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Biochemical Analyses and Nutritional Content of Four Castes of Subterranean Termites, *Macrotermes subhyalinus* (Rambur) (Isoptera: Termitidae): Differences in Digestibility and Anti-nutrient Contents among Castes

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Abstract

Termite is used in many parts of the world but little is known about their biochemical properties and nutritional value as food. Protein digestibility and anti-nutrients of four castes (queen, worker, soldier and winged (alate)) of subterranean termite, *Macrotermes subhyalinus* were determined to evaluate the protein bioavailability and confirm nutritional value as food for human and animal consumption. Physico-chemical properties of the oil of each caste were also determined to reveal the potential uses of the oil and its stability in storage using standard analytical methods. The queen was the most easily digestible (84.72%) of the castes while the soldier was the least with a digestibility value 81.10%. The queen was significantly more digestible than the other castes. In general, all the four castes have low anti-nutrient (oxalate, phytate and tannin) values. Among the castes, worker has significantly higher anti-nutrient levels than the other castes. In general, worker and soldier have relatively higher digestibility values and significantly higher anti-nutrient contents as well as relatively higher values for most of the physico-chemical parameters tested, suggesting that they are the least edible caste. Overall, the results showed that all four castes of *M. subhyalinus* have very low levels of anti-nutrients tested for, are easily digested and produce edible oil. These findings confirm that all four castes of *M. subhyalinus*, and possibly other termite species, are safe for human and animal consumption, and therefore recommended for inclusion in human foods and livestock feeds.

Keywords: physico-chemical properties, anti-nutrient, *In vitro* digestibility, *Macrotermes subhyalinus*

1. Introduction

Insects have served as a food source for people for thousands of years. Balanced diet is quite expensive in developing countries, especially for poor people. This therefore leads malnutrition; because the only readily available diet for the masses is carbohydrates, due to their abundance and low cost (Defoliart, 1989). To supplement and provide other components of food like proteins, vitamins and minerals, insects are popular foods in many developing regions of Central and South America, Africa, and Asia (Fasoranti & Ajiboye, 1993). There are over a thousand or more species of insects in more than 370 genera and 90 families that are or have been used for food somewhere in the world either for man or animals, especially in Central and Southern Africa, Asia, Australia and Latin America (Defoliart, 1989). Some of these insects eaten as food include termites, crickets, grasshoppers, locusts, beetles, ants, bee brood, moth and beetle larvae, bamboo worm etc. Insects are high in proteins ranging between 60%-70% (Mba, 1986), energy and various vitamins and minerals, and can form 5 – 10% of the annual animal protein consumed by certain indigenous peoples (Redford & Dorea, 1984; Defoliart, 1989). In Nigeria, larvae of palm weevils and silk worms, grasshoppers, locusts, termite (alates), crickets are in use as foods. Termite is the most widely accepted insect as food in Nigeria (Fasoranti & Ajiboye, 1993).

Termites, popularly but erroneously called white ant are small insects, found chiefly in the tropical areas (Umeh, 2000). They are serious pests of wood and wood products (Hornby, 1989). Termites are known to be of economic importance in the tropics. They mix soil matter and thereby increase the fertility (Umeh, 2003). However the affinity of termites for cellulose materials makes them deleterious to many plant species including timber, agricultural products and buildings. Feeding on termite will in addition to nutritional benefit for man and animal, serve as biological means to reduce the population build-up in the environment and thus suppress damage of wood and wood products.

Digestibility of protein determines protein quality. Not all proteins consumed are digested, assimilated and utilized maximally. This may be as a result of inherent differences in the nature of food protein, presence of some anti-nutritional factors or processing conditions that alter the enzymatic activities (Sandrez-Vioque et al., 1999). The shelf-life of any product of animal or plant origin depends on the nature of its chemical components. In this study, biochemical analysis was carried out to determine the level of protein digestibility, some anti-nutrient components and physico-chemical properties of the oil of the four castes of the African termite, *Macrotermes subhyalinus.* Due to the reported differences in the nutritional physiology, chemical content and behavioural ecology among termite castes (Roisin, 2000; Suarez & Thorne, 2000), it was hypothesized that workers and soldiers will have relatively lower nutritional value (i.e. lower digestibility and higher anti-nutrient content) than the other castes.

2. Materials and Methods

2.1 Collection of Termite Castes

The queen, worker and soldier castes used in this study were obtained through excavation of termitaria from various locations in Akure, using digger, hoe, cutlass, broom and containers. Workers and soldiers were on occasions also collected by a method described by Imhasly and Leuthold (1999). Winged (alate) termite was collected during seasonal flight.

2.2 In-vitro Protein Digestibility

In-vitro digestibility test was carried out according to standard method (Hsu et al., 1977). An aliquot of 50 ml of an aqueous suspension of each sample (6.25 mg sample/ml) in distilled water was adjusted to pH 8.0 with 0.1M HCl and/ or 0.1M NaOH while stirring in a 37^{o}C water bath. The multi-enzymes solution used which comprised 1.6mg trypsin, 3.1mg chymotypsin and 1.3 mg peptidase per ml was adjusted to pH 8.0 with 0.1M HCl and/ or 0.1M NaOH and maintained in an ice bath. Glass distilled water was used for the preparation of all the solutions. A 5 ml of the multi-enzyme solution was added to each sample suspension with constant stirring at 37^{o}C. The pH of each suspension was recorded 10min after the addition of the multi-enzyme solution. The pH value obtained was substituted in regression equation of Hsu et al. (1977) to calculate protein digestibility of each caste.

2.3 Anti-nutrient Content

Phytate, oxalate and tannin contents of the queen, worker, soldier and winged were determined. Phytate was determined according to the method of Wheeler and Ferrel (1971). The determination of oxalate was done according to the procedure of Day and Underwood (1980). The procedure of Makkar et al. (1993) was followed for the gravimetric determination of tannins.

2.4 Physico-chemical Properties

The oil extracts was obtained using Soxhlet apparatus. The physico-chemical properties (density, refractive index, peroxide value, acid value, FFA, iodine, saponification value and unsaponification matter) were determined. The methods of AOAC (1990) were used for iodine value, peroxide value, saponification value, unsaponifiable matter, acid value, density and refractive index. Free fatty acid was determined according to the method of Pearson (1976).

2.5 Analysis

Analysis of variance (ANOVA) was carried out on the data obtained and the means separated at 5% level of significance with New Duncans' Multiple Range Test (NDMRT).

3. Results

3.1 Protein Digestibility

In vitro multi-enzyme digestibility of protein of four castes of *M. subhyalinus* revealed significant differences between the castes ($X^2 = 21.323$, df = 3, $P < 0.005$). The result is shown in Figure 1. The digestibility value of the four castes ranged from 81.10% to 84.72%. The highest percent digestibility value (84.72%) was recorded in queen caste. Worker and winged castes had 82.37% and 83.41% digestibility, respectively. Soldier had the least value of 81.10%.

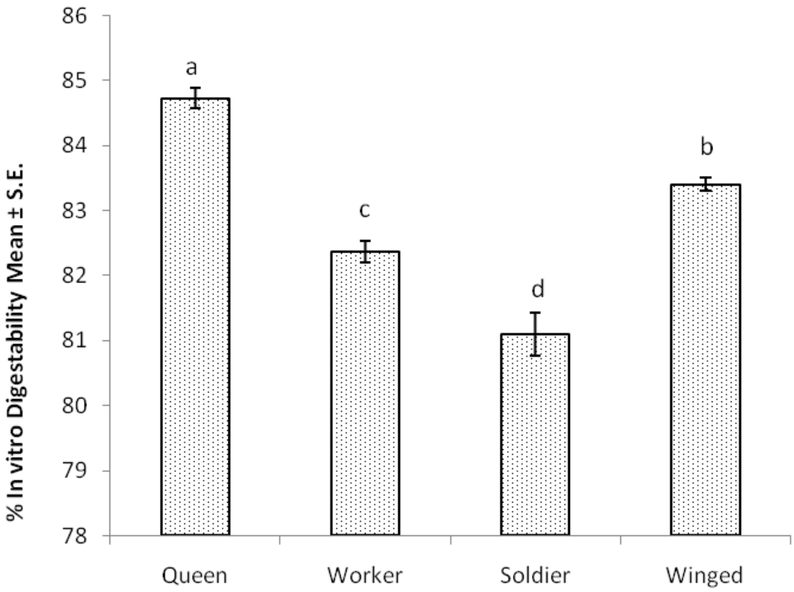

Figure 1. *In vitro* digestibility of protein of four castes of the subterranean termite, *Macrotermes subhyalinus.* Error bar indicates SE

3.2 Anti-nutritional Contents

The levels of anti-nutrients in termites are presented in Table 1. Oxalate content of the termites ranged between 0.005% in winged and 0.012% in worker. Queen, worker, soldier and winged had 0.018%, 0.130%, 0.029% and 0.016% phytate, respectively. The tannin content were 4.7 $\times 10^{-8}$mgTA/100g, 5.5 $\times 10^{-8}$mgTA/100g, 4.8 $\times 10^{-8}$mgTA/100g and 4.8 $\times 10^{-8}$mgTA/100g respectively. Significant differences were recorded among the castes in oxalate (F = 229.430, df = 3, P = 0.05), phytate (F =278.012, df = 3, P = 0.05), and tannin (F =120.250, df = 3, P = 0.05) values (Table 1). Worker had significantly higher oxalate, phytate and tannin contents than the other castes.

Table 1. Antinutrient content of four castes of the termite, *Macrotermes subhyalinus*

Termite caste	Oxalate (%)	Phytate (%)	Tannin (mgTA/100g)
Queen	0.0068^{c}±0.00	0.0175^{c}±0.00	4.7 $x10^{-8b}$±0.00
Worker	0.0117^{a}±0.00	0.1300^{a}± 0.00	5.5 $x10^{-8a}$±0.00
Soldier	0.0094^{b}±0.00	0.0286^{b}±0.00	4.8 $x10^{-8b}$±0.00
Alate	0.0054^{d}±0.00	0.0156^{c}±0.00	4.8 $x10^{-8b}$±0.00

Means (±S.E.) followed by the same letter within the columns are not significantly different. (P > 0.05) using New Duncan's' Multiple Range Test.

3.3 Physico-chemical Properties of the Oils

The oil obtained in queen, worker, soldier and winged castes is 27.37%, 5.55%, 4.80% and 47.03% respectively. The results of the physical and chemical analyses carried out on the oils of termite castes are presented in Table 2. Refractive index at 20^{o}C ranged from 1.462 in queen oil to 1.465 in winged. The specific gravity of the oil extracts of worker, soldier and queen was not detected because the oils solidify at room temperature, while that of the winged was 0.9222. Soldier had the highest acid value (73.39mgKOH/g) while the lowest value (42.95mgKOH/g) was recorded in queen caste. The peroxide values of the oil of the various samples ranged between 0.44 and 2.49mg/KOH/g. Iodine value ranged between 118.45 and 208.07. The highest saponification value of 338.58mg/KOH/g was obtained in soldier oil while 108.45mg/KOH/g was recorded in winged. Unsaponifiable matters in percentage ranges from 1.95% in oil of soldier to 2.25% in oil of winged. Soldiers had the highest acid value (as oleic acid) of 3.67mgKOH/g.

Table 2. Physicochemical properties of the oil of four castes of subterranean termite, *Macrotermes subhyalinus*

Termite castes	Refractive index (20°C)	Density (29°C)	Colour	Acid value (mgKOH/g)	Peroxide value (mgKOH/g)	Iodine value (mg/g)	Saponification value (mgKOH/g)	Free fatty acid (mgKOH/g)	Unsaponifiable matter (%)
Queen	1.46^{b}± 0.00	N.D	Brownish	1.08^{c} ± 0.01	0.44^{d} ± 0.00	138.21^{c}± 0.00	2.15^{d} ± 0.00	157.18^{c} ±0.00	2.22^{b} ±0.00
Soldier	1.46^{b}± 0.00	N.D	Brownish	1.83^{a} ±0.01	2.49^{a} ±0.00	208.07^{a}±0.00	260.69^{b} ±0.00	3.67^{a} ±0.00	2.10^{c} ±0.00
Worker	1.46^{b} ±0.00	N.D	Golden yellow	1.81^{a} ±0.01	2.20^{b} ±0.01	143.23^{b}±0.00	338.58^{a} ±0.00	3.63^{b} ±0.00	1.95^{d} ±0.00
Alate	1.47^{a} ±0.00	0.92±0.00	Golden yellow	1.46^{b} ±0.01	0.78^{c} ±0.01	118.60^{d}±0.00	108.45^{d} ±0.00	2.95^{c} ±0.00	2.25^{a} ±0.00

Means (±S.E.) followed by the same letter within the columns are not significantly different. (P > 0.05) using New Duncan's' Multiple Range Test. *N.D = Not Detected.

4. Discussion

The results show that protein in the four castes of *M. subhyalinus* is highly digestible; however protein digestibility varies by castes. The queen has the highest digestibility value, while workers and soldiers are the least digestible. The observed difference in the digestibility of the four castes is of biological significance in terms of ecological niche. The queen caste functions mainly in production of eggs and is nourished for that purpose. The recorded highest digestibility value obtained in queen caste may be as a result of abundance of eggs in the body and the nourishment received from the worker caste. The alate caste is the potential reproductive in the colony and the body would equally be enriched with nutrient in preparation for the next flight and establishment of new termitaria. Workers accumulate food out of which other members are fed and required to have reserves in the body. Soldier works with workers in search for food and again defends the colony. In the course of these activities, worker and soldier make use of the nutrients more than others in the colony. This may have contributed to the low protein digestibility obtained in them.

In general, very low anti-nutrients were obtained for *M. subhyalinus* in this study. However, anti-nutrient values varied significantly by caste. Worker and soldier had relatively higher anti-nutrient content than other castes. This can be attributed to the role performed in the colony. Worker functions in exploring food and water sources, regurgitating food for soldiers and parents, caring for eggs and younger siblings, nest building, chemically guarding against fungi and other micro-organisms (Watson et al., 1985; Malaka, 1996). Soldiers are sterile workers. They are responsible for the protection of the nest and often becoming suicidal for the good of the colony (Kamble, 1991). Usually only the workers and soldiers visit feeding sites. The reproductive almost always stays in the nest while the queen remains permanently in royal chamber with king caste (Creffield, 1991). As a result of these activities, worker and soldier feed more than other castes. Termites are phytophagous insect, and plants contain secondary metabolites that hinder or influence the bioavailability of some nutrients in animal system hence the higher anti-nutrients recorded in worker and soldier.

The protein digestibility and anti-nutrient levels of *M. subhyalinus* recorded in this study are lower than those obtained in some food crops. The protein digestibility values of all the castes of *M. subhyalinus* were higher than those reported for some grain legumes like *Phaseolus vulgaris* (78.5%), *Lentil esculenta* (80.3%), *Cajanus cajan* (59.9), *Carnavalia ensiformis* (78.8%) (Hsu et al., 1977; Mba, 1986; Oshodi et al., 1999). The digestibility values recorded in this study are equally higher than those reported for protein concentrates of animal source such as fish meal (74.7%) and casein (78.4%) (Aletor, 2007). The high protein digestibility presents termite as a good protein source for humans and animals. The digestibility of protein and bioavailability of its constituent amino acids are crucial factors that determine protein quality. Not all proteins consumed are digested, assimilated and utilized maximally. This may be as a result of inherent differences in the nature of food protein, presence of some anti-nutritional factors or processing conditions that alter the enzymatic activities (Sandrez-Vioque et al., 1999). Similarly, the anti-nutrient levels of *M. subhyalinus* are lower than those reported for some animal and plant food products. For instance, Akinyede et al. (2005) reported phytic acid and tannin contents of full fat *Dioclea reflexa* to be 0.34 and 0.20% (mg/kg), Tannin and oxalate contents of plantain were 0.28% and 0.82%, respectively (Adeniji et al., 2007), compared to relatively lower tannin (4.7×10^{-8} – 5.5×10^{8}% mgTA/100g) obtained for *M. subhyalinus* in this study. Tannins have been reported to interfere with digestion by displaying anti-trypsin, anti-proline and anti-amylase activity in higher animal (Helsper et al., 1993; Omotosho and Adedire, 2008). Tannins can also form insoluble complex with proteins thereby reducing the absorption as proteins in the system due to phenolic hydroxyl groups which produces unstable radicals (Feeny, 1969). Young and Greeves (1940) reported that phytic acid, oxalic acid and fatty acids form insoluble salts with calcium and therefore interfere with absorption of calcium in the body. In general, the low level of anti-nutrients recorded for the four castes of *M. subhyalinus* confirms that this termite species is safe for human and animal consumption.

The results of the physico-chemical analyses show that the termite castes oils will be good for domestic and industrial purposes as well. It will equally be stable in storage provided the precautions for effective storage of oil (prevention of water, light and oxygen) are followed. The data obtained in this study support our hypothesis that all the castes are safe for human and animal consumption and of immense nutritional benefit to the body due to high protein digestibility.

References

Adeniji, T. A., Sanni, L. O., Barimalaa, I. S., & Hart, A. D. (2007). Anti-nutrient and heavy metals in some new plantain and banana cultivars. *Nigerian Food Journal*, *25*(1), 171-177. http://dx.doi.org/10.4314/nifoj.v25i1.33666

Akinyede, A. I., Amoo, I. A., & Eleyinmi, A. F. (2005). Chemical and functional properties of full fat and defatted *Dioclea reflexa* seed flours. *Journal of Food, Agriculture and Environment, 3*(2), 112-115.

Aletor, O. (2007). Physico-chemical and nutritive characteristics of some conventional and alternative protein concentrates. Ph.D. Thesis, Department of Chemistry, Federal University of Technology Akure. pp 112-113.

Amoo, I. A., & Moza, L. (1991). Extraction and physico-chemical properties of oil from *Bauchinia racemosa* seeds. *La Rivista Italliana Delle Sostanze Grasse*. LXXVI Settembre.

AOAC (Association of Official Analytical Chemists). (1990). Official Methods of Analysis, 15th Ed. Association of Official Analytical Chemists. Washington DC.

Day, R. A., & Underwood, A. L. (1980). *Quantitative analysis*. Prentice-Hall. *Inc., Eablewood Cliffs M. J.,* 586-588.

Defoliart, G. R. (1989). The human use of insect as food and as animal feed. *Bulletin of the Entomological Society of America, 35*, 22-35.

Fasoranti, J. O., & Ajiboye, D. O. (1993). Some edible insects of Kwara State Nigeria. *American Entomologist, 93*, 113-116.

Feeney, P. P. (1969). Inhibitory effect of oak leaf tannins on the hydrolysis of proteins by trypsin. *Phytochemistry, 8*, 2119-2126. http://dx.doi.org/10.1016/S0031-9422(00)88169-8

Helsper, J. P. F. G., Hoogendijk, J. M., VanNorel, A., & Burger-Meyer, K. (1993). Antinutritional factors in faba beans (*Vicia faba*) as affected by breeding toward the absence of condensed tannin. *Journal of Agriculture and Food Chemistry, 41*(7), 1058-1061. http://dx.doi.org/10.1021/jf00031a008

Horn, D. J. (1989). Ecological Approach to Pest Management 285. London, Elsvier.

Hsu, H. W., Varak D. L., Satterlee L. D., & Miller G. A. (1977). A multienzymes technique *of International workshop on wood Preservation at FRIN, Ibadan, Nig*. 7th-8yh Nov. For estimating protein digestibility. *Journal of Food Science, 42*, 1269-1273. http://dx.doi.org/10.1111/j.1365-2621.1977.tb14476.x

Imhasly, P., & Leuthold, R. H. (1999). Intraspecific colony recognition in the termites *Macrotermes subhyalinus* and *M. bellicosus* (Isoptera: Termitidae). *Insectes Sociaux., 46*(2), 35-38

ISO. (1998). Animal fats and Vegetable oil: determinationof saponificationvalue, ISO, 3657, McGraw Hill, p. 1-2.

Makkar, H. P. S., Blummed, M., Bowwy, N. K., & Becken, K. (1993). Determination of tannins and their correlation with chemical and protein precipitation method. *Journal of Food Science and Agriculture, 61*, 161-185. http://dx.doi.org/10.1002/jsfa.2740610205

Manzoor, M., Farooq, A., Tahira, V., & Bhanger, M. (2007). Physico-chemical characterization of *Moringa concanensis* seeds and seed oil. *Journal of the American Oil Chemists' Society, 84*(5), 413-419.

Mba, A. U. (1986). Chemical composition of some local sources of protein foods for man. *Nigerian Journal of Nutrition Sciences, 1*, 142-147.

Ngoddy, P. O., & Ihekoronye, A. I. (1985). Integrated Food Science and Technology. International College Edition. Macmillan publisher Ltd, pp 250-365.

Omotoso, O. T., & Adedire, C. O. (2008). The potential domestic and industrial uses and the levels of certain anti-nutrients in the developmental stages and the quality of oil extracts of palm weevil, *Rhynchophorus phoenicis* F. (Coleoptera: Rhynchophorinidae). *Pakistan Journal of Nutrition, 6*(3).

Oshodi, A. A., Ogungbenle, H. N., & Oladimeji, M. C. (1999). Chemical composition, nutritionally valuable minerals and functional properties of benniseed (*Sesamin radiation*), pearl millet (*Permisetuin typhoides*) and quinoa (*Chenopodium quinoa*) flours. *International Journal of Food Science and Nutrition, 50*, 325-331. http://dx.doi.org/10.1080/096374899101058

Pearson, D. (1976). Chemical Analysis of Foods. 7th ed. Churchill Livingstone, London. pp. 7-11.

Phelps, R. J., Struthers, J. K., & Moyo, S. J. L. (1975). Investigations into the Nutritive Value of *Macrotermes falciger* (Isoptera: Termitidae). *Zoologica Africana, 10*, 123-132.

Redford, K. H., & Dorea, J. G. (1984). The Nutritional Value of Invertebrates with Emphasis on Ants and Termites as Food for Mammals. *Journal of Zoology, 203*, 385-393. http://dx.doi.org/10.1111/j.1469-7998.1984.tb02339.x

Roisin, Y. (2000). Diversity and evolution of caste pattern. *Termites: Evolution, Sociality, Symbioses, Ecology* (ed. By T. Abe, D. E. Bignell and M. Higashi), pp. 95-120. Kluwer Academic Publishers. The Netherlands.

Sandrez-vioque, R., Clement, A., Vioque, J., & Millian, F. (1999). Protein isolates from chickpea (*Cicer arietinum*) chemical composition, functional properties and characterization. *Food Chemistry, 64*, 243-273.

Suarez, M. E., & Thorne, B. L. (2000). Rate, amount, and distribution pattern of alimentary fluid transfer via trophallaxis in three species of termites (Isoptera: Rhinotermitidae, Termopsidae). *Annals of the Entomological Society of America. 93*, 145-155. http://dx.doi.org/10.1603/0013-8746(2000)093%5B0145:RAADPO%5D2.0.CO;2

Umeh, V. C. (2000). Advances in the control of termite pests of some tropical crops using naturally occurring pesticides. Paper presented at 14th Africa Association of Insect Scientists and the 9th Crop Protection society of Ethiopia joint conference, June 4-8 2001, Ethiopia an Agricultural Research organization Addis Ababa.

Umeh, V. C. (2003). An appraisal of farmers' termite control methods in some traditional cropping system of West Africa. *Nigerian Journal of Entomology, 20*, 25-39

Vane-Wright, R. I. (1991). Why Not Eat Insect? *Bulletin of Entomological Research, 81*, 1-4. http://dx.doi.org/10.1017/S0007485300053165

Wheeler, B. L., & Ferrel, R. E. (1971). A method for phytic acid determination in wheat fractions. *Cereal Chemistry, 48*, 312-316.

Young, S. M., & Greeves, J. S. (1940). Influence of variety and treatment on phytin contents of wheat. *Food Research, 5*, 103-105. http://dx.doi.org/10.1111/j.1365-2621.1940.tb17171.x