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Original Article

Do the Tissue Concentrations of Accumulated Arsenic, Calcium, Iron, Magnesium, Manganese, Potassium and Zinc Become Uniform throughout the Hijiki Plant Body with Growth?

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Summary

Hijiki (*Sargassum fusiforme*[***]) plant having genital organs was harvested on the Hime Coast, Kushimoto, Wakayama Prefecture, Japan, and the concentrations of accumulated arsenic, calcium, iron, magnesium, manganese, potassium, and zinc were determined. The fresh plants were washed thoroughly and cut at a length of 10 cm along the stalk from the bottom to the top of the plants. The twigs of the lower, middle and upper portions of the respective sections were separated to twig-stalks, twigs' large leaves and twig's small leaves. The genital organs of each section were also separated. The respective samples were weighed and stored under －40℃ until freeze-dried. The lyophilized samples were decomposed with conc HNO_3 and $HClO_4$ on an electric furnace, and the respective elements were determined with an atomic absorption spectrophotometer. These plants, having genital organs contained rather constant levels of calcium, 11.7 ± 1.4[*] (genital organs), ～ 12.2 ± 0.9[*] (leaves) mg *Ca*/g of dried tissues. They also contained rather constant levels of arsenic, 83.6 ± 12.07[*] (stalks) ～ 110 ± 27.2[*] (leaves) μg *As*/ g dry weight of tissues. Genital organs of some sections showed lesser arsenic accumulation than in most other sections, suggesting that accumulation started later in these sections. The concentrations of magnesium, manganese and zinc have not become uniform yet. However, the correlation coefficients between the accumulated magnesium and manganese were 0.93 to 0.69, and those between the accumulated manganese and zinc were 0.59 to 0.67. Thus, these elements seem to be accumulating more abundantly. Iron accumulation was, in average, 100.3 to 134.0 μg *Fe*/ g dry weight of tissues, but seems to be further continuing, as their accumulated concentrations had not become uniform yet. ([*] : average ± standard deviation)

Keywords: Hijiki (*Sargassum fusiforme*) plants; arsenic (*As*); calcium (*Ca*); iron (*Fe*); magnesium (*Mg*); manganese (*Mn*); potassium (*K*); zinc (*Zn*).

Introduction

In the previous papers[2-4)], we reported accumulation processes of arsenic, calcium, iron, magnesium, manganese and zinc during the growing period from November through April. Among the elements, the concentrations of calcium arrived at a constant value by April, but manganese and zinc showed various values, suggesting their later stabilization during growth. Thus, we attempted to determine these elements in more matured Hijiki plants having many genital organs.

Materials and methods

1. Samples of Hijiki plants

Hijiki, [*Sargassum fusiforme*, (Harvey) Setchell[***]], a family of Brown algae, grows on rocks on the sea-coast of Japan, bathed by the Kuroshio Current stream.

The embryos of Hijiki are fixed on rocks and germinate in summer[5-7)]. In early winter, they grow to the primary-leaf stage through the germlings in autumn. Thereafter, Hijiki grows to become adult plants, and drift away leaving their filamentous holdfasts in early autumn.

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*** Newly proposed taxonomic name of *Hizikia fusiformis* (Harvey) Okamura[1)].

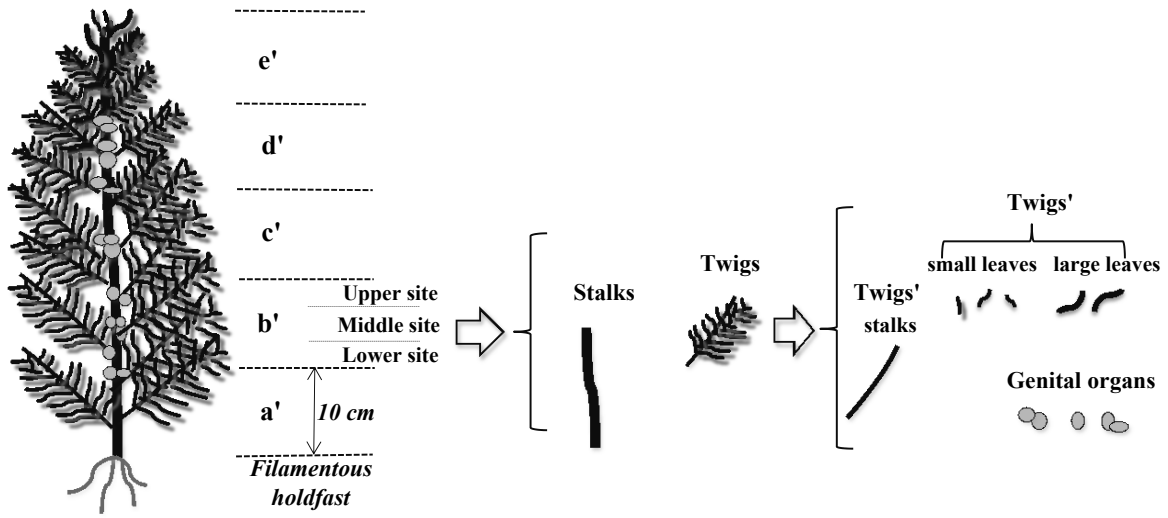

Fig. 1 Fractionation of Hijiki, *Sargassum fusiforme*, plants.
After washing, the sample plants were cut into pieces of 10 cm length, and separated to respective tissues, a stalk and twigs. The twigs were fractionated to upper-site, middle-site and lower-site positions. The twigs were separated to twigs' stalks, twigs' small and large leaves, and genital organs. The samples were stored under −40℃ until lyophilized.

Hijiki plants were harvested on the Hime Coast of Kushimoto District, Wakayama Prefecture, Japan, at the time of the lowest tide in April, 2013. The fresh plants had many genital organs at the bases of most of twigs, showing that they are much more matured than the samples of 2009-Apr, which had no genital organs[5-7] yet. The samples were brought back in an ice-cold box to the laboratory.

2. Preparation of Hijiki plant samples for analysis

One fresh plant was washed thoroughly with artificial sea-water three times and then with purified distilled water[8] three more times, and blotted each time with filter paper. The harvested plant was cut at a length of 10 cm along the stalk from the bottom to the top of the plants, and each section was designated as a', b' c' and so on from the bottom.

Twigs from the stalks were designated as lower-, middle- and upper-site twigs, being separated on the stalk by 3.3 cm. From the respective twigs, positioned at the lower-, middle- or upper-site, small and large leaves were taken off and collected according to the positions (Fig. 1). The genital organs, located at the joint of the twigs to the stalks, were also collected. They were weighed and stored under −40℃ until lyophilization.

3. Determination of arsenic (*As*); calcium (*Ca*); iron (*Fe*), magnesium (*Mg*), manganese (*Mn*), potassium (*K*) and zinc (*Zn*)

The samples were ashed in conc HNO_3 – $HClO_4$ on an electric heater for a few hours, and made up to a constant volume. ***As***, ***Fe***, ***Mn***, and ***Zn*** in HNO_3 solution were determined by the flame-less method on a pyro-coated graphite tube connected to an atomic absorption spectrophotometer (Shimadzu AA-7000 with ASC-7000 and GFA-7000). For ***As*** determination, 10 mg/L of palladium nitrate was added.

Ca, ***Mg***, and ***K*** were determined in acetylene gas flame with an atomic absorption spectrophotometer (Shimadzu AA-7000 with ASC-7000).

The respective samples were duplicatively and/or triplicatively determined.

4. Reagents

The reagents were of the JIS Special Grade or its equivalent. The standard solutions of calcium, iron, magnesium, manganese and zinc were of the JCSS grade, in 0.1 mol/L HNO_3, and those of arsenic (pH 5.0 with HCl) and potassium (in water) were of the JCSS grade. The palladium nitrate solution was of the AAS grade.

5. Statistic treatment

The respective values were expressed as average ± standard deviations, with sample numbers (n) in parenthesis. The correlation between the minerals' accumulation was expressed as Pearson's correlation coefficients one-tailed test, with p values. For multiple comparison test between the respective harvest times were done by Scheffe's F test. The statistic calculations were made by a built-in-function in Microsoft Excel 2011 (Mac version) and Statcel 3 (SSRI Co.), add-in forms on Excel.

Results

1. Water contents in the plants holding genital organs

Water contents in the respective tissues were at similar levels, indicating a similar maturity (Table 1). However, the water contents were somewhat lower in the stalks than in the twig's leaves.

2. Concentrations of potassium (*K*)

In general, the potassium concentrations in genital organs were less than half of those in the leaves, and those in the stalks were in-between (Table 2).

3. Accumulation convergence of arsenic (*As*)

Arsenic levels in the present samples (Table 3) were 83.6 ± 12.7 (stalks; n = 15) to 110.4 ± 27.2 (leaves; n = 22)

Table 1 Water (***H₂O***) contents, expressed as g ***H₂O***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	0.879			0.898	0.905	0.848	**
d'	0.866	Upper site	0.857	0.885	0.892	0.804	
		Middle site	0.850	0.882	0.896	0.812	
		Lower site	0.857	0.887	0.904	0.810	
c'	0.845	Upper site	0.853	0.890	0.899	0.803	
		Middle site	0.848	0.893	0.898	0.796	
		Lower site	0.846	0.892	0.903	0.814	
b'	0.831	Upper site	0.862	0.892	0.904	0.863	
		Middle site	0.865	0.820	0.906	0.816	
		Lower site	0.860	0.905	0.911	0.845	
a'	0.802	All twigs	0.866	0.904	0.908	0.841	

* Sections were designated from the lower site (filamentous holdfast site) to the upper site of the Hijiki plant as a', b', c', etc. Most of respective sections had twigs holding large and small leaves, positioned at lower site, middle site and upper site. The data columns were arranged in the order of the top to the bottom sections of the stalks.
** The section e' had no twigs and held leaves directly to the stalk.

Table 2 Potassium (***K***) contents, expressed as mg ***K***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	97.931			131.983	165.016	62.873	**
d'	96.129	Upper site	76.813	123.472	172.329	69.007	
		Middle site	60.984	119.813	150.571	41.697	
		Lower site	73.814	91.197	146.839	25.324	
c'	99.580	Upper site	78.423	136.894	166.356	46.472	
		Middle site	82.617	138.407	156.447	58.846	
		Lower site	85.406	141.471	158.923	64.120	
b'	87.630	Upper site	122.577	153.294	158.388	71.362	
		Middle site	114.919	144.330	198.604	62.402	
		Lower site	103.496	145.321	180.666	97.985	
a'	65.781	All twigs	75.354	150.245	184.012	72.903	

*, ** The explanations are as described in Table 1.

Table 3 Arsenic (***As***) contents, expressed as μg ***As***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	60.776			100.005	101.947	92.737	**
d'	81.607	Upper site	81.797	124.553	1.345	120.993	
		Middle site	74.156	135.476	123.101	89.564	
		Lower site	79.643	92.621	133.114	45.049	
c'	93.827	Upper site	90.667	121.757	125.617	103.533	
		Middle site	91.596	107.879	114.334	110.161	
		Lower site	82.118	113.289	125.585	116.214	
b'	76.000	Upper site	93.321	110.988	113.618	109.988	
		Middle site	109.930	135.714	122.441	0.884	***
		Lower site	94.705	102.851	112.077	109.372	
a'	80.570	All twigs	62.721	97.604	113.834	102.026	

*, ** The explanations are as described in Table 1.
*** Cited in the text of Results.

μg *As*/g dry weight of the tissues (Table 9). Comparison with the values of the samples harvested in 2008-2009[4] is described in Discussion.

Arsenic concentrations in the genital organs were 106.1 ± 9.7 (n = 9) μg *As*/g dry weight of the tissues, when the extraordinary low values of 2 specimens were excluded.

The above values had smaller standard deviations than those of the 2008-2009 samples, suggesting that the arsenic concentrations are approaching a uniform value.

4. Accumulation of calcium (*Ca*)

The average calcium levels in the leaves, stalks and genital organs were 11.9 ± 1.1 (n = 15), 12.2 ± 0.9 (n = 22), and 11.2 ± 2.1 (n = 11) mg *Ca*/g dry weight of the tissues (Table 4 and 9). One genital organ sample (the section **d'** of Lower site) showed the lowest level of calcium.

This sample also contained lower concentrations of ***Fe***, ***Mn***, ***Zn*** and ***As*** than the other samples.

5. Accumulation of iron (*Fe*) (Table 5)

Average ***Fe*** concentrations were more increased compared to the previous samples (2009-April)[3], but the differences between the individual sections did not become small.

This may suggest that iron accumulation continues further for a longer time.

Higher concentrations of ***Fe*** (their average was higher than 130 μg *Fe*/g dry weight of the tissues) were found in many genital organs than in the other tissues, but two samples out of the genital organs showed a lower iron accumulation (the section **b'** of Middle site and the section **d'** of Lower site).

6. Accumulation of magnesium (*Mg*)

The respective sections accumulated, in average, 7.75 ± 2.06 (stalks; n = 15), 7.69 ± 1.20 (leaves; n = 22) and 8.16 ± 10.82 (genital organs; n = 11) mg *Mg*/g dry weight of tissues (Table 6).

Although the respective sections accumulated various concentrations of ***Mg***, the accumulation of ***Mg*** had a strong correlation with that of ***Mn***. This correlation may suggest a biochemical basis for the accumulated concentrations.

Table 4 Calcium (***Ca***) contents, expressed as mg ***Ca***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	12.240			11.421	12.465	12.456	**
d'	14.099	Upper site	12.347	12.040	12.781	13.687	
		Middle site	12.354	13.471	12.400	10.923	
		Lower site	11.979	9.704	12.853	6.279	
c'	12.633	Upper site	12.153	12.969	12.386	12.060	
		Middle site	11.066	11.245	12.142	12.927	
		Lower site	11.669	11.615	12.020	13.239	
b'	12.015	Upper site	12.010	11.733	13.279	10.517	
		Middle site	10.800	11.828	13.160	10.824	
		Lower site	11.724	12.237	13.160	10.587	
a'	11.588	All twigs	9.034	11.138	11.282	9.172	

*, ** The explanations are as described in Table 1.

Table 5 Iron (***Fe***) contents, expressed as μg ***Fe***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	70.835			80.907	68.430	146.998	**
d'	88.166	Upper site	247.726	42.174	2.619	199.658	
		Middle site	333.820	84.721	52.956	301.684	
		Lower site	179.509	66.873	66.536	94.158	
c'	75.014	Upper site	108.945	100.535	163.479	151.947	
		Middle site	115.263	108.910	136.878	153.044	
		Lower site	100.579	163.033	189.051	163.760	
b'	26.319	Upper site	44.653	121.352	439.909	124.797	
		Middle site	26.143	82.876	60.513	5.7292	***
		Lower site	31.695	227.171	186.531	78.932	
a'	22.034	All twigs	34.060	38.867	35.868	53.712	

*, ** The explanations are as described in Table 1.
*** Cited in the text of Results.

7. Accumulation of manganese (*Mn*)

The plant body accumulated, in average, 4.35 ± 3.10 (stalks; n = 15), 5.36 ± 2.79 (leaves; n = 22) and 4.62 ± 2.03 (genital organs; n = 11) μg *Mn*/g dry weight of tissues (Table 7). Out of the genital organs, a sample (marked *** in the Table 7) showed lowest levels of accumulation of ***Zn*** (1.12 μg *Zn*/g dry weight of tissues) and ***As*** (Table 3).

8. Accumulation of zinc (*Zn*)

The plant accumulated 11.61 ± 8.57 (stalks; n = 15), 8.72 ± 6.08 * (leaves; n = 22), and 16.91 ± 12.35 (genital organs; n = 10) μg *Zn*/g dry weight of tissues, although variation of ***Zn*** accumulation levels among the respective sections was remarkable. In general, the stalks and leaves showed lower average values than the genital organs (Table 8). Those average values were lower because some sections have not yet achieved enough accumulation of ***Zn***.

Table 6 Magnesium (***Mg***) contents, expressed as mg ***Mg***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	7.654			7.288	7.462	8.818	**
d'	9.776	Upper site	11.623	7.588	7.411	10.936	
		Middle site	13.760	7.905	7.063	11.432	
		Lower site	10.650	5.824	7.192	4.747	
c'	9.648	Upper site	9.011	8.351	8.205	7.981	
		Middle site	8.790	7.870	8.929	8.660	
		Lower site	8.279	8.340	9.235	8.704	
b'	7.438	Upper site	8.035	7.468	11.447	7.355	
		Middle site	7.304	7.296	6.865	7.739	
		Lower site	6.722	6.406	8.583	7.152	
a'	7.424	All twigs	5.090	5.932	6.425	6.267	

*, ** The explanations are as described in Table 1.

Table 7 Manganese (***Mn***) contents, expressed as μg ***Mn***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	2.522			3.535	2.861	5.192	**
d'	4.616	Upper site	8.417	3.015	—	5.950	
		Middle site	12.505	4.061	3.601	8.274	
		Lower site	7.498	3.482	4.023	2.646	
c'	4.130	Upper site	5.227	5.347	6.367	5.379	
		Middle site	5.544	5.388	6.523	6.038	
		Lower site	5.129	7.887	8.192	5.577	
b'	1.491	Upper site	1.423	4.956	14.051	3.742	
		Middle site	1.617	5.586	4.285	0.075	***
		Lower site	1.748	5.444	10.024	3.675	
a'	1.902	All twigs	1.546	1.921	2.047	4.238	

*, ** The explanations are as described in Table 1.
*** Cited in the text of Results.

Table 8 Zinc (***Zn***) contents, expressed as μg ***Zn***/g dried weight of tissues.

Sections *	Stalks	Twigs	Twig's stalks	Twig's small leaves	Twig's large leaves	Genital organs	Note
e'	10.745			19.343	4.941	10.737	**
d'	3.943	Upper site	25.429	2.531	1.401	31.620	
		Middle site	25.360	6.772	4.555	47.462	
		Lower site	9.355	7.340	15.381	22.495	
c'	3.326	Upper site	9.824	6.908	17.185	10.609	
		Middle site	9.787	4.013	4.160	7.434	
		Lower site	29.627	7.492	6.540	10.497	
b'	6.669	Upper site	4.564	16.571	26.386	15.345	
		Middle site	3.408	5.416	8.148	1.123	***
		Lower site	8.185	6.785	6.345	17.781	
a'	4.762	All twigs	19.115	5.891	7.643	10.900	

*, ** The explanations are as described in Table 1.
*** Cited in the text of Results.

Table 9 The average values and the standard deviations of the determined values of the concentrations of accumulated calcium (*Ca*)*, arsenic (*As*)* and iron (*Fe*)** during the growth of Hijiki.

Years	***Ca*** (mg *Ca*/g dried weight of tissues)			***As*** (μg *As*/g dried weight of tissues)			***Fe*** (μg *Fe*/g dried weight of tissues)		
	Stalks	Leaves	Genital organs	Stalks	Leaves	Genital organs	Stalks	Leaves	Genital organs
2008-Nov	—	6.6 ± 1.66[e] (n = 5)	—	—	10.52 ± 2.87[k] (n = 5)	—	—	93.9 ± 17.7 (n = 5)	—
2009-Feb	4.39 ± 1.19[a, c] (n = 8)	5.97 ± 1.07[e] (n = 8)	—	13.39 ± 6.46[i] (n = 8)	3.46 ± 1.97[k] (n = 8)	—	64.2 ± 41.9 (n = 8)	40.6 ± 27.0 (n = 8)	—
2009-Mar	6.05 ± 0.88[a, d] (n = 12)	5.87 ± 1.27[e, g] (n = 12)	—	7.19 ± 3.01[i] (n = 12)	28.36 ± 12.93[k] (n = 12)	—	61.4 ± 59.0 (n = 12)	90.3 ± 38.2 (n = 12)	—
2009-Apr	6.73 ± 1.33[a, d] (n = 12)	7.37 ± 1.48[e, h] (n = 10)	—	8.56 ± 5.03[i] (n = 12)	20.16 ± 12.54[k] (n = 10)	—	87.9 ± 88.3 (n = 12)	108.0 ± 94.8 (n = 10)	—
2013-Apr	11.85 ± 1.08[b] (n = 15)	12.15 ± 0.89[f] (n = 22)	11.15 ± 2.12 (n = 11)	83.56 ± 12.73[j] (n = 15)	110.44 ± 27.21[m] (n = 22)	91.79 ± 34.68 (n = 11)	100.32 ± 90.15 (n = 15)	114.55 ± 90.15 (n = 22)	134.04 ± 78.60 (n = 11)

The values are expressed as average ± standard deviation.
* The *Ca* and *As* values of 2008-Nov, 2009-Feb, 2009-Mar and 2009-Apr were described in Reference 4.
** The *Fe* values of 2008-Nov, 2009-Feb, 2009-Mar and 2009-Apr were described in Reference 3.
By Scheffe's F test, there are significant differences between a and b, c and d, e and f, g and h, i and j, and k and m ($p < 0.01$), and between g and h ($p < 0.05$).

9. Correlation coefficients among the accumulation levels of arsenic (*As*), calcium (*Ca*), manganese (*Mn*), magnesium (*Mg*), and zinc (*Zn*)

In the 2013 samples, the correlation coefficient between ***As*** and ***Ca*** accumulation in genital organs was 0.5022 (p value = 0.049).

Strong correlations were observed between ***Mn*** and ***Mg*** accumulation as well as between ***Mn*** and ***Zn*** accumulation.

The correlation coefficients of ***Mg*** and ***Mn*** accumulation were 0.9286 ($p = 2.9 \times 10^{-7}$) in stalks, 0.8145 ($p = 0.02$) in leaves and 0.6922 ($p = 0.0063$) in genital organs.

The correlation coefficients of ***Mn*** and ***Zn*** accumulation were 0.5894 ($p = 0.0063$) in stalks, 0.6465 ($p = 0.00077$) in leaves, and 0.6051 ($p = 0.0185$) in genital organs.

Out of the genital organs, one section (the section **b'** of Middle site, marked as*** in Table 3) contained the least concentration of ***As***, 0.884 μg *As*/g dry weight of tissues, in contrast to the other sections, which had 84 to 110 μg *As*/g dry weight of tissues. This section also accumulated smallest amounts of ***Fe*** (several percent of those in the other sections; *** in Table 5), ***Mn*** (a fortieth to eightieth of those in the other sections; *** in Table 7), and ***Zn*** (a tenth of those in the other sections; *** in Table 8). However, ***Ca***, ***Mg***, and ***K*** were of usual levels as in the other sections.

It is noteworthy that another section containing a rather lower accumulation level of ***As*** (the section **d'** of Lower site, Table 3) contained also a small amounts of ***Fe*** (Table 5), ***Mn*** (Table 7) and ***Zn*** (Table 8).

Discussion

Growth of Hijiki plants and accumulation of arsenic (*As*) and calcium (*Ca*):

The factors affecting the growth and mineral accumulation in Hijiki plants have not been identified yet, but the recent rising geological temperature of ocean water may affect the growth rate as well as the minerals' accumulation rates in Hijiki plants. Maturation of the present samples[5-7] seems to be advanced in comparison to the previous (2009) plants[2-4], because of their morphological features such as genital organs appearing at almost all of the sections[5-7].

The hitherto analyzed values of arsenic in the Hijiki plants harvested at various districts showed different concentrations in the respective sections, between the different stocks or between the districts of harvesting[4, 9-12]. The differences of arsenic concentrations in different sections in one plant suggest that accumulated arsenic may not be easily transferable between the tissues. Those plants on the way to arsenic accumulation during their growing period may have different rates of accumulation in their respective sections.

In comparison with ***As*** accumulation, ***Ca*** accumulation is different in the rate and manner, although both elements approach rather constant values, as shown in the 2013- samples; among the individual tissues (Table 9), less discrepancy of the ***Ca*** or ***As*** concentrations was observed. The Scheffe's F test indicate that the respective increments of ***As*** accumulation as well as ***Ca*** accumulation in leaves and stalks of 2013-April was significant in comparison with those of 2009-April ($p < 0.01$), respectively (Table 9).

Accumulations of iron (*Fe*):

The rate of ***Fe*** accumulation seems to vary according to the environmental and/or growing conditions. A marine algae, *Sargassum* sp.[13], harvested in Margarta Islands, Venesuela, accumulated ***Fe*** in the tissues ten times more than Hijiki plants.

The accumulation level of ***Fe*** in the Hijiki plants growing in the Kushimoto coast reached higher than 100 μg *Fe*/g dry weight of tissues, but these accumulation levels differed greatly among the respective samples. Our results in Table 5 (the section **b'** of Middle site-genital organs) showed much lower levels of ***Fe*** accumulation. The discrepancies of the ***Fe*** content among the 2013-samples suggest that ***Fe*** accumulation continues further. On the other hand, it is probable that in some ocean districts there are generally some factors delaying ***Fe*** accumulation in Hijiki tissues.

For a long time, dried Hijiki products in Japan have been considered as a ***Fe***-rich foodstuff[14]. However, some of the recent commercial products of Hijiki available in Japan contained extremely low levels of *Fe**. It seems necessary to investigate the reason for this by studying the growing conditions or circumstances of Hijiki plants as well as the processes of dried Hijiki production in factories. These must be performed in the global scale, because of the greater ratio of dried Hijiki being imported recently to Japan from foreign countries.

The magnesium (*Mg*) accumulation:

The lower level of ***Mg*** accumulation than in the previous samples should be investigated in correlation with the chlorophyll contents and/or with ***Mg*** compounds other than chlorophylls.

The ***Mg*** content ascribable to chlorophylls in a seaweed, *Ulva* leaves, was 0.13 mg/g dry weight of the tissues[15], a similar level to those in the terraneous plants such as wheat cultivars[16]. In the tissues of Hijiki plants (Table 6), the ***Mg*** accumulation could be five to ten times higher. Some compounds like magnesium-phytate, as found in brown rice, could contribute to the high contents of ***Mg***[3].

The lower levels of ***Mg*** in the 2013-samples may be ascribable to the lower contents of ***Mg*** compounds other than chlorophylls. Differences in various biochemical activities between the respective sections of lower or higher ***Mg*** accumulation levels are to be investigated.

Correlations of manganese (*Mn*) and magnesium (*Mg*):

In the 2013-samples, the accumulated ***Mn*** in the tissues had a strong correlation with the ***Mg*** contents.

These data suggest that there may be accumulation mechanisms of various elements underlying their biochemical relationships.

Manganese (*Mn*) and zinc (*Zn*) accumulation:

In the samples of April 2009, the accumulation rates of ***Mn*** and ***Zn*** showed strong correlations[2], even though their concentrations in the respective sections greatly differed. In the genital organs of the samples of April 2013, one section accumulating an extraordinary low concentration of ***Mn*** has a low concentration of ***Zn*** (Table 7 and 8, marked ***). This may indicate a biological significance in the relationship of both elements even at the lower accumulation levels.

Conclusion: From the view point of utilization as foodstuffs, younger Hijiki plants will be better than older ones, because of their less accumulation of arsenic in younger plants. By this time of growth, calcium is arriving at their peak concentration, but iron is still accumulating in Hijiki plants.

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