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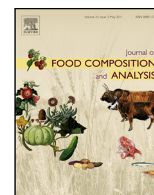

Original Research Article

Nutritional and toxicological studies of New Zealand *Cookia sulcata*

Susan L. Mason [a,*], Jinlin Shi [a], Alaa El-Din Bekhit [b], Ravi Gooneratne [a]

[a] Faculty of Agriculture & Life Sciences, Lincoln University, PO Box 85084, Canterbury, New Zealand
[b] Department of Food Science, Division of Sciences, University of Otago, PO Box 56, Dunedin, New Zealand

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ABSTRACT

Interest in snails as a source of protein and as a delicacy is increasing in many countries. The present study investigated selected nutritional (proximate, amino acid, fatty acid, vitamin E, cholesterol and macro- and trace minerals) and toxic (toxic elements and organochlorine) concentrations of small and large (≤60 and >60 g whole animal weight, respectively) Captain Cook snails (*Cookia sulcata*). The major amino acids in *C. sulcata* muscle were glutamic (13.9 g/100 g protein), arginine (10.2 g/100 g protein), glycine (9.5 g/100 g protein) and taurine (9.5 g/100 g protein). There was no difference in the amino acid profiles related to the snail size. *C. sulcata* had relatively high amounts of saturated fatty acids (44.4%) and polyunsaturated fatty acids (34.3%), and lesser amounts of mono-unsaturated fatty acids. The major fatty acids detected in *C. sulcata* were C16:0, C18:0, C20:4 and C22:5, which accounted for more than 60% of the total fatty acids. Snail size had a significant ($P < 0.05$) effect on the C16:0 and C18:3 concentrations. The only isoform of vitamin E present in *C. sulcata* was identified as α-tocopherol at 2.16 and 3.71 mg/100 g fresh weight for the small and large snails, respectively. The average cholesterol concentration in *C. sulcata* was 1.33 mg/100 g fresh weight. The results indicated that none of the toxic elements, including Al, Ni, As and Pb of *C. sulcata*, were over the maximum concentration allowed in the Australia New Zealand Food Standard; and the organochlorine pesticides concentrations in *C. sulcata* were below the detection limit (<0.0005 mg/kg). *C. sulcata* could, therefore, be utilized for special dietary applications requiring higher amounts of Fe, Zn, taurine and tryptophan.

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1. Introduction

Production of the major sources of meat protein for humans; poultry, pork, beef, lamb and fish is currently declining due to natural disasters including persistent droughts and the resultant high cost of feed resulting from habitat changes and animal diseases. The imbalance between demand and supply is further exacerbated by the rapid increase in human populations as well as an increase in demand for animal proteins (Cao and Li, 2013; IFarm, 2012; Knudsen et al., 2010; Maday, 2013; USDA, 2013). Finding new sources of protein that are low in fat to supplement meat protein is a challenge for the world's food industries and requires a major effort.

The potential for using molluscs has been explored and research on the nutritional value of molluscs has attracted some attention (Özden and Erkan, 2011; Çağıltay et al., 2011). Molluscs include univalve organisms such as snails and slugs, bivalves such as mussels, oysters and cockles, and cephalopods such as octopus, squid and cuttlefish (Copper and Knower, 1991). Although molluscs are predominantly aquatic, some are terrestrial. Humans have consumed snails for thousands of years, and edible snails still have a place on the menu in many European and some Asian countries. The greatest quantities of snails are consumed in France (Milinsk et al., 2003), while snails are considered a delicacy in Korea, China and Japan (Qun et al., 2004). Interest in local snails has been developing in Nigeria (Udoh et al., 1995; Adeyeye and Afolabi, 2004) and Brazil (Milinsk et al., 2003). Snails may provide a supplemental food to improve the nutrition of those in poverty. Commercial production of edible snails has developed in China and Brazil because of historical interest, favourable weather and environmental conditions (Xia et al., 2007; Milinsk et al., 2006). Information regarding the nutritional value of snails is mostly limited to land snails (Hamzat et al., 2002; Fagbuaro et al., 2006) with *Helix*, *Archachatina*, and *Achatina* being the most commonly studied species.

The meat of land snails was found to be low in fat and rich in protein, and it contained other nutrients beneficial to a healthy diet

* Corresponding author. Tel.: +64 34230639.
E-mail address: Sue.Mason@lincoln.ac.nz (S.L. Mason).

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(Ademolu et al., 2004; Çağıltay et al., 2011; Fagbuaro et al., 2006; Milinsk et al., 2003, 2006). Nutritional data for marine molluscs including mussels, scallops and oysters show them to be rich in calcium, sodium, zinc and iron (Hughes et al., 1980; Karakoltsidis et al., 1995). Compared with domestic meats such as beef, lamb and chicken, these molluscs have a similar protein content but less fat (Vlieg, 1988; Karakoltsidis et al., 1995).

However, nutritional data on marine snails are scarce (Özden and Erkan, 2011). This is particularly significant for New Zealand which, as an island nation, has rich marine resources with a diverse range of species of marine snails and the potential to harvest or 'farm' them. *Cookia sulcata* belongs to the family of *Turbinidae* and is an abundant marine snail species found at beaches throughout New Zealand. These snails feed on algae and seaweed and, along with other turban snails, are consumed mostly by Maori, New Zealand's native people.

The present study was designed to assess nutritional aspects of *C. sulcata* in New Zealand. In addition, heavy metals and organochlorine pesticide concentrations were assessed to determine the toxicological safety of this snail meat.

2. Material and methods

2.1. Materials, sample collection and preparation

Fatty acid methyl ester standards (purity >99%), α and γ-tocopherol (purity >96%), and δ-tocopherol (purity >90%) standards were obtained from NU-Check, Elysian, Minnesota, USA and Sigma Aldrich, Bellefonte, Pennsylvania, USA, respectively. All other chemicals were analytical grade and solvents were HPLC grade and were purchased from Thermofisher Ltd, North Shore City, NZ.

Cookia sulcata snails were collected for both nutritional and toxicological analyses from Te Oka Bay (43°51′S, 172°47′E), Canterbury, New Zealand between the mid- and low-tide marks in summer 2006–2007 ($n = 94$). Live snails of varying sizes were collected, stored in plastic containers filled with seawater and kept on ice during transport to the laboratory. Snails were frozen in plastic bags and stored at −18 °C until analysis. Snails were divided into two groups based on the weight of the entire snail: small (≤60 g whole animal weight) and large (>60 g whole animal weight). The edible portion of each snail was removed, homogenized and used for analyses. Snails were analyzed individually unless otherwise stated in the methods. The sex of the snails was not determined because there was no expertise available to identify the sex and moreover this is not a criterion for human consumption of snails.

2.2. Chemical analyses

Proximate analysis (moisture, crude protein, fat and ash) of the edible portion of individual snails was determined using standard methods (AOAC, 2000).

Most amino acids (AA) were analyzed after acid hydrolysis and oxidation with performic acid for cysteine and methionine analysis, as described earlier (Bekhit et al., 2009a), while alkaline hydrolysis was used for tryptophan (Yust et al., 2004). Amino acid analysis was performed on an Agilent 1100 series HPLC (Agilent Technologies, Waldbronn, Germany) controlled by Chemstation Software (version 09.01). A Lichrospher 100 NH2 (250 mm × 4 mm, particle size 5 μm) column coupled with a Lichrocart 4-4 guard column was used to separate amino acids and fluorescence detection was employed.

For fatty acid (FA) analysis, lipid was extracted using a modification of the method of Folch et al. (1957). Fatty acids were methylated using 1% sulphuric acid in methanol, and extracted into hexane according to the method of Hughes et al. (1980). Fatty acid methyl esters were separated and quantified using a Shimadzu gas chromatograph (GC) model 2010 (Shimadzu, Kyoto, Japan) equipped with a flame ionization detector (FID) and 30 m 0.25 mm ID JW-Innowax column with polar ethylene glycol stationary phase (0.25 μm film thickness), and GC Solution software (version 2.3000 SU4). Fatty acids were identified by comparison of retention times of the fatty methyl esters with standards (GLC-68E, GLC-68F GLC-411, Nu-Check).

Extraction and measurement of vitamin E was carried out as described by Bekhit et al. (2009a), with modifications. Edible snail meat was extracted with ethanol containing 0.2% BHT and saponified under nitrogen using KOH (1.5%) at 70 °C for 20 min. After cooling and addition of NaCl, sterols were extracted into hexane containing 10% (v/v) ethyl acetate, dried under nitrogen and stored at −20 °C until analysis. Vitamin E was analyzed using the same column and Agilent HPLC, as described above. A mobile phase of heptane:tert-butymethyether:tetrahydrofuran:methanol (79:20:0.98:0.02, v/v/v/v) at a flow rate of 1.2 ml/min and fluorescent detection at 294 nm (excitation) and 320 nm (emission) were used. Standard mixtures of α-tocopherol, δ-tocopherol and γ-tocopherol (Sigma Aldrich) were used to construct standard calibration curves, and to identify and quantify the tocopherols.

Cholesterol was measured in the same extract using Liberman-Burchard reagent by the method of Sabir et al. (2003).

Mineral analysis was carried out, as described by Bekhit et al. (2009a). Samples (0.5 g) of freeze dried edible snail meat were dissolved in a mixture of HNO_3 (65%) and H_2O_2 (30%) (3:1, v/v) and subjected to a microwave digestion system (ETHOS SEL/plus, Milestone Srl, Sorisole, Italy). The elemental concentrations were determined by inductively-coupled plasma (ICP) emission spectrometry using an AXIAL Varian 720 OES Series ICP spectrometer with a SP3 Auto Sampler (Varian Inc., Palo Alto, California, USA).

Total organochlorine pesticides (OCPs) on pooled *C. sulcata* (3 × 5 snails) were analyzed by an accredited laboratory (Hill Laboratories, Hamilton, New Zealand). The samples were extracted with hexane:acetone (3:2) by sonication and cleaned using solid phase extraction and gel permeation chromatography before separation and detection of OCPs by GC mass spectrometry (GCMS QP5050; Shimadzu) with a DB-5 M5 fused silica capillary column (30 m, 0.25 mm i.d.).

2.3. Statistics

The data obtained were subjected to analysis of variance (ANOVA) to test the effect of snail size on the composition of the snails. The data were analyzed using the general linear model protocol in MINITAB® software (Release15, Minitab Inc., Pennsylvania, USA). Significant differences between means were determined by Tukey Tests ($P < 0.05$).

3. Results

Harvested *C. sulcata* were divided into two groups; small (≤60 g) and large (>60 g), based on the weight of the whole animal including the shell. The average weights of these groups were 42.7 ± 8.9 g and 102.4 ± 27.2 g, respectively. The edible portion of the snail constituted 23.9 ± 2.9 and 23.6 ± 3.0% of the snail weight for the small and large snails, respectively.

3.1. Proximate analyses

The proximate compositions of *C. sulcata* are reported on a dry matter (DM) and fresh weight (FW) basis in Table 1 for comparison with the literature where no consistent basis has been used to report proximate analysis results. The moisture content of *C. sulcata* ranged from 75.5 to 80.0%. The protein content (% FW) was 17.5 ± 0.9 and 17.6 ± 1.5% for small and large snails, respectively,

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