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A preliminary study on the estimation of nutrients and anti-nutrients in *Oedaleus abruptus* (Thunberg) (Orthoptera: Acrididae)

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Natural protein is being overexploited gradually because of a huge population boom. Grasshoppers may be a good food resource to overcome this problem. The present study evaluated the nutritional quality and anti-nutritional factors that is, the proximate composition, vitamins, minerals, fatty acids and amino acid contents of *Oedaleus abruptus*. Results revealed that this species contains about 60% crude protein and nearly 587 kcal/100 g of energy. A total of six minerals were detected, where calcium and magnesium had the highest content. Among the fatty acids, palmitic acid, oleic acid and eicosenoic acid was present in quite good amount and a very high amount of linoleic and linolenic acid was also detected. Eighteen amino acids have been reported, among which the values of threonine, proline and tyrosine were more than 10%. Results of vitamins were also encouraging as a good amount of retinol, ascorbic acid and niacin were detected. The anti-nutritional factors showed a very low value. The results indicated the insects to be a good source of food that could be considered as an alternative to fish and meat and/or a supplement for both human and livestock consumption.

Key words: Nutrients, anti-nutrients, alternative food, *Oedaleus abruptus*.

INTRODUCTION

Most developing and under developed countries are having difficulties to provide sufficient food for their people, and consequently an insufficient intake of protein is leading to malnutrition (Aylward and Morgans, 1995). Sometimes calories are considered to be more important than protein supplements. However, in some instances, authors like Ramos-Elorduy et al. (1984) opined that the deficiency of high quality of protein might be the major problem. In the developing countries of Asia, Latin America and Africa, a gradual “population boom” is an additional problem that is causing over-exploitation of natural food resource and importing more and more of food is quite expensive (Breman and Debrah, 2003). In this scenario, there is an urgent need to conduct experiments for the search of indigenous alternative protein supplements worldwide. Various researchers reported that insects could be an attractive alternative as they are natural food of many vertebrates including human (DeFoliart, 1999).

Among these edible insects, short-horned grasshoppers of the order *Orthoptera* are proving to have immense potential. Some of them are known to contain high amount of protein, fat, carbohydrate and energy (Anand et al., 2008a), and many of them are easy to culture

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in mass scale (Haldar et al., 1999; Anand et al., 2008b), and are suitable for building “acridid farms” as they could yield a huge annual biomass with a very low maintenance cost (Anand et al., 2008b). Hence, farming these grasshoppers in controlled conditions could constantly supply a low cost alternative protein rich diet for human and livestock consumption.

Although in various parts of the world, insects are considered as an important protein source, literature is almost absent in the Indian point of view. In this context, Anand et al. (2008a) did a preliminary study on the proximate composition and mineral content of four acridid species commonly found in India. However, further studies are needed to look for the most suitable species throughout the world. Moreover, for a complete picture on the suitability of any material as food, there is an obvious need to further explore the contents of fatty acids, amino acids, vitamins and anti-nutritional factors. Keeping this in mind, the present study aimed to explore the nutritional value in terms of proximate composition, minerals, vitamins, fatty acids, amino acids, energy and anti-nutritional factors of a multivoltine grasshopper species: *Oedaleus abruptus* (Thunberg) from family Acrididae under the order *Orthoptera*.

MATERIALS AND METHODS

Collection and preparation of sample species

Nine hundred eighty seven (987) adult individuals of *O. abruptus* were collected from the nearby grasslands and croplands of Santiniketan (23° 39'N, 87° 42'E), Birbhum, West Bengal, India by sweeping technique, using standard insect nets. They were freeze killed prior to the estimation of wet body weight. Then they were oven dried (Indian instrument manufacturing company, Kolkata, India) at 60°C until the dry body weight became constant. Moisture was estimated using the formula:

Moisture (M%) = 100 - [(dry weight × 100)/wet weight]

Legs and wings were removed from the dried matter to get rid of excess chitin, and after that they were crushed to powder form before subjected to further analyses.

Estimation of proximate composition of the acridids

Nutrient composition was estimated by standard procedures according to Helrich (1990) on dry matter basis. Percentage nitrogen content was estimated by Micro-Kjeldahl method using Tecator Kjeltec system (Sweden). Nitrogen content was converted into crude protein (%) using the factor $N \times 6.25$. Crude fat was estimated by ether extraction method, using soxhlet apparatus (Indian instrument manufacturing company, Kolkata, India). Percentage content of crude fibre was chemically determined by repeated treatment of dilute H_2SO_4 and dilute NaOH, and washed with distilled water. Ash contents were obtained by keeping the dried samples in a muffle furnace (Indian instrument manufacturing company, Kolkata, India) at 550°C for 6 h. Nitrogen free extract (NFE) and carbohydrate content (%) was calculated by difference method. Energy was estimated using oxygen bomb calorimeter (Rajdhani Scientific Instruments Co., New Delhi, India) and expressed as kcal/100 g.

Estimation of minerals, amino acids, fatty acids and vitamins

Mineral contents such as Ca, Fe, Zn, Mg, Cu and Mn were estimated by Varian Techtron atomic absorption spectrophotometry (Victoria, Australia) using standard reference chemicals according to Anand et al. (2008b). The extraction of fatty acids from the acridid samples and their methyl ester preparation were performed according to the method of Bettelheim and Landesberg (1997). The purified methyl esters of fatty acids were subjected to Agilent 6890N gas chromatographic (Palo Alto, USA) analysis. Percent compositions of the samples were computed from the GC peak areas. Amino acids were analyzed according to Ghosh et al. (1995) and Wang et al. (2007). Samples were first hydrolyzed with 6 N HCl containing 1% phenol for 22 h at 105°c, then the amino acid contents were determined by a PICO.TAG system according to PICO.TAG operation manual (Waters, USA) at 38°C. Quantitative estimation of tryptophan could not be done by afore mentioned method, so it was determined by AIMIL Photochem colorimeter (Photochem Electric Instruments, Jodhpur, India) following the strategies proposed by Fischl (1960). Vitamins like retinol, ascorbic acid, niacin, riboflavin and thiamin were also estimated by colorimetric method according to Helrich (1990).

Estimation of anti-nutritional factors

Anti-nutritional factor of phenolic polymers like tannin was determined chemically with vanillin-HCl reagent and catechin solution according to Gupta et al. (1988). Content of oxalate was determined by simple titration using methyl red as indicator following the procedures proposed by Gupta et al. (1988). Titration was again used to measure the amount of phytin phosphorus using ferric chloride ($FeCl_3$) as indicator according to Agbede and Aletor (2004). Phytin content was calculated with a multiplication of the value of phytin phosphous by 3.55 (Agbede and Aletor, 2004).

Statistical analysis

Data are presented as means ± standard deviation (SD). The experiments were carried out in three replicates. For completely randomized designs, all the data were statistically analyzed by one-way analysis of variance (ANOVA) using ‘S Plus’ (version 4.0) software. Results were subjected to Duncan’s multiple range test (DMRT) to understand the significant difference between the data within a sample group.

RESULTS

Proximate composition revealed about 70% moisture content in *O. abruptus* (Figure 1). The species had a quite high level of crude protein (about 60%). On the other hand, the percentage of crude fat, crude fiber and ash were reasonably low (less than 10%), however, nitrogen-free extract (NFE) and carbohydrate contents were of moderate level (nearly 20 and 30%, respectively). The energy content of the species was 587.48 ± 2.14 kcal/100 g (mean ± SD) and the protein to energy (P/E) ratio was 100.97 ± 1.08 mg protein/kcal (mean ± SD), and these were quite high in this acridid (data not presented in graphical or tabulated forms).

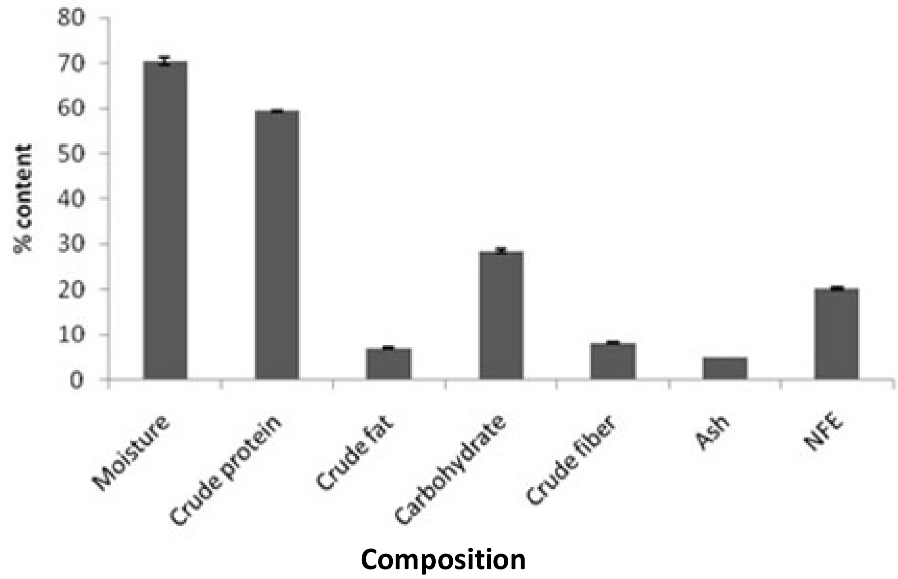

Figure 1. Proximate composition of *O. abruptus*. Data are presented as means ± SD.

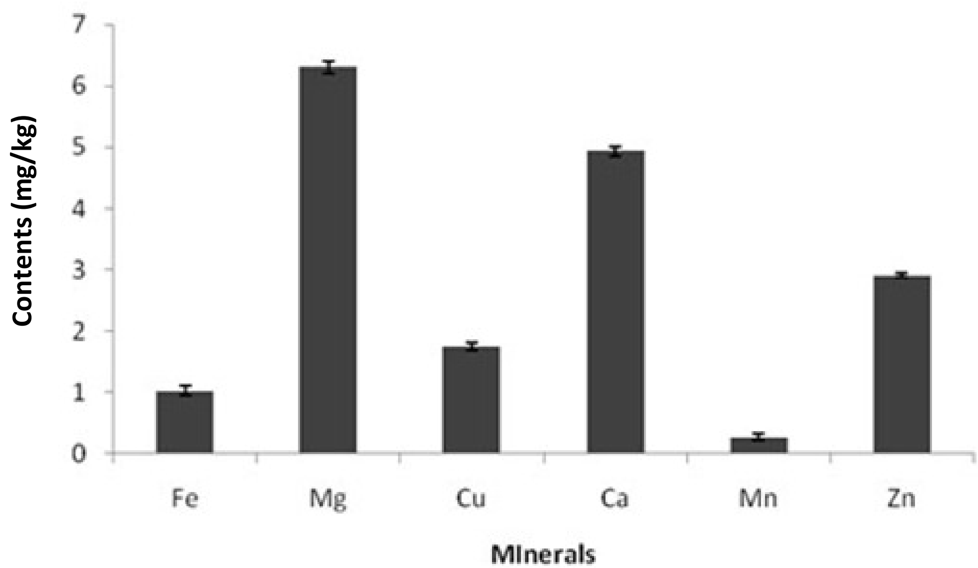

Figure 2. Mineral contents (mg/kg) in the body tissues of *O. abruptus*. Data are presented as means ± SD.

A total of six minerals were estimated. Among them, magnesium (Mg) was present in the highest amount (more than 6 mg/kg), followed by calcium (Ca) and zinc (Zn) (Figure 2). Iron (Fe) and copper (Cu) contents were moderately low (1.03 and 1.74 mg/kg, respectively), and manganese (Mn) was found in the lowest amount (about 0.27 mg/kg) (Figure 2). Eight fatty acids were detected in *O. abruptus* (Table 1). The content of linolenic acid was found to be highest (39.45%), followed by linoleic acid (15.34%). The content of palmitic acid, oleic acid and eicosenoic acid were present in good amounts (4.55, 4.27 and 7.91%, respectively), but myristic acid was extremely low (0.37%).

Among the 18 amino acids detected in this study, threonine, proline and tyrosine were present in very high amount (more than 10%), whereas aspartic acid and cysteine were detected to be very low (0.65 and 0.48%, respectively). Five vitamins, namely: retinol, thiamine, riboflavin, niacin and ascorbic acid were detected in the body tissues of the selected acridid (Table 1). Thiamine

Table 1. Fatty acid, amino acid and vitamin contents of the acridid species.

Property	*O. abruptus*
Fatty acid (%)	
Myristic acid	0.37
Palmetic acid	4.55
Oleic acid	4.27
Stearic acid	1.12
Arachidonic acid	1.62
Eicosenic acid	7.91
Linoleic acid	15.34
Linolenic acid	39.45
Amino acid (%)	
Aspartic acid	0.65
Glutamic acid	4.51
Serine	5.08
Glycine	7.92
Histidine	7.58
Arginine	8.03
Threonine	15.99
Alanine	3.02
Proline	15.34
Tyrosine	10.31
Valine	6.24
Methionine	1.98
Cysteine	0.48
Isoleucine	1.45
Leucine	5.21
Phenylalanine	4.10
Lysine	2.04
Tryptophan	2.25
Vitamin (mg/100 g)	
Retinol	4.64
Thiamine	0.50
Riboflavin	1.01
Niacin	6.00
Ascorbic acid	6.25

was found to be present in very low amount (0.50 mg/100 g), whereas retinol, niacin and ascorbic acid were quite high (4.64, 6.00 and 6.25 mg/100 g, respectively).

Four anti nutritional factors, tannin, oxalate, phytin bound phosphorus (phytin P) and phytin were analyzed for the acridid. All of them were found to be present in very low amount. Among the analyzed anti-nutritional factors, tannin was present in the highest amount, which was around 2.45%, followed by oxalate (around 0.6%). On the other hand, phytin P and phytin was around 0.025 and 0.085%, respectively (data not presented in graphical or tabulated forms).

DISCUSSION

Insects are rich in protein, and many scientists have reported the protein qualities of insects from different parts of the world. Chen and Feng (1999) reported that in China, nearly hundred kinds of edible insects had been analyzed till date. According to them, at the egg, larva, pupa and adult stages, the raw protein content generally is 20 to 70%, with average higher protein content in the order *Hemiptera* (around 42 to 73%). Banjo et al. (2006) analyzed fourteen edible insect species from south-western Nigeria and found that twelve of them have protein content of 20% and above, where the highest amount of protein (29.62%) was observed in *Analeptes trifasciata* (Fabricius, 1775). Ramos-Elorduy (1998) worked on the edible insects of Mexico and found those insects to have very high crude protein content. According to this study, the highest amount of protein was found in the red legged locusts (75.30%), whereas most of them contained around 40 to 60% crude protein. Our results revealed that the selected acridid species was also high in protein content (about 60%) and have a very high P/E ratio (more than 100 mg of protein/kcal) which proved this group of insects to be a good protein and energy supplement.

Protein is composed of mainly twenty amino acids. Among these, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, valine and histidine are essential for human adults (Young, 1994). Analysis of more than hundred edible insects in China showed that they contain all the necessary amino acids (Chen et al., 2009). Wang et al. (2007) reported a total of seventeen amino acids in the Chinese acridid *Acrida cinerea* (Thunberg, 1815). Comparing their data with the acridid species of our interest revealed that nine of those seventeen amino acids were present in higher amount in *O. abruptus*. Two of them were almost similar and six were present in lower amounts. Although edible insects are good in amino acid content, reports indicated that some of them are deficient in some amino acids. As an example, studies on other edible insects like mormon cricket (*Anabrus simplex* Haldeman 1852) and house cricket (*Acheta domesticus* Linnaeus 1758) showed that they were deficient in methionine (DeFoliart et al., 1982; Finke et al., 1985; Nakagaki et al., 1987). Similarly, as reported by Landry et al. (1986), the essential amino acids of six lepidopteran species were also found to be deficient in the same amino acid. Our result corresponds with the above stated reports as the content of methionine, cysteine and lysine were found to be quite low (below 3%). Therefore we support the conclusion of Wang et al. (2007) that insects might be unsatisfactory as the only source of dietary protein due to limiting amino acids, but would be extremely beneficial as a supplement.

Fat is one of the major nutrients for any living organism. Authors like Feng et al. (2000a, b), Chen and Feng (1999) and He et al. (1999) reported that insects are higher in fat content at the larva and pupa stages, while adults contained relatively lower amount of fat. Among grasshoppers, the fat content of adult *Oxya chinensis* (Thunberg 1815) was found to be only 2.2% (Chen et al*.,* 2009); likewise Melo et al. (2011) reported that the same of *Sphenarium purpuracens* (Charpentier 1841-45) was also quite low (5.75%). Our study also showed lower fat content (nearly 7%) in the experimental acridid. Though the fat content of most of the edible insects is lower than other animal food sources but still they could play an important role for human nutrition.

Chen and Feng. (1999) reported that unlike other animal fat, edible insects have a higher essential fatty acid content which is necessary for human body, and hence edible insect fat has a high nutritive value. The results of fatty acid content of our species when compared to that of the variegated grasshopper *Zonocerus variegatus* (Linnaeus 1758), it was revealed that *O. abruptus* contained a total of eight fatty acids compared to five of *Z. variegatus* (Adeyeye, 2011), however in the fatty acids *Z. variegatus* is richer in palmitic acid (13.5%), stearic acid (5.7%), oleic acid (4.27%) and linoleic acid (9.4%). On the other hand, Wang et al. (2007) also reported the existence of eight fatty acids in *A. cinerea*. The palmitotelic and lauric acid that they found was not observed in our results; instead we detected arachidonic and eicosenoic acids. According to Chen and Feng (1999), edible insects are rich in protein and fat but not so rich in carbohydrate content; however the authors also added that carbohydrates of edible insects differ from species to species ranging from 1 to 10% in China. On the other hand, larva of *Cirina forda* (Westwood 1849) which is relished as a human food in Nigeria contains about 38% of carbohydrate (Akinnawo and Ketiku, 2000). Our species of interest also showed a moderately high amount of carbohydrate content (about 30%).

The gross amount of energy content of the food comes mainly from protein, fat and carbohydrate. As edible insects are quite rich in these three ingredients, in general they mostly contain quite high energy. For example, Sogbesan and Ugwumba (2008) reported energy value of 587.13 kcal/100 g in the sexual forms of the African termite (*Macrotermes subhyalinus* Rambur 1842), while Oyarzun et al. (1996) reported 688 kcal/100 g of energy in the arboreal termites, *Nasutitermes* spp. Braide et al. (2010) reported a calorific value of 433.8 kcal/100 g for the caterpillar *Bunaea alcinoe* (Stoll 1780) in Nigeria. Literature survey showed that the energy content of most of the edible insects varied from nearly 217 to 777 kcal/100 g (Ramos-Elorduy, 2008). Energy content of *O. abruptus* also supported this observation which was more than 550 kcal/100 g.

Edible insects have been found to contain minerals also. Among them, Na, K, Ca, Zn, Fe and Mg are prevalent in literature (Akinnawo and Ketiku, 2000; Ramos-Elorduy et al., 2002; Ojewola and Udom, 2005; Ekop et al., 2010). Ramos-Elorduy (2005) reported that edible insects under the orders *Orthoptera*, *Lepidoptera* and *Hymenoptera* have much lower variation in mineral contents. The author continued stating that these insects are mostly low in Na, and sometimes high in Ca, Zn, Fe, K and Mg. Our results supported this view as Mg is present in the highest amount (more than 6 mg/kg), followed by Ca and Zn. Fe was observed in a moderate amount.

Though Na and K were not detected, a small trace of Mn (less than 1 mg/kg) was obtained. There were also reports that indicated edible insects to be rich in vitamin contents too. For example, Kodondi et al. (1987) reported rich vitamin content in some edible caterpillar in Zaire. Ramos-Elorduy et al. (1988) observed that edible insects are rich in vitamin B group such as thiamine, riboflavin and niacin. The presence of these three vitamins was also observed in case of *O. abruptus*, where niacin was present in high amount but the rest of the two were a little lower. Furthermore, two other vitamins, that is ascorbic acid and retinol were also present in quite high amount.

Up to this level, it has been discussed that the grasshopper of our interest is nutritionally comparable to the other available food insects. However, it is also important to know whether this species can compete with the other conventional protein sources like fish, meat, soybean etc. Surveying literature in this regard showed that the protein content of *O. abruptus* is higher than that of grain and soybean and almost comparable to fishmeal but lower than meat (Hasanuzzaman et al., 2010; Baker et al., 2011; Adeniyi et al., 2011). Hasanuzzaman et al. (2010) and Adeniyi et al. (2011) also reported crude fat content of fish and meat varies between 4 and 9% which is also similar to that of *O. abruptus*. On the other hand, Baker et al. (2011) observed crude fat of soybean meal to be about 1 to 1.5%, quite lower than that obtained in the present study.

According to Koumi et al. (2011), the carbohydrate content of fish and soybean could be around 23 and 27%, respectively, again very close to the value of the carbohydrate content of nearly 30% observed in the grasshopper of the this study. Among vitamins, this insect has a higher proportion of thiamine than wheat germ, peas, bread, beans, rice, soybean, milk and egg; whereas for riboflavin it is richer than aged cheese, bread, beef liver, milk, eggs, yogurt, pork, spinach, trout and chicken and niacin is in greater amount than bread, grouper fish, peas, beans, corn, wheat, milk, bacon and eggs, and in general is poor in terms of ascorbic acid (Ramos-Elorduy and Pino, 2001). On the other hand, mineral contents were quite lower in comparison with meat, fish, soybean or corn (Batal et al., 2010; Koumi et al., 2011; Adeniyi et al., 2011).

Anti-nutritional factors are usually present in plant materials but many phytophagous insects have been identified to retain these materials in quite a good amount (Berenbaum, 1993). Hence, it is recommended to analyze these anti-nutrients if a phytophagous insect is being considered as food, though this kind of study is not much frequent in the literature. Among the anti-nutritional factors, tannin, oxalate and mineral bound phytate (that is, phytin P and phytin) were detected in the grasshopper of our interest. Adeduntan (2005) reported tannin percentage in grasshoppers of Ondo state of Nigeria to be 1.05%, which is a little lower than the amount (about 2.5%) observed in *O. abruptus.* However, this amount is itself quite lower than cereal grains and other plant food materials for example in tannin content *Pachira glabra* (Bombacaceae) (3.53%) and in *Afzelia africana* (Fabaceae) (2.65%) (Ogunlade et al., 2011; Hassan et al., 2011). On the other hand the percentage of phytate was reported to be quite high in grasshoppers by Adeduntan (2005), which was about 1.1% compared to quite lower amount of nearly 0.025% of phytin P and 0.09% of phytin in the present study.

We could not find any literature describing the oxalate content of any grasshopper species but it has been reported in some edible beetles such as adults of *Oryctes monoceros* (Olivier 1789) and *Cirina forda* (Omotoso, 2006; Ifie and Emeruwa, 2011). Both of these papers revealed that those beetles contain less than 0.005% oxalate, much lower than the insect of our interest. However, this amount is also under the tolerance limit because much higher amounts have been observed in various plant food materials (Kalita et al., 2007).

Conclusion

Nutrient composition of the present study reveals that *O. abruptus* is a nutritious insect with high protein and energy. Amino acids, fatty acids, and vitamins were present in high amount. Though anti-nutritional factors were present, they were found to be in very low amount and within the tolerance limit. It has been also discussed that this insect is nutritionally comparable to fish, soybean and various other conventional food ingredients in terms of proximate composition and/or vitamins. Thus it can be safely stated that acridids could be a good alternative source of not only protein and energy but also important as source of vitamins. This proves that an effort should be made so that these grasshoppers could be mass reared in controlled conditions in farms as a mini-livestock. This insect could be used as a supplement in formulated feed for various animal species such as fish, chickens, ostriches, pigs or some pets and for humans, it could be used in the preparation of snacks, creams, salads, cookies, casseroles, flours, breads, desserts, sauces, seasonings, soups, etc.

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ABBREVIATIONS

NFE, Nitrogen free extract; **P/E,** protein to energy ratio.

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