



## IN VIVO NUTRITIONAL ASSESSMENT OF FERMENTED CASSAVA SEMOLINA (ATTIÉKÉ) FORTIFIED WITH SOY FLOUR ON GROWING RAT (WISTAR)

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### ABSTRACT

This study aimed to contribute to the enhancement of *attiéké* nutritional balance by the incorporation of soy flour. Thirty rats teamed up into five per group, were fed during 21 days with six diets. These diets were a protein-free diet, a diet containing classic *attiéké*; three diets containing *attiéké* fortified with 5%, 9% and 12% of soy flour; and a control diet. At the end of experimental period, nutritional values of soy-fortified *attiéké* were determined, based on their biochemical composition, growth parameters and digestibility parameters of rats. The variation of biochemical components of soy-fortified *attiéké* indicates that protein (from 4.06 to 14.58%), fatty matters (from 1.92 to 8.51%), ashes (from 1.10 to 1.70 %) and fibers (from 4.00 to 13.50 %) contents increase significantly with the incorporated soy flour quantity. On the contrary, it shows a decrease in total carbohydrate content from 85.92 to 69.81 %. The nutritionnal parameters such as weight gain, dietary consumption index, dietary and protein efficiency coefficients of rats fed with the test diets were correlated with protein content. Their values were lower than those of control diet. In addition, the comparison biological values of diets, shows that there is no significant difference between classic *attiéké*, 5% soy-fortified *attiéké* and 9% soy-fortified *attiéké*. These diets have lower biological values than 12% soy-fortified *attiéké*. This study revealed that the consumption of *attiéké* fortified with 9% and 12% soy promotes a good growth of rats.

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### INTRODUCTION

Cassava tuberous roots (*Manihot esculenta* Crantz) plays an important role in human nutrition in Côte d'Ivoire. It's the second food mostly consumed by Ivorians with a production of 4.5 million tons in 2016 (MIADER, 2017). However, cassava has three major disadvantages: (i) toxicity associated with the presence of cyanogenic compounds; (ii) enormous post-harvest losses; and (iii) low protein content (Mlingi *et al.*, 1993). Though the first two constraints mentioned above have been resolved through innovative processing technologies and scientific research, the one related to protein value remains a major concern because of the persistence and the increase of protein-energy malnutrition in Africa (Stupak *et al.*, 2006; Guira, 2013). In Côte d'Ivoire, cassava was transformed into ten dishes of which the most known are *attiéké*, *placali*, *gari*, *attoukpou* and *tapioca* (Caransa, 1980; Diallo *et al.*, 2013).

*Attiéké* is a food product based on cassava mostly consumed in Côte d'Ivoire, by all social strata and at any age (Djéni *et al.*, 2014).

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Thus, the annual production of fresh *attiéké* in Côte d'Ivoire, was estimated to 30 000 tonnes and its consumption was 29 kg per inhabitant (Krahi *et al.*, 2015). *Attiéké* is known for its high starch, low protein and low micronutrient contents (Guira, 2013). The high consumption makes *attiéké* an ideal food-vector for protein fortification. Protein fortification of food products is one of the strategies to fight protein-energy malnutrition. It consists of protein incorporation into a widely used and consumed staple food to improve its nutritional balance (Berger, 2003, WHO / FAO, 2011).

The fortification of *attiéké* with vegetable protein sources, such as soybeans, could improve its nutritional quality. Indeed, soybean is an excellent source of proteins (40%), lipids (20%) (Zannou-Tchokoi *et al.*, 2011) and dietary fibers (18%) (Tu, 2010). The protein content of these seeds largely covers the need for essential amino acids of the human organism. Soybeans are among the oilseeds rich in polyunsaturated fatty acids, accounting for 54 to 72% of total lipids (Lecerf, 2011). Among them, linoleic acids (omega 6) and alpha-linolenic (omega 3) are the main essential fatty acids for the body because they cannot be synthesized (Demaison and Moreau, 2002). The nutritional composition could validly replace animal proteins (Lokuruka, 2010).

Several studies have been carried out on the improvement of cassava products nutritional properties such as gari, tapioca and cassava flour; but little on *attiéké*, except the studies of Essia *et al.* (2003) and Kouakou *et al.* (2018). This work aims to assess the nutritional value of *attiéké* with the soy flourbased on their biochemical composition and nutritional tests investigated on growing rats.

## MATERIALS AND METHODS

### Raw materials

Fresh cassava roots (*Manihot esculenta* Crantz) and yellow soybeans (*Glycine max* L. Meril) were used in this study. Cassava and soybeans were obtained from market of Bonoua (Côte d'Ivoire) and National Center of Agricultural Research (Côte d'Ivoire) respectively.

### Animal material

The animal material consisted of 30 growing rats of species *Rattus norvegicus* (*wistar*) weighing  $55 \pm 3$  g. They were obtained from the pet shop of Nutrition and Pharmacology Laboratory (UFR Biosciences of Felix Houphouët-Boigny University), Abidjan, Côte d'Ivoire.

### Methods

**Preparation of soy-fortified *attiéké*:** Soy-fortified *attiéké* was prepared according to the method described by Kouakou *et al.* (2018). The tuberous roots of cassava were peeled, were defibrated, were cut, were crushed and were mixed with the ferment (10% of cassava pulp; cooked cassava roots for 10 min and fermented for 48 hours). The pre-fermented dough for 2h was packed in the synthetic fibre bags and then wringed with a screw press until a mass dough was obtained. To this dough was added soy flour, which values were 5%, 9% and 12% of cassava pulp. The mixture of cassava-soy was fermented for 15h at room temperature to allow the development of aroma and taste as well as texture of soy-fortified *attiéké*. After the fermentation period, the dough was granulated, manually sieved and then partially sun dried. Two other sieves were carried out. The semolina was winnowed and steamed for 30 min in a couscous pot. The soy-fortified *attiéké* samples were dehydrated at 45°C for 60h.

**Biochemical Analysis:** Dry matter, ash, protein, fat and dietary fiber contents were determined using the AOAC methods (AOAC, 1990). Dry matter was determined by oven drying at 105°C for 24h and ash using a muffle furnace at 550°C for 24h. Crude protein was determined using the Kjeldahl method. Fat content was determined according to the Soxhlet method using hexane as solvent. Total carbohydrates were determined by difference of total material to other biochemical compounds. The energy value was calculated using the Atwater's calorie conversion factors: 4kcal/g for crude proteins, 9kcal/g for crude fats and 4kcal/g for carbohydrates. Dietary fibers were determined by acid and alkaline hydrolysis.

**Elaboration of diets:** Diets were prepared according to Pellett and Young (1980), with modifications (Table 1). Six (6) diets were developed: a protein-free diet based on cassava (RSP); a diet containing classic *attiéké* (RAN); three diets containing *attiéké* fortified with 5% soy (RAC<sub>5</sub>), 9% soy (RAC<sub>9</sub>) and 12% soy (RAC<sub>12</sub>) and a control diet based on fish flour (RFP).

**Table 1** Composition of rat diets

Ingredients (g)	RSP	RAN	RAC <sub>5</sub>	RAC <sub>9</sub>	RAC <sub>12</sub>	RFP
Soy-fortified <i>attiéké</i>	-	978	978	978	978	-
Fish flour	-	-	-	-	-	209
Cassava starch	948	-	-	-	-	739
Sunflower oil (ml)	30	-	-	-	-	30
Vitamin / mineral mixture	2	2	2	2	2	2
Sugar	20	20	20	20	20	20
Dry matter (g)	1000	1000	1000	1000	1000	1000
Protein (g/100g)	0	4	9	11	14	14

**Animal experimentation:** Five (5) rats per diet, were distributed based on their weight in individual metabolic cages. These cages were arranged into the room temperature and at the relative humidity. They make it possible to retain, upstream, the remains of food and feces (which prevents these two from polluting the urine). The urine was collected downstream in jars arranged under removable funnels attached to base of cages. The investigation of experiment was carried out according to the method reported by Adrian *et al.* (1998). It made over a period of twenty-one (21) days including two (2) days of adaptation period, where the animals are fed with a standard food (pellets) and a growth period of fourteen (14) days. In the last five (5) days, the nitrogen balance was measured. At the time of their distribution; the various diets of semolina were rehydrated in couscous (60 ml of water / 100 g of diet). The dry matter of reconstituted diets is measured daily. The next day, before making the distribution, the refusals were collected and were weighed. Clean water, frequently renewed, as well as food is served ad libitum to rats. Quantities of consumed food were obtained by difference between given quantities and refused quantities. These quantities are converted into ingested dry matters for each diet. Rats were weighed at the beginning of the experiment and then at two (2) days interval. The last weighing was carried out at the end of the experimental period. During the nitrogen balance period, consumption and growth measurements as well as faeces and urine collection were performed individually on rats. The total nitrogen was determined on the rat's faeces of diets and on the urine.

**Growth parameters of rats fed with the diets:** The ingested dry matter and the measured rat weight permit to establish three values: the consumption index (ICons); the dietary / protein efficiency coefficients (DEC / PEC) and the net dietary / protein efficiency coefficient (NDEC/NPEC) (Adrian *et al.*, 1998).

**Weight gain (WG):** The weight gain is the difference between final mass and initial mass of rats.

$$WG (g/j) = \frac{fm-im}{Nd} \quad (1)$$

im: initial mass

fm: final mass

Nd: number of days of experimentation

**Ingested total dry matter (ITDM):** The total ingested dry matter represents the total quantity of ingested food as dry matter consumed by the rat during the experimental period.

$$ITDM (g/j) = \frac{\sum IFDM}{Nd} \quad (2)$$

IFDM: ingested food as dry matter

Nd: number of experimentation days

**Consumption index (ICons):** The consumption index represents the ratio of food-ingested quantity and the weight gain of the rat. It reflects the overall food efficiency and the ration yield.

$$ICons = \frac{IFDM}{WG} \quad (3)$$

IFDM: ingested food as dry matter (g)  
WG: weight gain (g)

**Ingested total proteins (PTI):** The ingested total proteins represents the ingested dietary proteins quantity during the experimental period.

$$PTI (g/j) = \frac{IFDM \times PC}{Nd} \quad (4)$$

IFDM: ingested food as dry matter  
Nd : number of experimentation days  
PC : protein content of diet(g)

**Dietary efficiency coefficient (DEC):** The dietary efficiency coefficient reflects the performance of assimilated food.

$$DEC = \frac{WG}{IFDM} \quad (5)$$

WG: weight gain (g)  
IFDM: ingested food as dry matter (g)

**Protein efficiency coefficient (PEC):** The Protein efficiency coefficient represents the yield of diet protein utilization.

$$PEC = \frac{WG}{PTI} \quad (6)$$

WG: weight gain (g)  
PTI: Ingested total proteins(g)

**Net dietary efficiency coefficient (NDEC):** The net dietary efficiency coefficient value takes into account the weight loss of rats fed with protein-free diet.

$$NDEC = \frac{WG+WL}{IFDM} \quad (7)$$

WG: weight gain (g)  
WL: weight loss of rats fed with protein-free diet (g)  
IFDM: ingested food as dry matter (g)

**Net protein efficiency coefficient (CEPN):** The calculation of the net protein efficiency coefficient (NPEC) takes into account the weight loss of rats fed with protein-free diet.

$$NPEC = \frac{WG (g)+WL (g)}{PTI (g)} \quad (8)$$

WG: weight gain (g)  
WL: weight loss of rats fed with protein-free diet (g)  
PTI: Ingested total proteins(g)

### **Digestibility parameters of diets**

**Digestive utilization coefficient (CUD):** The digestive utilization coefficient is the ratio of absorbed proteins and ingested proteins when the metabolic faecal proteins are included in the calculation. The apparent digestive utilization coefficient (CUDa) and the true digestive utilization coefficient (CUDr) were determined according to the following formulas:

$$CUDa (\%) = \frac{PTI (g) - F (g)}{PTI (g)} \times 100 \quad (9)$$

$$CUDr (\%) = \frac{PTI (g) - (F - Fm)}{PTI (g)} \times 100 \quad (10)$$

CUDa: apparent digestive utilization coefficient  
CUDr: true digestive utilization coefficient  
PTI: Ingested total proteins(g)  
F: dietary faecal protein  
Fm : metabolic faecal protein

**Net protein utilization (UPN):** The net protein utilization represents the proportion of dietary protein that is retained by the organism.

$$UPN (\%) = \frac{PTI - (F - Fm) - (U - Um)}{PTI} \times 100 \quad (11)$$

PTI: Ingested total proteins(g)  
F: dietary faecal proteins(g)  
Fm: metabolic faecal proteins(g)  
U: dietary urinary proteins(g)  
Um: metabolic urinary proteins(g)

**Biological value (VB):** The biological value was determined according to the following formula:

$$VB (\%) = \frac{PTI - (F - Fm) - (U - Um)}{PTI - (F - Fm)} \times 100 \quad (12)$$

F: dietary faecal proteins(g)  
Fm: metabolic faecal proteins(g)  
U: food urinary proteins(g)  
Um: metabolic urinary proteins(g)  
PTI: Ingested total proteins(g)

### **Statistical analysis of data**

The statistical package in IBM SPSS STATISTICS 22.0 computer program was used. Data obtained were subjected to Analysis of Variance (ANOVA). Differences between means were evaluated using Duncan's test and significance accepted at  $\alpha=0.05$  level.

## **RESULTS AND DISCUSSION**

### **Biochemical composition of soy-fortified attiéké**

Table 2 shows the biochemical composition of soy-fortified *attiéké*. The dry matter content varies between 93.00 and 94.60 %. These values are higher compared to those of Yao *et al.* (2006) who found 89.40% for dehydrated *attiéké*. The low moisture content of dehydrated semolina inhibits the growth of microorganisms (Gnagne *et al.*, 2016), which promote better stability and extend the shelf life of food.

Protein, fat and fiber contents of soy-fortified *attiéké* increased significantly ( $p<0.05$ ) with incorporated soy flour. The values vary between 4.06 and 14.58 %; 1.92 and 8.51 %; 1.10 and 1.70 % and 4.00 and 13.50 %, respectively. The progressive increase of soy flour could explain observed changes because of its high protein, fat and dietary fiber contents. These results are in accordance with those of Ezinwanyi and Ndaeyo (2017) who reported an increase in protein content with addition of the soy flour.

According to Labat (2013), soybean contains proteins of good biological value. Thus, the incorporation of soybeans could improve the content and the quality of fortified *attiéké* proteins. The increase in protein content of *attiéké* could promote good growth of children. In addition to providing energy, consumption of the soy-fortified *attiéké* could have other advantages. As reported to Demaison and Moreau

(2002), omega-type fatty acids present in soybean are responsible of the cardiovascular and immune balance. The presence of lipids in a food is essential to prevent the deficiency of fat-soluble vitamins; particularly vitamin A. Inadequate lipid intake may affect the intestinal absorption, availability and metabolism of vitamin A. The dietary fibers facilitate intestinal transit and faecal evacuation. Moreover, fibers decrease blood cholesterol, glycemia and prevent colon cancer (Barakat and Ghazal, 2016).

The ash content reflects the mineral composition of a food. Ash contents of classic *attiéké* (1.10 %) and *attiéké* fortified with 5% soy (1.20 %) were not significant different between them and were lower than those of *attiéké* fortified with 9% soy (1.60 %) and *attiéké* fortified with 12% soy (1.70 ± 0.10%) which were not significant different between them. However, the fortification of *attiéké* results into a decrease of the total carbohydrate content, from 85.92 to 69.81 %. As for energy values of studied diets, values were ranged between 377.20 and 414.15 kcal/100 g. Carbohydrates are the body's first calorie source. All samples are excellent calorie sources because *attiéké* is a good source of starch and soybean is a good source of fat (Kouakou *et al.*, 2018).

**Table 2** Biochemical composition of the soy-fortified *attiéké*

Samples	Classic <i>attiéké</i>	AC <sub>5</sub>	AC <sub>9</sub>	AC <sub>12</sub>
	Content in g / 100 g of dry matter			
Dry matter	93,00±0,00 <sup>a</sup>	93,80±0,00 <sup>b</sup>	93,90±0,10 <sup>b</sup>	94,60±0,00 <sup>c</sup>
Protein	4,06±0,44 <sup>a</sup>	9,01±0,43 <sup>b</sup>	11,23±0,25 <sup>c</sup>	14,58±0,00 <sup>d</sup>
Fat	1,92±0,04 <sup>a</sup>	4,65±0,44 <sup>b</sup>	5,51±0,28 <sup>c</sup>	8,51±0,02 <sup>d</sup>
Ash	1,10±0,10 <sup>a</sup>	1,20±0,00 <sup>a</sup>	1,60±0,10 <sup>b</sup>	1,70±0,10 <sup>b</sup>
Carbohydrates	85,92±0,05 <sup>d</sup>	78,94±1,12 <sup>c</sup>	75,56±0,50 <sup>b</sup>	69,81±0,44 <sup>a</sup>
Dietary fiber	4,00±1,00 <sup>a</sup>	9,75±0,25 <sup>b</sup>	11,25±1,25 <sup>b</sup>	13,50±0,00 <sup>c</sup>
Energy value	377,20±0,60 <sup>a</sup>	393,65±2,20 <sup>b</sup>	396,73±1,82 <sup>c</sup>	414,15±0,30 <sup>d</sup>

Values are expressed as means±standard deviation; means with different letters are significantly different (p< 0.05) along the lines.

AC<sub>5</sub> : *attiéké* fortified with 5% soy; AC<sub>9</sub> : *attiéké* fortified with 9% soy; AC<sub>12</sub> : *attiéké* fortified with 12% soy

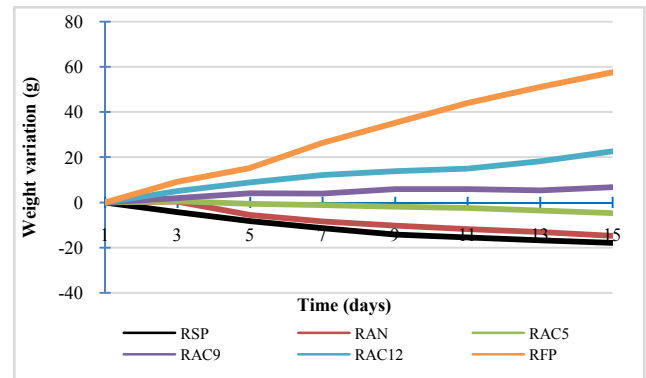
**Parameters of rat growth fed with the diets**

Results of growth parameters of rats fed with diets are summarized in the table 2. There were significant differences between control diet (RFP), protein-free diet (RSP) and test diets (RAN, RAC<sub>5</sub>, RAC<sub>9</sub> and RAC<sub>12</sub>).

Weight evolution of rats fed during 15 days with diets is shown in figure 1. The weight variation was significantly different (p<0.05) with the control diet (3.85 g / rat / day), followed by RAC<sub>12</sub> diet (1.51 g / rat / day), RAC<sub>9</sub> diet (0.45 g / rat / day), RAC<sub>5</sub> diet (-0.32 g / rat / day), RAN (-0.98 ± 0.08 g) / rat / day) and RSP diet (-1.20 ± 0.15 g / rat / day). The weight losses were not significant different between RSP and RAN diets.

Growth results highlight that RSP and RAN diets cause weight loss and food intake loss; test diets (RAC<sub>9</sub> and RAC<sub>12</sub>) increase the weight and food intake of rats. Indeed, rats fed with low protein diets (RSP and RAN) consumed less food than control rats (RFP) and rats fed with RAC<sub>9</sub> and RAC<sub>12</sub> diets. The decrease of the consumption of protein-poor diets seems to be a consequence of this weight loss. Furthermore, the high consumption of RAC<sub>9</sub>, RAC<sub>12</sub> and RFP diets led to significant weight gain. These performances could be explained by the protein content higher than 9% of these diets. According to Adrian *et al.* (1998), protein requirements of rat are between 9 and 18% of proteins. These results were in

accordance with those of Dally *et al.* (2010) who observed that the highest weight gain was obtained with the food having the highest protein content.



**Figure 1** Weight evolution of rats fed with diets for 15 days of experimentation Values correspond to mean of five independent measurements (n=5).

RSP : protein-free diet ; RAN : diet containing classic *attiéké* ; RAC<sub>5</sub> : diet containing *attiéké* fortified with 5% soy ; RAC<sub>9</sub> : diet containing *attiéké* fortified with 9 % soy ; RAC<sub>12</sub> : diet containing *attiéké* fortified with 12 % soy ; RFP : control diet based on fish flour.

The ingested food as dry matter (IFDM) and ingested total protein (PTI) are shown in table 3. Rats fed with RSP and RAN diets have the IFDM of 4.24 g / day for RSP and 4.04 g / day for RAN, that are not significantly different. These values are lower those of rats fed with RAC<sub>5</sub> diet (5.72 g / day), RAC<sub>9</sub>diet (6.99 g / day), RAC<sub>12</sub>diet (8 g / day) and control diet (10.32 ± 0.62 g / d). With the classic *attiéké*, ingested protein content displays lower values (0.16 g / d) than those of RAC<sub>5</sub> diets (0.52 g / day), RAC<sub>9</sub> (0.77 g / day), RAC<sub>12</sub> (1.23 g / day) and control diet (1.45 g / day). There are significant differences (p< 0.05) between these values.

The IFDM and the PTI of rats fed with test diets and control diet increase significantly with the protein content (p< 0.05). These results are in accordance with those of Serna-Saldivar *et al.* (1999) and Bouafou *et al.* (2011) who reported that higher consumption values obtained with rats fed with control diet compared to those receiving diets containing classic *attiéké* and soy-fortified *attiéké*. These results are in contradiction with those of Méité *et al.* (2008) who did not report a significant difference (p> 0.05) between quantities of consumed diests IFDM and PTI.

Food consumption depends of several factors, including the physiological state of organisms and related factors to food characteristics, such as aroma, flavor and chemical composition (Dally *et al.*, 2010). However, very low values that were measured on rats fed with the protein-free diets (RSP), confirm that the nutritional balance between protein and energy is an essential factor to control consumption and weight gain (Bouafou *et al.*, 2007).

The consumption index (ICons) reflects the overall food efficiency and quantifies the ration yield. The weakest result reflects the better performance. Thus, the control diet (2.73) has the lowest ICons, followed by RAC<sub>12</sub>diet (5.18) and finally by RAC<sub>9</sub>diet (14.00). There are no significant differences (p> 0.05) between RAC<sub>12</sub> diet and control diet. However, ICons alone cannot predict the efficiency of proteins because the consumed quantity is not always the assimilated quantity. For this reason, other calculations are made using the ingested and used protein content.

Table 3 gives the dietary efficiency coefficient (DEC), net dietary efficiency coefficient (NDEC), protein efficiency coefficient (PEC), net protein efficiency coefficient (NPEC) values of studied diets. The highest DEC values were obtained with rats fed with the control diet (0.37). The incorporation of soy flour resulted an improvement of the DEC values of RAC<sub>9</sub> diet (0.07 ± 0.01) and RAC<sub>12</sub> diet (0.20 ± 0.02) compared to the classic *attiéké* diet and RAC<sub>5</sub> diet (- 0.23 and -0.04). The DEC ranged in decreasing order is RFP> RAC<sub>12</sub>> RAC<sub>9</sub>.

The control diet has the highest PEC (2.63) compared to test diets. Diets containing soy-fortified *attiéké* have high PEC than the classic *attiéké* diet (-5.73). PEC values of the RAC<sub>9</sub> diet (0.66) and the RAC<sub>12</sub> diet (1.39) have significant differences (*p*<0.05) between them, but are both greater than the RAC<sub>5</sub> diet (-0, 39). NDEC values of test diets represent 8.51%, 31.91%, 48.94% and 65.96% of control group for classic *attiéké*, RAC<sub>5</sub>, RAC<sub>9</sub> and RAC<sub>12</sub> diets, respectively. However, differences between the NPEC values of RAC<sub>5</sub>, RAC<sub>9</sub> and RAC<sub>12</sub> diets are not significant (*p*<0.05).

The DEC permits a better appreciation of the ingested food utilization yield. The efficiency of proteins for growth can be estimated by the calculation of NPEC which takes into account the weight loss of rats fed with the protein-free diet (RSP) (Kenfack, 2010). Obtained values with test diets represent more than 50% of NPEC compared to the control diet (50% for RAC<sub>5</sub>, 61.01% for RAC<sub>9</sub> and 66.69% for RAC<sub>12</sub>), except classic *attiéké* diet, but there are not significantly different (*p*<0.05). These results are lower to those of Méité *et al.* (2008) with bread fortified with cucurbits. The nature of fortified food (*attiéké*) could be at the origin of these lower values compared to the bread. Indeed, *attiéké* (4% of proteins) is very low in protein compared to the bread (9.42% of proteins).

**Table 3** Measurement parameters of rat growth

Diets	RSP	RAN	RAC <sub>5</sub>	RAC <sub>9</sub>	RAC <sub>12</sub>	RFP
WG (g / j)	-1,20 <sup>a</sup> ±0,15	-0,98 <sup>a</sup> ±0,08	-0,32 <sup>b</sup> ±0,02	0,45 <sup>c</sup> ±0,11	1,51 <sup>d</sup> ±0,11	3,85 <sup>e</sup> ±0,56
IFDM (g / j)	4,24 <sup>a</sup> ±0,13	4,04 <sup>a</sup> ±0,27	5,72 <sup>b</sup> ±0,23	6,99 <sup>c</sup> ±0,72	8,80 <sup>d</sup> ±0,20	10,32 <sup>e</sup> ±0,62
PTI (g / j)	0,00 <sup>a</sup> ±0,00	0,16 <sup>b</sup> ±0,01	0,52 <sup>c</sup> ±0,02	0,77 <sup>d</sup> ±0,08	1,23 <sup>e</sup> ±0,03	1,45 <sup>f</sup> ±0,08
ICons	-	-4,37 <sup>b</sup> ±0,27	-35,25 <sup>c</sup> ±17,37	14,00 <sup>d</sup> ±1,57	5,18 <sup>e</sup> ±0,51	2,73 <sup>f</sup> ±0,29
DEC	-	-0,23 <sup>a</sup> ±0,01	-0,04 <sup>b</sup> ±0,02	0,07 <sup>c</sup> ±0,01	0,20 <sup>d</sup> ±0,02	0,37 <sup>e</sup> ±0,03
PEC	-	-5,73 <sup>a</sup> ±0,37	-0,39 <sup>b</sup> ±0,23	0,66 <sup>c</sup> ±0,06	1,39 <sup>d</sup> ±0,14	2,63 <sup>e</sup> ±0,26
NDEC	-	0,04 <sup>a</sup> ±0,03	0,15 <sup>b</sup> ±0,02	0,23 <sup>c</sup> ±0,00	0,31 <sup>d</sup> ±0,02	0,47 <sup>e</sup> ±0,03
NPEC	-	0,87 <sup>a</sup> ±0,78	1,68 <sup>b</sup> ±0,28	2,05 <sup>b</sup> ±0,08	2,25 <sup>b</sup> ±0,16	3,36 <sup>c</sup> ±0,24

Values expressed as means±standard deviation; means with different letters are significantly different (*p*< 0.05) along the lines.

WG : weight gain, IFDM : ingested food as dry matter, PTI : protéines totales ingérées, ICons : indice de consommation, DEC : dietary efficiency coefficient, PEC : protein efficiency coefficient, NDEC : net dietary efficiency coefficient, NPEC : net protein efficiency coefficient.

RSP: protein-free diet; RAN: diet containing classic *attiéké*; RAC<sub>5</sub>: diet containing *attiéké* fortified with 5% soy ; RAC<sub>9</sub> : diet containing *attiéké* fortified with 9% soy ; RAC<sub>12</sub> : diet containing *attiéké* fortified with 12% soy ; RFP : control diet based on fish flour

***In vivo* digestibility of diets**

Figure 2 shows results of apparent digestibility coefficient (CUDA), true digestibility coefficient (CUDr), net protein utilization (UPN) and biological value (VB). These results indicate that the control diet (RFP) has the highest CUDA (86.22) compared to the test diets. The CUDA of RAN diet (71.99 %) and RAC<sub>9</sub> diet (74.67 %) do not show any significant difference and are significantly lower (*p*<0.05) than those of RAC<sub>5</sub> diet (77.50 %) and RAC<sub>12</sub> diet (82.08 %). True digestibilities (CUDr) are higher than those of apparent digestibilities (ranged from 77.68 to 86.28 against from 71.99 to 86.22%, respectively) because of corrections made by the

faeces nitrogen of rats fed with the RSP diet. Thus, protein digestibilities of the control diet, RAC<sub>12</sub> and RAN diets are significantly (*p*<0.05) higher than those of RAC<sub>5</sub> and RAC<sub>9</sub> diets. The lowest value was obtained with the RAC<sub>9</sub> diet.

It appears that the control diet based on fish flour (RFP) had nutritional qualities higher than those of test diets. Values of CUDr (87.82 %) and VB (84.97 %) of control diet (14% of proteins) are lower than those of Bouafou *et al.* (2007) who obtained CUDr values of 89% and VB of 87%. They are also lower than those of Bouafou *et al.* (2008) and Méité *et al.* (2017) that used the casein diet as the control (10% protein). This superiority of casein diet is explained by its good biological value. However, the control diet (RFP) has a better biological value compared to test diets. Indeed, fish proteins have balanced essential amino acids that have high digestibility (Apfelbaum *et al.*, 2004).

The digestive utilization coefficient (CUD) characterizes the ability of a protein to be hydrolyzed by proteolytic enzymes and to be absorbed through the intestinal mucosa (Adrian *et al.*, 1998). The nutritional value of a protein is estimated by the percentage of ingested nitrogen used for protein synthesis. Soybeans are rich in dietary fibers. These fibers could have two opposite effects, firstly to reduce the enzymatic activity in the digestive tract and secondly, to facilitate the intestinal transit of consumed food. Kenfack (2010) stated in his study that fibers have a high hydration capacity. At very large quantities, they reduce intestinal absorption by increasing endogenous losses.

The net protein utilization (UPN) measures the proportion of ingested protein that is retained for tissue synthesis. It takes into account both the digestive and metabolic utilization of dietary proteins (Pellet and Young, 1980). The histogram in figure 2 shows that the UPN of diets varies from 54.97 % (RAN group) to 65.44% (RAC<sub>9</sub> group). The control diet has a high value of UPN (74.43 %). The UPN values of diets containing soy-fortified *attiéké* fortified are higher than that of the classic *attiéké* diet, except the RAC<sub>5</sub> diet. The control diet had the highest biological value (84.76 %) compared to diets containing classic *attiéké* and soy-fortified *attiéké*. The comparison of biological values is no significant but shows difference between RAN, RAC<sub>5</sub> and RAC<sub>9</sub> diets. In contrary, the results show differences between RAN, RAC<sub>5</sub> and RAC<sub>9</sub> diets, RAC<sub>12</sub> diet and the control diet (RFP) (*p*<0.05). These observations confirm that the nutritional value of dietary proteins depends both on their availability and on their essential amino acid balance (Apfelbaum *et al.*, 2004).

Previous studies on the protein sources (fish and soybean) indicated that they had balanced essential amino acid contents (Labat, 2013). However, soy contains antinutrients that may prevent protein absorption (Habtamu and Negussie, 2014). In this study, soy flour was submitted to treatments like fermentation, drying and cooking at the same time as cassava. Under these conditions, the anti-nutritional factors should not play a major role in protein digestibility during *in vivo* test.

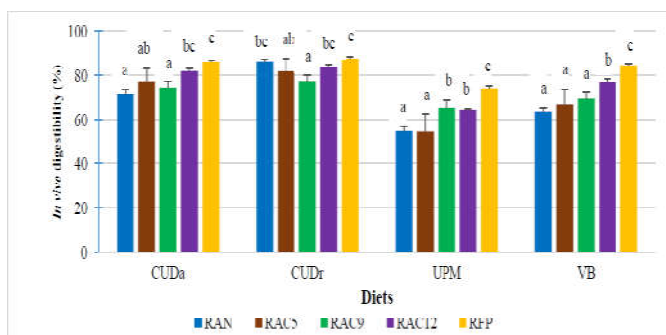


Figure 2 In vivo digestibility of diets

Error bars indicate the standard deviation of five determinations (n = 5). Values followed by different letters are significantly different (p<0.05).

CUDa : apparent digestive utilization coefficient, CUDr : true digestive utilization coefficient, UPM : net protein utilization et VB : biological value.

RSP : protein-free diet ; RAN : diet containing classic attiéké ; RAC<sub>5</sub> : diet containing attiéké fortified with 5% soy ; RAC<sub>9</sub> : diet containing attiéké fortified with 9 % soy ; RAC<sub>12</sub> : diet containing attiéké fortified with 12 % soy ; RFP : control diet based on fish flour.

### Conflict of Interest

The authors have no conflicts of interest to declare.

### CONCLUSION

Attiéké (semolina of cassava) fortified with proteins by incorporation of the soy flour showed improved balance of protein, fat, ash and fiber contents. In vivo study on growing rats indicated that diets containing attiéké fortified with 9% soy and 12% soy diets promote a good rat growth as much as the control diet. These results also suggested that without intake of another protein source, consumption of classic attiéké led to weight loss in rats. The 5% soy-fortified did not allow the growth of rats, but allowed the maintenance of their weight. The data of food intake highlighted the increase of consumption index with the protein content of diets. In vivo digestibility parameters of these diets were lower than of control diet.

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