



# KNOWLEDGE UPDATE ON MACROALGAE FOOD AND FEED SAFETY

based on data generated in the period 2014-2019 by the Institute of Marine Research, Norway



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Kunnskapsoppdatering på makroalger som mat og fôr

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## Summary (English):

**It has previously been addressed that some species of macroalgae may contain elevated levels of iodine, cadmium and inorganic arsenic. With the increased amount of data in the present report it is now possible to discriminate between individual species that have high levels of these components and others that are within the normal range. Among the updates are also new data on microbiology and iodine reduction that may contribute to a better understanding of this food group that is new to most Europeans.**

This report follows up a previous report from 2016 pointing out knowledge-gaps in the area of food and feed safety regarding macroalgae. The levels of iodine, cadmium and inorganic arsenic were pointed out as the main challenges for macroalgae as food. Since 2016 a substantial number of samples of Norwegian macroalgae have been analysed, with data from 27 species and 14 of these with five or more samples. The report is based on about 400 analyses of cadmium, mercury, lead and iodine and 332 analyses of inorganic arsenic. This allows for a more detailed evaluation of many individual species compared to the previous report. The present report answers to a request from the Norwegian Food Safety Authority for updates on filling of the knowledge gaps from the previous report.

The main updates are:

- New data on iodine and metals with increased resolution at the species level.
- New data on inorganic arsenic identifies a group of macroalgae that hold substantially higher inorganic arsenic concentration than the normal range.
- Bioavailability of 73-78 % of iodine from sugar kelp was found in a rat model study
- Imported species with Asian origin had similar levels of iodine and heavy metals to closely related species from Norwegian waters.
- New results demonstrate iodine reduction in kelp through drying, boiling and frying.
- New data on kainic acid in dulse shows relatively low levels also in Norwegian dulse.
- New knowledge on microbiology shows that for products that have been heat treated some attention as to cold storage due to the possible presence of spore forming bacteria should be given, similar to what applies for other heat treated products as for example dairy products. Spore formers pose a low challenge for fresh or dried seaweed used directly.
- Data on macro- and microminerals are presented.
- Experimental use of macroalgae for fish feed via insects shows that macroalgae provides marine nutrients into the feed chain, but also that there is a risk that some batches of seaweed are exceeding the maximum levels for cadmium and arsenic in animal feed

The variation of inorganic arsenic concentrations is particularly large, both between and within species. Four species showed clearly higher levels than the rest. Oar weed (*Laminaria digitata*) showed a range from very low to very high concentrations, with more than 50 % of the samples showing high levels. The highest concentrations are at the level of the Asian produced hijiki (*Sargassum fusiforme*), that was also analysed. Several countries around the world have issued warnings for hijiki. Two local relatives of hijiki also show high concentrations, but more samples are needed to confirm this. A few samples of the close relative to oar weed, tangle (*Laminaria hyperborea*), show low concentrations of inorganic arsenic. More samples are needed to confirm this, but tangle may provide an alternative to oar weed for wild harvest.

In accordance with the previous report, cadmium concentrations are highest in the brown and red algae, but without a distinct group of macroalgal species with higher level than the rest as for inorganic arsenic.

Iodine levels are highest among the brown algae, in accordance with the previous report. The clearly highest levels are found in the kelp species in the *Saccharina* and *Laminaria* genera, with typical values between 2000 and 6000 mg/kg dry weight within both genera. Perennial brown algae, like wracks and others that do not shed the blade during winter, are lower, while green and red algae, including the popular sushi seaweed Nori, are relatively low in iodine.

The levels of lead in the various macroalgae are generally low. The levels of mercury are also low, and the proportion of the toxic form, methyl mercury, also seems to be low.

The higher number of data in this update also allows more precise consideration of the main commercial farmed species in Norway, sugar kelp (*Saccharina latissima*) and winged kelp (*Alaria esculenta*). Neither species are among the high-level species regarding inorganic arsenic and both are intermediate in cadmium concentrations. Regarding iodine, sugar kelp is among the high-level species and winged kelp intermediate.

For some of the species, some information is available on effects of biotic and abiotic factors on the content of

cadmium, iodine and inorganic arsenic. However, large variations are found in metal concentrations, and more knowledge of the factors causing such variation is needed to allow more predictable product quality for the seaweed industry.

Recent data on radioactivity in macroalgae is reviewed and the Norwegian monitoring programme, which includes macroalgae, is described. In general, and in accordance with the previous report, no radioactivity levels of concern with respect to food safety is found in seafood, including macroalgae.

Regarding microbiology, data on winged kelp and sugar kelp are described. Low microbial numbers for total aerobic count were found in all samples as well as low incidence of cold adapted bacteria and spore-forming bacteria. Furthermore, there were no detection of indicators of faecal contamination as enterococci and coliforms, nor pathogenic vibrios or *Listeria monocytogenes*. However, in several of the examined samples, spore forming *Bacillus* spp. were isolated and seems able to pose a challenge if not processing and storage conditions take their possible presence into account. To avoid revival and growth of *Bacillus* spores in heat treated products, continuous chilling is necessary. *Bacillus* spores possess a low risk for dried products and other products that are not heated.

Results are presented from a project on the iodine and metals in sugar kelp. Health effects of high iodine intake as well as bioavailability of iodine from sugar kelp was studied in a 13 weeks rodent trial. No harmful effects of high iodine or other content in the kelp could be found in any of the study groups. Healthy rats have high tolerance for iodine even at the high level in the present study, and the rat model was hence not suited for evaluation of harmful effects of high iodine intake. On the other hand, the high tolerance of the iodine intake allowed to conclude that none of the other components of the kelp had a negative impact on the health of the rats. The high tolerance of iodine in rats also made it possible to study the bioavailability of iodine from kelp at very high concentrations. Iodine availability was lower, but still high, from kelp (80 %) compared to iodine added as potassium iodide (95 %), and the kelp fed rats had more iodine in feces. The project also studied the geographical variation in iodine and metals in sugar kelp along the Norwegian coast in a standardised growth trial. The results showed no geographical trend in iodine or inorganic arsenic, while a clear increase in cadmium was seen from south to north. Another study examined the effect of dehydration and cooking on iodine in sugar kelp. Iodine is reduced through drying, boiling and frying processes, with prolonged simmering showing a substantial reduction in total in kelp and stock.

Results on nutrients with trace metals (iron, zinc and selenium) and macro minerals (calcium, potassium, magnesium, sodium and phosphorus) are described.

A SWOT analysis showed the potential of seaweed as a source of protein and other nutrients for salmon feed but emphasises the same challenges with anti-nutrients and accessibility of nutrients as for other non-animal protein sources. An alternative method for using macroalgae biomass in fish feed is via insect larvae, which overcomes the problem of high carbohydrate content in the macroalgae. Results from an international project showed that a seaweed-enriched media resulted in a more "marine" nutritional profile of the insect larvae. However, there is a risk that some batches of seaweed are exceeding the maximum levels for cadmium and arsenic in animal feed.

The process of submitting data on macroalgae to the EFSA database is described, and codes for the various species and products are given.

More knowledge is still needed in the field of food and feed safety regarding macroalgae. In particular, more data is needed for the species with low number of analyses in the present report, and in particular the species with high levels of inorganic arsenic and cadmium relative to the normal range. More data on inorganic arsenic in tangle should be acquired to explore the indication that the levels are drastically lower than the closely related oar weed. More data is also needed in general on seasonal and geographical variation as well as on the bioavailability of cadmium and inorganic arsenic relative to other food and feed sources.

## Summary (Norwegian):

**Det har tidligere blitt påpekt at noen arter av makroalger kan ha høye nivåer av jod, kadmium og uorganisk arsen. Med et langt høyere antall data i denne rapporten er det nå mulig å skille mellom arter som har høye nivåer av disse elementene og andre som er innenfor normalområdet. Oppdateringen inneholder også data på mikrobiologi og jodreduksjon som kan bidra til bedre forståelse av denne gruppen av matvarer som er ny for de fleste europeere.**

Denne rapporten følger opp en tidligere rapport fra 2016 der det ble pekt på kunnskapshull i forhold til makroalger og mattrygghet og som trygt får. Innhold av jod, kadmium og uorganisk arsen ble utpekt som hovedutfordringer for bruk av makroalger som mat. Siden 2016 har det blitt analysert et betydelig antall prøver av norske makroalger, med data fra 27, arter hvorav 14 av disse har fem eller flere prøver. Denne rapporten er basert på rundt 400 analyser av

kadmium og jod og 332 analyser av uorganisk arsen. Dette gjør det mulig med en mer detaljert vurdering av mange enkelt-arter sammenliknet med forrige rapport. Rapporten svarer opp forespørsel fra Mattilsynet der det bes om ny kunnskap som kan fylle kunnskapshullene fra den forrige rapporten.

De viktigste oppdateringene er:

- Nye data på jod og metaller med økt oppløsning på artsnivå.
- Nye data på uorganisk arsen viser en liten gruppe makroalger som har betydelig høyere konsentrasjoner enn normalnivå hos de andre artene.
- Biotilgjengelighet av jod på 80 % ble funnet i en studie med rottemodell.
- Importerte arter med asiatisk opprinnelse hadde nivåer av jod og tungmetaller tilsvarende nært beslektede arter fra Norge.
- Nye data viser reduksjon av jodinnhold i tare ved tørking, koking og steking.
- Nye data på kainsyre i søl viser relativt lave nivåer også i norsk søl.
- Ny kunnskap i forhold til mikrobiologi viser at det bør tas forholdsregler som god kjøling av varmebehandlede produkter av makroalger, tilsvarende det som er gjeldende for andre varmebehandlede produkter som for eksempel melkeprodukter, som følge av mulig forekomst av sporedannende bakterier. Sporedannende bakterier utgjør liten utfordring for ferske eller tørkede produkter brukt direkte.
- Data på makro- og mikromineraler er presentert.
- Eksperimentell bruk av makroalger som fôr til fisk via insekter viser at makroalger bidrar med viktige marine næringsstoffer til fôrkjeden, men også at det kan være risiko for at noen partier av makroalger kan overstige grenseverdiene for kadmium og arsen i dyrefôr.

Variasjonen i konsentrasjoner av uorganisk arsen var spesielt høy, både innen og mellom arter. Fire arter hadde klart høyere konsentrasjoner enn de resterende artene. Fingertare (*Laminaria digitata*) hadde et spenn i konsentrasjoner fra svært lave til svært høye verdier, der over 50 % av prøvene hadde høye konsentrasjoner. De høyeste konsentrasjonene i fingertare var på nivå med den asiatisk produserte hijiki (*Sargassum fusiforme*) som også ble analysert. Flere land rundt om i verden har gitt ut advarsler mot å spise hijiki. To lokale slektninger av hijiki hadde også høye konsentrasjoner, men det bør tas flere prøver for å bekrefte dette. Noen få prøver av stortare (*Laminaria hyperborea*), som er nært beslektet til fingertare, hadde svært lave konsentrasjoner av uorganisk arsen. Det bør tas flere prøver for å bekrefte dette, men stortare kan være et alternativ til fingertare for villhøsterne.

I samsvar med den forrige rapporten var konsentrasjonene av kadmium høyest i brunalger og rødalger, men uten en adskilt gruppe arter med høyere nivåer enn resten, slik det ble funnet for uorganisk arsen.

Innhold av jod var høyest blant brunalgene, i samsvar med forrige rapport. De klart høyeste konsentrasjonene ble funnet innenfor slektene *Saccharina* og *Laminaria*, med typiske konsentrasjoner mellom 2 000 og 6 000 mg/kg tørrvekt innenfor begge slektene. Flerårige brunalger, som tangsorter med flere som ikke mister bladene om vinteren, har lavere konsentrasjoner, mens grønналger og rødalger, inkludert den populære sushi-arten Nori, har relativt lavt innhold av jod.

Innholdet av bly i makroalger er generelt lavt. Nivåer av kvikksølv er også lave, og andelen av den toksiske formen metylkvikksølv ser også ut til å være lav.

Basert på den økte mengden data i denne oppdateringen er det nå mulig med en grundigere vurdering av sukkertare (*Saccharina latissima*) og butare (*Alaria esculenta*), som er de viktigste artene som dyrkes i Norge. Ingen av disse er blant artene med høyt innhold av uorganisk arsen og begge har middels høye konsentrasjoner av kadmium. Når det gjelder jodkonsentrasjoner er disse høye hos sukkertare og middels høye hos butare.

For noen av artene har vi nå informasjon om effekt av biotiske og abiotiske faktorer på innholdet av kadmium, jod og uorganisk arsen. Det er imidlertid stor variasjon i disse dataene, og det er behov for mer kunnskap om faktorer som er opphavet til denne variasjonen, slik at produktkvaliteten kan bli mer forutsigbar for næringen.

Nyere data på radioaktivitet i makroalger er presentert og det norske overvåkningsprogrammet, som inkluderer makroalger, er beskrevet. Generelt, og i samsvar med den forrige rapporten, er det ikke funnet nivåer av radioaktivitet som er problematisk i forhold til mattrygghet, i sjømat, inkludert makroalger.

Data på mikrobiologi i sukkertare og butare er gjennomgått. Lave verdier for totalt aerobt kimtall samt lav forekomst av kuldeadapterte og sporedannende bakterier ble funnet i alle prøvene. Videre ble det ikke detektert indikatorer på fekal kontaminering som enterokokker og coliforme bakterier, eller patogene vibrio eller *Listeria monocytogenes*. Det ble imidlertid isolert sporedannende *Bacillus* spp. fra flere av de undersøkte prøvene, noe som kan gi en utfordring om ikke forhold under prosessering og lagring tar hensyn til disse sporedannerne. For å unngå aktivering og vekst av *Bacillus* sporer i varmebehandlede produkter er det nødvendig med kontinuerlig kjøling. *Bacillus* sporer er et lite

problem for tørkede produkter og produkter som ikke er varmebehandlet.

Rapporten presenterer resultater fra et prosjekt på jod og metaller i sukkertare. Helseeffekter av høyt jodinntak samt biotilgjengelighet av jod fra sukkertare ble studert i et 13-ukers rotteforsøk. Ingen negative effekter av høyt jodinnhold eller andre komponenter i taren ble funnet i noen av forsøksgruppene. Friske rotter har høy toleranse for jod, selv ved det høye inntaket i dette forsøket, og rottemodellen var dermed ikke egnet til å vurdere effekter av høyt jodinntak. På den annen side gjorde denne toleransen det mulig å konkludere at heller ingen av de andre komponentene i tare hadde negative helseeffekter hos rottene. Den høye toleransen for jod gjorde det også mulig å vurdere biotilgjengeligheten av jod fra tare i høye konsentrasjoner. Tilgjengeligheten var lavere, men fortsatt høy, fra tare (80 %) sammenliknet med jod tilsatt som kaliumjodid (95 %), og rottene som fikk tare i fôret hadde mer jod i feces. Prosjektet undersøkte også geografiske variasjoner i innhold av jod og metaller i sukkertare langs norskekysten i et standardisert dyrkingsforsøk. Resultatene viste ingen geografiske trender i jod eller uorganisk arsen, mens en klar økning i konsentrasjoner av kadmium fra sør til nord ble funnet. Det ble også sett på effekt av tørking tilberedning av sukkertare på innhold av jod. Innholdet av jod ble redusert både ved tørking, steking og småkoking, og vedvarende småkoking viste en vesentlig nedgang i tare og kraft til sammen.

Rapporten presenterer også data på næringsstoff med spormineraler (jern, sink og selen) og makromineraler (kalsium, kalium, magnesium, natrium og fosfor).

En SWOT analyse viste potensialet til makroalger som en kilde til protein og andre næringsstoff til laksefôr, men fremhever de samme utfordringene i forhold til antinæringsstoff og tilgjengelighet av næringsstoff som for andre ikke-animalske proteinkilder. En alternativ metode for å bruke makroalger til fiskefôr er via insektlarver, noe som omgår utfordringen med høyt innhold av karbohydrater i makroalgene. Resultater fra et internasjonalt prosjekt viste at medium tilsatt makroalger ga en mer «marin» ernæringsprofil til insektlarvene. Det er imidlertid en risiko for at grenseverdien for kadmium og arsen i dyrefôr overstiges i noen partier med makroalger.

Proessen med innsending av data til EFSA databasen er beskrevet, og koder for de ulike artene og produktene er oppgitt.

Det er behov for mer kunnskap innen feltene mattrygghet og trygt fôr når det gjelder makroalger. Det er behov for mer data for arter med færrest antall analyser i denne rapporten, og spesielt for arter med høye verdier av uorganisk arsen og kadmium i forhold til normalnivåene. Mer data på uorganisk arsen i stortare er nødvendig for å undersøke videre indikasjonene på at nivåene i denne arten er så mye lavere enn den nært beslektede fingertaren. Det er også behov for mer data generelt på sesongmessig og geografisk variasjon og i tillegg mer kunnskap om tilgjengelighet av kadmium og uorganisk arsen fra makroalger i forhold til andre mat- og fôrressurser.

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# 1 - Introduction

In 2016, a technical report on potential risks posed by macroalgae for application as feed and food, from a Norwegian perspective was published by the Institute of Marine Research ([Duinker et al., 2016](#)). The Norwegian Food Safety Authority (NFSA) has now requested the Institute of Marine Research (IMR) for an updated knowledge status on macroalgae, with special emphasis on data on iodine and metals and new data that could change the conclusions from the 2016 report. Other data on macroalgal food and feed safety generated by the IMR since 2016, that might fill some of the knowledge gaps pointed out in the 2016 report, are also requested.

One of the main conclusions from the 2016 report was that macroalgae, in particular brown algae, may contain elevated levels of inorganic arsenic, total arsenic and cadmium. Further, the amount of data on Norwegian seaweed was too low, allowing only general conclusions. In 2018, EFSA made a call for data in the commission recommendation (EU) 2018/464 on the monitoring of metals and iodine in seaweed, halophytes and products based on seaweed, including both local and imported products.

Since 2016 the amount of chemical data generated by the IMR on Norwegian as well as imported macroalgae has increased substantially, and a monitoring programme was started according to EFSA's requirements. The main updated conclusion is that it now can be discriminated between individual species that have high levels of these components and others that are within the normal range. The 2016 report also revealed a lack of information on seasonal and geographical variation, which is still lacking except for a few species as discussed below. Some work has also been done on the presence of bacteria on kelp and will be summarised here. Studies on bioavailability of metals from kelp have been performed together with studies on the effect of cooking on iodine content, and ongoing student work on iodine is also described. Work on the use of macroalgae in fish feed is summarised as well as analyses of minerals. Finally, the process of submitting occurrence data to EFSA is described and discussed.



## 2 - Material and methods

Samples were retrieved by field work by IMR staff, kelp growers and wild harvesters during the period 2014-2019. Samples were freeze dried and homogenised before further analysis. The analyses are ISO accredited for most elements and otherwise analysed in our accredited labs, hence fulfilling the request in EFSA's call for data that the analyses should be carried out in accordance with Annex III to Regulation (EC) No 882/2004. Samples of arame, kombu, hijiki, wakame and nori are imported from production in Asia.

An overview of the species collected is given in Table 1.

Table 1 . List of species collected in the period 2014-2019 with Latin, English and Norwegian names.

	Latin name	English name	Norwegian name
Brown algae	<i>Alaria esculenta</i>	Winged kelp	Butare
	<i>Ascophyllum nodosum</i>	Rockweed	Grisetang
	<i>Chorda filum</i>	Dead man's rope	Martaum
	<i>Eisenia bicyclis</i>	Arame	Arame
	<i>Fucus serratus</i>	Toothed wrack	Sagtang
	<i>Fucus spiralis</i>	Spiral wrack	Kaurtang
	<i>Fucus vesiculosus</i>	Bladderwrack	Blæretang
	<i>Halidrys siliquosa</i>	Halidrys siliquosa	Skolmetang
	<i>Himanthalia elongata</i>	Thongweed	Remtang
	<i>Laminaria digitata</i>	Oar weed	Fingertare
	<i>Laminaria hyperborea</i>	Tangle	Stortare
	<i>Pelvetia canaliculata</i>	Channelled wrack	Sauetang
	<i>Saccharina latissima</i>	Sugar kelp	Sukkertare
	<i>Saccharina spp</i>	Kombu	Kombu
	<i>Sargassum fusiforme</i>	Hijiki	Hijiki
	<i>Sargassum muticum</i>	Wireweed	Japansk drivtang
<i>Undaria pinnatifida</i>	Wakame	Wakame	
Green algae	<i>Codium fragile</i>	Green sea fingers	Pollpryd
	<i>Ulva intestinalis</i>	Gutweed	Tarmgrønske
	<i>Ulva lactuca</i>	Sea lettuce	Havsalat
	<i>Ulva spp</i>	Green nori	Green nori
Red algae	<i>Chondrus crispus</i>	Irish moss	Krusflik
	<i>Palmaria palmata</i>	Dulse	Søl
	<i>Porphyra purpurea</i>	Purple laver	Purpurfjærehinne
	<i>Porphyra spp</i>	Nori	Nori
	<i>Porphyra umbilicalis</i>	Pink laver	Vanlig fjærehinne
	<i>Vertebrata lanosa</i>	Wrack siphon weed	Grisetangdokke

*Determination of metals (including Cd, Hg, Pb, Fe, Zn and Se) by ICPMS (IMR method 197)*

Two parallels were weighed from each sample. The metals were determined by inductively coupled plasma-mass spectrometry (ICP-MS) after decomposing in microwave oven as described by Julshamn et al. (2007). The method is accredited for cadmium (Cd), mercury (Hg), lead (Pb), zinc (Zn) and selenium (Se).

*Determination of macro minerals (Na, Mg, K, Ca and P) by ICPMS (IMR method 382)*

Two parallels were weighed from each sample. The concentrations of the macro minerals (Na, Mg, K, Ca and P) were determined by inductively coupled plasma-mass spectrometry (ICP-MS), after acid wet digestion in a microwave oven. The concentrations were determined using an external calibration (standard curve) and the method is accredited according to ISO 17025.

*Determination of inorganic arsenic by HPLC-ICPMS (IMR method 261)*

The sample was added 10 ml 0.07 mol/l  $\text{HNO}_3$  in 3 %  $\text{H}_2\text{O}_2$  and extracted in microwave oven for 20 minutes at 90 °C. The sample was then cooled, centrifuged and filtrated prior to analysis. Inorganic arsenic was selectively separated from other arsenic compounds by anion exchange HPLC and determined as  $\text{As}^{5+}$  by ICP-MS. The method is accredited according to iso-17025.

*Determination of iodine*

Samples were added 1 ml tetramethylammonium hydroxyide (TMAH) and 5 ml deionized water before extraction at  $90 \text{ }^\circ\text{C} \pm 3 \text{ }^\circ\text{C}$  for 3h. The samples were then diluted and centrifuged. Prior to quantification, the samples were filtered through a 0.45  $\mu\text{m}$  single use syringe and disposal filter. Tellurium which was used as an internal standard in order to correct for instrument drift. Iodine concentration in the samples was determined by inductively coupled plasma-mass spectrometry (ICP-MS).

## 3 - Results and discussion

### 3.1 - Occurrence of inorganic arsenic, iodine, cadmium, lead and mercury in macroalgae

Both fresh and dried macroalgae have been sampled and analysed. Dried products show higher concentrations than fresh, since dry matter percentage typically range from 10 to 30 % (see appendix) resulting in 3-10 times higher concentrations of metals after drying. As an example, median iodine concentration in 141 samples of fresh sugar kelp (*Saccharina latissima*) was 410 mg/kg wet weight while 16 samples of dried products had median concentration of 3650 mg/kg dry weight. For comparison purposes, all concentrations are converted to dry weight basis in the following review. 27 species have been sampled and analysed, and 14 of these have five or more samples.

The tables include both mean and median values, since in many instances the mean values are affected by a few atypically high values, and median values are more representable for the typical values. The 25 % quartile range is presented in addition to the minimum and maximum values for the same reason, and the upper 75 % quartile value is more suited for comparison of typically high values, not affected by a few extreme values.

At present there are no maximum levels (MLs) for minerals in seaweed as food in the EU, except for mercury, and a call for data for the period 2018-2021 has been made to generate a basis for evaluating establishment of MLs. As stated in the Commission Recommendation (EU) 2018/464 on the monitoring of metals and iodine in seaweed, halophytes and products based on seaweed: "For arsenic, cadmium and lead, maximum levels (MLs) for various foodstuffs are established under Commission Regulation (EC) No 1881/2006. However, currently no MLs are established for these substances in seaweed and halophytes, except for the MLs established under this Regulation for food supplements consisting exclusively or mainly of seaweed or products derived from seaweed". Concentrations in fresh seaweed have been converted to dry weight concentrations for comparisons with dry seaweed products. Dry weight percentages are presented in the appendix.

#### 3.1.1 - Inorganic arsenic

Content of inorganic arsenic show a particularly high variation (Table 1). Among the types of seaweed that have been sourced in Norwegian waters from mainly wild harvest, but also from cultivation, the oar weed (*Laminaria digitata*) shows the highest concentrations. The variation spans from the lowest concentrations to the highest concentrations in Table 1, and the highest concentrations are at the level of the Asian produced hijiki (*Sargassum fusiforme*), which several European countries (including Norway), USA and Canada have issued warnings for due to the high level of inorganic arsenic and increased risk of cancer. 50 % of the samples of oar weed have concentrations of inorganic arsenic above 24 mg/kg dry weight, and the three highest samples have concentrations between 63 and 79 mg/kg dry weight. Also cultivated oar weed showed very high concentrations. In a small study, inorganic arsenic was seen to increase substantially in oar weed between March and June (Duinker, 2014), but samples submitted by the industry in 2018 showed an opposite pattern. The concentration in one sample of Hijiki was 59 mg/kg dry weight, and levels from literature typically vary between 50 and 90 mg/kg dry weight (Rose et al., 2007; Yamashita, 2014). A related species to hijiki, the invasive wireweed (*Sargassum muticum*) also showed high concentration of inorganic arsenic in four samples with concentrations between 48 and 68 mg/kg dry weight. A native species, *Halidrys siliquosa* (sea oak, family *Sargassaceae* also including the *Sargassum* species), is also related to Hijiki and had concentrations between 2.4 and 42 mg/kg dry weight. These two latter species are not commonly used at the moment. On the other side, a species that is closely related to oar weed with quite similar morphology, *Laminaria hyperborea* (tangle), showed concentrations in the lower end of all seaweed species between 0.03 and 0.04 mg/kg dry weight in four

samples analysed. Several companies report a shift from *L. digitata* to *L. hyperborea* in their products. More samples are needed of both the *Sargassum* related species and *L. hyperborea* to confirm these findings.

The species with high concentrations of iAs range in median concentrations from 22 to 59 mg/kg dry weight, and there is then a distinct gap down to the other species where median concentrations are 100 times lower from 0.23 and below.

Table 2. Inorganic arsenic in macroalgae, mg/kg dry weight. Species presented in order by decreasing median concentrations. Cell colouring corresponds to brown, green and red algae.

Latin name	Common name	N	Mean	Median	Min-max	25 % Quartiles
<i>Sargassum fusiforme</i>	Hijiki	1	59	59		
<i>Sargassum muticum</i>	Wireweed	4	54	51	48-68	49-60
<i>Halidrys siliquosa</i>	Sea oak	8	12	7.6	2.4-42	4.7-13
<i>Laminaria digitata</i>	Oar weed	40	24	21	0.06-79	6.9-39
<i>Chondrus crispus</i>	Irish moss	2	0.23	0.23	0.21-0.25	0.21-0.25
<i>Ulva spp</i>	Green nori	1	0.21	0.21		
<i>Saccharina latissima</i>	Sugar kelp	77	0.17	0.16	0.03-0.67	0.11-0.22
<i>Chorda filum</i>	Dead man's rope	2	0.15	0.15	0.03-0.27	0.03-0.27
<i>Vertebrata lanosa</i>	Wrack siphon weed	19	0.27	0.15	0.04-1.04	0.11-0.24
<i>Codium fragile</i>	Green sea fingers	2	0.14	0.14	0.07-0.21	0.07-0.21
<i>Ulva intestinalis</i>	Gutweed	6	0.18	0.13	0.02-0.44	0.05-0.33
<i>Alaria esculenta</i>	Winged kelp	33	0.77	0.11	0.03-2.7	0.08-0.22
<i>Fucus serratus</i>	toothed wrack	18	0.14	0.1	0.01-0.56	0.06-0.18
<i>Fucus vesiculosus</i>	Bladderwrack	23	0.2	0.1	0.02-1.64	0.07-0.19
<i>Pelvetia canaliculata</i>	Channelled wrack	2	0.1	0.1	0.09-0.12	0.09-0.12
<i>Ulva lactuca</i>	Sea lettuce	10	0.14	0.09	0.03-0.45	0.06-0.12
<i>Palmaria palmata</i>	Dulse	23	0.22	0.09	0.02-1.03	0.04-0.28
<i>Ascophyllum nodosum</i>	Rockweed	22	0.11	0.04	<0.01-1.21	0.02-0.12
<i>Fucus spiralis</i>	Spiral wrack	2	0.04	0.04	0.03-0.05	0.03-0.05
<i>Himantalia elongata</i>	Thongweed	4	0.04	0.04	0.01-0.05	0.02-0.05
<i>Porphyra purpurea</i>	Purple laver	3	0.09	0.04	0.03-0.19	0.03-0.19
<i>Laminaria hyperborea</i>	Tangle	4	0.036	0.037	0.03-0.041	0.032-0.04
<i>Undaria pinnatifida</i>	Wakame	5	0.03	0.03	<0.01-0.06	0.03-0.04
<i>Porphyra spp</i>	Nori	11	0.08	0.03	0.01-0.3	0.02-0.13
<i>Eisenia bicyclis</i>	Arame	1	0.02	0.02	0.02-0.02	0.02-0.02
<i>Saccharina spp</i>	Kombu	4	0.03	0.02	0.02-0.05	0.02-0.04
<i>Porphyra umbilicalis</i>	Pink laver	5	0.03	0.02	0.01-0.06	0.02-0.05

### 3.1.2 - Cadmium

Regarding cadmium, the levels are in accordance with the 2016-report (Duinker et al., 2016), but the available information of specific species is improved. There is no group of species that show drastically elevated concentrations like for inorganic arsenic, and the concentration ranges overlap (Table 2). Red and brown algae have the highest concentrations, while green algae are very low in cadmium. Sugar kelp has the highest concentrations for the northern localities as discussed below (“Geographic variation of cadmium in farmed sugar kelp”).

Table 3. Cadmium concentrations in macroalgae, mg/kg dry weight. Species presented in order by decreasing median concentrations. Cell colouring corresponds to brown, green and red algae.

Latin name	Common name	N	Mean	Median	Min-max	25 % Quartiles
<i>Vertebrata lanosa</i>	Wrack siphon weed	18	3.4	3.3	2.1-5	2.7-3.9
<i>Undaria pinnatifida</i>	Wakame	5	2.7	3.1	0.72-4	2.6-3.2
<i>Sargassum fusiforme</i>	Hijiki	1	2.3	2.3	2.3-2.3	2.3-2.3
<i>Fucus serratus</i>	Toothed wrack	19	1.9	1.8	0.88-3.3	1.5-2.4
<i>Porphyra spp</i>	Nori	11	1.7	1.5	0.41-3.4	0.87-2.3
<i>Alaria esculenta</i>	Winged kelp	40	1.5	1.3	0.3-4.8	1-1.7
<i>Fucus vesiculosus</i>	Bladderwrack	27	1.4	1.2	0.41-3.1	0.79-2
<i>Saccharina latissima</i>	Sugar kelp	148	0.94	0.65	0.16-3.1	0.41-1.4
<i>Eisenia bicyclis</i>	Arame	1	0.6	0.6	0.6-0.6	0.6-0.6
<i>Himanthalia elongata</i>	Thongweed	5	0.78	0.56	0.39-1.8	0.45-0.66
<i>Porphyra umbilicalis</i>	Pink laver	6	0.57	0.49	0.19-1.3	0.39-0.56
<i>Fucus spiralis</i>	Spiral wrack	3	0.67	0.47	0.45-1.1	0.45-1.1
<i>Saccharina spp</i>	Kombu	4	0.46	0.47	0.15-0.75	0.29-0.63
<i>Laminaria hyperborea</i>	Tangle	1	0.82	0.82	0.82	0.82
<i>Porphyra purpurea</i>	Purple laver	3	0.67	0.39	0.17-1.5	0.17-1.5
<i>Sargassum muticum</i>	Wireweed	2	0.37	0.37	0.09-0.64	0.09-0.64
<i>Pelvetia canaliculata</i>	Channelled wrack	3	0.3	0.3	0.24-0.36	0.24-0.36
<i>Ascophyllum nodosum</i>	Rockweed	24	0.29	0.28	0.16-0.47	0.23-0.32
<i>Chorda filum</i>	Dead man's rope	2	0.27	0.27	0.07-0.47	0.07-0.47
<i>Palmaria palmata</i>	Dulse	26	0.37	0.25	0.05-1.6	0.15-0.37
<i>Laminaria digitata</i>	Oar weed	33	0.38	0.22	0.033-1.9	0.17-0.53
<i>Chondrus crispus</i>	Irish moss	2	0.21	0.21	0.14-0.28	0.14-0.28
<i>Halidrys siliquosa</i>	Sea oak	2	0.19	0.19	0.09-0.28	0.09-0.28
<i>Ulva intestinalis</i>	Gutweed	7	0.24	0.18	0.08-0.55	0.14-0.28
<i>Ulva lactuca</i>	Sea lettuce	12	0.17	0.15	0.08-0.34	0.12-0.22
<i>Ulva spp</i>	Green nori	1	0.08	0.08	0.08-0.08	0.08-0.08
<i>Codium fragile</i>	Green sea fingers	2	<0.06	0.06	<0.06-0.06	<0.06-0.06

### 3.1.3 - Iodine

The concentrations of iodine are in accordance with the 2016 report (Duinker et al., 2016), but with more detailed information at the species level. The kelp species in the *Saccharina* and *Laminaria* families, both in Norway and Asia, have the highest levels with typical concentrations between 3 000 and 4 000 mg/kg dry weight and maximum concentrations around 10 000 mg/kg dry weight (Table 3), which is higher than any other food group. Winged kelp and the related Wakame from Asia, however, have a concentration range one tenth or less of the *Laminaria* and *Saccharina* species, which is part of the reason that winged kelp is getting more popular for cultivation in Norway these days. In general, the brown perennial species are intermediate, and the green and red algae are relatively low in iodine concentrations. One exception here is *Vertebrata lanosa* (wrack siphon weed) which grows as a symbiont on *Ascophyllum* and has ten times higher iodine concentrations compared to the other red algae.

Table 4. Iodine concentrations in macroalgae, mg/kg dry weight. Species presented in order by decreasing median concentrations. Cell colouring corresponds to brown, green and red algae.

Latin name	Common name	N	Mean	Median	Min-max	25 % Quartiles
<i>Laminaria digitata</i>	Oar weed	33	5 100	5 000	1400-10000	3600-6400
<i>Laminaria hyperborea</i>	Tangle	1	4 200	4 200	4200-4200	4200-4200
<i>Saccharina latissima</i>	Sugar kelp	150	3 700	3 500	670-10000	2600-4600
<i>Saccharina spp</i>	Kombu	4	2 800	2 600	2100-4000	2100-3500
<i>Vertebrata lanosa</i>	Wrack siphon weed	18	2 500	2 200	710-6200	1900-3000
<i>Chorda filum</i>	Dead man's rope	2	850	850	120-1600	120-1600
<i>Alaria esculenta</i>	Winged kelp	30	840	740	70-2400	450-1100
<i>Halidrys siliquosa</i>	Sea oak	2	690	690	670-710	670-710
<i>Ascophyllum nodosum</i>	Rockweed	24	710	670	320-1500	510-800
<i>Fucus serratus</i>	Toothed wrack	20	650	620	280-1000	530-760
<i>Sargassum fusiforme</i>	Hijiki	1	490	490	490-490	490-490
<i>Eisenia bicyclis</i>	Arame	1	450	450	450-450	450-450
<i>Fucus vesiculosus</i>	Bladderwrack	27	380	310	140-830	210-520
<i>Sargassum muticum</i>	Wireweed	2	300	300	120-480	120-480
<i>Chondrus crispus</i>	Irish moss	2	260	260	200-330	200-330
<i>Palmaria palmata</i>	Dulse	26	300	260	15-790	130-430
<i>Pelvetia canaliculata</i>	Channelled wrack	3	210	200	200-220	200-220
<i>Undaria pinnatifida</i>	Wakame	5	150	160	39-280	110-170
<i>Fucus spiralis</i>	Spiral wrack	3	150	150	140-150	140-150
<i>Ulva intestinalis</i>	Gutweed	7	130	130	29-240	41-220
<i>Ulva lactuca</i>	Sea lettuce	12	110	100	37-290	53-120
<i>Ulva spp</i>	Green nori	1	92	92	92-92	92-92
<i>Porphyra purpurea</i>	Purple laver	3	67	79	22-100	22-100
<i>Porphyra umbilicalis</i>	Pink laver	6	68	69	14-140	15-100

Latin name	Common name	N	Mean	Median	Min-max	25 % Quartiles
<i>Himanthalia elongata</i>	Thongweed	5	90	59	41-230	58-61
<i>Porphyra spp</i>	Nori	11	51	37	8-100	32-85
<i>Codium fragile</i>	Green sea fingers	2	23	23	17-29	17-29

For adults, recommended daily intake of iodine is 150 µg and maximum daily intake is 600 µg. Nori sheets contained about 60 µg per sheet, and between 2 and 10 sheets should cover the range 150-600 µg iodine. 1 ml (1/5 teaspoon) of dried kelp flakes weighing approximately 100 mg correspond to approximately 400 µg of iodine, given a typical concentration of 4 000 mg/kg dry weight. Similar calculations could make producers able to advice intake to the consumers.

### 3.1.4 - Lead

There are generally low levels of lead in seaweed (Table 5), although large variation is seen within each species with a few relatively high values 5-10 times higher than the 75% quartile for some of the species. Such high values should be followed up in future studies.

Table 5. Lead concentrations in macroalgae, mg/kg dry weight. Species presented in order by decreasing median concentrations. Cell colouring corresponds to brown, green and red algae.

Latin name	Common name	N	Mean	Median	Min-max	25 % Quartiles
<i>Sargassum fusiforme</i>	Hijiki	1	1.6	1.6		
<i>Codium fragile</i>	Green sea fingers	2	1.4	1.4	0.4-2.3	0.4-2.3
<i>Undaria pinnatifida</i>	Wakame	5	0.76	0.93	<0.22-1.1	0.63-0.99
<i>Ulva spp</i>	Green nori	1	0.85	0.85		
<i>Ulva intestinalis</i>	Gutweed	7	0.89	0.67	0.21-3	0.36-0.82
<i>Vertebrata lanosa</i>	Wrack siphon weed	18	0.96	0.59	0.24-3.3	0.36-1.3
<i>Chorda filum</i>	Dead man's rope	2	0.37	0.37	<0.21-0.52	0.21-0.52
<i>Chondrus crispus</i>	Irish moss	2	0.35	0.35	0.3-0.41	0.3-0.41
<i>Fucus spiralis</i>	Spiral wrack	3	0.33	0.32	0.27-0.4	0.27-0.4
<i>Fucus serratus</i>	Toothed wrack	19	0.44	0.29	<0.13-1.7	0.2-0.5
<i>Halidrys siliquosa</i>	Sea oak	2	0.28	0.28	0.024-0.54	0.024-0.54
<i>Sargassum muticum</i>	Wireweed	1	0.28	0.28		
<i>Ulva lactuca</i>	Sea lettuce	12	0.62	0.27	<0.2-2.8	0.22-0.73
<i>Porphyra umbilicalis</i>	Pink laver	6	0.38	0.26	<0.22-0.84	<0.22-0.67
<i>Laminaria hyperborea</i>	Tangle	1	<0.25	<0.25		
<i>Alaria esculenta</i>	Winged kelp	38	0.66	0.24	<0.055-4.4	0.21-0.86
<i>Pelvetia canaliculata</i>	Channelled wrack	3	0.4	0.24	<0.21-0.75	<0.21-0.75
<i>Saccharina latissima</i>	Sugar kelp	148	0.33	0.24	<0.22-5.7	<0.22-0.27
<i>Fucus vesiculosus</i>	Bladderwrack	26	0.49	0.23	<0.077-3.3	0.19-0.34
<i>Palmaria palmata</i>	Dulse	25	0.33	0.23	<0.039-1.1	<0.21-0.41
<i>Porphyra purpurea</i>	Purple laver	3	0.24	0.23	0.055-0.44	0.055-0.44



Latin name	Common name	N	Mean	Median	Min-max	25 % Quartiles
<i>Ascophyllum nodosum</i>	Rockweed	24	0.34	0.22	<0.052-1.9	0.099-0.29
<i>Porphyra spp</i>	Nori	11	0.28	0.22	<0.21-0.8	<0.21-0.24
<i>Eisenia bicyclis</i>	Arame	1	<0.21	<0.21		
<i>Saccharina spp</i>	Kombu	4	<0.21	<0.21		
<i>Himanthalia elongata</i>	Thongweed	5	<0.2	<0.2		
<i>Laminaria digitata</i>	Oar weed	33	0.15	0.19	<0.021-0.64	0.065-0.21

### 3.1.5 - Mercury

The levels of mercury are generally low. Most mercury concentrations were below the limit of quantification (loq), and only 100 of 406 samples were above loq. Only five of the species had more than 50% of mercury concentrations above loq so that mean concentrations could be presented (Table 6). However, this does not reflect higher concentrations of mercury as loq was quite variable. Samples with high total mineral content were diluted more than other samples, resulting in higher loq, and some species had hence more concentrations above loq but lower concentrations than other species.

The highest max concentrations were found in sugar kelp, oar weed and wrack siphon weed with 0.08, 0.07 and 0.07 mg/kg dry weight, respectively.

*Table 6. Mercury concentrations, mg/kg dry weight. Number of samples above loq and total number of samples are presented. Upper-bound mean concentration is given when more than 50% of samples are above loq. For species with concentrations above loq, max concentration is given as the highest concentration above loq, although the highest loq is above this concentration for most species. Cell coloration is according to brown, green and red algae.*

Latin name	Common name	N>LOQ / total N	Mean	Median	Min-max
<i>Alaria esculenta</i>	Winged kelp	6/38		<0.044	<0.0043-0.054
<i>Ascophyllum nodosum</i>	Rockweed	12/24		<0.031	<0.0078-0.033
<i>Chorda filum</i>	Dead man's rope	0/2		<0.048	<0.045-<0.051
<i>Eisenia bicyclis</i>	Arame	0/1		<0.052	
<i>Fucus serratus</i>	toothed wrack	9/19		<0.015	<0.0045-0.015
<i>Fucus spiralis</i>	Spiral wrack	2/3	0.02	<0.008	<0.046-0.005
<i>Fucus vesiculosus</i>	Bladderwrack	14/27	0.028	0.022	<0.0065-0.022
<i>Halidrys siliquosa</i>	Halidrys siliquosa	1/2			<0.042-0.006
<i>Himanthalia elongata</i>	Thongweed	0/5		<0.043	<0.0049-<0.051
<i>Laminaria digitata</i>	Oar weed	17/33	0.03	0.024	<0.0059-0.067
<i>Laminaria hyperborea</i>	Tangle	0/1		<0.051	
<i>Pelvetia canaliculata</i>	Channelled wrack	1/3		<0.035	<0.033-0.042
<i>Saccharina latissima</i>	Sugar kelp	17/148		<0.047	<0.0098-0.081
<i>Saccharina spp</i>	Kombu	0/4		<0.052	<0.051-<0.053
<i>Sargassum fusiforme</i>	Hijiki	0/1		<0.056	
<i>Sargassum muticum</i>	Wireweed	0/2		<0.044	<0.04-<0.048
<i>Undaria pinnatifida</i>	Wakame	0/5		<0.053	<0.053-<0.054
<i>Codium fragile</i>	Green sea fingers	0/2		<0.05	<0.042-<0.058
<i>Ulva intestinalis</i>	Gutweed	3/7		<0.045	<0.005-0.011
<i>Ulva lactuca</i>	Sea lettuce	4/12		<0.045	<0.0035-0.009
<i>Ulva spp</i>	Green nori	0/1		<0.053	
<i>Chondrus crispus</i>	Irish moss	2/2	0.006	0.0059	0.005-0.007
<i>Palmaria palmata</i>	Dulse	4/26		<0.043	<0.0042-0.005

<i>Porphyra purpurea</i>	Purple laver	2/3	0.006	0.005	<0.005-0.007
<i>Porphyra spp</i>	Nori	0/11		<0.052	<0.048-<0.055
<i>Porphyra umbilicalis</i>	Pink laver	1/6		<0.043	<0.005-0.007
<i>Vertebrata lanosa</i>	Wrack siphon weed	5/17		<0.047	<0.017-0.068

### 3.1.6 - Commercially cultivated species: Sugar kelp and winged kelp

The higher number of data in this update also allows more precise consideration of sugar kelp and winged kelp, which are the most common species for both cultivation and wild harvest in Norway at the moment. Neither of these species are among the high concentration species regarding inorganic arsenic. Sugar kelp is number seven and winged kelp number 12 in the list based on decreasing median concentrations of inorganic arsenic (Table 2). For the winged kelp one extreme value of inorganic arsenic was seen with more than ten times higher concentration than the 75 % percentile. This could be associated with epiphyte algae as suggested below (3.2), and this should be followed up with further studies. Winged kelp and sugar kelp have intermediate cadmium concentrations present as number six and eight on the list with decreasing median values (Table 3). Regarding iodine, sugar kelp is among the species with the highest concentrations and winged kelp intermediate (Table 4). The higher concentrations of cadmium in sugar kelp are found in more northern areas as discussed below (“Geographic variation of cadmium in farmed sugar kelp”).

## 3.2 - Factors affecting the levels of metals in kelp

As discussed below (“Geographic variation of cadmium in farmed sugar kelp”), cadmium concentrations in farmed sugar kelp showed a clear increase from south to north in Norway. The same section also discusses how iodine is reduced during cooking and lack of seasonal variation for farmed kelp.

During a master thesis study of wild and cultivated sugar kelp and winged kelp (Kleppe, 2016), cadmium decreased with increasing size of the plants, which suggests that fast growing individuals have lower concentrations of cadmium than slow growing individuals. Cadmium concentrations also decreased from the stipes and growth zone towards the tip. Iodine showed a similar trend within the plants, but no clear correlation with size. Inorganic arsenic showed no consistent variation but was usually highest in the mid-section of the blades of both species. Fouling was found to increase metal concentrations with higher cadmium concentrations in areas of sugar kelp covered with the bryozoan *Membranipora membranacea*, while an unknown filamentous alga increased concentrations of inorganic arsenic substantially in wild winged kelp. Differences were seen between wild and cultivated plants, but the differences were inconsistent between the two species and between the different minerals.

The amount of data on season, geography, exposure etc. are still scarce and more studies are needed. The variation even within the same locality (data not shown) for cultivated kelp is surprisingly high, and more knowledge of the factors causing such variation is needed to allow more predictable product quality for the seaweed industry.

## 3.3 - Radioactivity

Macroalgae are known to effectively concentrate radionuclides from seawater and are therefore widely used as a bio-indicator for radioactive pollution in the marine environment (e.g. Keogh, 2006; Kershaw et al., 2005). For example, *Ascophyllum nodosum* are known to have a high uptake of the radionuclide <sup>99</sup>Tc (Sjøtun et al., 2011), with increasing concentrations from young to older growth segments (Heldal and Sjøtun, 2010). Nevertheless, few publications describe food safety aspects of radionuclides in macroalgae. Tuo et al. (2016) found no higher

levels of either natural ( $^{238}\text{U}$ ,  $^{226}\text{Ra}$ ,  $^{228}\text{Ra}$ ,  $^{40}\text{K}$ ) or anthropogenic ( $^{137}\text{Cs}$ ) radionuclides in different seaweed species compared to other seafood, vegetables or meat products. Similarly, Moreda-Piñeiro et al. (2011) concluded that radiation levels in typical Japanese and Korean foodstuff, which include seaweed, are safe and at the same level as other countries.

The Norwegian marine monitoring programme Radioactivity in the Marine Environment (RAME) ([www.dsa.no](http://www.dsa.no)) charts trends of radionuclides in the marine environment. The results show that levels of the beta emitter  $^{99}\text{Tc}$  in *F. vesiculosus* collected along the Norwegian coast in the period 2012-2014 did not exceed 70 Bq/kg dry weight (d.w.) (Skjerdal et al., 2017). Further, the levels of the gamma emitter  $^{137}\text{Cs}$ , in *F. vesiculosus* collected in the same area and period ranged from 0.17 Bq/kg (d.w.) to 3.2 Bq/kg (d.w.). Levels of the alpha emitter  $^{239,240}\text{Pu}$  were below the detection limit in samples collected in the same area and period (Skjerdal et al., 2017). There are no maximum permitted levels for  $^{99}\text{Tc}$  and  $^{239,240}\text{Pu}$  in foodstuffs in Norway. The maximum permitted level for  $^{137}\text{Cs}$  in foodstuffs set by Norwegian authorities after the Chernobyl accident is 600 Bq/kg. The  $^{137}\text{Cs}$ -levels in macroalgae along the Norwegian coast are far below this and should be of little or no concern to seafood consumers. In general, no radioactivity levels of concern with respect to food safety is found in seafood. Levels of natural radionuclides are generally higher than levels of anthropogenic radionuclides. For example, Tuo et al. (2016) found that the natural radionuclide  $^{40}\text{K}$  accounted for around 87% of the total dose from the radionuclides  $^{238}\text{U}$ ,  $^{226}\text{Ra}$ ,  $^{228}\text{Ra}$ ,  $^{40}\text{K}$  and  $^{137}\text{Cs}$  in foodstuff from coastal areas of China.

### 3.4 - Other components

A few samples of kelp have been analysed for **dioxins and PCB's** and show low concentrations, in accordance with the 2016 report. 13 samples of dulse were analysed for **kainic acid** at the Danish Technical University and showed a range from 5 to 180  $\mu\text{g/g}$  dry weight and hence confirm relatively low levels of this toxin. The toxicity of kainic acid has not yet been determined. However, in order to reach the hazardous levels of kainic acid similar to what was dosed in mice and rat experiments, a total amount of about 30 kg dry dulse of the 130  $\mu\text{g/g}$  dulse would have to be eaten (Mouritsen et al., 2013). Recently, higher values up to 500  $\mu\text{g/g}$  dry weight has been found in Danish dulse, but still almost 10 kg would be needed for toxic responses.

### 3.5 - Microbiology

Since 2016 IMR worked with microbiology in an industry project. Blikra et al. (2019) describe the food quality and microbial safety of the two brown macroalgae winged kelp (*Alaria esculenta*) and sugar kelp (*Saccharina latissima*) harvested and processed in Norway. Samples included raw and frozen kelp. The authors found for all samples low microbial numbers (1–3 log colony forming units/g) for total aerobic count, cold adapted bacteria and spore-forming bacteria. Furthermore, there were no detection of indicators of faecal contamination as enterococci and coliforms, nor pathogenic vibrios or *Listeria monocytogenes*.

However, in several of the examined samples, *Bacillus* spp. were isolated and seems able to pose a challenge if not processing and storage conditions take their possible presence into account.

Bacteria in the genus *Bacillus* may grow under aerobic and anaerobic conditions. The bacteria and their spores are widely distributed in the environment and has been isolated from a wide variety of foods, especially of plant origin, but also from meat, fish, and dairy products. The most important species in food microbiology is *B. cereus*, and bacteria in this group have been involved in several food borne infections and intoxications.

Most strains of *B. cereus* are able to grow in low-acid foods at temperatures down to 10 °C and up to 55 °C (optimum 30 to 40 °C). During the last decades, some strains of *B. cereus* have been found able to grow at

temperatures down to 4 °C. *Bacillus* species has been isolated from *sous vide* cod fillets at 5 °C and in many other *sous vide* products. Food containing more than 10<sup>4</sup> *B. cereus* cells per g may not be safe for consumption. This number is far above what was seen by Blikra et al., 2019.

Several foods like milk and rice are also known to contain *Bacillus* spp., and it is common routine that heat-treated food products containing milk or rice are stored cold to prevent *Bacillus* spp. from growing. Heat treatment usually gives a temperature high enough to kill vegetative bacterial cells and any competitive microbiota, but not *Bacillus* spores. After such heat treatment, spores may be reactivated and give multiplication of *Bacillus* without competing bacteria present. Control of *Bacillus cereus* is efficiently obtained by chilling, except for some few cold-adapted strains that mainly pose a challenge for dairy products. The same precautions should hence be taken for heat treated products containing macroalgae due to possible presence of *Bacillus* spp. spores.

### 3.6 - Bioavailability, geographical variation and effect of cooking on sugar kelp composition

This project has not yet been finalised and reported, but summary of the findings this far, is given below. The project as financed by the Norwegian Seafood Research Fund (FHF) and carried out in collaboration with the Technical University of Denmark (DTU) and the kelp producer Ocean Forest.

Original project title: «På sporet av ny mat»

#### *Experimental diet for rats added sugar kelp (*Saccharina latissima*)*

We have performed a 13-week feeding trial with female Wistar IGS rats to compare the health effects and bioavailability of iodine from farmed sugar kelp with potassium iodine. No signs of toxic responses were found in animals with 0.5 and 5% sugar kelp or comparable amounts of potassium iodine in the diet. No difference in body weight, feed intake, heart-, kidney- or live weight were observed in the animals fed dried kelp in the diet at 0.5 and 5%. Biomarkers for liver and kidney damage measured in plasma were not increased in rats given potassium iodide or iodine from sugar kelp. No significant differences in serum concentrations of the thyroid stimulating hormone (TSH) and thyroid hormone T3 concentrations were observed. However, a small but significant reduction in serum T4 was observed in the rats fed the highest concentration of sugar kelp and potassium iodide.

Collectively, the observed high tolerance of iodine in rats agree with previous studies (Calil-Silveira et al., 2016; Yoshida et al., 2014). Yoshida et. al (2014) reported no significant effects on serum T3 and T4 concentration in rats despite an iodine intake up to 3500 µg iodine per day from kelp.

Based on available literature and the results from our experiment, it seems clear that rats have a high tolerance for iodine, even at the high intake of sugar kelp (5% of the diet) in the present study. The rat model used was hence not suited for evaluation of negative effects of iodine. On the other hand, the high tolerance of the iodine allowed to conclude that none of the other components of the kelp had a negative impact on the health of the rats. The high tolerance of iodine in rats also made it possible to study the bioavailability of iodine from kelp at very high concentrations.

#### *Bioavailability of iodine from sugar kelp in rats*

From previous studies both in animals (Yoshida et al., 2014) and humans (Aquaron et al., 2002; Combet et al., 2014), based on excretion of iodine in urine, there are indications that the bioavailability of iodine is lower from kelp compared to the salt form of iodine, potassium iodide. Most of absorbed iodine is excreted in the urine, and only a small fraction is retained in the thyroid gland or found in circulation. The amount of iodine excreted in urine is hence used as a measure for the amount of absorbed iodine over a time period. By monitoring urinary iodine excretion in rats from the 13-week feeding trial, the bioavailability of iodine from sugar kelp was evaluated. Urine excretion of iodine reflects the dietary intake in all experimental groups. 94-95 % of total iodine intake was excreted in urine in both the low and high dose of potassium iodide. Urine excretion in rats given iodine from sugar kelp, show a significantly lower excretion level in urine with 73-78% bioavailability of iodine. This is still relatively high, but significantly lower compared to potassium iodide. These results are in agreement with data reported on bioavailability of iodine in humans using both potassium iodide (96.4%) and seaweed (93 and 75%) (Aquaron et al., 2002). A lower bioavailability of iodine from seaweed were also observed when comparing three iodine-rich foods in a human intervention pilot study. Iodine excretion in urine were 86%, 87% and 60% with 36 hours collection after consumption of fish, milk or seaweed, respectively (Redway et al., 2018). The rat model used in our study was probably well suited for evaluation of iodine availability, although it was not

suited for evaluation of toxic effects as discussed above.

In contrast to earlier reports we quantified iodine in feces. A lower urine excretion of iodine in rats given sugar kelp was accompanied by a significant increase in iodine found in feces. Diets supplemented with potassium iodide resulted in 0.5-1 % of the iodine in feces on average. In contrast, rats given kelp in the diet had on average 8% of total iodine in feces, representing iodine not absorbed in the intestine from the kelp diet.

#### *Bioavailability of total and inorganic arsenic, cadmium and copper from sugar kelp in rats*

More than 70 % of ingested total arsenic in rats fed the a diet with 5 % sugar kelp was found in the 24 hours feces collection. This is in accordance with a high proportion of arsenic being organic, hence having a high availability. For cadmium, the amounts in the 24 hours feces collection were almost identical to the ingested amount, indicating very low availability. This is in accordance with a generally low availability of inorganic metals forms. Some cadmium was assimilated, though, as seen from the significant increase in cadmium in liver and kidney in the 5% diet group. Regarding copper, the concentrations in the kelp were too low to give differences in the feed between the groups, and there were no differences in the liver or kidney samples.

Regarding inorganic arsenic, rats and other rodents are not suitable models since these animals metabolise and excrete inorganic arsenic at high rates. Inorganic arsenic was only detected in the 5 % kelp group feed, and no concentrations above limit of quantification (LOQ) could be found in the liver or kidneys samples from any of the groups.

#### *The effect of cooking on iodine levels in sugar kelp*

Iodine is water soluble, and iodide ( $I^-$ ), probably the main form in kelp, reacts with water forming the volatile hydrogen iodide. Several forms of cooking were performed to assess the effect of iodine content of the sugar kelp. Boiling in water for 15 minutes released 50-90 % of iodine to the water, and of this iodine 50% was released to air. Continued simmering of the stock reduced further 50 % over 15 minutes. Frying released in average 50 % (25-80) of the iodine. Drying of sugar kelp increases concentrations of all elements ten times just by removing the water, which makes 90 % of the wet weight. However, when comparing iodine concentrations on dry weight, it was seen that about 25 % of the iodine evaporated during drying.

#### *Geographic variation of cadmium in farmed sugar kelp*

In cooperation with SINTEF (Macrosea project), a standardised growth trial was performed on localities all along the coast of Norway, and samples were analysed for metals at the IMR. The results showed a clear trend with increasing concentrations of cadmium from south to north. The levels in the north (68-70N) were above 1.0 mg/kg dry weight which is the limit for inclusion in fish feed, and more than four times higher than the southern samples (58-60N). Inorganic arsenic and iodine did not show a similar trend.

### 3.7 - Student projects on iodine

There are three relevant master theses at the IMR with main focus on iodine. Two of these are based on purchased seaweed products from Norwegian web shops. The products are analysed for iodine, total arsenic, mercury, lead and cadmium. This work also includes blood and urine samples from some seaweed eaters and are analysed for iodine status and thyroid markers. The third study has included four of the purchased seaweed products in a vegan week menu. These daily menus are analysed with and without seaweed for iodine and metals. All three master theses were defended in June 2020 and the main results will be published in peer reviewed journals in 2020-21.

### 3.8 - Nutrient analyses

The trace minerals, iron (Fe), zinc (Zn) and selenium (Se), were analysed together with the heavy metals and metalloids and hence with the same number of analyses. The macro minerals, calcium (Ca), potassium (K), magnesium (Mg), sodium (Na) and phosphorus (P), were analysed separately with in total 104 analyses. Seven species had three or more samples. The results are presented in Table 7 and Table 8.



Table 7. Selenium (Se), iron (Fe) and zinc (Zn) medians ( $\pm$  25% percentiles) concentrations in macroalgae, mg/kg dry weight. Species are presented in order by decreasing median concentrations. Cell colouring corresponds to brown, green and red algae.

		N	Se			N	Fe			N	Zn	
<i>Vertebrata lanosa</i>	Wrack siphon weed	18	0.83	(0.69-1.2)	Hijiki	1	820		Wrack siphon weed	18	95	(18-100)
<i>Sargassum fusiforme</i>	Hijiki	1	0.22		Green nori	1	480		Tangle	1	76	
<i>Porphyra spp</i>	Nori	11	0.21	(0.16-0.25)	Wrack siphon weed	18	340	(260-540)	Toothed wrack	18	65	(19-86)
<i>Ulva spp</i>	Green nori	1	0.2		Irish moss	2	260	(190-330)	Pink laver	6	59	(6-70)
<i>Chondrus crispus</i>	Irish moss	2	0.19	(0.14-0.23)	Gutweed	5	230	(160-580)	Hijiki	1	39	
<i>Undaria pinnatifida</i>	Wakame	5	0.17	(0.16-0.23)	Nori	11	160	(90-180)	Wakame	5	37	(5-41)
<i>Palmaria palmata</i>	Dulse	23	0.16	(0.14-0.36)	Wireweed	2	160	(32-280)	Bladderwrack	27	36	(27-60)
<i>Porphyra purpurea</i>	Purple laver	3	0.16	(0.046-0.39)	Wakame	5	150	(120-160)	Oar weed	33	36	(33-41)
<i>Sargassum muticum</i>	Wireweed	2	0.14	(0.08-0.2)	Spiral wrack	3	130	(120-240)	Winged kelp	37	35	(38-49)
<i>Ulva intestinalis</i>	Gutweed	5	0.14	(0.11-0.38)	Sea lettuce	10	120	(90-300)	Thongweed	5	35	(5-39)
<i>Laminaria hyperborea</i>	Tangle	1	0.13		Channelled wrack	3	120	(25-300)	Dead man's rope	2	30	(2-47)
<i>Fucus serratus</i>	Toothed wrack	18	0.12	(0.089-0.12)	Purple laver	3	110	(89-330)	Rockweed	24	29	(24-47)
<i>Codium fragile</i>	Green sea fingers	2	0.12	(0.1-0.13)	Pink laver	6	110	(58-110)	Dulse	23	28	(26-33)
<i>Ulva lactuca</i>	Sea lettuce	10	0.12	(0.068-0.22)	Dulse	23	100	(84-160)	Channelled wrack	3	25	(3-36)
<i>Alaria esculenta</i>	Winged kelp	37	0.11	(0.09-0.15)	Toothed wrack	18	78	(46-110)	Sugar kelp	146	24	(148-32)
<i>Chorda filum</i>	Dead man's rope	2	0.11	(0.09-0.13)	Bladderwrack	27	68	(41-130)	Nori	11	22	(11-35)
<i>Eisenia bicyclis</i>	Arame	1	0.1		Rockweed	24	62	(33-95)	Wireweed	2	19	(2-29)
<i>Saccharina spp</i>	Kombu	4	0.1	(0.098-0.1)	Winged kelp	37	56	(44-68)	Arame	1	16	
<i>Fucus spiralis</i>	Spiral wrack	3	0.099	(0.095-0.11)	Halidrys siliquosa	2	56	(16-96)	Green nori	1	13	
<i>Pelvetia canaliculata</i>	Channelled wrack	3	0.095	(0.051-0.098)	Green sea fingers	2	51	(36-65)	Gutweed	5	12	(7-18)
<i>Saccharina latissima</i>	Sugar kelp	146	0.095	(0.074-0.12)	Sugar kelp	146	44	(31-65)	Sea lettuce	10	11	(12-14)
<i>Himanthalia elongata</i>	Thongweed	5	0.091	(0.049-0.094)	Oar weed	33	39	(18-78)	Spiral wrack	3	7	(3-8)
<i>Fucus vesiculosus</i>	Bladderwrack	27	0.089	(0.071-0.11)	Kombu	4	33	(20-41)	Green sea fingers	2	5.9	(2-5.9)

		N	Se			N	Fe			N	Zn	
<i>Ascophyllum nodosum</i>	Rockweed	24	0.084	(0.044-0.096)		Arame	1	32		Irish moss	2	5.6 (2-49)
<i>Halidrys siliquosa</i>	Halidrys siliquosa	2	0.08	(0.034-0.13)		Dead man's rope	2	23 (9.2-37)		Kombu	4	5.6 (4-7)
<i>Porphyra umbilicalis</i>	Pink laver	6	0.077	(0.067-0.098)		Tangle	1	20		Purple laver	3	2 (3-18)
<i>Laminaria digitata</i>	Oar weed	33	0.065	(0.042-0.091)		Thongweed	5	20 (14-22)		Halidrys siliquosa	2	2 (2-17)

Table 8. Calcium (Ca), potassium (K), magnesium (Mg), Sodium (Na) and phosphorus (P) concentrations medians ( $\pm$  25% percentiles) in macroalgae, mg/kg dry weight. Sorting order by decreasing median concentrations. Cell colouring: Brown, green and red algae.

		N	Ca		N	K		N	Mg		N	Na		N	P					
<i>Sargassum muticum</i>	Wireweed	2	22 000	(22000-22000)	Dulse	6	99 000	(43000-140000)	Green sea fingers	1	31 000		Green sea fingers	1	170 000		Winged kelp	12	3 700	4100
<i>Ulva intestinalis</i>	Gutweed	2	22 000	(14000-29000)	Sugar kelp	51	91 000	(73000-130000)	Sea lettuce	2	26 000	(25000-28000)	Sugar kelp	51	48 000	(43000-56000)	Dulse	6	3 200	3700
<i>Alaria esculenta</i>	Winged kelp	12	19 000	(17000-22000)	Oar weed	5	80 000	(40000-82000)	Wireweed	2	11 000	(7200-14000)	Winged kelp	12	46 000	(40000-65000)	Purple laver	3	3 200	3300
<i>Fucus vesiculosus</i>	Bladderwrack	4	18 000	(15000-24000)	Winged kelp	12	62 000	(57000-66000)	Gutweed	2	9 900	(8700-11000)	Oar weed	5	44 000	(39000-56000)	Green sea fingers	1	2 700	
<i>Fucus spiralis</i>	Spiral wrack	2	18 000	(17000-18000)	Wireweed	2	61 000	(48000-75000)	Winged kelp	12	9 800	(8800-12000)	Thongweed	1	39 000		Irish moss	1	2 400	
<i>Himanthalia elongata</i>	Thongweed	1	18 000		Thongweed	1	47 000		Thongweed	1	9 400		Wrack siphon weed	1	36 000		Oar weed	5	2 300	3600
<i>Codium fragile</i>	green sea fingers	1	18 000		Wrack siphon weed	1	47 000		Irish moss	1	9 000		Toothed wrack	1	32 000		Sea lettuce	2	2 300	2400
<i>Ascophyllum nodosum</i>	Rockweed	6	16 000	(15000-17000)	Halidrys siliquosa	1	36 000		Rockweed	6	8 500	(8000-9500)	Rockweed	6	31 000	(27000-35000)	Gutweed	2	2 100	2500
<i>Fucus serratus</i>	toothed wrack	1	16 000		Toothed wrack	1	30 000		Sugar kelp	51	8 100	(7300-8900)	Bladderwrack	4	25 000	(20000-31000)	Sugar kelp	51	2 000	2600
<i>Halidrys siliquosa</i>	Halidrys siliquosa	1	16 000		Sea lettuce	2	30 000	(27000-34000)	Spiral wrack	2	7 900	(7500-8200)	Wireweed	2	25 000	(20000-29000)	Pink laver	2	2 000	2600
<i>Saccharina latissima</i>	Sugar kelp	51	15 000	(12000-17000)	Irish moss	1	30 000		Channelled wrack	1	7 900		Spiral wrack	2	24 000	(20000-27000)	Thongweed	1	1 500	

		N	Ca		N	K		N	Mg		N	Na		N	P					
<i>Laminaria digitata</i>	Oar weed	5	15 000	(13000-15000)	Bladderwrack	4	27 000	(21000-30000)	Toothed wrack	1	7 400		Channelled wrack	1	23 000		Wireweed	2	1 500	1700
<i>Pelvetia canaliculata</i>	channelled wrack	1	14 000		Purple laver	3	26 000	(15000-31000)	Oar weed	5	7 200	(6500-9600)	Sea lettuce	2	19 000	(8300-30000)	Wrack siphon weed	1	1 400	
<i>Chondrus crispus</i>	Irish moss	1	13 000		Spiral wrack	2	25 000	(21000-28000)	Bladderwrack	4	7 100	(6200-8100)	Irish moss	1	18 000		Bladderwrack	4	1 300	1700
<i>Vertebrata lanosa</i>	Wrack siphon weed	1	7 200		Pink laver	2	22 000	(13000-31000)	Wrack siphon weed	1	6 700		Pink laver	2	18 000	(5300-30000)	Halidrys siliquosa	1	1 100	
<i>Ulva lactuca</i>	Sea lettuce	2	6 000	(5100-6900)	Rockweed	6	17 000	(16000-25000)	Halidrys siliquosa	1	6 200		Dulse	6	15 000	(4100-20000)	Spiral wrack	2	960	1100
<i>Porphyra purpurea</i>	Purple laver	3	4 800	(2300-5400)	Channelled wrack	1	17 000		Pink laver	2	5 000	(4400-5700)	Halidrys siliquosa	1	13 000		Toothed wrack	1	760	
<i>Palmaria palmata</i>	Dulse	6	4 300	(2500-18000)	Gutweed	2	16 000	(12000-20000)	Purple laver	3	4 700	(3600-17000)	Purple laver	3	7 200	(6200-100000)	Rockweed	6	720	830
<i>Porphyra umbilicalis</i>	Pink laver	2	2 900	(2200-3500)	Green sea fingers	1	14 000		Dulse	6	2 800	(1200-3300)	Gutweed	2	7 000	(5500-8500)	Channelled wrack	1	700	

### 3.9 - Seaweed as a salmon feed resource

IMR conducted a SWOT analysis of the use of macroalgae in fish feed (Lock and Belghit, 2018). Although seaweeds are taxonomically not plants, many parallels between seaweeds and plants exist. Both can be a valuable source of nutrients that can be used by animals higher up the food chain, but both can also contain anti-nutritional factors, preventing them from being preyed on. The effect of a plain soybean meal on the development of enteritis in Atlantic salmon is well known and similar effects are seen of peas and other vegetable products. In commercial diets, it is highly processed protein concentrates of these plant products that are used today in Norway. Processing removes or reduces some of the anti-nutritional factors and simultaneously concentrates the protein content of the product. Salmon is a carnivorous fish that requires protein, lipid and micronutrients for healthy growth, while the requirement for carbohydrates is very low. Seaweed is mainly made-up of carbohydrates that cannot be used by the fish. This raises the obvious questions about post-harvest processing of seaweed to make nutrients more accessible and remove anti-nutritional factors, which currently are underdeveloped or non-existing. A fractionation of the seaweed biomass is needed where high-end products (e.g. alginates) can offset a large part of the production and processing costs. The lack of seaweed processing and diversification of the processing is the major hurdle for the use of seaweed in aquafeed. The SWOT analysis elaborates on the strengths and weaknesses of using seaweeds in feed for fish and pinpoints future changes that could stimulate (opportunities) or raise barriers (threats) in the application of marine macroalgae in aquafeed.

The high content of carbohydrates in seaweed limits the use of seaweed as a major component in fish feed. The use of insect larvae for processing seaweed is a way to overcome this problem by increasing the protein and lipid concentrations and decreasing the carbohydrate content. The insect larvae are then a promising feed source for fish aquaculture that was examined in the project Aquafly.

Larvae of the black soldier fly (*Hermetia illucens*; BSF) were reared on plant-based media enriched with the brown alga *Aschophyllum nodosum* (rockweed), in increasing percentages (from 0 to 100 % seaweed inclusion) (Liland et al., 2017). All the tested substrates allowed for BSF larval growth and development, although the best growth performance, nutrient utilization and retention were observed in larvae fed up to 50 % seaweed inclusion in the medium (Liland et al., 2017). The larvae fed seaweed-enriched media had a more "marine" profile than the control larvae, with the eicosapentaenoic acid (20:5n-3) (EPA), iodine and vitamin E introduced in the larvae from the seaweed in the media. The nutritional profile of the larvae fed seaweed better suited the dietary requirements of Atlantic salmon for these nutrients (NRC, 2011), compared to larvae fed media without seaweed.

The legislation in the European Union (EU) has set maximum levels (MLs) for undesirable substances in feed and food stuff (EC Directive 2002/32; Commission Regulation (EC) No 1881/2006). When seaweed is used as feeding substrate for insect larvae, levels of heavy metals and arsenic in the medium should be within MLs set by the legislation, as insects are considered full-fledged "farmed animals" by the EU (Regulation (EC) No 1069/2009; Commission Regulation (EU) 2017/893). Mixing seaweed biomass (rockweed) with the plant-based medium for BSF larvae, increased the concentrations of heavy metals (cadmium, lead and mercury) and arsenic in the media for the larvae, when more seaweed was included (Biancarosa et al., 2018).

Accumulation of cadmium and total arsenic in the insect larvae fed with different batches of rockweed was sometimes higher than the MLs. In a feeding trial, insect meal had concentrations above ML for arsenic, while whole feed concentrations were below ML (Biancarosa et al., 2019). Salmon were fed increasing levels of insect meal replacing fish meal, and levels of cadmium, mercury and lead in the feed were reflected in the salmon fillets. Interestingly, arsenic levels were similar in all feed groups, but total arsenic decreased with

increasing insect meal in the diet, suggesting low availability of the forms of arsenic in the insect meal.

## 4 - EFSA data submission

Initial work was done during the autumn of 2019 attempting to submit data on macroalgae to the EFSA database, using the new SSD2 format. However, for most of the species the necessary codes needed for the submission was not established and added to the database until after the deadline for submission in October 2019. Upon request a list of seaweed species were added to the FOODEX database catalogue, but not in the reporting hierarchy directly. These species can then be specified as sources within the different types of seaweed, like Sea belt as "Brown algae, SOURCE = Sea belt (as organism)" as seen in the table below.

Table 9. Codes for macroalgae species for reporting to the EFSA FOODEX database.

Common name	Latin name	Code	Text
Winged kelp/dabberlocks	<i>Alaria esculenta</i>	A00VK#F01.A18BQ	Brown algae, SOURCE = Dabberlocks (as organism)
Rockweed	<i>Ascophyllum nodosum</i>	A170Z	Rockweed
Irish moss	<i>Chondrus crispus</i>	A00VG	Carrageen mosses
Dead man's rope	<i>Chorda filum</i>	A00VK#F01.A18BK	Brown algae, SOURCE = Dead man's rope (as organism)
Green sea fingers	<i>Codium fragile</i>	A00VB#F01.A18BJ	Green algae, SOURCE = Green sea fingers (as organism)
Toothed wrack	<i>Fucus serratus</i>	A00VK#F01.A0B6S	Brown algae, SOURCE = Toothed wrack (as organism)
Spiral wrack	<i>Fucus spiralis</i>	A00VK#F01.A18BL	Brown algae, SOURCE = Spiral wrack (as organism)
Bladderwrack	<i>Fucus vesiculosus</i>	A00VK#F01.A0CRR	Brown algae, SOURCE = Bladder wrack (as organism)
Halidrys siliquosa	<i>Halidrys siliquosa</i>	A00VK#F01.A18BM	Brown algae, SOURCE = Sea oak (as organism)
Thongweed	<i>Himanthalia elongata</i>	A00VN	Sea spaghetti
Oar weed	<i>Laminaria digitata</i>	A00VK#F01.A0B6M	Brown algae, SOURCE = Tangle (as organism)
Tangle	<i>Laminaria hyperborea</i>	A00VK#F01.A0B6N	Brown algae, SOURCE = North European kelp (as organism)
Dulse	<i>Palmaria palmata</i>	A00VJ	Dulse
Channelled wrack	<i>Pelvetia canaliculata</i>	A00VK#F01.A18BH	Brown algae, SOURCE = Channelled wrack (as organism)
Purple laver	<i>Porphyra purpurea</i>	A00VH	Laver (species-specific code is missing)
Pink laver	<i>Porphyra umbilicalis</i>	A00VH	Laver (species-specific code is missing)
Sugar kelp	<i>Saccharina latissima</i>	A00VK#F01.A0B6Q	Brown algae, SOURCE = Sea belt (as organism)
Gutweed	<i>Ulva intestinalis</i>	A00VB#F01.A0B6F	Green algae, SOURCE = Hollow green nori (as organism)
Sea lettuce	<i>Ulva lactuca</i>	A00VD	Sea lettuce
Wrack siphon weed	<i>Vertebrata lanosa</i>	A00VE#F01.A18BG	Red algae, SOURCE = Wrack siphon weed (as organism)

Codes for dried seaweed should be reported in the following format as informed by EFSA secretary October 2019:

A00ZQ#F01.A0B6Q\$F27.A00VK	Dried vegetables, SOURCE = Sea belt (as organism), SOURCE-COMMODITIES = Brown algae
A00ZQ#F01.A05MJ\$F27.A00VB	Dried vegetables, SOURCE = Sea lettuce (as organism), SOURCE-COMMODITIES = Green algae
A00ZQ#F01.A05MP\$F27.A00VE	Dried vegetables, SOURCE = Dulse (as organism), SOURCE-COMMODITIES = Red algae

In addition, codes for wild and cultivated seaweed should be found to address the call for data. Data for 353 samples of macroalgae were submitted by the IMR in time for the deadline of October 1, 2020.

## 5 - References

- Aquaron, R., Delange, F., Marchal, P., Lognone, V., Ninane, L., 2002. Bioavailability of seaweed iodine in human beings. *Cell. Mol. Biol.* 48, 563-569.
- Biancarosa, I., Liland, N.S., Biemans, D., Araujo, P., Bruckner, C.G., Waagbo, R., Torstensen, B.E., Lock, E.J., Amlund, H., 2018. Uptake of heavy metals and arsenic in black soldier fly (*Hermetia illucens*) larvae grown on seaweed-enriched media. *Journal of the Science of Food and Agriculture* 98, 2176-2183.
- Biancarosa, I., Sele, V., Belghit, I., Ornsrud, R., Lock, E.J., Amlund, H., 2019. Replacing fish meal with insect meal in the diet of Atlantic salmon (*Salmo salar*) does not impact the amount of contaminants in the feed and it lowers accumulation of arsenic in the fillet. *Food Additives and Contaminants Part a-Chemistry Analysis Control Exposure & Risk Assessment* 36, 1191-1205.
- Blikra, M.J., Løvdaal, T., Vaka, M.R., Roiha, I.S., Lunestad, B.T., Lindseth, C., Skipnes, D., 2019. Assessment of food quality and microbial safety of brown macroalgae (*Alaria esculenta* and *Saccharina latissima*). *Journal of the Science of Food and Agriculture* 99, 1198-1206.
- Calil-Silveira, J., Serrano-Nascimento, C., Laconca, R.C., Schmiedecke, L., Salgueiro, R.B., Kondo, A.K., Nunes, M.T., 2016. Underlying Mechanisms of Pituitary-Thyroid Axis Function Disruption by Chronic Iodine Excess in Rats. *Thyroid* 26, 1488-1498.
- Combet, E., Ma, Z.F., Cousins, F., Thompson, B., Lean, M.E.J., 2014. Low-level seaweed supplementation improves iodine status in iodine-insufficient women. *Br. J. Nutr.* 112, 753-761.
- [Duinker, A., 2014. Report from the project: Alger: mat - forskning- formidling: Mineraler og tungmetaller i alger fra Lindesnes. NIFES report., 11 pp. pp.](#)
- [Duinker, A., Roiha, I.S., Amlund, H., Dahl, L., Lock, E.-J., Kögel, T., Måge, A., Lunestad, B.T., 2016. Potential risks posed by macroalgae for application as feed and food - a Norwegian perspective. NIFES-report, 24 pp.](#)
- Heldal, H.E., Sjøtun, K., 2010. Technetium-99 (Tc-99) in annual growth segments of knotted wrack (*Ascophyllum nodosum*). *Sci. Total Environ.* 408, 5575-5582.
- Keogh, S., 2006. Carbon-14 and iodine-129 in the Irish marine and terrestrial environment. PhD thesis. National University of Ireland, Dublin. 101 p.
- Kershaw, P., McMahon, C., Rudjord, A., Smedley, C., Leonard, K., 2005. Spatial and temporal variations in concentration factors in NW European Seas - secondary use of monitoring data. *Radioprotection* 40 (1), 93-99.
- [Kleppe, M., 2016. Kadmium, uorganisk arsen og jod i dyrket og viltvoksende butare \(\*Alaria esculenta\*\) og sukkertare \(\*Saccharina latissima\*\). Master thesis. University of Bergen.](#)
- Liland, N.S., Biancarosa, I., Araujo, P., Biemans, D., Bruckner, C.G., Waagbo, R., Torstensen, B.E., Lock, E.J., 2017. Modulation of nutrient composition of black soldier fly (*Hermetia illucens*) larvae by feeding seaweed-enriched media. *Plos One* 12.
- Lock, E.J., Belghit, I., 2018. Seaweed as feed ingredient in aquafeed – a SWOT analysis. Rapport fra



Havforskningen, 18 pp.

Lu, Y., Suliman, S., Hansen, H.R., Feldmann, J., 2006. Iodine excretion and accumulation in seaweed-eating sheep from Orkney, Scotland. *Environ Chem* 3, 338-344.

Moreda-Piñeiro, A., Peña-Vázquez, E., Bermejo-Barrera, P., 2011. Significance of the Presence of Trace and Ultratrace Elements in Seaweeds, in: Kim, S.K. (Ed.), *Handbook of Marine Macroalgae*. John Wiley & Sons, Ltd, pp. 116-170.

Mouritsen, O.G., Dawczynski, C., Duelund, L., Jahreis, G., Vetter, W., Schroder, M., 2013. On the human consumption of the red seaweed dulse (*Palmaria palmata* (L.) Weber & Mohr). *J. Appl. Phycol.* 25, 1777-1791.

NRC, 2011. *Nutrient requirements of fish and shrimp*. The National Academies Press, Washington, DC.

Redway, M., Bouga, M., Combet, E., 2018. Impact of the food matrix on iodine bioavailability. *Proc. Nutr. Soc.* 77.

Rose, M., Lewis, J., Langford, N., Baxter, M., Origgi, S., Barber, M., MacBain, H., Thomas, K., 2007. Arsenic in seaweed--forms, concentration and dietary exposure. *Food Chem. Toxicol.* 45, 1263-1267.

Sjøtun, K., Heldal, H.E., Brakstad, D.S., 2011. Differential concentration of Technetium-99 (Tc-99) in common intertidal molluscs with different food habits. *Mar. Pollut. Bull.* 62, 2420-2426.

Skjerdal, H., Heldal, H.E., Gwynn, J., Strålberg, E., Møller, B., Liebig, P.L., Sværen, I., Rand, A., Gäfvert, T., Haanes, H., 2017. NRPA. Radioactivity in the marine environment 2012, 2013 and 2014. Results from the Norwegian National Monitoring Programme (RAME). *StrålevernRapport 2017:13*. Østerås: Norwegian Radiation Protection Authority, 2017.

Tuo, F., Zhang, Q., Zhou, Q., Xu, C., Zhang, J., Li, W., Zhang, J., Su, X., 2016. Measurement of <sup>238</sup>U, <sup>228</sup>Ra, <sup>226</sup>Ra, <sup>40</sup>K and <sup>137</sup>Cs in foodstuffs samples collected from coastal areas of China. *Appl. Radiat. Isot.* 111, 40-44.

Yamashita, Y., 2014. Method of removing inorganic arsenic from dried hijiki seaweed products. *Nippon Suisan Gakkaishi* 80, 973-978.

Yoshida, M., Mukama, A., Hosomi, R., Fukunaga, K., Nishiyama, T., 2014. Serum and Tissue Iodine Concentrations in Rats Fed Diets Supplemented with Kombu Powder or Potassium Iodide. *J. Nutr. Sci. Vitaminol. (Tokyo)*. 60, 447-449.

## 6 - Appendix

Table 10. Dry weight percentages in samples of fresh and dried macroalgae (imported products sold dry). Species are presented with decreasing median values. Cell colouring according to brown, red and green algae.

Latin name	Common name	N	Mean	Median	Min-max	25 % Quartiles
<i>Saccharina spp</i>	Kombu, dried	4	97	97	95-99	95-98
<i>Eisenia bicyclis</i>	Arame, dried	1	96	96		
<i>Porphyra spp</i>	Nori, dried	10	95	95	83-99	94-97
<i>Undaria pinnatifida</i>	Wakame, dried	5	94	94	93-95	94-94
<i>Ulva spp</i>	Green nori, dried	1	94	94		
<i>Sargassum fusiforme</i>	Hijiki, dried	1	90	90		
<i>Ascophyllum nodosum</i>	Rockweed	17	30	31	18-37	28-34
<i>Pelvetia canaliculata</i>	channelled wrack	1	31	31		
<i>Fucus vesiculosus</i>	Bladderwrack	20	31	28	19-94	25-30
<i>Halidrys siliquosa</i>	Halidrys siliquosa	8	25	24	22-35	22-24
<i>Fucus serratus</i>	toothed wrack	16	23	23	17-28	21-24
<i>Fucus spiralis</i>	Spiral wrack	1	20	20		
<i>Laminaria digitata</i>	Oar weed	28	19	18	14-25	15-21
<i>Vertebrata lanosa</i>	Wrack siphon weed	15	16	17	8.6-22	13-18
<i>Alaria esculenta</i>	Winged kelp	28	15	16	7.9-22	12-18
<i>Laminaria hyperborea</i>	Tangle	4	16	16	14-17	15-16
<i>Ulva lactuca</i>	Sea lettuce	11	16	16	9.4-28	11-18
<i>Palmaria palmata</i>	Dulse	20	17	16	12-34	13-20
<i>Himanthalia elongata</i>	Thongweed	2	15	15	14-16	14-16
<i>Sargassum muticum</i>	Wireweed	4	13	13	11-15	12-15
<i>Chorda filum</i>	Dead man's rope	1	13	13		
<i>Saccharina latissima</i>	Sugar kelp	156	13	12	6-26	10-15
<i>Porphyra purpurea</i>	Purple laver	1	11	11		
<i>Ulva intestinalis</i>	Gutweed	5	10	8	7.3-13	7.9-13
<i>Codium fragile</i>	Green sea fingers	2	5	5	4.8-5.2	4.8-5.2



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