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Inactivation of Surface Microorganisms of Moth bean (Vigna aconitifolia) by Micronization and Changes in its Physico Chemical and Rheological Properties

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Abstract
Moth bean seeds (*Vigna aconitifolia*) were micronized at different temperatures viz 150, 250, 300, 350 and 400^0 C to study the efficacy of treatment to check the surface microorganisms. Surface microbes were found to decrease by 90 % when micronized at 300^0 C with more than 85% germination at this temperature. Functional and rheological properties were not affected much up to the treatment of the seeds at 300^0 C. Above this temperature, significant changes ($p\leq0.05$) in functional and rheological properties were observed. Antinutritional factors viz phytic acid, trypsin inhibition activity (TIA), tannins and saponins decreased significantly ($p\leq0.05$). Total phenols and total flavonoids also found to decrease slightly but significantly ($p\leq0.05$) during micronization which correlated with the decrease in antioxidant activity as observed by DPPH radical scavenging methods. Micronization of moth bean at 300^0 C can be used effectively for the surface inactivation of microbes without affecting the seed quality deleteriously.
Keywords: Moth bean, micronization, surface inactivation, rheology, physico-chemical changes

1. Introduction

Grain legumes are the major food sources for humans across the world, as it is one of the important sources of protein among the vegetarian population. Most of the food grains are processed into end products before consumption or is consumed with minimal processing. Food grains are often contaminated with bacteria, virus and other microorganisms during harvesting, transportation and improper storage conditions (Hocking, 2003). In order to ensure that the grain and its end product are healthy and safe, it becomes necessary to enhance its safety from production to consumption. Even though the moisture content of the dried grain does not allow the growth of microorganisms, but the surface contamination through various species of microorganisms and also the risk associated with improper storage of grains in the field, contamination from pests, rodents cannot be ruled out. It has been reported that more than 25 species of different fungi are known to invade stored grains and legumes (Duan et al., 2007) . Invasion of stored grains by fungi and other microbes causes reduction in cooking, produce undesirable odour and colour and reduces its market value as well. Further, mycotoxins produced from the fungi makes the product unacceptable for edible purpose (Selcuk et al., 2008). Chemical treatment using insecticides has been constantly used for the disinfestations of the agricultural products. Due to the increased concern about the hazardous effects of the existing disinfestation practices, various alternate non hazardous thermal treatments such as microwave heating (Zain and Ooi 1994), radio frequency energy (Wang et al., 2010) and hot air methods (Fields 1992, Dowdy 1991, Dosland et al., 2006) have been employed for the disinfestation purpose. One such possible methods of thermal treatment drawing greater attention during recent years is the use of micronization, as this technique offers several advantages such as short time treatment, versatile, safe, uniform heating with better quality of finished products, and high efficiency as compared to other methods of heating (Rastogi, 2012). As micronization is a surface phenomenon, the rays will fall rapidly and effectively, heat the surface of the grain without penetrating deep in to the substance to be heated thereby causing less deterioration to the internal quality of the grains by leaving no residues on the grain.

Moth bean is one of the most important legume crop consumed in almost all parts of India in different forms. But surface contamination of grain from microbes during harvesting, storage and infestation from various insects and pests affects its quality by reducing its nutritional value thus making it unfit for consumption. Effect of insecticides, pesticides and also storage in cloth bags were studied to control infestation in moth bean during storage (Ananymous, 2005). Several grains and legumes were subjected to plasma (Selcuk et al., 2008, Butscher et al., 2015) and infrared treatments (Uchino et al 2000) to reduce surface contamination. However no work has so far been undertaken to decontaminate moth bean through micronization to ensure microbial safety by surface treatment with minimum changes in nutritional and functional properties.

Therefore the present work was undertaken to investigate the effect of micronization on the inactivation of surface microorganisms and to study the effect of different micronization temperatures on the physico chemical and rheological changes of moth bean and to check the suitability of a particular micronization temperature to extend its shelf life with minimum deterioration in its quality.

2.0 Materials and methods
Moth bean seeds were procured from the local market of Mysuru, Karnataka, India. The seeds were carefully cleaned by removing foreign particles, broken and damaged seeds and stored at room temperature until micronization.

2.1 Micronization of moth bean.
2 kg of sound and cleaned moth bean seeds were passed through Infrared (IR) heaters mounted with electrically powered ceramic heating elements (1000 watt) having the capacity of 40 kg/h (Therman Heating Technologies Pvt Ltd, Bangalore, India) and can be operated up to 900°C. Moth bean seeds were micronized at fixed IR temperatures of 150°C, 250°C, 300°C, 350°C and 400°C respectively for approximately 50±2 Sec. After micronization, surface temperature of seeds was measured using hand-held infrared temperature gun thermometer (Mode: DT-8550, Frontier, M/s Sunray Industries, Mysore) and spread on trays and cooled to room temperature. Raw (non micronized) seeds served as a control. Micronized and non- non micronized samples of moth bean seeds were ground to pass through 0.250 mm sieve, packed in polypropylene (75 μ) pouches and stored at room temperature for further analysis.

2.2 Analysis
Moisture, total ash, crude protein, crude fat, crude fiber of raw and micronized moth bean seeds were determined by the methods described by (AOAC, 1984). Water and oil absorption capacities were measured using centrifugation technique of Sathe et al., (1982). The solubility index and swelling capacity of the samples were determined based on the method of Leach et al., (1959). Swelling index of the sample was estimated according to Abu et al., (2005). Sedimentation value of raw and micronized samples of moth bean was measured as per the method of Ghadge et al., (2008). Antioxidant activity was measured according to the method of Gorenstein et al., (2004). Total phenolics and flavonoids were estimated by the method described by Gerard and Roberts (2004). Total tannins were determined colorimetrically as described in AOAC (1990). Saponins were estimated according to the method suggested by Dorothy and Oakenfull (1981). Phytate content was determined using the method described by Haugh and Lantzsch (1993). Trypsin inhibitor activity was measured according to the method of Kakade et al., (1969) using BAPA as the substrate. In vitro protein digestibility (IVPD) was determined by multi enzyme technique as described by Hsu et al., (1977). In vitro starch digestibility was determined by Singh et al., (1982). Microbiological analysis was carried out as per APHA (1992) methods.

2.3 Statistical analysis
The reported values are the mean of three replicates each and statistical analysis was carried out by using statistical software (Statistica, Ver 7.0 of Stat Soft Incorporation, Tulsa OK, USA). Experimental results were subjected to one way analysis of variance (ANOVA) for significance ($p \leq 0.05$) using Duncan's multiple range tests.

3.0 Results and discussion
3.1 Effect of micronization on the proximate composition of moth bean seeds
Table- 1 shows the proximate composition of raw as well as micronized moth bean seeds treated at varying temperatures. IR treated moth bean samples at different temperatures have not undergone major changes in the crude protein, crude fat, total ash and crude fibre contents with respect to the untreated raw sample, may be due to the heat treatment given for shortest time of approximately 50±2 sec. Crude protein content varied from 23.40 to 23.82%, crude fat content from 0.80 to 1.04%, total ash from 3.01 to 3.18%, crude fibre from 3.79 to 3.92%. Carbohydrate calculated by difference varied from 58.22 to 59.31% with an energy value ranging from 335.40 to 339.76 Kcal. However, with the increase in temperature from 150 to 400^{0}C, the moisture content was found to decrease slightly. The raw seed which has exhibited a moisture content of 9.88% was decreased to 9.86, 9.81, 9.76, 9.49 and 9.34% when seeds were treated at 150, 250, 300, 350, 400^{0}C respectively.

3.2 Effect of micronization on the functional properties of moth bean seeds
The changes in functional properties of raw and IR treated moth bean seeds at different temperatures are indicated in the Table-2. Water and oil absorption capacities have shown an insignificant ($p \geq 0.05$) increase in all the micronized samples as compared to the untreated ones except the samples treated at 400^{0}C which has shown a slight but, significant increase ($p \leq 0.05$) in water absorption capacity only. The increase in water absorption is due to the increase in gelatinization of starch and protein denaturation upon cooking (Lin et al 1974).The dry heating of the seeds might have resulted in slight starch gelatinization and therefore water absorption capacity increased slightly from 1.94 to 2.16 g/g. Oil absorption capacity remained almost unchanged and it increased by 0.13 units wrt to the raw sample. Our study has resulted in a slight increase in the sedimentation value during micronization of moth bean at various temperatures. It increased from 28 (in untreated raw sample) to 29, 29, 30,

31, 32 ml in irradiated samples at 150, 250, 300, 350, 400^0C respectively. Increase in sedimentation value indicates better dispersibility of the particulate matter without much damage to starch and protein during cooking (Ghadge et al., 2008). Swelling index which indicates swelling of starch during gelatinization reduced by one unit during micronization. Raw sample has shown a swelling index of 4.09, reduced to 3.10 in the sample micronized at 400^0 C. Water solubility index as well as swelling power has shown an increase after the treatment of the seeds above 150^0C, later at all the temperatures of micronization, it remained constant. The results of the above study revealed only slight changes in the functional properties of moth bean when micronized at various temperatures from 150 to 300^0 C.

3.3 Effect of micronization on total phenols, total flavonoids and antioxidant activity of moth bean seeds

The result in the Table- 3 summarizes total phenols, total flavonoids and antioxidant activity of raw and micronized moth bean at varying temperatures of treatment. Treatment of the seeds above 150^0C, have shown less but significant ($p \leq 0.05$) reduction in both phenols and flavonoid contents. Untreated raw legume showed 301.27mg/100g total phenols, decreased to 258.80 mg/100g upon micronization at 400^0 C and flavonoid content showed a decrease from 22.00 to 16.15 mg/100g at the end of micronization. Ramkrishna et al., (2006) also reported a decrease in total phenol content during different processing treatments in Indian beans. The antioxidant activity of micronized moth bean samples as reported by DPPH radical scavenging methods implies that, with the progression of micronization temperature, the radical scavenging activity was decreased though slightly, but significantly ($p \leq 0.05$) as compared to the untreated raw sample. The observed decrease in antioxidant activity might be due to the decrease in total phenol and total flavonoid contents. The radical scavenging activity was decreased maximum by 4.51 units when seeds were micronized at 400^0 C.

3.4 Effect of micronization on the antinutritional factors of moth bean seeds

The effect of micronization on the anti-nutritional factors such as phytic acid, trypsin inhibition activity, tannins and saponins are presented in the Table-4. Treatment of moth bean at various micronization temperatures have shown less but significant ($p \leq 0.05$) reduction of phytic acid and trypsin inhibition activity. Raw sample reported 146.57 mg/100g of phytic acid and samples treated at 150, 250, 300, 350, 400^0C has shown 141.93, 141.20, 139.65, 139.40, 131.80 mg/100g phytic acid contents. The decrease in phytic acid upon thermal treatment may be either due to the formation of insoluble phytate- protein and phytate- protein mineral complexes or inositol hexa phosphate hydrolyzed to penta and tetra phosphates (Siddaraju and Becker, 2001). Our study revealed lesser phytic acid contents in comparison to the data reported by Kokkhar and Chauhan (1986) for different varieties of moth bean. Trypsin inhibition activity showed a reduction from 171 TIU/g to 160 TIU/g. Treatment of the seeds at varying temperatures significantly reduced ($p \leq 0.05$) tannins from 3.77 to 2.18 mg/100g and saponins from 3.03 to 2.02%. The values of saponins and trpysin inhibition activity of the moth bean correlated with the findings of Kokkhar and Chauhan (1986).

3.5 Effect of micronization on the pasting properties of moth bean seeds

The pasting properties of raw and micronized moth bean seeds are shown in the Table- 5. Raw seeds have shown 205, 899, 246 and 84 RVU of breakdown, final, set back viscosities and pasting temperatures respectively. Break down viscosity which measures the susceptibility of the starch to disintegrate increased significantly ($p \leq 0.05$) from 205 RVU in raw seeds to 405 RVU in the seeds treated at 400^0C, but not affected much when micronized up to 300^0 C. Final viscosity, a parameter related to define a particular sample quality indicating the ability of a material to form a paste or gel also has shown an increase from 899 to 935 RVU up to 300^0 C, but increase was found more rapid after the treatment of the seeds above 300^0 C. Set back viscosity which indicates the reordering or retrogradation of starch was found to increase from 246 to 381 RVU during micronization of the seeds. Micronization has not revealed any major changes in pasting temperature and it ranged from 81 to 87 RVU at various temperatures of micronization. Xie et al., (2013) has also observed changes in rheological properties of potato starch granules when microwave treated for different intervals of time. Micronization up to 300^0 C did not show major changes in the rheology of moth bean seeds but affected slightly and significantly ($p \leq 0.05$) when the temperature of micronization was raised above 300^0 C.

3.6 Effect of micronization on the germination potential of moth bean seeds

The germination studies carried out in raw as well as micronized moth bean at different temperatures is indicated in Table 6. Raw seeds showed 97% germination. Micronization of moth bean up to 150^0C, has not changed its germination potential. Seeds micronized at 250 and 300^0 C have shown 90 and 88% germination respectively. Micronization of the seeds at 350^0 C has reduced its germination capacity significantly ($p \leq 0.05$) to 79% less than the expected rate of germination for the good quality seed. Moth bean micronized at 400^0 C has reduced its germination potential to almost half indicating the seed quality deterioration at this temperature of treatment.

3.7 Effect of micronization on invitro protein and starch digestibility of moth bean seeds

Invitro protein and starch digestibility carried out in raw as well as micronized samples at various temperatures are represented in the Figure 1. Micronization has not improved the protein digestibility of the seeds. It remained constant and showed an average of 73.54% digestibility upto 350^{0}C. It enhanced only by 1.37% after the treatment of the seeds at 400^{0}C. However, as far as the digestibility of the starch is concerned, raw samples of moth bean showed 28.82% digestibility and micronization at each temperature has significantly improved the digestibility of the starch. Starch digestibility has enhanced max by 24.83% after the treatment of the seeds at 400^{0}C. It enhanced from 28.82 (Raw, untreated) to 29.99% at 150^{0}C, 32.31% at 250^{0}C, 34.10% at 300^{0}C, 36.11% at 350^{0}C and 38.34% at 400^{0}C. Increase in starch digestibility is due to the hydrolysis of starch as a result of heat treatment and also some investigators have reported that cooking enhances digestion through starch gelatinization (Yu-hui 1991, Mbofung et al., 1999, Rehman et al., 2001).

3.8 Effect of micronization on the surface inactivation of microorganisms

The effect of micronization on the inactivation of surface microorganisms at various temperatures of treatment is shown in the Figure 2. Increase in temperature of micronization has shown a significant ($p \leq 0.05$) reduction in surface microbial population. When the samples were micronized at 150^{0}C, surface inactivation was achieved only by 20%, as the surface temperature of the grain was 35^{0}C, slightly above the room temperature. When the temperature of micronizaton was increased to 250^{0}C, the surface temperature of the seeds raised to 56^0 C with 60% decrease in microbial load. More than 90% reduction in microbial population was observed at 300 and 350^0 C, with the corresponding surface temperatures of 69 and 78^{0}C respectively. Uchino et al., (2000) also reported a significant decrease in the survival rate of surface microorganisms when wheat grains were micronized at different temperatures. Even though the micronization of the seeds at 400^{0}C has decreased the microbial load by 96%, but temperature of the seed surface was raised to 90^{0}C, which deteriorated seed quality by diminishing the surface colour as well as its physico-chemical and rheological characteristics. Samples were also tested for few pathogens like *E. coli, Salmonella* and *S. aureus* and found to be absent in all the micronized samples.

4.0 Conclusions

Micronization of moth bean seeds up to 300^0 C, has not significantly affected the quality of the seeds in terms of changes in its physico chemical and rheological attributes. Eventhough, there were slight changes in the rheology of the seeds, but their pasting temperature remained almost constant indicating that the cooking quality of moth bean is not affected by micronization when the seeds were treated up to 300^0 C showing good germination capacity of 88%. The surface microorganisms decreased significantly ($p \leq 0.05$) with the increase in micronization temperature and 90 % reduction in microbial load was achieved upon treatment at 300^0 C. Thus micronization, a short time heating technique, a non chemical thermal treatment has a great potential to inactivate surface microorganisms without deteriorating seed quality significantly.

5.0 References

1. Abu, J.O. Muller, K. G. and Minnar, A. 2005. Functional properties of cowpea flours and pastes as affected by gamma irradiation. Food Chemistry, 93:103-111.
2. Anonymous, 2005. Karnataka Journal of Agricultural Sciences, 18: 174-176.
3. AOAC. 1984. Official methods of analysis, Association of Official Analytical Chemists.14th edition. Arlington, Virginia.
4. AOAC. 1990. Official methods of analysis, Association of Official Analytical Chemists. 15th edition. Washington, DC.
5. APHA. 1992. Compendium of methods for the microbiological examination of foods. 2nd Edition. Speck ML (Ed). American Public Health association, Washington DC.
6. Butscher, D. Schlup, T. Rotho, C. Fischer, N.M. Demarchi, C.G and Van Rohr, P.R. 2015. Inactivation of microorganisns on granular materials, Reduction of Bacillus amyloliquifaciens endospores on wheat grains in a low pressure plasma circulating fluidized bed reactor. Journal of Food Engineering, 159: 48-56.
7. Dorothy, F. E. and Oakenfull, D. 1981.Saponin content of soya beans and some commercial soya bean products. Journal of Science of Food and Agriculture, 32: 273–278.
8. Dosland, Q. Subramanyam, B.H. Sheppard, K. and Mahroof R. 2006. Temperature modification for insect control. In: Heaps J (ed) Insect management for food storage and processing, 2nd edition, American Association of Clinical Chemistry, St Paul. MN 2006. pp 89-103.
9. Dowdy, A.K. (1999). Heat sterilization as an alternative to methyl bromide fumigation in cereal processing plant. In: Jin, Z. Liang, Q. Liang, Y. Tan, X. Guan, L. (Eds) Proceedings of 7th International working conference for stored products protection, Beijing, China, pp. 1089-1095.

10. Duan, C.X. Wang, X.M. Zhu, Z.D. and Cue, XF. 2007. Treating of seed borne fungi in wheat germ plasma conserved in the national crop genebank of China. Agricultural Sciences of China, 6:682-687.
11. Fields, P.G. 1962. The control of stored product insects and mites with extreme temperatures. Journal of Stored Products Research, 28: 89-118.
12. Gerard, K.A. and Roberts, J.S. 2004. Microwave heating of apple mash to improve juice yield and quality. Lebensmittel-Wissenschaft Technol, 37 : 551-557
13. Ghadge, P. N. Shewalkar, S.V. and Wankhede, D. B. 2008. Effect of processing methods on qualities of instant whole legume: pigeon pea (*Cajanus cajan* L.). Agriculture Engineering International. The CIGR Journal, 10: 01-08.
14. Gorinstein, S. Haruenkit, R. Yong, S.P. Jung, S.T. Zachweija, Z. and Jastrzehski, Z. 2004. Bioactive compounds and antioxidant potential in fresh and dried Jaffa sweeties, a new kind of citrus fruit. Journal of Science of Food and Agriculture, 84: 1459-1463.
15. Haugh, W. and Lantzsch, H. J. 1993. Sensitive method for determination of phytate in cereals and cereal products. Plant Foods for Human Nutrition, 44: 261–266.
16. Hocking, A.D. 2003. Microbiological facts and frictions in grain storage. Proceedings of the Australian postharvest technical conference, Canberra. pp 25-27
17. Hsu, H.W. Vavak, D.L. Satterlee, L.D. and Miller, G.A. 1977. A multienzyme technique for estimating protein digestibility. Journal of Food Science, 42: 1269-1271.
18. Kakade, M. L. Simsons, N. and Liener, I. E. 1969. An evaluation of natural V/s synthetic substrates for measuring the antitryptic activity of soy bean samples. Cereal Chemistry, 46: 518-528.
19. Kokkar, S. and Chauhan, B.M. 1986. Antinutritional factors in moth bean; varietal differences and effects of methods of domestic processing and cooking .Journal of Food Science, 51: 591-594.
20. Leach, H.W. Mc Cowen, L.D. and Schoch, T.J. 1959. The structure of starch granules. I. Swelling and solubility patterns of various starches. Cereal chemistry, 36: 534-544.
21. Lin, M. J. Y. Humbert, E. S. and Sosulski, F. 1974. Certain functional properties of sunflower meal products. Journal of Food Science, 39: 368-372.
22. Mbofung, C.M.F. Rigby, N. and Waldion, C.K. 1999. Use of two varieties of hard to cook beans and cowpeas in the processing of Koki, a steamed legume product. Plant Food for Human Nutrition, 54: 131-150.
23. Ramkrishna, V. Jhansi rani, P. and Ramkrishna rao, P. 2006 . Nutritional quality of storage proteins during germination of Indian bean (*Dolichos lablab*). Crop Science, 40 : 125-126
24. Rastogi, N.K. 2012. Recent trends and developments in infrared heating in food processing, Critical Review in Food Science and Nutrition, 52: 737-760.
25. Rehman, Z.U. Salariya, A.M. Yasin, M. and Zafar, S.I. 2001. Thermal heat processing effects on invitro protein digestibility of chick pea (*Cicer arietinum).* Pakistan Journal of Scientific Research, 53:75-77.
26. Sathe, S. K. Deshpande, S. S. and Salunke, D. K. 1982. Functional properties of lupin seeds (*Lupinus muta bilis*) protein and protein concentrate. Journal of Food Science, 47: 491-497.
27. Selcuk, M. Oksuz, L. Basaran P. 2008. Decontamination of grain legumes infested with Aspergillus spp and Pencilium spp by cold plasma treatment. Bioresource Technology, 99: 5104-5109.
28. Siddaraju, P and Becker, K. 2001. Effect of various domestic processing methods on antinutrients and invitro protein and starch digestibility of two indigenous varieties of Indian pulses, *Mucuna pruries Var utilis*. Journal of Agricultural Food Chemistry, 49: 3058-3067.
29. Singh, U. Kherdekar, M. S. and Jambunathan, R. 1982. Studies on Desi and Kabuli chickpea (*Cicer arietinum L*) cultivars. The levels of amylase inhibitors, levels of oligosaccharides and *in vitro* starch digestibility. Journal of Food Science, 47: 510-512.
30. Uchino, T. Hamanoka, D. and Hu, W. 2000. Inactivation of micro organisms on wheat grains by using infra red radiations. Proceedings of international workshop on Agricultural Engineering and Agro Products: Processing Towards mechanization and modernization in agriculture and rural areas.
31. Wang, S. Tiwari, G. Jiao, S. Johnson, J.A. and Tang, J. 2010. Developing postharvest disinfestations treatments for legumes using radio frequency energy. Biosystems Engineering, 105: 341-349.
32. Xie, Y. Yan, M. Yuan, S. Sun, S. and Huo. Q. 2013. Effect of microwave treatment on the physico chemical properties of potato starch granules. Chemistry Central Journal, 7: 113-116.
33. Yu-Hui, 1991. Effect of hard to cook defect and processing on protein and starch digestibility of cow peas. Journal of Cereal Chemistry, 68: 413-418.
34. Zain, A.M. and Ooi, L.H. 1994. Functional properties of stored grains after microwave treatment. Proceedings of 6th International working conference on stored product protection, 2: 712-714.

Table 1: Proximate composition of raw and micronized moth bean seeds at varying temperatures (%)

Temp (^{0}C)	Moisture	Crude Protein	Crude Fat	Total Ash	Crude Fibre	Carbohydrate (By Difference)	Energy (Kcal)
Raw	9.88^a	23.50^a	0.81^a	3.09^{ab}	3.92^a	58.79^a	336.45^a
150	9.86^a	23.82^a	0.87^a	3.18^a	3.85^a	58.22^a	335.40^a
250	9.81^a	23.40^a	0.80^a	3.15^{ab}	3.79^a	59.05^a	337.00^{ab}
300	9.76^a	23.40^a	0.93^b	3.06^{ab}	3.82^a	59.03^a	338.09^{bc}
350	9.49^b	23.41^a	0.95^b	3.05^{ab}	3.91^a	59.19^a	338.95^{bc}
400	9.34^b	23.42^a	1.04^b	3.01^b	3.88^a	59.31^a	339.76^c

Values are the mean of three determinations ± SD
Values with different superscript in column differ significantly ($p \leq 0.05$)

Table 2: Changes in functional properties of raw and micronized moth bean seeds at varying temperatures

Temp (^{0}C)	WAC (g/g)	OAC (g/g)	Sedimentation value (ml)	Solubility index (%)	Swelling index	Swelling power
Raw	1.94^a	1.75^a	28^a	24.29^a	4.09^a	1.62^a
150	1.94^a	1.82^a	29^a	24.29^a	4.08^a	1.70^{ab}
250	2.00^a	1.82^a	29^a	25.08^b	3.58^b	1.90^{bc}
300	2.02^a	1.83^a	30^{ab}	25.50^c	3.42^b	1.90^{bc}
350	2.01^a	1.87^a	31^{ab}	25.53^c	3.39^b	1.95^c
400	2.16^b	1.88^a	32^b	25.70^c	3.10^b	1.99^c

Values are the mean of three determinations ± SD
Values with different superscript in column differ significantly ($p \leq 0.05$)

Table 3: Changes in antioxidant properties of raw and micronized moth bean seeds at varying temperatures

Temperature (^{0}C)	Total Phenols (mg/100g)	Total Flavonoids (mg/100g)	DPPH (% Inhibition)
Raw	301.27^a	22.00^a	23.37^a
150	300.26^a	21.90^a	22.85^{ab}
250	292.79^b	21.13^b	22.52^b
300	288.16^c	19.18^c	21.68^c
350	269.80^d	17.98^d	20.05^d
400	258.80^e	16.15^e	18.86^e

Values are the mean of three determinations ± SD
Values with different superscript in column differ significantly ($p \leq 0.05$)

Table 4: Changes in anti-nutritional factors of raw and micronized moth bean seeds at varying temperatures

Temperature (^{0}C)	Phytic acid (mg/100g)	Trypsin inhibition activity (TIU/ g)	Tannins (mg/100g)	Saponins (%)
Raw	146.57^a	171^a	3.77^a	3.03^a
150	141.93^b	170^a	3.79^a	3.02^a
250	141.20^b	168^{ab}	3.58^a	3.01^a
300	139.65^c	165^{ab}	3.38^a	2.82^a
350	139.40^c	165^{ab}	2.38^b	2.27^b
400	131.80^d	160^b	2.18^b	2.02^b

Values are the mean of three determinations ± SD
Values with different superscript in column differ significantly ($p \leq 0.05$)

Table 5: Changes in pasting properties of raw and micronized moth bean seeds at varying temperatures

Temperature (^{0}C)	Break down (RVU)	Final Viscosity (RVU)	Set back (RVU)	Pasting temperature (RVU)
Raw	205^a	899^a	246^a	84^a
150	214^a	915^{ab}	254^{ab}	83^a
250	224^{ab}	926^{bc}	269^{bc}	82^a
300	235^b	935^c	280^c	87^a
350	376^c	1045^d	364^d	82^a
400	405^d	1154^e	381^e	81^a

Values are the mean of three determinations ± SD
Values with different superscripts in column differ significantly ($p \leq 0.05$)

Table-6 changes in surface temperature and germination rate in raw and micronized moth bean seeds

Micronization temperature (^{0} C)	Surface temperature (^{0} C)	Germination rate
Raw	39±2^a	97±2^a
150	35±2^a	96±1^a
250	56±1^b	90±1^b
300	69±1^c	88±1^b
350	78±2^d	79±2^c
400	90±2^e	50±3^d

Values are the mean of three determinations ± SD
Values with different superscripts in column differ significantly (p≤0.05)

Figure 1: Changes in in-vitro protein and starch digestibility of raw and micronized moth bean seeds at varying temperatures

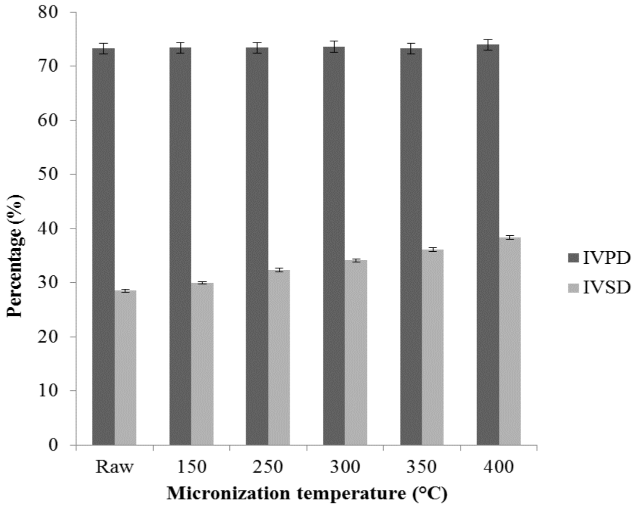

Figure-2 Effect of micronization on the surface micro organisms at varying temperatures