NUTRITIONAL VALUE AND EFFECT OF COOKING, DRYING AND STORAGE PROCESS ON SOME FUNCTIONAL PROPERTIES OF *RHYNCHOPHORUS PHOENICIS*

HILAIRE MACAIRE WOMENI[1*], BERNARD TIENCHEU[1], MICHEL LINDER[2], ERIC MARTIAL CHOUATCHO NABAYO[1], NOEL TENYANG[1], FELICITE TCHOUANGUEP MBIAPO[1], PIERRE VILLENEUVE[3], JACQUES FANNI[2] AND MICHEL PARMENTIER[2]

[1] Laboratory of Biochemistry of Medicinal Plants, Food Science and Nutrition, Faculty of Science, University of Dschang, P.O. Box 67 Dschang, Cameroon,
[2]Laboratoire d'Ingénierie des Biomolécules, ENSAIA-INPL, 2 avenue de la Forêt de Haye, 54500, Vandoeuvre-les-Nancy cedex, France,
[3]Laboratoire de Lipotechnie, CIRAD, UMR IATE, TA 40/16, 73 rue JF Breton, 34398 Montpellier cedex 5, France.

ABSTRACT

The proximate composition, amino acid composition, and effects of drying, storage and cooking processing on physicochemical properties of *Rhynchophorus phoenicis* (RP) flour consumed in Cameroon were determined. Results showed that the proximate composition fluctuated in samples with a protein content of 67.09 % for larvae flour. The moisture content recorded for this insect was about 60%. The ash and carbohydrate contents of defatted flours were 2.7% and 17.56% respectively. The hyperbolic kinetics of dehydration showed that the sun was a slow process of dehydration (6 days were required for the complete elimination of water) while the electric drying at 50°C and smoking were relatively rapid processes of dehydration. Processing of insects greatly increases protein solubility. The analysis of functional properties revealed that roasting and grilling of larvae led to a decrease in water absorption capacity. In contrast to the boiling and grilling which maintained water absorption capacity at 3.5 ml/g, culinary treatments (smoking) increased the solubility of proteins, and decreased oil absorption capacities to values that are still higher than 0.7 and 0.9 ml/g needed in food formulation. The effect of pH (1-12) on the proteins showed that a majority of proteins are soluble in alkaline pH and there are more than two proteins in RP larvae flours with iso-electric point of 4 and 8. Protein solubility was pH dependent, and increased at pH 7and 8.

Key words: *Rhynchophorus phoenicis,* Edible insect, Culinary treatment, Drying, Storage, Functional properties

1. INTRODUCTION

The larva of *Rhynchophorus phoenicis (R.P.)*, a Coleoptera of Curculionidae family is used as traditional food in several countries. It is a delicious meal in many parts of Cameroon and other countries in Africa where it is found.The high cost of animal protein has directed interest towards several insects as potential sources of proteins for humans. Among the insects species, *R. phoénicis* larvae are considered the major sources of dietary lipids and proteins. They are consumed world wide, especially in developing and under developed countries where consumption of animal protein may be limited because of economic, social, cultural or religious factors (Sanchez *et al.*, 1996; Cerda *et al.*, 1999). African insects are rich in protein and usually processed to tasty food products which are used as flavour intensifiers in soups and stews and also

add protein to protein-poor diets. Ordinarily, insects are not used as emergency food sources during shortages, but are included as a planned part of the diet throughout the year or when seasonally available (Inyang and Iduh, 1996; Lin et al., 1974).

Among the people living in south of the Sahara, the spectrum of hunger is endemic. This makes the insects, food unconventionally interesting to study because they remain under exploited and not recovered. However, the physical and chemical properties of their proteins in food systems during processing, storage, preparation and consumption is affected (Fennema, 1996). This insect is rich in protein, inexpensive and underutilized by the industries, meanwhile it offers the same benefits as other meat products with less fat when deffated. They contain in this delipidated form over 80% of high quality protein with high content of essential amino acids (Basha and Pancholy, 1982; USDANAL, 2005) and can be useful in many food applications (Prinyawiwatkul et al, 1993).

The defatted flour of insects, despite an excellent amino acid profile, could find a marginal use in the food industry because of its functional properties. Thus, the development of new proteins product such as insect proteins concentrate from defatted meal is more important because it could provide to food industries, a new high protein (over 80 g protein / 100 g) food ingredient in the formulation of products and the enrichment of foods (Wu et al., 2007). This is critically needed in many developing countries, because animal protein is becoming more expensive and beyond the reach of many people in developing countries. Insects will be less expensive sources of protein used for this purpose.

The functional properties of food proteins play an important role in food processing and formulation of food products. These properties are affected by many factors. For end users, pH, temperature and ionic strength of food systems are important factors to consider. For producers, procedures and conditions for protein extraction and downstream processing of extracted proteins such as cooking, drying and cold are factors that must be brougt up (Fukushima, 2000).

In order to conclude whether insects can be used as intermediate flour with good functional characteristics for food product development, it was necessary to evaluate some nutritional properties of some insects for their transformation into a flour that can be used in food formulations, in order to promote, their use and therefore contribute to food safety.

2. MATERIALS AND METHODS

2.1. Materials
Life larvae of R. phoenicis were collected in Mvog-Mbi market of Yaoundé (Central region of Cameroon) and transported in palm grove. The species were specifically identified in the Biology Department of University of Dschang (Western region of Cameroon).

2.2. Proximate composition
To analyze the chemical composition, the larvae were killed by freezing for 5 minutes (Finke et al., 1989; Adedire and Aiyesanmi, 1999). After thawing at room temperature, they were dried in an oven at 50°C for a period of 72 hours. The dried samples were reduced to powder and stored in a desiccator for food-science analysis. Moisture, Crude protein, fat, ash and total carbohydrate contents were assayed by the Association of the Official Analytical Chemists (AOAC,1988).

2.3. Determination of amino acid composition
Amino acid analysis was done by ion exchange chromatography(IEC) (FAO/WHO, 1991), using the Technicon Sequential Multisample (TSM) Amino Acid Analyser (Technicon Instruments Corporation, Dublin, Ireland). About 2.0 g of each sample was defatted using chloroform/methanol and then hydrolysed using 6M HCl. The hydrolysate was then injected into the amino acid analyser for separation and characterisation. Tryptophan was not evaluated. The total essential amino acids (TEAA), the percentage of the total essential amino acids in the total amino acids (%TEAA), total acid amino acids (TAAA), total sulphur amino acids (TSAA) were calculated and the predicted protein efficiency ratio (P-PER) was determined using the equations of Alsmeyer, Cunningham and Hapich (1974) as adapted by Adeyeye (2009) (i.e. P-PER = -0.468 + 0.454(Leu) -0.105(Tyr)).

2.4. Cooking and conservation treatments

2.4.1. The Culinary or cooking treatments

Collected samples were separated into two batches; one for the culinary treatments and other for conservation treatments. Common ways of cooking were used. Boiling was performed at approximately 100°C (with 200 g of sample in 1000 ml water) for 20 min by using an adjustable thermostat diver. Two hundred grams of grilled insects were prepared in a Black and Decker griller model G48 (on a stainless steel grill (Balay, Zaragoza, Spain) with the thermostat set at 135°C. After the set temperature was attained the RP were grilled for 10 min (5 min on each side). The insects RP brochettes (kebab skewer) were deep roasted in charcoal for 10 min. The roasting temperature was around 95° C. Samples of raw RP, an untreated sample of zero processing time (sample kept fresh and raw) were immediately homogenized and used as control or as reference.

2.4.2. Treatments of dehydration

Smoked dried samples were obtained after spreading 200g of insects on a rack and exposition to smoke heat for 6 hours. The kinetics of mass loss was determined during this 6 hours of smoking treatment. This process was done in a traditional manner and with fuel wood and soot.

The sun and oven dried samples were obtained after spreading approximately 200g of sample in single layer under the sun for 14 days or in an electric dryer (ventilated oven thermostated at 50° C for 48 hours). The kinetics of mass loss was estimated after the drying process. Boiling, as described previously, was also combined to these treatments of dehydration.

2.4.3. Cold treatments

For cold treatments, 6 groups of 200 g of *R. Phoenicis* larvae sample were wrapped in plastic papers and arranged in batches of three and placed in the freezer (-18°C) and refrigerator (4°C) for durations of 1, 3 and 7 days and 7, 37 and 90 days respectively. After these periods, samples were removed, allowed to thaw before characterization and analysis.

2.5. Lipid extraction or defatted method

After different culinary, storage and conservation treatments of insects, samples were deffated (lipids were extracted) according to the method described by Bligh and Dyer (1959).

2.6. Functional properties
2.6.1. Measurement of protein solubility

The protein solubility was determined according to the modified method of Ige *et al*. (1984) and Oshodi and Ekperigin (1989). Two hundred milligrams of each deffated flours were dissolved in 5 ml of distilled water and the pH was adjusted to the desired value with 0.1 mol/L HCl or NaOH. The tubes containing the mixture were then centrifuged at 4000 rpm and the OD (optical density) expressed in terms of absorbance unit (UA)) of the supernatant was determined using the Biuret method.

2.6.2. Effect of pH on proteins

The methods described by Ige *et al*. (1984) and Oshodi and Ekperigin (1989) was used on raw insects flours (deffated residues) to determine the different classes of proteins which precipitate at the pH range 1-12.

2.6.3. Oil absorption capacity(OAC) and water absorption capacity (WAC)

They were determined using the method described by Lin *et al. (1974)*. One gram of protein sample was vortex-mixed with 10 ml of sunflower oil or distilled water for 30 s. The emulsion was incubated at room temperature (about 20 °C) for 30 min and then centrifuged at 13,600g for 10 min at 25°C. The supernatant was decanted and drained at 45°C for 20 min. The volume of oil or water absorbed was divided by the weight of the protein sample to obtain the oil absorption capacity of the sample.

2.6.4. Determination of the density

Fifty gram sample of flour were introduced into a 100 ml test tube. The sample was continually packed until a constant volume of flour in the sample. The density in g/cm^3 was calculated as the weight (g) of flour divided by the volume (cm^3) of flour (Kinsella, 1976).

2.7. Statistical analyses.

The tests were performed in duplicate and results are representative of the mean ± standard deviations. All results were submitted to the analysis of variance (ANOVA) at 0.05%

probability level. The Dunnett test was used to compare means using the software Graphpad InStat, 2000.

3. RESULTS AND DISCUSSION

3.1. Chemical composition

The chemical composition of RP larvae is presented in Table 1. It appears that these larvae are composed mostly of water like most livingthings. This table revealed that larvae of *R. Phoenicis* larvae are also good sources of fat in the dried state (68.61% of dry weight). A high level of ash (16.35 ± 0.08) was obtained indicating that larvae foods are rich in minerals. The larvae flours were important sources of proteins when delipidated. We find that the water content of larvae was found to be 61%. This content is not very different from that found by Ekpo and Onigbinde (2005) for the larvae of *R. phoenicis*, but superior to that reported by Okaraonye and Ikewuchi (2009) on larvae of *O. Rhineceros* (16.73 ± 0.49%) and *Acheta domesticus* (73%) by Pennino *et al*. (1991). This protein content was lower than that reported on *Anafi venata* (6.61%) by Ashiru (1988). The high water content of these insects may predispose them to chemical and microbiological alterations; hence the need for drying. The ash content (2.37%) was higher compared to other meat products indicating that larvae contain many minerals. This ash was comparable to that reported by Pennino et al. (1991) on *Tenebrio molitor* (2.5%) and Ashiru (1988) on *A. venata* (3.21%).

Table 1 also presents the value of lipid content. The results show that this insect is a good source of fat, because these values are greater than 50%. The crude fat content are more than 50% higher than the range of lipids from 1.5 to 31.40%, previously reported for the various forms of Lepidoptera, Coleoptera and Orthoptera in edible insects in southwestern Nigeria (Banjol *et al*., 2006). In addition, this value is within the range of lipid from 4.2 to 77.2% reported for the seventy-eight different edible insects of Oaxaca State, Mexico (Ramos-Elorduy *et al*., 1997; Ekpo and Onigbinde, 2005). These values are higher than the values found in most foods such as beef, chicken, eggs, herring, mackerel and milk (Pyke, 1979), and are considered responsible for its acceptable taste when roasted or fried. Malnutrition in developing countries is equally a problem of lack of caloric intake (De-Foliart, 1992). The fat content means that samples of 100 g of larvae satisfy caloric requirements in most developing countries (Davidson *et al*., 1973). Indeed lipids are necessary in food because they increased palatability and retain the flavor of food (Aiyesanmi and Oguntokun, 1996). They also play a structural and physiological role. The carbohydrate content was lower compared to values reported by Ajakaiye and Bawo (1990) on termites, Okaraonye and Ikewuchi (2009) on the larvae of *O. Rhineceros* (19.47% - 33.52%).

Protein content was equally low, but when these insects were completely defatted, flours obtain were concentrated in proteins. However, the protein content was higher than that found by Mercer (1994) on *R. ferrugineus* (6.10%), but comparable to values reported by Ekpo and Onigbinde (2005). However, the values plotted on meal tracked Imbrasia (70-90%) remains higher than this. Compared with beef or fish, insects possess high fat content, defatted flours had high protein content, and are therefore extremely energetic.

Table 1: *Proximate composition of larvae of R. phoenicis*

Component	% Fresh weight	g/100 g dry weight	% Flours (defatted residues)
Moisture	61.85±0,2	-	-
Ash	2.10±0,10	5.53±0.17	16.66±0,17
Lipids	25.30±0,22	66.61±0.35	-
Proteins	8.21±0,35	21.06 ±0,22	67.09 ±0.28
carbohydrates	2.97 ±0,01	7.63 ±0,12	17.56 ±0,02
Energy (Kcal)	684.81	714.25	338.60

3.2 Amino acids composition

Amino acid composition of larvae of R. phoenicis is illustrated in Table 2. Results showed that this larva contained at least 18 known amino acids with almost all the essential amino acids (EAA) except tryptophan and cysteine that were not evaluated.

Table 2: *Amino acid profile of R. phoenicis larvae (mg/g protein).*

Amino acid	R. Phoenicis	Egg
aspartic Acid	104.41	82.20
glutamic Acid	155.05	121.30
Serine	41.23	67.20
Glycine	39.68	30.20
Histidine	24.00	20.90
Arginine	34.44	57.00
Threonine	23.91	44.70
Alanine	54.96	50.30
Proline	64.00	33.80
Tyrosine	25.15	38.10
Valine	27.64	54.20
Methionine	22.97	28.10
Tryptophane and cysteine	NE	17.2 et 19.00
Isoleucine	67.33	48.80
Leucine	96.02	81.10
Phenylalanine	31.59	48.20
Lysine	54.84	65.90
Total (mg)	**867.20**	**915.20**
AAE	**394.29**	**388.2**
% EAA	**45.46**	**40.54**
NEAA	**472.91**	**544.20**
EAA /NEAA	**0,83**	**0.68**
P-PER	**1.50**	**2.81**

NE: non evaluated

All the essential amino acids were almost found: lysine, valine, leucine, isoleucine, phenylalanine, threonine, methionine. Amino acid profile showed that the essential amino acids like lysine and threonine, normally deficient in grains and cereals, were in high concentrations but low when compared to references (FAO/WHO, 1990), while the other with the exception of tyrosine and phenylalanine were in high concentrations.

Tyrosine and methionine were present in low concentrations in the larvae. Almost all the essential amino acids had high chemical scores implying that these amino acids have high biological values as in animal protein compared to plant proteins that have no or less than 4 amino acids. Most of the proportions of these essential amino acids exceed those of the standard value required by FAO (Table 3).

Table 3 : *Essential amino acids score (%) of R. phoenicis compared with the FAO/WHO pattern (mg/g crude protein)*

Amino acid	RDA mg/ 70 kg body weight	FAO/WHO pattern	Amino acid scores
Isoleucine	1400	28	2,40

Leucine	2730	66	1,46
Lysine	2100	55	0,99
Méthionine +Cystéine	1050	25	0,92
Phénylalanine +Tyrosine	1750	60	0,94
Thréonine	1050	34	0,70
Tryptophane	280	10	/
Valine	1820	35	0,78

The amino acids aspartate, glutamate, proline, alanine, isoleucine and leucine were found in high proportions in larvae insects, while histidine, threonine, tyrosine and methionine were the most poorly represented in RP larvae. When comparing the amino acid composition of RP larvae and egg (Rao, 1994; Wang, 1991), we found that the EAAs of RP larvae (394.29 mg / g protein), were higher than those of eggs (388.2mg / g protein) and that reported by FAO / WHO (360 mg / g protein).

Amino acid scores (Table 3) revealed that the most limiting amino acids of the larvae were threonine and valine. The percentage of EAAs in the total amino acids was 45.46% in the larvae, greater than 42% found in the pulp of the silkworm Anthereae pernyl in China by Zhou and Yang (1993), greater than 40.54% in the egg and ranged within the reference value of 40% and 60 recommended by the FAO/WHO (1980). The ratio of EAA/NEAA is 0.83 higher than that of the egg (Zhou and Yang, 1993).

In addition, RP flours proteins in term of the percentage of taste amino acids (arginine and glutamic acid) and sweet amino acid (glycine and alanine) had lower values compared to total amino acids; while a value of 18, 8% and 11.0% respectively was found by Adeyeye and Afolabi (2004). This profile of EAA in Table 3 revealed that the level of EAA is comparable to the reference model of amino acid of the FAO/WHO (1980) established for humans.

Amino acids are important in healing processes and its composition in larva is similar to that in man and people can therefore acquire the essential and non-essential amino acids in abundant and proper balance by eating larvae or fish (Osibona et al., 2009). The essential amino acids cannot be manufactured in human bodies, but can be obtained from food. Eight amino acids are generally regarded as essential for humans: phenylalanine, valine, threonine, tryptophan, isoleucine, methionine, leucine and lysine (Young, 1994). Additionally, cysteine (sulphur-containing amino acids), tyrosine (aromatic amino acids), histidine and arginine are required by infants and growing children (Imura & Okada, 1998). Deficiency in these amino acids may hinder healing recovery process (Mat Jais et al.,1994). All of these essential amino acids are found to be present in the larvae samples investigated except tryptophan, which was not evaluated.

The predicted protein efficiency ratio of the larvae samples was 1.5. The P-PER of samples are generally near the P-PER of the corresponding fresh T. trachurus, but lower than that of boiled and roasted fish samples obtained by Oluwaniyi et al. (2010). The standard reference for the PER is based on casein, a cow milk protein, which has a PER of 2.5 (FAO/WHO/UNU, 1985) and the values obtained for larvae in this study was lower than 2.5, while the values for S. scrombus and H. harengus were slightly less than 2.5. Generally, a PER below 1.5 approximately describes a protein of low or poor quality (Friedman, 1996). According to Abdul-Hamid et al., (2002), the PER values of protein have no proportional relationship to one another or to the suitability of the protein source analysed for maintenance of protein nutrition. It also lacks precision, has poor reproductibility and is expensive. Nevertheless, the PER was predicted based on amino acid composition, for comparison purposes and the values obtained were found to be greater with the P-PER for cowpea (1.21) and lower than that of pigeon pea (1.82) (Salunkhe & Kadam, 1989).

The preliminary results presented here indicate that the defatted flour larvae of RP have great potential to reduce the deficit in amino acid known in the diets of developing countries. In China, more than 100,000 tons of insects are

L - 208

produced annually (Wei and Xu, 1999) and used as nutritional resources to reduce nutritional deficiencies. Intensive efforts for the popularization of entomophagy in Cameroon must therefore be encouraged.

3.3. Dehydration

Figure 1 represent the curves of mass loss during electrical drying, sun drying and smoking of larvae. It appears from the figures that during the first minutes or days of drying, the losses were low; this would correspond to a temperature setting of larvae. Beyond these first few minutes, all curves increased linearly, corresponding to the acceleration of dehydration. From more than 15 hours of electrical drying and 4 hours of smoking, the curves showed a constant phase, corresponding to a complete elimination of free water in larvae. The percentages of mass loss did not evolve anymore and remained close to the water content of insects (> 60%). Therefore, a period of 15 hours, 6 days and 4 hours respectively for electric heating, solar drying and smoking are needed for a complete dehydration of the insects for better conservation. Since moisture or high water content are described as catalysts of

reactions of spoilage (Karel, 1977), electric heating and smoking remain a faster process of dehydration. Less laborious and slowest process of dehydration was sun drying, but cheaper. Similar conclusions were obtained by Womeni et al. (2002); Tiencheu (2006) respectively during the drying of shea nuts and palm kernel. During smoking and drying in the sun, there are percentages of mass loss at the end of processes slightly exceeding the water content beyond 5 hours and 4 days respectively. This may be because there was a loss of fat during prolonged drying periods. These results corroborate with that of Edijala *et al*. (2009) in smoked *Orytes monoceros,* which state that smoking lowers the fat content. Ultimately, this loss of mass could be explained by the fact that, during the drying of food as described by Gautier and Bimbenet (1977), a difference of temperature and of partial pressure of water is established spontaneously between the foods and a hot-air. This leads to heat transfer from air to the product under the influence of temperature gradient. It also leads to a transfer of water (by evaporation) in the opposite direction because of the water concentration in air (Bimbenet *et al*., 1971).
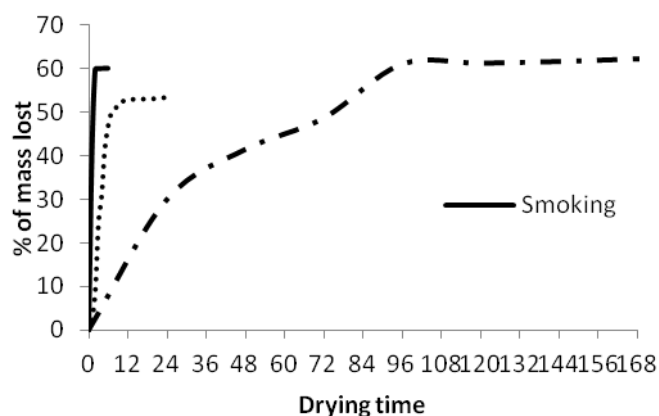

Figure 1: *Kinetics of dehydration of RP larvae during smoking, electrical and sun drying*

3.4. Effect of pH on proteins

The results of the effect of pH on protein solubility of larvae (Figure 2) shows that the highest solubilities appeared at basic pH (pH 7 and pH 9) while low values are obtained betwenn pH 1 to 4. Similar results were found by Omotoso and Adedire (2009) on the *R. phoenicis* larvae stage, but at pH 4 and 7. Similarly, high solubility

in an alkaline medium were found on grasshoppers by Omotoso (2005). This gives the possibility to use these cake larvae in food supplements and formulations. The existence of three isoelectric points (around 1; 4 and 8) shows that delipidated flours of *R phoenicis* larvae have more than one class of protein as decribed by Omotoso and Adedire (2006).
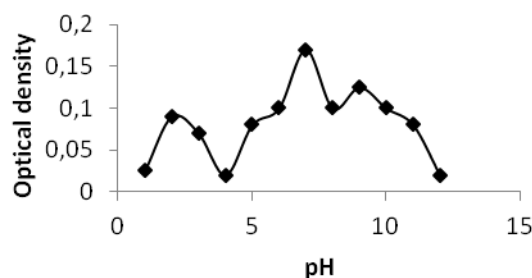
Figure 2: *Effect of pH on protein solubility of larvae*

3.5. Effects of cooking and conservation methods on water absorption capacity (WAC)
3.5.1. Effects of cooking methods

Water absorption capacity provides information on the degree of hydration of proteins. This will depend on the nature of amino acids and protein conformation. In short, the amount of water held in a molecule will depend on the fraction of channel charge, polar or nonpolar.

Figure 3 shows the evolution of water absorption capacity of the flours obtained after cooking treatments of larvae. It appears that roasting and grilling significantly low (P< 0.05) WAC (2.25 and 2.5 ml / g, respectively) compared to control; followed by boiling (3.25 ml / g). Grilling coupled with boiling did not affect significantly the WAC. Boiling, grilling-smoking and smoking would have favoured a low distortion of protein, that exposed more polar group of proteins on the surface. Contrary to the grilling and roasting which favoured the aggregation of proteins with more non-polar groups or cross-linking, which would have reduced the ability of water absorption. El Hassan *et al* (2008) proved this on boiled and fried locusts having respectively 2.93 and 2.47 ml/g as WAC. The reduction of WAC is caused by non-dissociation occurring because of low thermal denaturation. This could be maximized if a higher processing time was applied. The degree of WAC is considered an indicator in the formulation of various foods specifically those requiring a high viscosity (Circle and Smith, 1972). Then grilled-boiled, smoked and boiled can be retained as samples having good WAC.
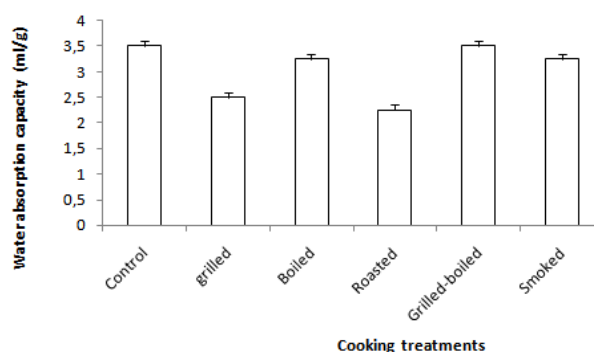

Figure 3 : *Water absorption capacity of the flours obtained after cooking treatments of larvae.*

3.5.2. Effects of conservation methods (dehydration and cold)

Figure 4 show the water absorption capacity of flours from dehydrated larvae. For these processes, retention capacities did not differ much with control sample in the flours from the boiling-smoking and sun drying processes.Treatments preceding boiling increase the ability of proteins to retain water (Chau and Cheung, 1998). Electrical drying and boiling-sun drying reduced significantly the WAC. Dehydration of meat causes hardening of texture and reduction on the ability to absorb water during rehydration. The residual water causes the formation of disulfide bridges and hydrogen bonds between proteins that adhere strongly to each other (Cheftel *et al*. 1983).
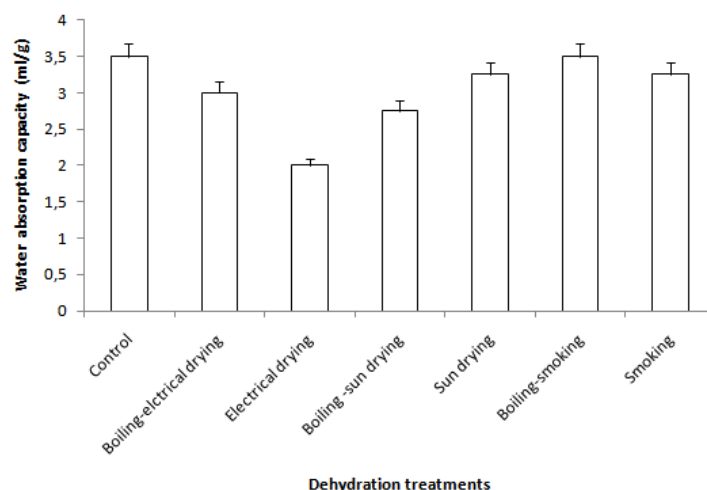
Figure 4: *Water absorption capacity of flours from dehydrated larvae*

Figure 5 revealed that cooling keeps the water absorption capacity, and the variations are less noticeable. Frozen storage after one week caused a reduction in this water retention due to its prolonged time, while refrigeration due to low storage time maintained the WAC as in control sample. This is due to deconfiguration and protein aggregation. Cell disruption by ice crystals has the same effect as estimated Cheftel *et al.* (1983), bringing about the liberation of fatty acids. By binding to proteins, they contribute in rendering them hydrophobic. The main consequence of this phenomenon is a decreased ability to absorb water. In general, cold treatments tend to reduce water absorption capacity. The heat treatments alter the three dimensional structure of the protein that reduce water retention capacity of certain flours or tuber meals (taro) and seedsas reported by Osundahunsi et al. (2003). Aletor et al. (1993) considered foods whose capacities range from 1.49 to 4.72 ml / g as viscous foods. These cakes can be recommended as thickeners in liquid or semi liquid formulations because they have the ability to absorb water and therefore better for improving the consistency of food. Refrigeration for less than 3 days and frozen for less than a week can maintain and preserve WAC of larvae flours.
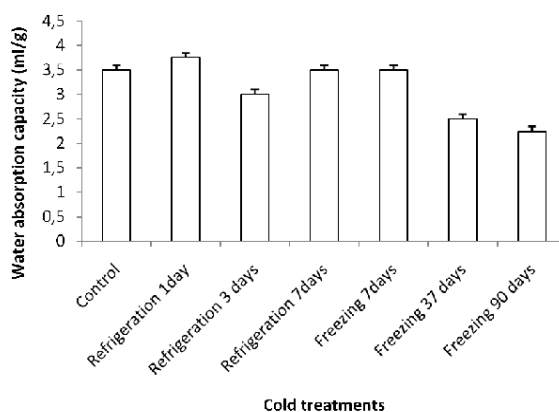

Figure 5 : *Water absorption capacity of flours from cold larvae*

3.6. Influence of culinary and conservation of larvae on oil absorption capacity (OAC) of the flours
3.6.1. Cooking

Oil-flour Interactions are very important in food systems due to their effects on the nutritional value and texture of the food. Figure 6 shows the OAC of flours obtained after culinary treatments of larvae. We note that, except for boiled samples that were not affected significantly, all other

L - 211

treatments lowered this capacity. These declines could be due to a distortion of the native structure of proteins leading to a more or less crosslinkings to form aggregates. These findings were contrary to those reported by Chau and Cheung (1998) that high temperatures increase the hydrophobicity by exposing hydrophobic groups. Kinsella (1976) emphasized that the ability of proteins to bind to lipids is an important phenomenon since the lipids

act as sensors of flavor. Certains OACs of larvae flours are superior to those of cérelac (2.15 ± 0.05) as found by Dangang (2009). OACs of all these treatments vary from 230% to 320%. They are far above the OAC of taro flour (190%) (Tagodoe and Nip, 1994) and the clustering of sweet potato flour (10 -12%) (Osundahunsi *et al.*, 2003) and similar to that reported by Mafokoué (2009) on cooked mackerel fish flour.
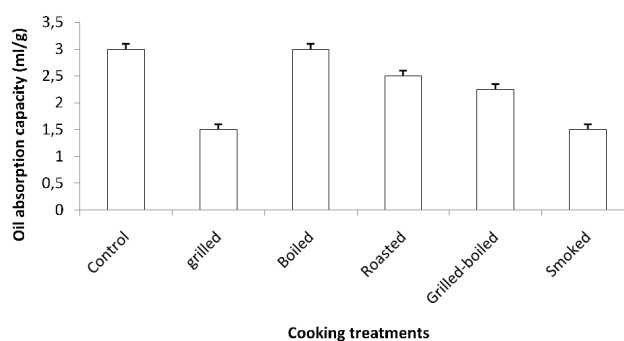

Figure 6 : *Oil absorption capacity of flour proteins obtained after culinary treatments of larvae.*

3.6.2. Conservation

Figures 7 and 8 show the variation of the oil absorption capacity of cakes, reduced to flours obtained after conservation treatments of larvae. It was found that smoked sample had the lowest oil retention. Given that during smoking food is not in direct contact with the heat source, distortion is not pronounced thereby reducing the exposure of nonpolar groups and the ability to absorb oil. However, all these values were much higher than those found by El Hassan et al. (2008) for boiled

and fried locusts (1.0 and 1.3 ml / g, respectively). Nevertheless all these values were low compared to the control, they remained higher than those found by El Hassan and al. (2008) and Dangang (2009) for the formulation of weaning food made from apples, beans and eggs (0.7 to 0.9 ml / g). The ability of protein meal to physically fix lipids by capillary attraction is important because lipids act by retaining or increasing the flavor of foods (Kinsella, 1976).
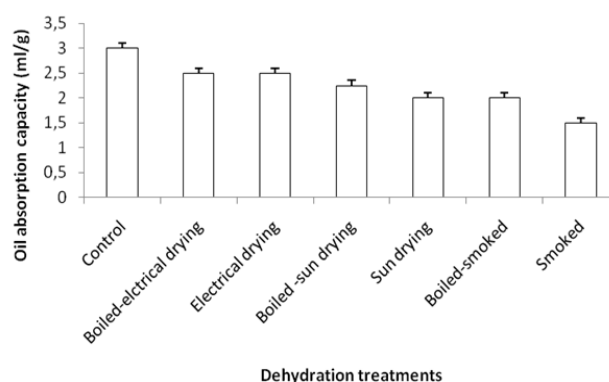

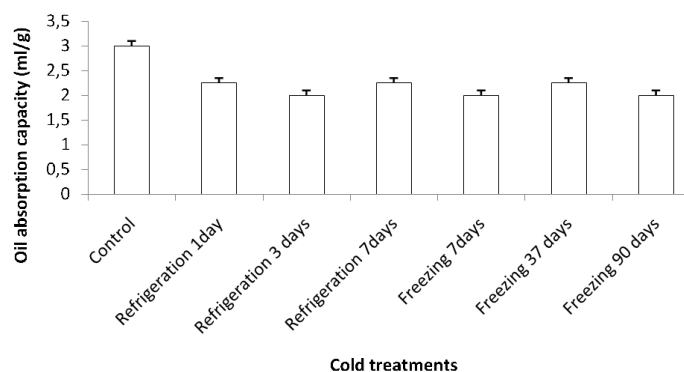**Figure 7** : *Oil absorption capacity of flours obtained after dehydration of larvae*

Figure 8 : *show the variation of the oil absorption capacity of flours obtained after cold treatments of larvae*

3.7. Effects of culinary and conservation or storage (dehydration and cold) methods on bulk density of flours
3.7.1. Effects of cooking

Figure 9 shows the densities of RP larvae. The larvae during cooking gave flours having densities ranging from 0.25 to 0.52 g/ml respectively for grilling and boiling .The culinary treatment increased the density with the exception of the grilling because of it high temperature. Indeed temperatures as those of the grilling (135 °C) gave fine grains, but larger than those from boiling. Lalude and Fashakin (2006) reported similar findings on sorghum, soybeans and peanuts. It is known that density is a function of particle size of flours and increases with the fineness of particles. Densities between 0.25 and 0.52 are within the range reported by Chau and Cheung (2008) on peanut flour hot pressed and cold.
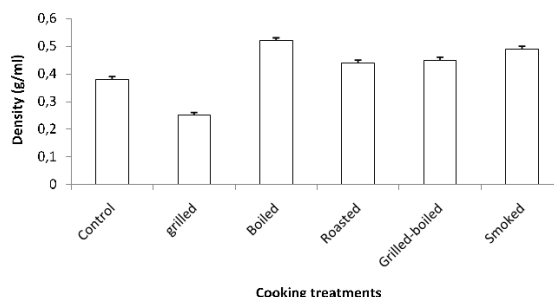

Figure 9 : *Densities of RP larvae flours after cooking treatments*

3.7.2. Effects of Conservation treatments

The densities of insects larvae flours after dehydration and cold treatments are presented in Figures 10 and 11. It shows that samples from electric heating, sun drying, smoking did not show strong density fluctuations. Smoking slightly increased the density of the flours; especially for the treatments of dehydration which was less severe than the treatment of cooking concerning thermal point of view. Freezing confered (one week) a higher density compared to control but when prolonged, they reduced the density. Refrigeration for less than 7 days had positive effect on flour density. To have acceptable densities, methods applied (temperatures) must be less severe. The values found in the context of this work were lower than those found by El Hassan et al. (2008) for boiled and fried grasshoppers (1.5 and 1.82 respectively). Flours with high densities are desirable because they help reduce the thickness of the dough, which is important in the diets of convalescent and children (Padmashree *et al.*, 1987). Although some densities were reduced, all were above 0.4 value reported by Ladude and Fashakin (2006) for food supplements.
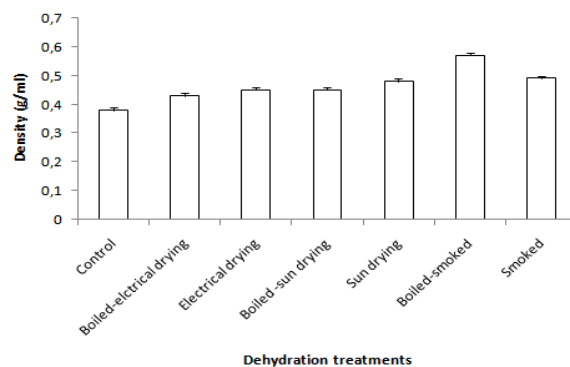
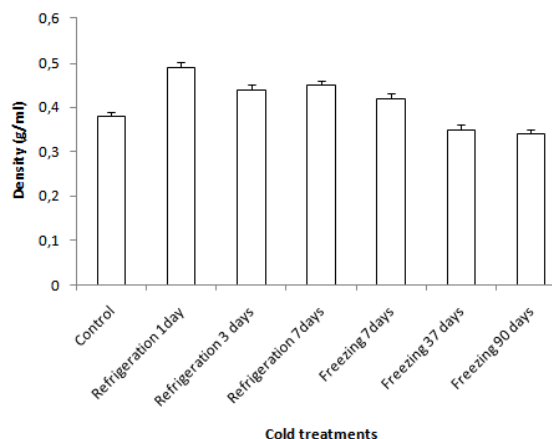**Figure 10** : *Densities of larvae flours after dehydration treatments*

Figure 11 : *Densities of larvae flours after cold treatments*

3.8. Effects of treatments on the protein solubility of RP flours

3.8.1 Cooking

It is clear from Figure 12 showing the influence of R. Phoenicis larval treatments on the solubility that, all culinary treatments (thermal) increased the solubility of proteins. This increase was more pronounced with the roasted and grilled samples. The heat treatments increased the protein solubility in KOH (pH 7 and 8); this could be explained by a structural and conformational change because internal portion of hydrophilic amino acids can open by denaturation and interaction as described by (Weiss et al., 2011). The increased solubility in alkaline medium can also be attributed to a change in the native conformation, which would increase the number of carboxyl and amino groups and, under these conditions, water-protein interactions would increase because of these electrostatic bonds. At acid pH the solubilities were low due to the fact that, this first protein exist at low concentration in the flours
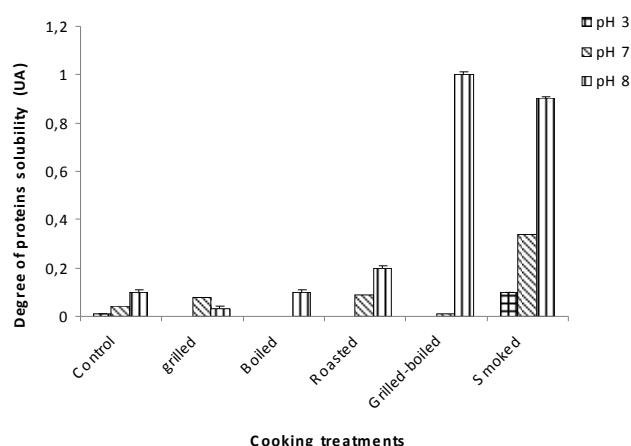
Figure12 : *Effects of cooking treatments on the protein solubility of larvae flours*

3.8.2 Conservation treatments

As for conservation treatments, Figure 13 shows that solubility also increased with temperature and was higher where the sample was boiled before drying. The highest solubilities were recorded for boiled, boiled-dried (sun or electrically) and smoked samples. Cold treatment (Figure 14) reduced the solubility of proteins. This decrease was more pronounced when the sample shelf life was prolonged after cold treatment. The proteins from cold treatments were less soluble. In any case, they were more soluble in alkaline pH. These solubility values ranged from 0.18 to 0.38 in contrast to other treatments involving heat, which increased the solubility up to 0.94 indicating that they are effective because their flour may be used for agro-industrial applications.
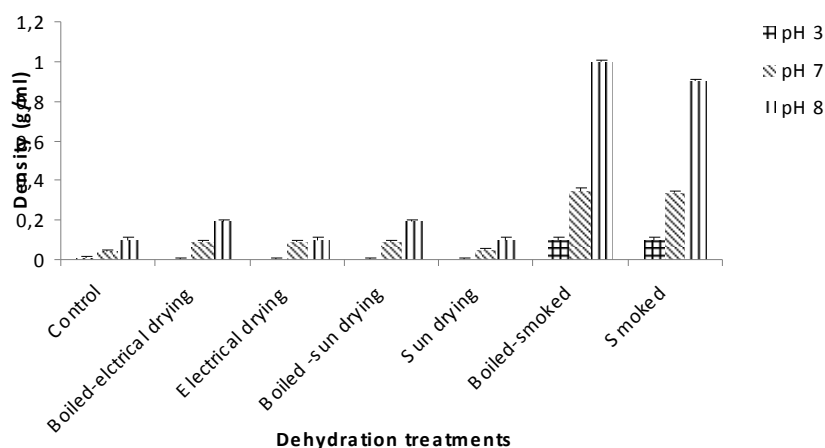

Figure 13 : *Effects of dehydration treatments on the protein solubility of RP larvae flours*
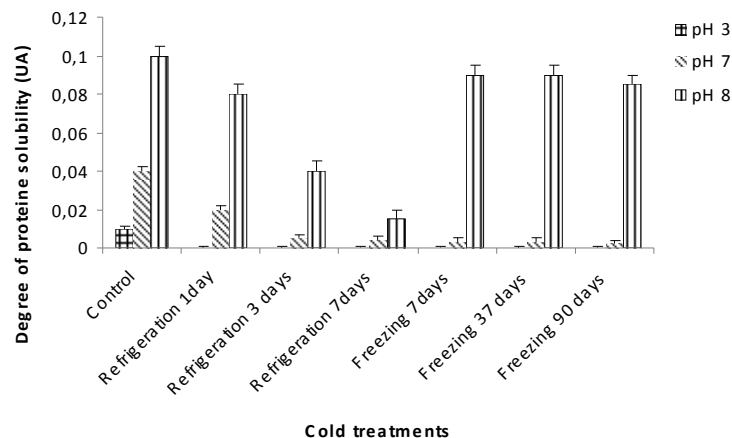
Figure14 : *Effects of cold treatments on the protein solubility of RP larvae flours*

CONCLUSION

These studies reveal that *R. phoenicis* larvae are good sources of nutrients. Indeed they can be used as food supplement especially in Africa and in Cameroon in particular to remedy nutritional deficiency (malnutrition).The analysis of functional properties reveals that roasting and grilling of RP larvae produce low water absorption capacity, in contrast to the boiling-grilling where capacity is maintained at 3.5ml/g. The culinary heat treatments increase the solubility of proteins especially when larvae are grilled, roasted or boiled before further treatment, and decrease the absorption of oils that are higher in spite of this (0.7 and 0. 9 ml/g), required for food supplements and formulations of weaning foods. The effect of pH 1-12 on proteins has shown that most proteins are soluble in alkaline pH and that there are more than two proteins in the larvae at isoelectric point. The density of flours ranged from 0.25 to 0.52 g/ml, in boiling, and grilling-boiling yield densities greater than 0.4 and can therefore be recommended in food supplements. The rich nutrient composition, protein solubility in alkaline, densities greater than 0.4 mg/ml, show that some treatments must be optimized, not only to preserve the food quality but also to define the agro-industrial and nutraceutical applications. Even if the lipid quality of larvae is bad, due to its degree of saturation, deffated flour residues possess many applications.

ACKNOWLEDGEMENTS

REFERENCES

1. Abdul E.l., Gengaihi S.E., Salem A, Bashandi S.A., Ibrahim N.A.and Abdel-Hamid S.R. (2002). Hypolipidemic effect of some vegetable oils in rats. *Food Agric. Environ.*, 2: 88-93.

2. Adedire CO., AiyesanmiA.F. (1999). Proximate and mineral composition of the adult and immature forms of the variegated grasshopper, *Zonocerusvariegatus* (L) (Acridoidea: Pygomorphidae). *Bioscience Research Communications*.11 (2): 121–126.

3. Adeyeye, E. I. (2009). Amino acid composition of three species of Nigerian fish: Clarias anguillaris, Oreochromis niloticus and Cynoglossus senegalensis. *Food Chemistry*, 113(1): 43–46.

4. AiyesanmiA.F., OguntokunM.O. (1996). Nutrient composition of *Diocleareflexa* seed an underutilized edible legume. *La RivistaItaliana DelleSostanze Grasse.*: 521–523.

5. Ajakaiye C.O. et Bawo A. (1990). Comparison of the chemical composition of *Trinervitermesgerminatus*(Wasmann) with that of its feed. *Nig. J. Entom*, 2: 90-99.

6. Aletor VA (1993). Allelochemicals in plant food and feedingstuffs: 1.Nutritional, biochemical and physiopathological aspects in animal production. *Vet. Hum. Toxicol.* 35(1): 57–67.

7. Alsmeyer, R. H., Cunningham, A. E., & Hapich, M. L. (1974). Equations to predict PER from amino acid analysis. *Food Technology*, 28: 34–38.

8. AOAC (1988) (Association of Official Chemists). Official and tentative methods of analysis of the AOAC; AOAC Washington, 978p.

9. Ashiru M. O. (1988). The food value of the larva of Anaphevenata Butler (Lepidoptera: Notodoutidae) *Ecol. Food Nutr,*22: 313 - 320.

10. Banjol A. D.; Laval O. A.; Songonuga E. A. (2005). The nutritional value of fourteen species of edible insects in southwestern Nigeria. *African journal of biotechnology.* 5 (3): 298-301.

11. Basha, S.M. and Pancholy, S.K. (1982). Composition and characteristics of basic proteins from peanut (*Arachishypogaea*L.). *J. Agricultural and Food Chemistry*, 30: 1176–1179.

12. Bimbenet J., Loncin M. and Brusset H., (1971). Heat and mass transfer during air drying of solide. Thécanadien. *J. Chem. Engny*. 49: 860 - 865.

13. Bligh EG, Dyer WJ (1959). A Rapid method for total lipid extraction and purification. *Can. J. Biochem. Physiol*. 37: 911-917.

14. Cerda, H., Martinez, R., Briceño, N., Pizzoferrato, L., Hermoso, D., and Paoletti, M. (1999). Cria, Analysis nutricional y sensorial del picudo del cocotero R.P (Coleoptera: Curculionidea), insecto de la dieta traditional indigena Amazonica. *Ecotropicos,* 12: 25-32.

15. Chau, C. F., , Cheung, Y. S. (1998). Functional properties of protein concentrates from three Chinese indigenous legume seeds. *Journal of Agricultural and Food Chemistry*, 45: 2500–2503.

16. Cheftel J., Cheftel H., et Besancon P. (1983). *Introduction à la biochimie et à la technologie des aliments*, vol.2. Technique et document, Paris, 420p.

17. Circle, S.J. and Smith, A.K. (1972). Functional propertiesof commercial edible soybean protein products. In: *Seed proteins*. E.G. Hnglet, Ed., Avi-Westport, Ct.: 242-254.

18. Dangang B.D. (2009). *Evaluation des aptitudes nutritionnelles et neutraceutiques des formulations de compléments à base d'aliments locaux*. Thèse de Master .Université de Dschang, 94p.

19. Davidson S.; Pasmore R.; Brock J. F.(1973). In: *Human nutrition and Dietetics* (5 edition). The English language Book Society. Churchill livingstone: 77.

20. De Foliart, G.R. (1992). Insects as human food. *Crop Protection* (11) 5: 395-399.

21. Edijala J. K. ,Egbogbo O., and Anigboro A. A., (2009). Proximate composition and cholesterol concentrationsof*Rhynchophorus phoenicis* and *Oryctesmonoceros*larvae subjected to different heat treatments. *African Journal of Biotechnology*. 8 (10): 2346-2348,

22. Ekpo k., Onigbinde A., (2005). Nutritional potentials of the larva of *Rhynchophorus phoenicis*. Ambrose Alli University, Ekpoma, Nigeria, *Pakistan journal of nutrition* 4 (5): 287-290.

23. El Hassan N; M.,. Hamed S. Y, Hassan A. B., Eltayeb M. M. and Babiker E. E. (2008). Nutritional Evaluation and Physiochemical Properties of Boiled and Fried Tree Locust. *Pakistan Journal of Nutrition* 7 (2): 325-329,

24. FAO WHO ,(1980). Technical report series no. 724. Geneva, Switzerland:

25. FAO/WHO (1991). Protein quality evaluation. Report of joint FAO/WHO expert consultation. FAO food and nutrition paper 51. Italy: Rome.

26. FAO/WHO/UNU (1985). Energy and protein requirement. Report of a Joint FAO/WHO/UNU Expert Consultation. World Health Organization, Geneva, Switzerland.

27. Fennema R.0. ,(1996). (ed) Food chemistry, 3rd edn. Marcel Dekker, Inc. 270 *Food Chemistry*, (3d edition): 365-369.

28. Finke M. D.; De Foliart G. R.; Benevenga N. J. (1989). Use of a Four-Parameter logistic model toevaluate the quality of proteins from three insect specieswhen fed to rats. *J. Nutr.*119: 864-871.

29. Friedman, M. (1996). Nutritional value of protein from different food sources. A review. *Journal of Agricultural and Food Chemistry*, 44(1): 6–29.

30. Fukushima, D. (2000).Soybean processing. In: Nakai, S. and Modler,H.W. Editors, *Food protein: Processing applications*, Wiley VCHInc, New York and Tronto: 309 – 442

31. Gautier et Bimberet J. (1977). le séchage dans l'industrie agricole et alimentaire : cahier du génie industriel alimentaire 29p.

32. Ige, M.N., Ogunsua A.O. and Oke O., (1984). Functional properties of the proteins of some Nigerian oilseeds, conophor seeds and three varieties of melon seeds. *Journal of Agricultural and Food Chemistry.*32(4):822–825.

33. Imura, K., & Okada, A. (1998). Amino acid metabolism in pediatric patients. *Nutrition,* 14(1) : 143–148.

34. Inyang, U.E. and Iduh, A.O. (1996). Influence of pH and salt concentration on protein solubility, emulsifying and foaming properties of sesame protein concentrate. *J. the American Oil Chemists' Society*, 73: 1663–1667.

35. Karel M., (1977). Development in food packaging protection based on control of water activity In :progrès et developpement de la technologiealimentaireC.P.C.I.A., Paris.

36. Kinsella J. E., 1976. Functional properties of protein foods. *Crit. Rev. Sci. Nutr*. 1: 219-229

37. Lalude L. O., Fashakin J.B., 2006.Development and nutritional assessment of a weaning food from sorghum and oil-seeds.*Pakistan Journal of Nutrition*. 5 (3): 257-260.

38. Lin M .J. Y., HumbertE.S. and SosulskiF.W., (1974).Certain functional properties of sunflower mealproducts. *J. Food Sci.,* 39: 368-370

39. Mafokoué C.(2009). Influence des procédés de cuisson sur la qualité du maquereau. 2009.Memoire de Master. Departement de biochimie, Université de Dschang 97p

40. Mat Jais, A. M., McCulloh, R., & Croft, K. (1994). Fatty acid and amino acid composition in haruan as a potential role in wound healing. *General Pharmacology*, 25: 947–950.

41. Mercer C.W.L. (1994). Sago grub production in Labu swamp near Lae-Papua New Guinea. Klinkii. *Journal of Forestry Papua New Guinea*. 5 (2) :30–34.

42. Okaraonye C.C. and IkewuchiJ.C. (2009). Nutritional Potential of *Oryctes rhinoceros* larva*Pakistan Journal of Nutrition* 8 (1): 35-38,

43. Oluwaniyi O.O, Dosumu O.O.,. Awolola G.V. Food Chemistry 123 (2010) 1000–1006 Effect of local processing methods (boiling, frying and roasting) on the amino acid composition of four marine fishes commonly consumed in Nigeria.

44. Oluwaniyi, O. O., & Dosumu, O. O. (2009). Preliminary studies on the effect of processing methods on the quality of three commonly consumed marine fishes in Nigeria. Biokemistri, 21(1), 1–7. of poorly digestible proteins supplemented with limiting amino acids in rats. Journal of Nutrition, 127(5): 758–764.

45. Omotoso O.T. and C.O. Adedire ⁽2009) Nutrient composition, mineral content and the solubility of the proteins of palm weevil, *Rhynchophorus phoenicis* f. (Coleoptera: Curculionidae).*African Journal of Pharmacy and Pharmacology* Vol. 3(2) : 051-057

46. Omotoso O.T., (2005). Nutritional quality, functional properties and anti-nutrient compositions of the larva of *Cirinaforda* (Westwood) (Lepidoptera: Saturniidae). Department of Zoology, University of Ado-Ekiti, Ado-Ekiti, Ekiti state, Nigeria 72p.

47. OmotosoO.T., (2006). Nutritional quality, functional properties and antinutrients compositions of larva of *Cirinaforda*(Westwood) (Lepidoptera:satuniidae), *J. ZhejiangUniv. Sci. B*., 7: 51-55.

48. Oshodi, A.A. et Ekperigin, M.M. (1989).Functional properties of pigeon bean pea (*Cajanuscajan*) flour. *Food Chemistry* 34: 187-199.

49. Osibona, A.O, Kusemiju K, Akande GR. (2009). Fatty acid composition and amino

acid profile of two freshwater species, African catfish (*Clarias gariepinus*) and tilapia (*Tilapia zillii*). *African Journal of Food, Agriculture, Nutrition and Development*, 9(1): 609-621.

50. Osundahunsi, O.F., Fagbemi, T.N., Kesselman, E. et Shimoni, E. (2003).Comparison of the physicochemical properties and pasting characteristics of flour and starch from red and white sweet potato cultivars. *Journal of Agricultural Food Chemistry* 51: 2232-2236.

51. Padmashree T.S., Vijayalakshmi L. etPuttaraj S. (1987). Effect of traditional processing on the functional properties of cowpea (*Vignacatjang*) Flour. *J. Food Sci. Tech.*, 24: 221-224.

52. Pennino M; Dierenfeld, ES; Behler, JL. (1991) inol, α-tocopheral and proximate nutrient composition of in food.

53. Prinyawiwatkul, W., Beuchat, L. R. and McWatters, K. H. (1993). Functional property changes in partially defatted peanut flour caused by fungal fermentation and heat treatment. J. Food Science, 58, 1318– 1323.

54. Pyke M. (1979). The Science of Nutrition. In: *Science in Nutrition*. John Murray (publishers) Ltd., London: 251-258.

55. Ramos-Elorduy J., Moreno J.M.P, Prado E.E.; Perez M.A., OteroJ.L. and de Guevara,O.L. (1997). Nutritional value of edible insects from the State of Oaxaca, Mexico, *Journal of Food Composition and Analysis*, 10 : 142 – 157.

56. Rao, P.U., (1994). Chemical composition and nutritional evaluation of spent silk worm pupae. *Journal of Agricultural and Food Chemistry*, 42 : 2201–2203.

57. Salunkhe, D. K., & Kadam, S. S. (1989). Handbook of world food legumes, nutritional chemistry, processing technology and utilization. Florida, USA: Boca Raton, CRC Press.

58. Sanchez P, Cerda H, Cabrera A, Caetano FH, Materán M, Sánchez F (1996). Secretory mechanisms for the male produced aggregation pheromone of the palm weevil *Rhynchophorus palmarum* L.(Coleoptera: Curculionidae). *J. Insect. Physiol.*, 42: 1113-1119.

59. Tagodoe, A. et Nip, W.K.,1994. Functional properties of raw and precooked taro *Colocasia esculenta* flour. *International Journal of Food Science and Technology*, 29: 457-482

60. Tiencheu B., (2006). Identification et amelioration des traitements critiques d'extraction industrielle d'huile de palmiste ; Memoire de Master. Departement de biochimie, Université de Dschang 97p.

61. USDA-NAL (2005). United States Department of Agricultural-National Agricultural Library. USDA Nutrient Database for Standard Reference, Release 18. Available from http://www.nal.usda.gov/fnic/foodcomp/ cgi-bin/list_nut_edit.pl.

62. Wang, G.Y., (1991). Table of Food Ingredients (Representative Values in China). People's Sanitation Press, Beijing: 78–79.

63. Wei, K.M., Xu, Q.Y., (1999). Situation and advantages of overall medicinalexploitation of silkworm in Zhejiang province. Zhejiang Journal ofTraditional Chinese Medicine 34 (11): 501–503.

64. Weiss, J., Kristbergsson, K., Kjartansson, G.T. (2011). Engineering Food Ingredients with High-Intensity Ultrasound. In *Ultrasound Technologies for Food and Bioprocessing*. Editors: Hao Feng, Gustavo V. Barbosa-Canovas and Jochen Weiss. Springer, New York. 665p.

65. Womeni, H. M., Tchagna D. T, Nkouenkeu R., Kapseu C., Mbiapo F. T., Lindes M., Fanni J. et Parmentier, (2002) : « influence des traitements traditionnels des graines et amandes de karité sur la qualité du beurre ». *Food Africa :improving in Sub – Saharan*

66. Wu, H.W., Wang, Q. and Zhou, S.M. (2007). Research progress on peanut protein and its functional properties, *China Oils and Fats* 32 (9): 7–11.

67. Young, V. R. (1994). Adult amino acid requirements: The case for a major revision in current recommendations. *Journal of Nutrition*, 124(8 Suppl), 1517S–1523S.

68. Zhou, C.Z., Yang, T. (1993). Overall exploitation and utilization of silkworm pupae. Bulletin of Biology 28 (10): 44–47.