PRELIMINARY STUDIES ON THE PROXIMATE COMPOSITION OF SOME SELECTED SEAWEEDS FROM MKOMANI AND KIBUYUNI, KENYA

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Abstract

The proximate composition of various seaweed species collected from Mkomani and Kibuyuni sites along the Kenya coast was determined The seaweed species had a moderate to high ash content. The seaweed with the lowest ash content (12.76±0.18% dw) was *Ulva reticulata* and the highest ash content (67.60±0.02% dw) was *Halimeda macroloba*. Protein contents were low to moderate, with *Hypnea musciformis* having the highest value (14.17±0.04 dw), followed by *Ulva reticulata* (11.87±0.10% dw). *Dictyota* sp. (variety 1) had the lowest protein content (1.71±0.04% dw). The crude fat contents were within the range mentioned for most seaweeds (<3% dw). The crude fibre contents for the selected seaweeds ranged between 4.00–18.06% dw. Based on the biochemical analysis, the composition of the studied seaweed species is comparable to that of several seaweeds traditionally used in human and animal nutrition thus may be utilized for animal and human consumption.

Key words: Seaweed, proximate composition, protein, nutrition, protein content, crude fiber, crude fat

1.0   Introduction

Seaweed also referred as marine macroalgae are classified based on anatomy, pigmentation, morphology and chemical composition as green (chlorophyta), brown (phaeophyta) and red (rhodophyta) algae (Dawczynski *et al.*, 2007) . About 386 Kenyan seaweeds species have been relatively well documented (Bolton *et al.*, 2007). Compared to terrestrial plants, there is limited literature on the chemical composition of seaweeds in Kenya and most of the available information only deals with traditional Japanese seaweeds (Fujiwara-Arasaki *et al.*, 1984; Nisizawa *et al.*, 1987). Knowledge of the nutrient composition of marine macroalgae is both important for the animal feeds (Hawkins & Hartnoll, 1983), possible human food (Abbott, 1988) and for the evaluation of potential sources of carbohydrate, protein and lipid for industrial use (Chapman & Chapman, 1980). Variations in chemical composition of seaweed species may be caused by climate and sea conditions (Burtin, 2003; Darcy-Vrillon, 1993). The aim of this study was to compare the proximate composition of selected seaweeds collected in Mkomani and Kibuyuni, Kenya.

2.0   Material and Methods

2.1   Seaweed Collection

The seaweeds *Hypnea musciformis*, *Ulva reticulatum*, *Laurencia intermedia*, *Enteromorpha muscoides* and *Ulva fasciata* were collected from Mkomani coastal region, and *Cystoseira trinodis*, *Eucheuma denticulatum* (phaeophyta), *Dictyota* sp. (variety 1), *Dictyota* sp. (variety 2), *Padina tetrastromatica*, *Sargassum* sp., and *Acanthophora spicifera* were collected from Kibuyuni, South Coast of Kenya. The seaweeds collected from both regions were *Chondrophycus papillosus*, *Halimeda macroloba*, *Gracilaria salicornia* and *Sargassum oligocystum*. Seaweed samples were picked by hand and immediately washed with seawater to remove the foreign particles, sand particles and epiphytes. The samples were kept in clean buckets and immediately transported to the laboratory and washed thoroughly using tap water to remove the salt on the surface of the samples. Seaweeds were spread on blotting paper to remove excess water before they were sundried to constant weight, and ground. The powdered samples were then refrigerated prior to chemical analysis.

2.2   Proximate Analysis

2.2.1   Moisture Content

Moisture was determined using the drying method, specification 950.46, method 925.10-32.10.03 (AOAC, 1995). Results were reported on the dry weight basis. About 5g of ground sample, accurately weighed into a moisture

dish was transferred in a hot-air oven previously heated to around 105°C and then drying done for one hour. Final weight of sample was taken after drying and cooling in a desiccator. The sample residue is to be taken as the total solids and lose in weight as the moisture content of the sample.

2.2.2 Crude Protein Content
Protein was determined using the semi-micro kjeldahl method, specification 950.46, method 20.87-37.1.22 (AOAC, 1995). Approximately 2g of sample was weighed into a digestion flask together with a combined catalyst of 5g K_2SO_4 and 0.5g of $CuSO_4$ and 15ml of concentrated H_2SO_4. The mixture was heated in a fume hood till the digest colour turns blue. This signified end of the digestion process. The digest was cooled, transferred to 100ml volumetric flask and topped up to the mark with deionized water. A blank digestion with the catalysts was made. 10ml of diluted digest was transferred into the distilling flask and washed with about 2ml of distilled water. 15 ml of 40% NaOH was added and washed with 2ml of distilled water. Distillation was done to a volume of about 60ml distillate. The distillate was titrated using 0.02N HCl to an orange colour of the mixed indicator, which signified the end point.

$$\% \text{ Nitrogen} = \left(V_1 - V_2\right) \times N \times f \times 0.014 \times \frac{100}{v} \times \frac{100}{S}$$

Where, V_1 = Volume (ml) of 0.02N HCl used in sample titration

V_2 = Volume (ml) of 0.02N HCl used in blank titration

N = Normality of HCl (0.02)

f = Factor of standard HCL solution

v = Volume of diluted digest taken for distillation (10ml)

S = Weight (g) of sample taken

% crude protein = % Nitrogen x protein factor (6.25).

2.2.3 Ash Content
Ash content was determined by incinerating in a muffle furnace (AOAC, 1995) method 923.03-32.1.05. Sample weights of about 5g were weighed in crucibles. First the sample was charred by a flame to eliminate smoking before being incinerated at 550^0C in a muffle furnace, to the point of white ash. The residues were cooled in a desiccator and the weights taken.

2.2.4 Crude Fat Content
Fat determination was done using the soxhlet method 920.85-32.1.13, (AOAC, 1995). This will give intermittent extraction of oil with excess of fresh condensed organic solvent to be used. Five grams of sample was weighed into extraction thimbles and initial weight of extraction flasks taken. Fat extraction was done using petroleum spirit in Soxhlet apparatus for 8 hours. The extraction solvents was rota-evaporated and the fat extracted dried in a hot air oven for 15 minutes before the final weight of flasks with extracted oil taken.

2.2.5 Crude Fibre Content
Crude fibre was determined according to method 920.86.32.1.15 (AOAC, 1995). Two grams of sample were weighed into a 500ml conical flask. About 200ml of boiling 1.25% H_2SO_4 was added and boiling done for 30 minutes under reflux condenser. Filtration was done in slight vacuum with Pyrex glass filter (crucible type) and the residue washed to completely remove the acid with boiling water. About 200ml of boiling 1.25% NaOH was added to the washed residue to and boiling under reflux for another 30 minutes. Filtration was done again using the same glass filter previously used with the acid. The residue was rinsed with boiling water followed by 1% HCl and again washed with boiling water to rinse off the acid from the residue. The residue was then be washed twice with alcohol and three times with ether. It was then dried in a hot- air oven at 105^0C in a porcelain dish to a constant weight (W_1). Incineration was done in a muffle furnace for 3 hours at 550^0C. The dish was then cooled in a desiccator and the final weight (W_2) taken.

$$\% \text{ Crude fibre} = \frac{\left(W_1 - W_2\right)}{W} \times 100$$

Where, W_1 = Weight (g) of digested sample before incineration

W_2 = Weight (g) of digested sample after incineration

W = Weight (g) of sample taken

2.3   Statistical Analysis

Proximate composition data of seaweeds found in both Mkomani and Kibuyuni were treated statistically by a one-way analysis of variance (ANOVA) with region as source of variance. For the different species in each region the proximate composition data was treated statistically by one-way analysis of variance (ANOVA) with species as the source of variance. Duncan Multiple Range Test (DMRT) was used to separate the means. Significance was determined at $p<0.05$.

3.0   Results and Discussion

The proximate composition of the selected seaweeds on dry weight basis are shown in Tables 1, 2 and 3. The proximate composition of the same seaweed species collected from Mkomani and Kibuyuni, varied with region (Table 1). The proximate composition of different seaweeds collected from Mkomani and Kibuyuni showed no significant difference for some proximate parameters (Table 2 and 3, respectively).

The crude protein content of chlorophytes such as *Halimeda macroloba* (both regions), *Enteromorpha muscoides* and *Ulva fasciata* ranged from 2.72–8.19% which was below the range of 10–26% observed for chlorophytes. However, *Ulva reticulatum*, a chlorophyte, had crude protein content of 11.87±0.10% and was within the range reported by Fleurence (1999). *Ulva fasciata* had crude protein content of 7.77±0.08% which was similar to that of *Ulva rigida* (Frikha *et al*., 2011) but was lower than the reported range of 10-26% for other *Ulva* species (Fleurence, 1999). The crude protein content of rhodophytes; *Laurencia* spp., *Gracilaria salicornia* (both regions), *Chondrophycus papillosus* (both regions), *Acanthophora spicifera* and *Cystoseira trinodis* ranged from 2.44–7.86% which was below the crude protein range of 10–47% for rhodophytes (Fleurence, 1999). *Hynea musciformis*, a rhodophte, had a crude protein content of 14.17±0.04% which was within the crude protein range of 10–30% as reported by Fleurence (1999). In addition, *Hynea musciformis* had protein content comparable to *Ceramium diaphanum*, a rhodophte as reported by Frikha *et al*. (2011). *Gracilaria salicornia* from Mkomani had crude protein content of 7.86±0.03% which was similar to that of the protein content in most *Gracilaria* species which ranges from 7–13% (Briggs and Smith, 1999). The crude protein content of phaeophytes; *Sargassum oligocystum* (both regions), *Sargassum* spp., *Padina tetrastromatica* and *Eucheuma denticulatum* ranged from 3.23–7.16% and were within range of 3–15% for phaephytes as reported by Darcy-Vrillon (1993). Variations in the protein content of seaweeds can be due to factors such as species, seasons and geographic area (Fleurence 1999, Sánchez-Machado *et al*. 2004).

The crude fat content of most seaweed species was generally less than 3.0% which was comparable to previous studies (Polat and Ozugal, 2008; Marsham *et al.,* 2007). The crude fat content of Gracilaria salicornia (both regions) was comparable to *Gracilaria caliculta* collected in Arabian Sea (Banaimoon, 1999). It is important to note that variations in crude fat content among the species are attributed to differences in species, habitat, seasons and geographical area (Fleurence 1999).

Ash contents of seaweeds ranged from 12.76–67.60% for those in Mkomani region and 18.36– 64.53% for those in Kibuyuni. High ash content was associated with the amount of mineral elements which is significantly high implying they are high in minerals due to their marine habitat, and the diversity of the minerals they absorb is wide. Important minerals, such as calcium, accumulate in seaweeds like *Halimeda macroloba* at much higher levels than in terrestrial foodstuffs. *Halimeda* macroloba is highly calcified seaweed hence high ash content and low in other proximate parameters as compared to other seaweeds (Table 1). These results are similar to previous studies on calcified seaweed species (Kaehler and Kennish, 1996). There was no significant difference in calcium, cadmium, potassium and phosphorous for *Chondrophycus papillosus* in both regions whereas for *Halimeda macroloba*, there was no significant difference in copper in both regions. The variations in ash content among the selected species is related to the capacity of the various species to accumulate minerals attributed to seasons, geographical area (Kaehler and Kennish, 1996) and environmental conditions (Polat and Ozugal, 2008).

Sargassum oligocystum had the highest fibre (Mkomani, 18.06±0.01%; Kibuyuni, 11.39±0.22%) contents in each regions. *Dictyota* spp. (variety 2) had the highest crude fibre content (14.10±0.02%), followed closely by *Dictyota* spp. (variety 1) (13.18±0.02%).

Table 1: Proximate composition of Gracilaria salicornia (GS), Chondrophycus papillosus (CP), Halimeda macroloba (HMa) and Sargassum oligocystum (SO) in Mkomani and Kibuyuni (% dry weight)

Seaweed	Moisture %	% dry weight (dw)				
		Crude Fat	Crude Fibre	Nitrogen	Crude Protein	Ash
GS[1]	16.51±0.08[b]	1.48±0.00[b]	12.37±0.01[b]	1.26±0.00[b]	7.86±0.03[b]	46.55±0.06[b]
GS[2]	3.96±0.29[a]	1.37±0.01[a]	7.73±0.01[a]	0.25±0.01[a]	1.71±0.05[a]	27.18±0.03[a]
HMa[1]	4.63±0.01[b]	1.80±0.00[a]	6.34±0.12[b]	0.79±0.01[b]	4.93±0.08[b]	67.60±0.02[b]
HMa[2]	3.44±0.03[a]	2.35±0.00[b]	4.00±0.12[a]	0.43±0.02[a]	2.72±0.10[a]	64.53±0.02[a]
CP[1]	14.25±0.00[b]	0.64±0.00[a]	12.54±0.01[b]	1.16±0.01[b]	7.27±0.04[b]	29.17±0.19[b]
CP[2]	9.83±0.06[a]	1.32 ±0.00[b]	10.52±0.05[a]	0.27±0.02[a]	1.68±0.12[a]	21.83±0.43[a]
SO[1]	15.50±0.13[b]	3.93±0.00[b]	18.06±0.01[b]	0.77±0.01[b]	4.84±0.07[b]	20.29±0.45[b]
SO[2]	13.40±0.11[a]	3.03±0.00[a]	11.39±0.22[a]	0.52±0.15[a]	3.23±0.15[a]	18.36±0.09[a]

[1] Mkomani region
[2] Kibuyuni region
Crude Protein = Nitrogen × 6.25
Values are expressed as mean±standard deviation, n=3
Values in the same column for same species with different superscripts letters are significantly different ($p<0.05$)

Table 2: Proximate composition of Laurencia intermedia, Enteromorpha muscoides (EM), Ulva reticulatum (UR), Ulva fasciata (UF) and Hypnea musciformis (HMu) in Mkomani (% dry weight)

Seaweed[1]	Moisture %	% dry weight (dw)				
		Crude Fat	Crude Fibre	Nitrogen	Crude Protein	Ash
LI	11.16±0.06[a]	1.72±0.01[c]	7.87±0.02[e]	1.24±0.01[a]	7.76±0.04[a]	24.17±0.25[c]
EM	13.27±0.05[b]	3.08±0.01[e]	4.56±0.30[b]	1.31±0.01[b]	8.19±0.07[b]	25.45±0.17[d]
UR	16.84±0.25[c]	1.16±0.00[a]	4.01±0.01[a]	1.90±0.02[c]	11.87±0.10[c]	12.76±0.18[a]
UF	17.66±0.00[e]	2.29±0.01[d]	5.38±0.16[c]	1.24±0.01[a]	7.77±0.08[a]	14.97±0.30[b]
HMu	17.10±0.02[d]	1.31±0.02[b]	5.43±0.01[c]	2.27±0.01[d]	14.17±0.04[d]	15.36±0.02[b]

[1]Mkomani region
Crude Protein = Nitrogen × 6.25
Values are expressed as mean±standard deviation, n=3
Values in the same column with different superscripts letters are significantly different ($p<0.05$)

Table 3: Proximate composition of Cystoseira trinodis (CT), Eucheuma denticulatum (brown) (EDb), Dictyota sp. (variety 1) (Dspr), Dictyota sp. (variety 2) (Dspp), Sargassum sp. (Ssp), Acanthophora spicifera (AS), Padina tetrastromatica (PT) in Kibuyuni (% dry weight)

Seaweed[2]	Moisture %	% dry weight (dw)				
		Crude Fat	Crude Fibre	Nitrogen	Crude Protein	Ash
CT	13.23±0.16[e]	3.17±0.02[e]	12.17±0.02[e]	0.39±0.01[b]	2.44±0.07[b]	25.04±0.28[d]
EDb	14.68±0.05[g]	2.79±0.01[d]	2.48±0.05[a]	0.52±0.01[c]	3.25±0.04[c]	22.82±0.01[c]
Dspr	8.91±0.07[a]	4.21±0.00[g]	13.18±0.02[f]	0.27±0.01[a]	1.71±0.04[a]	19.49±0.01[b]
Dspp	9.83±0.02[b]	4.04±0.00[f]	14.10±0.02[g]	1.08±0.00[e]	6.84±0.00[e]	22.70±0.18[c]
Ssp	14.17±0.20[f]	1.76±0.01[b]	11.03±0.03[d]	0.71±0.02[d]	4.41±0.13[d]	26.49±0.18[e]
AS	10.54±0.06[d]	1.27±0.00[a]	10.21±0.04[c]	1.14±0.01[f]	7.10±0.06[f]	27.60±0.01[f]
PT	10.22±0.15[c]	2.48±0.00[c]	9.86±0.05[b]	1.15±0.00[f]	7.16±0.01[f]	18.36±0.09[a]

[2] Kibuyuni region
Crude Protein = Nitrogen × 6.25
Values are expressed as mean±standard deviation, n=3
Values in the same column with different superscripts letters are significantly different ($p<0.05$)

4.0 Conclusion

The results obtained in the present study clearly showed that the proximate composition of various species from Mkomani and Kibuyuni (Kenya) varied slightly when compared to other seaweeds. Based on the biochemical analysis, the composition of the studied seaweed species (proteins, lipids and ash) is comparable to that of several seaweeds traditionally used in human and animal nutrition.

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