

Nutrient content of earthworms consumed by Ye'Kuana Amerindians of the Alto Orinoco of Venezuela

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For the Makiritare (Ye'Kuana) native people of the Alto Orinoco (Venezuela), earthworms (Anellida: Glossoscolecidae) are an important component of the diet. Two species in particular are widely consumed: 'kuru' (Andiorrhinus kuru n. sp.) and 'motto' (Andiorrhinus motto). We analysed eviscerated kuru body proper, and whole and smoked preparations of motto for their content of protein and amino acids, fatty acids and 20 minerals and trace elements.

The samples contained large amounts of protein (64.5–72.9% of dry weight), essential amino acids, calcium and iron together with notable quantities of other important elements, indicating that these earthworms contain potentially useful quantities of many nutrients that are critical to the health of the humans who consume them.

Keywords: edible earthworms; Glossoscolecidae; arachidonic acid; calcium; Alto Orinoco; Ye'Kuana

1. INTRODUCTION

Although Wallace (1853, 1889) reported more than 100 years ago the inclusion of earthworms in the diets of particular Amerindian populations of the Rio Negro of Brazil, little is known about the nutritive value of these edible invertebrates. Schlenker (1974) reported that the Makiritare (autodenomination is Ye'Kuana or Yekuana) people of the Alto Orinoco of southern Venezuela regard some earthworms as a highly desirable food. However, other ethnic groups in the region, including the Yanomamo, Curripaco, Hivi (or Guahibo), Baniwa, Piapoco and Hoti, do not consume earthworms (M. G. Paoletti, personal observation).

The Yekuana traditionally gather several different species of earthworm (Anellida: Oligochaeta: Glossoscolecidae), including the white worm called 'motto', which lives in the mud of streams and river-banks (figure 1), and a larger species they call 'kuru', which inhabits the floor of the highland forest (figure 2). The Piaroa of Caño Tigre also collect an earthworm named gua or wua (Zent 1992; Paoletti et al. 2000), which is probably similar to motto (Andiorrhinus motto; Righi & Araujo 1999). In 1996–1999, M. G. Paoletti and coworkers documented the collection of motto by local populations inhabiting the settlements of Toki and Guatamo, which are situated on the Padamo River in southern

Venezuela (Paoletti *et al.* 2000). They reported that both kinds of earthworm, *motto* and *kuru*, are consumed either fresh after heating in water at 60–80 °C (figure 3), or after they have been smoked over a wood fire. The smoked variety (figure 4) is considered a delicacy and has a high commercial value, commanding, on a weight basis, three times the price of smoked fish, game, pork or chicken.

Enquiries have revealed that among the inhabitants of Guatamo, Toki and Buena Vista earthworms are recommended by local healers for people with malaria or anaemia, or for women after parturition. In fact, among the Yekuana, tradition prescribes that for at least the first month following parturition the mother should consume a diet composed of only cassava and earthworms (Schlenker 1974; personal observations in Toki, Guatamo and Buena Vista villages—Paoletti, Cerda and Torres).

Reliance of these women on earthworms as the principal dietary protein source caused us to question the nutritional quality of earthworms, in particular their iron content. Because we had recently completed a study of the amino-acid, mineral, trace-element and essential-fatty-acid content of the mopane worm of southern Africa (Glew *et al.* 1999), we were in a position to conduct a similar analysis of the two widely consumed edible earthworms of the Alto Orinoco, namely *motto* and *kuru*. In this paper, we report the results of our analysis of the amounts of 18 of the common amino acids that constitute proteins, 20 minerals and trace elements and the nutritionally relevant ω -3 and ω -6 fatty acids, linoleic acid, α -linolenic

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mo, which are situated on the Padamo River in souther

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Figure 1. A *motto* specimen from Toki village (photograph: M. G. Paoletti).



Figure 2. A *kuru* specimen from Buena Vista village (photograph: M. G. Paoletti).

acid, arachidonic acid and docosahexaenoic acid in kuru, motto and smoked motto.

2. MATERIAL AND METHODS

(a) Source of earthworms

The motto samples (A. motto; Righi & Araujo 1999) were collected by M.G.P., H.C. and F.T. in Guatamo village (Rio Padamo, Municipio Alto Orinoco, Estado Amazonas, Venezuela) on 14 January 1999 near the village in small stream banks. Fresh specimens of adult motto weighed 16-49 g, and were 25-45 cm long. The smoked motto (figure 4) was purchased in Guatamo on the same day. The kuru samples (Andiorhinus kuru n. sp.; Moreno & Paoletti 2002) were collected in Buena Vista, Municipio Alto Orinoco, Estado Amazonas, Venezuela (3°41'23.5" N, 65°13'4.3" W) on 15 January 1999. Living adult specimens weighed 120-185 g and were 50-70 cm long. The two species were preserved in liquid nitrogen during the trip back to Caracas. However, the motto were first purged of soil content by squeezing the gut. As for the kuru, the internal organs and gut were completely removed. The gut organs are not consumed by the Yekuana. Once in Caracas, both motto and kuru were freezedried and later analysed. The smoked motto samples had been squeezed by the collectors to eliminate gut content and then smoked overnight by suspending them over a fireplace in Guatamo.

(b) Lipid extraction and fatty-acid analysis

The dried powdered specimens were extracted with chloroform-methanol (2:1, v/v) as described elsewhere (Chamberlain et al. 1993) and the solid non-lipid material was removed by filtration. The total extracted lipid material was recovered after solvent removal in a stream of nitrogen. The samples were then redissolved in anhydrous chloroform-methanol (19:1, v/v) and clarified by centrifugation at 10 000 xg for 10 min. Transmethylation was performed using 14% (w/v) boron trifluoride (BF₃) in methanol (Morrison & Smith 1964). We then transferred 50 ng of heptadecanoic acid (internal standard) and a 1 ml aliquot of each sample to a 15 ml teflon-lined screw-cap tube. After removal of the solvent by nitrogen gassing, the sample was mixed with 0.5 ml of the BF₃ reagent, placed in a warm bath at 100 °C for 30 min and cooled. After the addition of a saline solution, the transmethylated fatty acids were extracted into hexane. A calibration mixture of fatty-acid standards was processed in parallel.

Aliquots of the hexane phase were analysed by gas chromatography. Fatty acids were separated and quantified using a Hewlett-Packard gas chromatograph (5890 Series II) equipped with a flame-ionization detector. We injected 1-2 µl aliquots of the hexane phase in split-mode onto a fused-silica capillary column (Omegawax; 30 m × 0.32 mm internal diameter, Supelco, Bellefonte, PA, USA). The injector temperature was set at 200 °C, the detector at 230 °C, and the oven at 120 °C initially, then at 120-205 °C at 4 °C min⁻¹, and at 205 °C for 18 min. The carrier gas was helium and the flow rate was ca. 50 cm s⁻¹. Electronic pressure control in the constant-flow mode was used. The internal standard (heptadecanoic acid, 17:0) and calibration standards (Nu-chek, Elysian, MN, USA) were used for quantitation of fatty acids in the various lipid extracts. Solvents were purchased from EM Science, Gibbstown, NJ, USA. The fattyacid data reported represent the average of three determinations.

(c) Amino-acid analysis

Two separate samples of each earthworm specimen were analysed for amino acids. We weighed out 2–3 mg of the dry powdered specimen into two separate hydrolysis vials and digested the samples separately. The average of the two determinations is reported in table 1. Norleucine, an amino acid not commonly found in proteins, was the internal standard used in all determinations. After 1.0 ml of 6 N HCl was added, the samples were flushed with nitrogen, evacuated, sealed and placed in an oven at 110 °C for 24 h. Following hydrolysis, a 10 μl aliquot was withdrawn and subjected to derivatization.

Samples to be used for the determination of cysteine were first oxidized with performic acid (9 volumes 80% formic acid: 1 volume 30% hydrogen peroxide) for 18 h at room temperature (Hirs 1967). Performic acid was removed in an evaporative centrifuge and the samples were hydrolysed with HCl as described above.

The tryptophan content was determined separately by adding 450 μl of 4.67 M KOH containing 1% (w/v) thiodiglycol to each sample (Hugli & Moore 1972). Hydrolysis was performed in plastic tubes within an evacuated ampoule at 110 °C for 24 h. After allowing the hydrolysate to cool, 0.5 ml of 4.2 M perchloric acid and 50 μl of acetic acid were added to neutralize the solution. The samples were mixed thoroughly using a Thermolyne Maxi mixer, chilled on ice and centrifuged. We then



Figure 3. Kuru (above) and motto (below) preparation (Schlenker 1974).



Figure 4. Smoked motto from Guatamo village (photograph: M. G. Paoletti).

transferred 15 μl of the supernatant to 6 mm \times 50 mm glass tubes and dried the samples in a speedvac in preparation for derivatization. Duplicate lysozyme controls were analysed for quality-control purposes.

The samples were dissolved using 20 µl of ethanol-triethylamine-water (2:1:2, v/v) and derivatized with 20 μl phenylisothiocyanate reagent (ethanol-triethylamine-water-phenylisothiocyanate (7:1:1:1, v/v) for 20 min at room temperature. Excess reagent was removed in a speedvac. Derivatized and dried samples were dissolved in 100 µl of equilibration buffer.

Analysis of the amino acids was performed with a Waters C₁₈ column (3.9 mm × 150 mm). The gradient solution was the same as that described by Bidlingmeyer et al. (1984). The solvents used were the (a) sodium acetate buffer and (b) acetonitrile (300 ml acetonitrile, 200 ml water, 0.2 ml calcium disodium edetate). We injected 20 µl aliquots onto the column. Tryptophan analysis was performed according to Hariharan et al. (1993). Elution of the amino acids was achieved by increasing

the acetonitrile concentration in the eluent, causing individual amino acids to be eluted at predetermined times. Quantitation was achieved by monitoring the absorption of the column effluent at 254 nm and comparing the absorbance of individual peaks with that of the corresponding amino-acid standard.

(d) Mineral analysis

The powdered samples were dried overnight at 110 °C. Only a single sample of each motto and fresh kuru was available and seven different smoked motto specimen replicates were made. Aliquots containing ca. 0.1 g of each sample were weighed into 125 ml Phillips beakers and then digested using 20 ml of concentrated nitric acid and 1 ml of concentrated perchloric acid. The samples were covered with watch glasses and set on a hotplate at 120 °C for 1 h. The hotplate temperature was then increased to 150 °C and the samples were refluxed overnight. The watch glasses were removed and the samples taken to near dryness (ca. 1 ml) at the same temperature. At that point, the samples were removed from the hotplate and treated with 2.5 ml of nitric-perchloric acid (4:1, v/v) and a minimal amount of deionized water to rinse down the walls of the beakers. After cooling, the solutions were quantitatively transferred to graduated centrifuge tubes and diluted to 50 ml final volume with deionized water. The samples were analysed by inductively coupled argon plasma atomic emission spectroscopy (ICP-AES, Jarrel-Ash) for trace metal content as described elsewhere (Yazzie et al. 1994; Kim et al. 1997) and quantified against standard solutions of known concentrations that were analysed concurrently. This digestion technique makes no attempt to solubilize any silicate-based materials that may be in the samples.

3. RESULTS

(a) Protein content and amino-acid composition

The total protein content of the earthworms, calculated by summing the quantities of the individual amino acids,

Table 1. Amino-acid content of *motto* and *kuru* (mg g^{-1} dry weight). (The values reported are the means of two determinations on two different samples.)

amino acid	kuru body $(n = 2)$	kuru gut organs ^a $(n=2)$	motto body $(n = 2)$	motto smoked $(n = 2)$
asp	71.3	35.5	62.5	68.1
thr	34.2	23.2	30.1	32.4
ser	35.8	18.6	32.2	34.8
glu	124	66.2	107	109
pro	26.2	15.9	23.2	23.1
gly	49.4	28.2	39.1	34.2
ala	42.9	26.6	37.1	36.9
val	33.9	21.4	31.5	32.5
met	17.0	11.2	14.4	16.0
ile	33.8	25.3	29.6	30.5
leu	62.0	34.9	55.3	55.4
tyr	21.4	9.52	20.2	19.9
phe	28.9	18.3	26.7	27.3
his	18.7	11.1	15.8	14.7
lys	54.2	32.1	49.7	48.5
arg	60.5	32.9	55.7	53.5
cys	7.17	5.02	5.81	5.86
trp	8.23	5.54	8.51	9.64
total protein content	729	421	644	653

^a Parts not eaten.

Table 2. Essential-amino-acid content of motto and kuru compared with the WHO ideal protein.

	pe	percentage of (amino acid/ideal) × 100%			
amino acid	motto smoked	motto body	kuru body	kuru gut organs ^a	WHO ideal protein ^b
his	118	129	135	228	1.9
ile	167	164	165	214	2.8
leu	129	130	129	125	6.6
lys	128	133	128	131	5.8
met+cys	139	131	128	154	2.5
phe+tyr	115	116	109	105	6.3
thr	146	137	138	162	3.4
trp	134	120	103	119	1.1
val	142	140	133	145	3.5

^a Parts not eaten.

was very high. *Kuru* and *motto* (heated in water or smoked) contained 64.4–72.9% protein, and protein accounted for 42.1% of the dry weight of the gut organs (which are not eaten) of *kuru* (table 1). By comparing the essential-amino-acid content of a sample protein with that of the World Health Organization (WHO) standard protein, one can calculate the protein's chemical score and identify limiting amino acids in the nutritional source (table 2). In all cases, the earthworm proteins were of high quality, comparable with those of cows' milk and eggs.

(b) Lipid and fatty-acid analysis

Table 3 reports on a mg g^{-1} dry weight basis the fatty-acid compositions of the various earthworm specimens. The total fatty-acid content of the earthworm preparations was very low and ranged from 6.6 to 10.5 mg g^{-1} dry weight. When the fatty-acid data were expressed in terms of the mass of each fatty acid per gram of dry weight of tissue, the relatively small contribution fatty acids make to the overall mass of the earthworms became apparent. In

none of the four earthworm preparations reported in table 3 did fatty acids account for more than $10.5~{\rm mg~g^{-1}}$ dry weight of the specimen. Thus, fatty acids accounted for only 0.66-1.05% of the dry weight of the earthworms. The fatty-acid percentage compositions of the four specimens were similar.

There are several remarkable aspects of the fatty-acid composition, as shown in table 4. First, the proportion of polyunsaturated fat was relatively high (46.7–54.2%). Second, within the polyunsaturated class of fatty acids, there was a much higher proportion of ω -6 fatty acids than ω -3 fatty acids. In fact, except for a moderately high percentage (e.g. 0.30% in *kuru*) of the ω -3 fatty acid eicosapentaenoic acid, ω -3 fatty acids were not well represented in the overall fatty-acid profile. Third, arachidonic acid (20:4 ω -6) represented 33–45% of the polyunsaturated fatty-acid content and nearly one-quarter (15.7–23.0%) of the fatty-acid total. The content of arachidonic acid on a percentage basis in these edible earthworms is very high when compared with that of some common foods, such as

ь Per cent of total.

Table 3. Fatty-acid content of *motto* and *kuru* (mg g^{-1} dry weight). (Abbreviation: s.d., standard deviation.)

fatty acid		<i>kuru</i> body (<i>n</i> = 1)	kuru gut organs $(n = 1)$	motto body $(n = 1)$	<i>motto</i> smok	$ \begin{array}{c} \text{red } (n=7) \\ \text{s.d.} \end{array} $
saturated						
10:0	capric acid	0.01	0.03	0.01	0.07	0.04
12:0	lauric acid	0.00	0.02	0.00	0.00	0
14:0	myristic acid	0.05	0.09	0.07	0.07	0.024
15:0	pentadecanoic acid	0.05	0.03	0.07	0.06	0.007
16:0	palmitic acid	0.40	0.51	0.37	0.42	0.134
18:0	stearic acid	1.14	0.92	0.47	0.71	0.095
20:0	eicosanoic acid	0.05	0.05	0.04	0.05	0.007
22:0	behenic acid	0.14	0.10	0.07	0.12	0.022
24:0	lignoceric acid	0.06	0.09	0.03	0.02	0.019
subtotal		1.91	1.84	1.13	1.52	
monounsaturated						
14:1	myristoleic acid	0.01	0.01	0.00	0.00	0
16:1	palmitoleic acid	0.39	0.05	0.51	0.31	0.177
18:1 ω-9	oleic acid	1.50	1.02	0.39	0.96	0.348
$18:1 \omega - 7$	vaccenoic acid	0.44	0.39	0.49	0.70	0.119
18:1 ω-5	D13-octadecenoic acid	0.07	0.06	0.04	0.08	0.017
$20:1 \omega - 9$	D11-eicosenoic acid	0.79	0.80	0.90	1.14	0.132
$20:1 \omega - 7$	D13-eicosenoic acid	0.01	0.02	0.02	0.00	0.008
$22:1 \omega - 9$	euric acid	0.03	0.05	0.03	0.09	0.043
24:1	nervonic acid	0.00	0.07	0.00	0.00	0
subtotal		3.24	2.46	2.37	3.28	
polyunsaturated						
$18:2 \ \omega-6$	linoleic acid	0.97	0.91	0.51	0.80	0.413
18:3 ω-6	γ-linolenic acid	0.05	0.04	0.01	0.05	0.02
18:3 ω-3	α-linolenic acid	0.08	0.06	0.11	0.09	0.018
20:2 ω-6	D11.14-eicosadienoic acid	0.36	0.48	0.29	0.23	0.019
$20:3 \omega - 6$	dihomo-g-linolenic acid	0.72	0.66	0.53	0.71	0.07
20:4 ω-6	arachidonic acid	2.41	2.09	1.04	1.60	0.115
$20:5 \omega - 3$	eicosapentaenoic acid	0.30	0.15	0.25	0.17	0.019
$22:4 \omega - 6$	adrenic acid	0.32	0.50	0.24	0.41	0.039
22:5 ω-6	4.7.10.13.16-docosapentaenoic acid	0.03	0.08	0.04	0.04	0.017
22:5 ω-3	7.10.13.16.19-docosapentaenoic acid	0.05	0.13	0.05	0.02	0.017
22:6 ω-3	clupanodonic acid	0.02	0.00	0.04	0.03	0.029
subtotal	ciupanodonic acid	5.31	5.09	3.11	4.16	0.029
total		10.5	9.41	5.11 6.6	4.10 8.9	
total		10.5	9.41	0.0	8.9	

^a Parts not eaten.

chicken, turkey, eggs and pork (figure 5). The caterpillar Imbrasia ertli consumed by humans in Africa is one of the few food sources in which arachidonic acid contributes remarkably to the fatty-acid total (Santos Oliveira et al. 1976).

(c) Mineral and trace element content

Earthworms contain significant amounts of several minerals that are nutritionally important (table 5). Calcium, the main constituent of bones and teeth, and a mineral essential for blood clotting and muscle contraction, ranged from 1020 $\mu g \; g^{-1} \; dry \; weight \; in \; smoked \; \textit{motto} \; to$ 7070 $\mu g g^{-1}$ dry weight in *motto* meat. These amounts are comparable with the calcium contents of fresh cheese and cows' milk (figure 6) and are higher than those found in conventional meats. All of the earthworm preparations were excellent sources of iron, having iron contents that ranged from 1050 $\mu g g^{-1}$ in kuru meat to 2990 $\mu g g^{-1}$ in motto meat. We compared the iron contents of the earthworm preparations with those of some conventional meats (figure 7). In general, the two species of earthworm contained moderate amounts of copper, magnesium, potassium and phosphorus. Motto meat was the best source of manganese (74.6 μ g g⁻¹), containing four times more than kuru meat or smoked motto. Smoked motto contained the most sodium (2160 $\mu g g^{-1}$), more than twice as much as kuru meat or fresh motto meat. We do not know whether local people add salt during the smoking of motto. Kuru meat and smoked motto contained the highest amounts of selenium, 9.02 and 2.71 μg g⁻¹, respectively, whereas motto meat did not contain detectable selenium. Only small amounts of zinc were found in kuru meat, motto meat and smoked motto. Also noteworthy were the large amounts of aluminium contained in the earthworms, the

Table 4. Fatty-acid composition (weight %) of motto and kuru.

fatty acid	kuru body $(n = 1)$	kuru gut organs ^a $(n = 1)$	motto body $(n = 1)$	motto smoked $(n = 7)$
saturated				
10:0	0.1	0.3	0.2	0.8
12:0	0.0	0.2	0.1	0.0
14:0	0.5	0.9	1.1	0.7
15:0	0.5	0.4	1.1	0.6
16:0	3.8	5.4	5.5	4.6
18:0	10.9	9.8	7.1	7.9
20:0	0.5	0.6	0.6	0.6
22:0	1.4	1.0	1.0	1.3
24:0	0.6	1.0	0.5	0.2
subtotal	18.3	19.6	17.1	16.7
monounsaturated				
14:1	0.1	0.1	0.1	0.0
16:1	3.7	0.5	7.6	3.5
$18:1 \omega - 9$	14.3	10.9	5.9	10.6
18:1 ω-7	4.2	4.1	7.4	7.9
18:1 ω-5	0.7	0.6	0.7	0.9
20:1 ω-9	7.6	8.5	13.6	12.8
20:1 ω-7	0.1	0.2	0.2	0.1
$22:1 \omega - 9$	0.3	0.5	0.4	1.0
subtotal	31.0	25.4	35.9	36.7
polyunsaturated				
18:2 ω-6	9.2	9.7	7.6	8.7
18:3 ω-6	0.5	0.4	0.2	0.6
18:3 ω-3	0.8	0.6	1.7	1.1
20:2 ω-6	3.5	5.1	4.3	2.6
20:3 ω-6	6.9	7.0	8.0	8.0
20:4 ω-6	23.0	22.3	15.7	18.2
20:5 ω-6	2.9	1.6	3.8	1.9
22:4 ω-6	3.1	5.3	3.7	4.7
22:5 ω-6	0.3	0.9	0.6	0.4
22:5 ω-3	0.5	1.4	0.8	0.3
22:6 ω-3	0.2	0.0	0.6	0.4
subtotal	50.8	54.2	47.1	46.7
total	100	100	100	100

^a Parts not eaten.

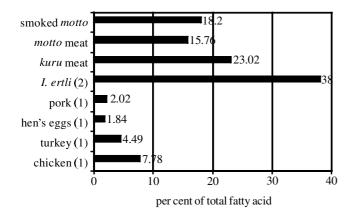


Figure 5. Arachidonic-acid content in the two earthworms *motto* and *kuru*, the African lepidopteran caterpillar *Imbrasia ertli* and some common foods (value expressed in per cent of the total fatty acid). (1), Carnovale & Marletta (1997); (2), Santos Oliveira *et al.* (1976).

gut organs of kuru (the parts not eaten) in particular (36 200 μ g g⁻¹). Kuru meat, kuru gut organs and motto meat contained substantial amounts of chromium (30.5–141 μ g g⁻¹). All specimens, except kuru gut organs, contained small but detectable amounts of lead.

4. DISCUSSION

The main overall finding of the present study was that the nutritional quality of two edible earthworms that play an important part in the diets of indigenous people of the Alto Orinoco of southern Venezuela is generally commendable. This generalization certainly applies to the protein and mineral components of *motto* and *kuru*. The protein content of the body tissue of the earthworms was high both in quantity (64.4–72.9%; table 1) and in quality: in fact, the amino-acid composition of these proteins indicates that they are suitable for human nutrition.

The earthworms of the Alto Orinoco are also capable of providing substantial quantities of minerals and trace elements that are essential in humans, in particular calcium and iron (table 5). Indeed, the iron content of the

Table 5. Mineral content of *motto* and *kuru* ($\mu g g^{-1}$ dry weight).

	kuru body	kuru gut	motto body	<i>motto</i> smoked $(n = 7)$	
mineral	(n=1)	organs ^a $(n = 1)$	(n=1)	mean	s.d.
aluminium	1430	36200	5220	2640	962
arsenic	0.91	0.53	1.41	1.41	0.23
calcium	2650	12900	7070	1020	260
chromium	30.5	141	90.1	1.67	0.56
copper	5.63	6.23	8.17	10.9	6.2
iron	1050	12000	2990	1080	121
potassium	3430	4510	897	6810	599
magnesium	527	457	792	730	52
manganese	17.9	29.8	74.6	22.6	2.7
molybdenum	0.61	1.6	1.41	0.29	0.05
sodium	997	1240	548	2160	116
nickel	10.6	53.2	38.6	0.64	0.14
phosphorus	3500	4220	3560	5620	326
lead	4.72	ND	2.5	4.17	1.43
selenium	9.02	ND	ND	2.71	0.41
strontium	7.43	27.2	28.8	4.12	1.07
vanadium	1.25	19.8	5.09	0.8	0.2
tungsten	1.49	0.92	1.31	1.51	0.33
yttrium	0	0.52	3.74	3.09	0.81
zinc	149	93.5	131	96.3	15.4

^a Parts not eaten; ND, not detectable.

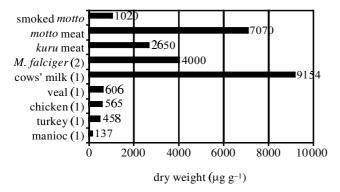


Figure 6. Calcium content in the two earthworms motto and kuru, one African termite Macrotermes falciger and some common foods ($\mu g g^{-1}$ dry weight). (1), Carnovale & Marletta (1997); (2), Malaisse & Parent (1997).

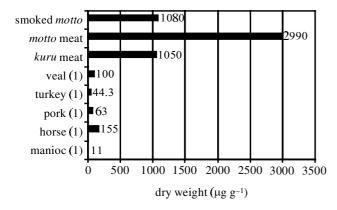


Figure 7. Iron content in motto and kuru and some common foods (μg g⁻¹ dry weight). (1), Carnovale & Marletta (1997).

two earthworm species was about 10 times higher than that of soybeans, and threefold higher than that of the mopane worm (Glew et al. 1999). This means that, assuming good bioavailability of the iron in the earthworms, they could be a useful food source to mitigate the risk of iron-deficiency anaemia among the Yekuana population of the Alto Orinoco who consume substantial quantities of these organisms. The earthworms also contain nutritionally significant amounts of calcium, which would be important for pregnant or lactating women, young children at risk of calcium-deficiency rickets and postmenopausal women in whom there might be concern about osteoporosis and fractures. The Yekuana consume quite a lot of earthworms. We estimated prudently a mean of $1.7-2 \text{ kg person}^{-1} \text{ y}^{-1}$ for all members of the population, including the children (Paoletti et al. 2000). The consumption by women after parturition can be several times higher. For instance when we went to Buena Vista the people collected about 18 specimens of kuru (fresh weight, 2.6 kg) and they were eaten by three persons the same day.

Assuming the *kuru* body contains 1 mg g^{-1} dry weight of iron, then 30 g of one dry specimen of kuru (fresh weight ca. 130 g) could satisfy the daily recommended allowance of iron for a pregnant woman.

The data resulting from the fatty-acid analyses point to several conclusions. First, the fact that fatty acids account for no more than 1% of the dry weight of the earthworms indicates that the organisms do not store very much fatty acid in the form of triacylglycerols. This conclusion is supported by the observation that the total fatty-acid composition of the organisms resembles more closely that of membrane phospholipids than that of storage lipids. Thus, it appears that these earthworms do not accumulate significant amounts of triacylglycerols, which are the type of lipid that many different cells and organisms use to store energy.

The second interesting result of the fatty-acid analyses was the finding of exceptionally high proportions of ω-6 fatty acids and arachidonic acid in particular. Arachidonic acid is an important component of the phospholipids found in the membranes of eukaryotes. In mammals, it is a precursor to prostaglandins. Along with the two essential fatty acids (linoleic acid and α-linolenic acid (ALA)) and docosahexaenoic acid, arachidonic acid is especially critical in the first six months of human life. It is useful to point out that, in the ω-6 family of fatty acids, the proportion of GLA in the earthworms was very low (0.051%), whereas that of dihomo-γ-linolenic acid (DGLA) was much higher (0.72%). In light of the fact that GLA is the immediate precursor to DGLA in the ω-6 desaturase-elongase pathway (Faas et al. 1996), these relative proportions of GLA and DGLA indicate that the elongase enzyme system in the earthworms that is responsible for catalysing the conversion of GLA to DGLA is very active relative to the 5-desaturase that converts DGLA into arachidonic acid.

Although the relative proportions of the essential fatty acids may be remarkable, the very low total fatty-acid contents of the various earthworm preparations means that they are not likely to contribute significantly to satisfying the daily requirement for these essential nutrients.

In conclusion, the nutritional analyses reported here provide quantitative evidence to support the assertion that the earthworms consumed by the Yekuana indigenous people of the Alto Orinoco are capable of satisfying a significant fraction of their daily requirements for essential amino acids and many of the trace metals, especially calcium and iron. However, the quantities of essential fatty acids (linoleic acid and ALA) and critical ω-6 and ω-3 fatty acids (e.g. arachidonic acid and docosohexaenoic acid) contained in the two earthworm species we analysed are so low as to suggest that the populations who consume them may need to look to other sources (e.g. fishes) to provide the amounts of the critical polyunsaturated fatty acids required for growth, development and maintenance of health. Finally, our data do not address the important issue of bioavailability. It remains to be seen to what extent the proteins and other nutrients in these edible earthworms are digested and absorbed.

The minilivestock, including earthworms (Hardouin & Stievenart 1993; Paoletti & Bukkens 1997; Paoletti et al. 2000; Cerda et al. 2001), could be the basis for a sustainable production of protein sources, especially in the tropics and in the Amazon, where conventional large animals (cows, sheep and goats), if grown on a large scale, can badly damage the forests resulting in grassland dominance and a reduction in the local biodiversity. Risks related to foodwebs of conventional large animals and their overexploitation are increasingly raising concern in Western European societies as well. A variety of small animals have been traditionally used as food in Amazonas; however, they are only in the early stage of domestication (e.g. the large rodents Agouti paca and Dasyprocta sp.), or have already been domesticated (e.g. the guinea pig Cavia porcellus). Additional research on the nutritional value of the larger invertebrates such as caterpillars, palmworms, termites, locusts, earthworms and many others is needed. Efforts are also needed to improve the local awareness and

knowledge of earthworms and other edible invertebrates as sources of protein, fat, minerals and vitamins in the Amazon.

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REFERENCES

- Bidlingmeyer, B. A., Cohen, S. A. & Tarvin, T. L. 1984 Rapid analysis of amino acid using pre-column derivatization. J. Chromatogr. 336, 93–104.
- Carnovale, E. & Marletta, L. 1997 Tabelle di composizione degli alimenti. Milan, Italy: Istituto Nazionale della Nutrizione ed Edra.
- Cerda, H., Martinez, R., Briceno, N., Pizzoferrato, L., Manzi, P., Tommaseo Ponzetta, M., Marin, O. & Paoletti, M. G. 2001 Palm worm: (*Rhychophorus palmarum*) traditional food in Amazonas, Venezuela—nutritional composition, small scale production and tourist palatability. *Ecol. Food Nutr.* 40, 13–32.
- Chamberlain, J., Nelson, G. & Milton, K. 1993 Fatty acid profiles of major food sources of howler monkeys (*Aloutta palliata*) in the neotropics. *Experientia* **49**, 820–823.
- Faas, F. H., Dang, A. Q., Pollard, M., Hong, X. M., Fan, K., Luckert, P. H. & Schutz, M. 1996 Increased phospholipid fatty acid remodeling in human and rat prostatic adenocarcinoma tissues. J. Urol. 156, 243–248.
- Glew, R. H., Jackson, D., Sena, L., VanderJagt, D. J., Pastuszyn, A. & Millson, M. 1999 Gonimbrasia belina (Lepidoptera: Saturniidae): a nutritional food source rich in protein, fatty acids and minerals. Am. Entomol. 45, 250-253.
- Hardouin, J. & Stievenart, C. 1993 Invertebrates (minilivestock) farming. In Proc. Seminar held at La Union, Philippines, November 1992. EEC-DGXII/CTAIFS/DMM MSU/ITM, Philippines.
- Hariharan, M., Naga, S. & VanNoord, T. 1993 Systematic approach to the development of plasma amino acid analysis by high-performance liquid chromatography with ultraviolet detection with precolumn derivatization using phenylisothiocyanate. *J. Chromatogr.* 621, 15–22.
- Hirs, C. W. H. 1967 Performic acid oxidation. *Meth. Enzymol.* 11, 197–199.
- Hugli, T. E. & Moore, S. 1972 Determination of the tryptophan content of proteins by ion exchange chromatography of alkaline hydrolysates. *J. Biol. Chem.* 247, 2828–2834.
- Kim, T. S., Pastuszyn, A., VanderJagt, D. J., Glew, R. S., Millson, M. & Glew, R. H. 1997 The nutritional composition of seeds from *Boscia senegalensis* (Dilo) from the Republic of Niger. J. Food Compos. Anal. 10, 73–81.
- Malaisse, F. & Parent, G. 1997 Minor wild edible products of the Miombo area. In *Se nourrir en foret africaine, approche* écologique et nutritionnelle (ed. F. Malaisse), p. 384. Belgium: Les presses agronomiques de Gembloux.
- Moreno, A. & Paoletti, M. G. 2002 Andiorhinus kuru n. sp., one giant earthworm (Oligochaeta: Glossoscolecidae) food resource for Makiritare Indians of the alto rio Padamo, Amazonas, Venezuela. Presented at the Seventh International Symposium on Earthworm Ecology, Cardiff, 6–12 September 2002.
- Morrison, W. R. & Smith, L. M. 1964 Preparation of fatty acid methyl esters and dimethylacetals from lipids with boron trifluoride-methanol. J. Lipid Res. 5, 600-608.

- Paoletti, M. G. & Bukkens, S. G. F. 1997 Minilivestock. Ecol. Food Nutr. 36 (Spec. Issue), 95-346.
- Paoletti, M. G., Dufour, D. L., Cerda, H., Torres, F., Pizzoferrato, L. & Pimentel, D. 2000 The importance of leaf- and litter-feeding invertebrates as sources of animal protein for the Amazonian Amerindians. Proc. R. Soc. Lond. B 267, 2247-2252. (DOI 10.1098/rspb.2000.1275.)
- Righi, G. & Araujo, Y. 1999 Andiorrhinus (Amazonidrilus) motto n. sp. and Rhinodrilus appuni pavoni n. subsp. (Oligochaeta, Glossoscolecidae) from the Venezuelan Amazonia. Miscellània Zoològica 22, 93-100.
- Santos Oliveira, J. F., Passos De Carvalho, J., Bruno De Sousa, R. F. X. & Madalena Simao, M. 1976 The nutritional value of four species of insects consumed in Angola. Ecol. Food Nutr. 5, 91-97.
- Schlenker, H. 1974 Makiritare (Venezuela, Orinoco-Quellgebiet) Sammeln, Zubereiten und Essen von Würmern. In Encyclopae-

- dia cinematographica (ed. G. Wolf), pp. 1-11. Gottingen, Germany: Institut fur den Wissenschaftlichen Film.
- Wallace, A. R. 1853 On the insects used for food in the Indians of the Amazon. R. Entomol. Soc. Lond. 2, 241-244.
- Wallace, A. R. 1889 (1972) A narrative of travels on the Amazon and Rio Negro. New York: Dover Publications Inc.
- Yazzie, D., VanderJagt, D. J., Pastuszyn, A., Okolo, A. & Glew, R. H. 1994 The amino acid and mineral content of baobab (Adansonia digitata L.). J. Food Compos. Anal. 7, 183-193.
- Zent, S. 1992 Historica and ethnographic ecology of the Upper Cuao River Votiha: clues for an interpretation of native Guianese social organization. PhD thesis, Columbia University, New York.

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