observe the comparative effectiveness of different thermal treatments on hydrogen cyanide (HCN) contents in flaxseed (*Linum usitatissimum* L.). **Methods:** For the purpose, flaxseed was procured from local market, Faisalabad-Pakistan. Then, it was characterized for nutritional composition and mineral profiling through respective methods. In addition, dietary fiber, fatty acids, lignans and hydrogen cyanide (HCN) content were assessed through Enzymatic-gravimetric methods, gas chromatography-mass spectrometry (GCMS), high performance liquid chromatography (HPLC) and alkaline titration, respectively. Moreover, different heat treatments *i.e.* autoclaving, microwave roasting, oven heating and water boiling were applied to mitigate the hydrogen cyanide contents. **Results:** Results of alkaline titration before heat treatment showed that hydrogen cyanide content was 376.0 mg/kg in flaxseed. The effect of heat treatments on hydrogen cyanide content of flaxseed was found to be highly significant. Hydrogen cyanide content was reduced to 22.33 mg/kg, 62.792 mg/kg, 204.33 mg/kg and 300.048 mg/kg by water boiling, microwave roasting, autoclaving and oven heating, respectively**. Conclusion:** Water boiling is most effectual technique (98% reduction) while oven heating is the slightest one (20.2% reduction).

INTRODUCTION

Flax belongs to *Linaceae* family and is principally grown for fiber and oil purposes [1-3]. Two varieties of flax seed-yellow and golden brown are prominent for having approximately identical nutritional value as well as components [4,5]. Flax seed is a supplement food that restrains high amount of essential omega, 3 polyunsaturated fatty acids (α-linolenic acid) and phytochemicals such as lignans [2,6,7]. Various factors such as variety, environment and method of analysis influence the composition of flax [8]. It contains oil, viscous lignan-rich fibers (mucilage), protein and minerals as major components. Protein present in flax seed is a good-quality protein and 58-66% of total seed protein are storage proteins i.e. albumins and globulin [9,10]. The prospective bioactive compound, lignan present in flaxseed is secoisolariciresinol diglucoside (SDG) which retards the development of type II diabetes allied with amplification in oxidative stress [11,12]. Though, the occurrence of certain anti-nutritional factors, such as cyanogenic glycosides, cadmium, trypsin inhibitors and phytic acid, makes flaxseed lethal and boost its negative effects on human health [13-15]. Cyanogenic glycosides are termed as glycosidic compounds formed from a simple sugar and another compound i.e. aldehydes or ketones by replacement of a hydroxyl group in the sugar molecule [16]. Their occurrence plays a vital role in the protection of flaxseed. Cyanogenic glycosides are hydrolysed as the plant tissues are damaged. As a result, hydrogen cyanide

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(HCN) is produced which is a determent of enzyme cytochrome oxidase and also an obstacle of cell respiration. HCN is produced by over 1000 species of plants, among which most important is flaxseed. HCN is a potent respiratory inhibitor. High levels of HCN are present in flaxseed [17,18]. Certain types of thermal methods such as water boiling, microwave roasting, oven heating and autoclaving are used to remove HCN from flaxseed. Such thermal treatments not only diminish the anti-nutritional compounds but also augment the efficacy of flaxseed against life style disorders [18]. Keeping in view all above characteristics of flaxseed, the present study has been designed to reduce the HCN content of flaxseed using certain heat treatments to make the commodity nutritionally valuable.

METHODS

Flaxseeds were procured from local market of Faisalabad, Pakistan. Seeds were cleaned to remove any debris or field dirt and sealed in polyethylene bags. The Hydrogen cyanide (HCN) contents of flaxseed were estimated by alkaline titration according to the AOAC Method 26.115. HCN was removed by four different types of heat treatments i.e. autoclaving, oven heating, water boiling and microwave roasting [18]. Flaxseed sample (50g) , at a temperature of 120°C, was autoclaved for 25 minutes and then allowed to cool down at room temperature. It was then dried in oven (40°C). Flaxseed sample (50 g) was heated in hot air oven (120°C for 20 min) and then cooled down at room temperature. Flaxseed sample (50 g) was boiled at 100°C (for 20 minutes) in a beaker containing 500ml of tap water. Water was drained after cooling down. The content in beaker was given 2 washes with tap water and then dried in oven (40°C). Flaxseed sample (50 g) was roasted for 2 min and the output power was 640 W. Then the sample was cooled in a desiccator. After heat treatments, the flaxseed was again analyzed for HCN content by using same estimation methods. Analyses were carried out in replicate for precision and accuracy. The obtained data were subjected to completely randomized design (CRD) using Statistical Package (Costat-2003, Co-Hort, v 6.1.). Levels of significance were determined (ANOVA) using 1-factor factorial CRD following the principles outlined by Steel *et al.* [19].

RESULTS

The present study indicated that autoclaving reduced about 29.7% HCN content of flaxseed as shown in Figure 1. The HCN content was decreased from 376.0 mg/kg Feng *et al.* [18] explicated that through autoclaving for 30min reduced the HCN level in flaxseed meal from 470–490mgkg–1 to negligible amounts. Figure 1 signified that microwave roasting reduced almost 83.3% of HCN content that showed its high effectiveness. The HCN content was lessened from 376.0 mg/kg heat treatment also reduced the HCNs content of flaxseed but the reduction was low i.e. almost 20.2%. The HCN content was reduced from 376.0 mg/kg to 300.048 mg/kg (Table 6). Water boiling reduced almost 98-100% hydrogen cyanide content of flaxseed.

Treatments	T_1	T_2	T_3	T_4
HCN	263.18±1.23	62.61±1.39	300.49±0.57	22.33±2.52

Table 1: Means for HCN contents, T1= Autoclaving, T2= Microwave roasting, T3= Oven heating, T4= Water boiling

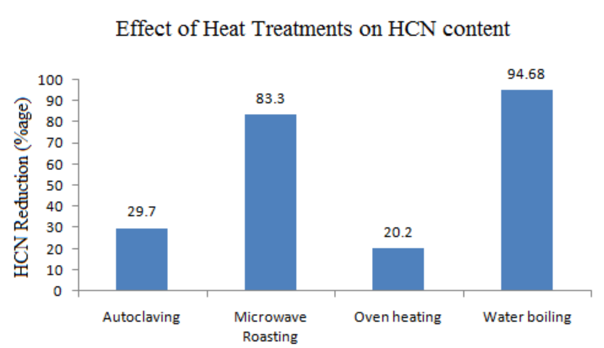

Figure 1: Effect of heat treatments on hydrogen cyanide content (%)

DISCUSSION

In the current study, the raw flaxseed had a hydrogen cyanide content of 376.0 mg/kg. It is strongly supported by the results of studies of Feng *et al.* [18] who reported that flaxseed contained 377.0 mg/kg HCN content. Moreover, Mandokhot & Singh [20] probed 470-490 mg/kg HCN in flaxseed. Roseling [21] evaluated that flaxseed contained 104-312 mg/kg HCN content. Studies of Bhatty [22] elaborated that cyanide content in the flaxseed was 79-98 µg/kg and the method used was colorimetric. A study [23] explicated 124-196 mg/kg HCN content in flaxseed determined by high performance liquid chromatography. Peculiarity cyanide in flaxseed takes place mainly owing to cultivar diversity, their expression in different compounds and differences in detection methods. Effect of heat treatments on HCN content of flaxseed was highly significant. This larger difference among the effectiveness of all heat treatments was due to many factors such as heating temperature, time, pressure, cooling temperature and so many other conditions. Comparative study of the effectiveness of heat treatments showed that HCN content

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was reduced by both autoclaving and oven heating but effectiveness of autoclaving was more than oven heating. In the same way, results showed that although all treatments were effective in reducing the HCN content but treatment water boiling possessed more pronounced effect while treatment oven heating was least effective among all heat treatments. Effect of microwave roasting was greater than autoclaving and oven heating but not from water boiling. This was due to the strong heat induction power of microwave. The studies of Feng *et al*. [18] showed almost similar results. Their results revealed that autoclaving reduced the HCN content from 377.0 mg/kg to 265.0 mg/kg. Microwave roasting reduced the HCN content in flaxseed from 377.0 mg/kg to 63.5 mg/kg. It was very effective treatment among all heat treatments. They explicated that oven heating reduced the hydrogen cyanide content from 377.0 mg/kg to 316.0 mg/kg. This treatment was not very effective.

CONCLUSION

Flaxseed (Locally grown) is found to be very high-quality reservoir of fat, protein, fiber and lignans and tremendous resource of unsaturated fatty acids including linolenic acid, linoleic acid, oleic acid etc. As a promising source of unsaturated fatty acids, flax seeds should be exploited for therapeutic and health enhancing food products. The effect of heat treatments on hydrogen cyanide content of flaxseed was found to be highly significant. The thermal treatment (water boiling) exhibited highest HCN reduction by 94.68% followed by microwave roasting 83.3%, autoclaving 29.7% and oven heating 20.0%, correspondingly. So it is concluded that highest positive response on HCN reduction was observed in water boiling followed by microwave roasting, autoclaving and oven heating.

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