Bioconcentration Analysis of Iodine Species in Seaweed (*Eucheuma cottonii*) from Maluku Marine as Alternative Food Source

Yeanchon H. Dulanlebit, Nikmans Hattu, Gloria Bora

Abstract**—**Seaweed is a type of macro algae which are good source of iodine and have been widely used as food and nutrition supplement. One of iodine species that found in ocean plant is iodate. Analysis of iodate in seaweed (*Eucheuma cottonii*) from coastal area of Maluku has been done. The determination is done by using spectrophotometric method. Iodate in sample is reduced in excess of potassium iodide in the presence of acid solution, and then is reacted with starch to form blue complex. The study found out that the highest wavelength on determination of iodate species using spectrophotometer analysis method is 570 nm. Optimum value to yield maximum absorption is used in this research. Contents of iodate in seawater from coastal area of Ambon Island, Western Seram and Southeast Maluku are 0.2655, 0.2719 and 0.1760 mg/L, respectively. While in seaweeds from Ambon Island, Western Seram, Southeast Maluku-Taar, Ohoidertawun and Wab are 6.3122, 6.3293, 6.2333, 3.7406 and 4.4207 mg/kg in dry weight. Bioconcentration (enrichment) factor of iodate in seaweed (*Eucheuma cottonii*) from the three samples (cluster) is different; in Coastal area of Ambon Island, Western Seram and Southeast Maluku respectively are 23.78, 23.28 and 27.26.

Keywords**—**Bioconcentration, *Eucheuma cottonii*, iodate, iodine, seaweed.

I. Introduction

IODINE is essential element for living beings although vertebrates only have thyroid gland and iodinated hormone. In humans about 60-80% of the total iodine is present in extra thyroidal tissue and is not involved with the function of thyroid gland. Lack of iodine causes iodine deficiency disorder (IDD), while excessive iodine dietary intake can result in serious pathological problems [1], [2]. Mapping Survey of IDD in Indonesia on 1998 showed that 87 million of Indonesian lives in risk area lacking of iodine. About 20 million people in Indonesia are estimated to have goiter, meanwhile 290.000 have cretin and 9.000 babies are born cretin every year [3].

Maluku is one of archipelago provinces in Indonesia with the high exploiting of the marine product, but prevalence of total goiter rate (TGR) is also high. Goiter prevalence is calculated based on all magnification stadiums of goiter gland such as touch stadium and visible stadium. TGR in Maluku is 33.3% for children (6-12 years old). Prevalence in the Middle of Maluku District is 40.9% and 16.0% in Ambon City District [3]. The TGR is often used to determine level of IDD endemic in the target location.

Iodine is an essential element for human beings. Therefore, recommended iodine intake for adults is between 140 and 160 µg per day [1], [2]. In order to prevent IDD, Government has designed programs to supply drinking water, oil, and/or table salt with iodide or iodate. Indonesian Standard (SNI 01-3556-2000) has obliged salt manufacturers to add 30 ppm potassium iodate (KIO_3) as iodine source. Unfortunately, 36% of salts in Indonesia were lower than standard content, while 6% do not contain KIO_3 [4].

National Survey Data for Iodized Salt in Indonesia by Central Bureau of Statistics (Government Institute for Mapping and Survey) indicate that the percentage of iodine salt which has been consumed by household in whole region in the country from 1996 to 2002 was only 62 to 68% [4]. The BPS's data in 2002 also showed that the percentage of household iodine salt consumption in Maluku was 69.68% for good category, 10.46% for less category, and 19.86% for poor category [4]. Besides this problem, there are many controversies about effectiveness of salt iodization. This controversy is about instability of iodine in salt which has been fortificated during the storage or food processing [5]-[7]. Therefore, request to exploit marine food product is wise alternative. Products of marine food have high iodine content. One of which is seaweeds. Seaweed consumption could increase the number of iodine in the body, even if it is consumed in small amount [8]-[13]. Most of published works report total iodine contents in seaweed and there are few works related with the iodine species. Some seaweeds are applied for therapy of IDD and urinary disease [13].

Iodine will be found in all seafoods. As micronutrient, iodine content in seafood like fish, cockle shells, squid or seaweed is 0.0002%. 90% of iodine consumption in human body is from foods, and the rest is from water [2].

Eucheuma cottonii is one of the seaweed commodities having economic value and planted in many places. Distribution of this species is wide in Maluku and they could be harvested in short time (45 to 50 days), making this commodity as one of excellent fishery commodities. Besides as food and carrageenan precursors, the primary and secondary metabolites of this species could be useful in pharmaceutical industry and cosmetics [8], [10], [13].

The concentration of iodine in terrestrial plants is quite low (< 1 mg/kg), while a higher concentration of iodine is found in marine organisms. The concentration of iodine in *Laminaria japonica* can reach 0.9% of dry weight, while the iodine

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concentration in seawater is only 6 x 10^{-8} g/L. The enrichment factor of *Laminaria japonica* for iodine reaches high. Despite the lack of information about the biological function of iodine and its concentration mechanism in seaweed, the possible mechanism could be plasma membrane diffusion [14], [15].

The content of iodine in seaweeds is according to the seaweed species and the season. Iodine in the seaweed samples are predominantly iodide, with small fraction of iodine present as iodate and organic iodine in the form of monoiodotyrosine and diiodotyrosine [9]-[15].

The aims of this research are to determine content of iodate species in seaweed (*Eucheuma cottonii*) and seawater, and to determine enrichment factor (bioconcentration) of iodate species in seaweed. *Eucheuma cottonii* seaweed was used in this research because of its wide distribution in the Coast area of Ambon Island, Western Seram and Southeast Maluku. Maluku ocean is located at Eastern Indonesia. Spectrophotometric method is used for all measurements. This method is based on modification of iodometric titration by forming color complex of starch-iodine from redox reaction. Due to the simplicity, this method can be potentially applied.

II. Experimental

Equipment

A Hewlett Packard double-beam spectrophotometer equipped with 10 mm quartz cells was used for all UV-Visible measurements. Sayota blender was applied to refine samples. Other equipment is used such as analytical balance, magnetic stirrer and glass equipment.

Chemicals and Samples

All chemicals used in this research are pure chemicals, including potassium iodate, potassium iodide, sodium chloride, phosphoric acid 85%, and starch solution. Seaweed (*Eucheuma cottonii*) and seawater were collected from coast area of Ambon Island, Western Seram and Southeast Maluku (Maluku - East of Indonesia). Sampling locations are shown in Fig. 1. Samples were dried and refined before both of extraction and determination steps.

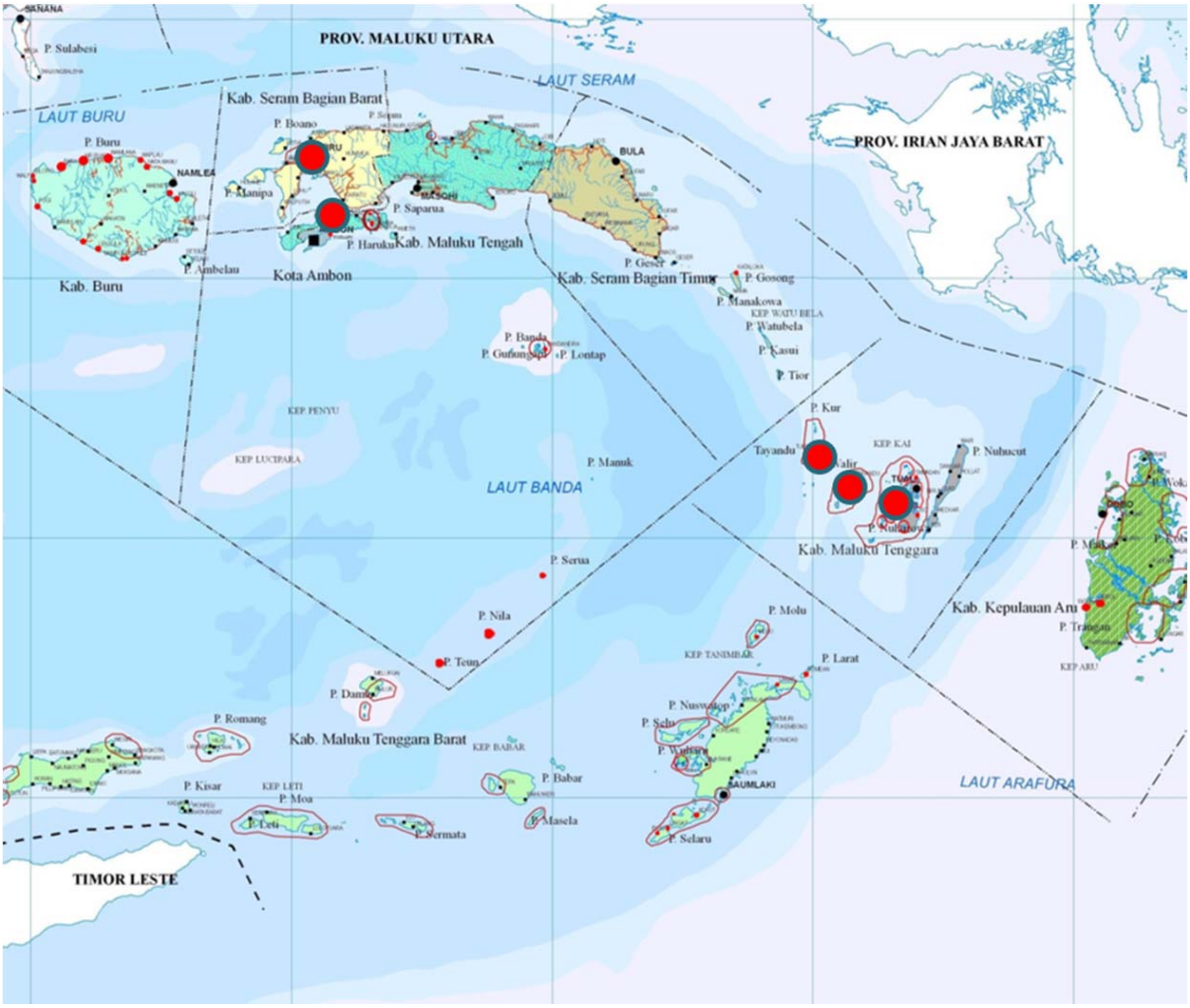

Fig. 1 Sampling Location

Methods

Absorption Spectra and Determination of λ_{max}

1.25 mL KIO_3 30 ppm, 0.5 mL KI 10%, 0.5 mL H_3PO_4 85%, and 0.5 mL starch 1% were filled into 25 mL volumetric flask. Aquadest was added into the flask until the limit mark then the solution was homogenized. Absorbance is measured at 300-800 nm using aquadest as blank.

Optimization of Measurement Conditions

The optimization of color stability with the addition of 85% H_3PO_4, 10% KI, 1% starch, and 20% NaCl needs to obtain the optimal conditions for measurements. The optimize effect of the color stability is done by reacting 5 mL KIO_3 30 ppm, 2 mL H_3PO_4 85%, 1.5 mL KI 10%, 2 mL starch 1% and diluting by distill water in 100 mL volumetric flask. The solution is homogenized and absorbance was measured for one until 15 minutes. The absorbance is measured at λ_{max}. Optimization of the effect of adding 85% H_3PO_4, 10% KI, 1% starch, and NaCl 20% is done in the same way. Optimization of H_3PO_4 is done by varying 1-5 mL of H_3PO_4 85% at optimum conditions of color stability. The optimization of the addition of KI is done by varying 1-5 mL KI 10% at optimum conditions of color stability and optimum condition of adding 85% H_3PO_4. The optimization effect of adding NaCl is done by varying 1-10 mL of NaCl 20% at optimum conditions of color stability, 85% H_3PO_4, 10% KI, and 1% starch. Optimization of the measurement conditions is done to get the maximum absorbance at λ_{max}.

Calibration Curve and Iodate Determination

0.25 mL of KIO_3 30 ppm, 1 mL of H_3PO_4 85%, 1 mL of KI 10%, 3 drops of starch 1% and 0.75 mL NaCl 20% are filled into 25 mL volumetric flask. Aquadest was added into the flask until the limit mark then the solution was homogenized. The absorbance was measured by using spectrophotometer at λ_{max} with blank solution without KIO_3. Measurement is done at optimum time when it forms blue color. Concentrations of KIO_3 solution are various. Calibration curve is made between absorbance obtained and concentration of KIO_3. Samples (seaweed and seawater) after extraction steps are measured like procedure above.

III. Results and Discussion

Absorption Spectra and Determination of λ_{max}

Determination of iodine is represented as iodate generally using volumetric method (iodometric titration). The principle of this method is based on reducing KIO_3 in a sample with excessive KI in acid condition to form iodine that can be titrated with sodium thiosulfate ($Na_2S_2O_3$) using starch as an indicator. This analytical method can be developed by using visible spectrophotometric method. Starch reacts with iodine to form starch-iodine complex as:

$$I_2 + H_2O \Leftrightarrow H^+ + I^- + HOI$$
$$I_2 + I^- \Leftrightarrow H^+ + I_3^-$$
$$I_3^- + \text{starch} \Leftrightarrow \text{starch - } I_3^- \text{ (blue color)}$$

The method is used to analyse iodine in iodate species (IO_3^-), when the iodate in sample reduces with addition of potassium iodate in acid condition.

$$IO_3^- + 5I^- + 6H^+ \rightarrow 3I_2 + 3H_2O$$

Iodine was determined as the representation of iodate species because it is a stable compound. Iodide species (KI) is unstable and volatile. The interaction between starch (amylase component) and iodine forms stable blue complex. Amylopectine gives unstable red complex with iodine. Visible absorption spectrum in determination of iodate is shown in Fig. 2. The spectrum shows two absorption peaks at 346 nm and 570 nm wavelengths. The absorption peak used for the quantitative determination is 570 nm because this peak was included in visible wavelength, while the peak of 346 nm was included in ultraviolet wavelength.

Triiodide ion formed by reaction between KIO_3 reduction with excess KI in acid condition produces maximum absorption in visible wavelength on 570 nm.

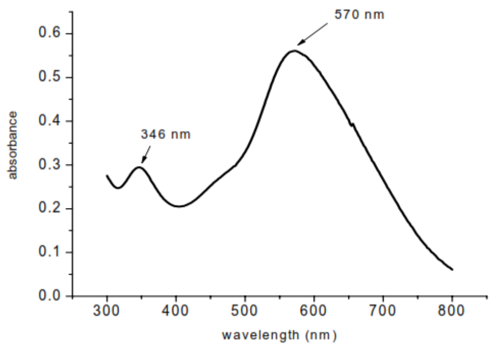

Fig. 2 The Absorption Spectrum of KIO_3

Optimization of Measurement Conditions

Optimization of measurement conditions was done to obtain maximum absorbance. Optimization of the parameter was measured by using the color solution stability to time, the effect of KI 10%, H_3PO_4 85% and NaCl 20% addition. Effect of the color stability to time was determined when the solution formed blue color for the first time. It was obtained at four minutes. The color stability influenced the obtained absorbance. At the optimum time, complex form stayed at highest absorption.

Addition of KI 10% on the determination of KIO_3 refers to reduction of iodate to iodine, to allow reaction with starch to form the color complex. The effect of KI 10% to absorbance value was obtained on addition of 1 mL KI 10%. Reduction of KIO_3 to iodine species is done in the presence of acid. Phosphoric acid 85% is used in this experiment. The optimum volume of H_3PO_4 85% to yield optimum absorbance is obtained on addition 1 mL H_3PO_4 85%.

Iodide optimization was done to reduce iodate to iodine. The addition of iodide was required to dissolve and prevent iodine evaporation. On the other hand, the optimization of phosphoric acid was required because in acid condition iodide can reduce iodate to iodine. On the other hand, acid was required to prevent the hydrolysis of I_2 and to keep the influence of metal.

Matrix in the sample was taken from the sea when the NaCl content was dominated. Therefore, optimization of matrix content composition is necessary to be done. The effect of NaCl 20% to produce the maximum absorbance was obtained in addition of 0.75 mL. 3 drops of indicator solution (starch 1%) were used in this experiment.

Optimization of sodium chloride is done by adding NaCl 20% to optimize the concentration of iodide and acid. The addition of NaCl could affect the result of absorbance. The increase of NaCl concentration could decrease the absorbance result. This is caused by the attack of chloride ions into iodine and producing ICl or I_2Cl^-, so that I_2 reacts with I^-. On the other hand, Schlieren effect is caused by the difference of refractive index between standard and sample containing NaCl.

Calibration Curve and Iodate Determination

Absorbance value from various concentrations of standard solution was used to make calibration curve on iodate determination. Calibration curve is shown in Fig. 3.

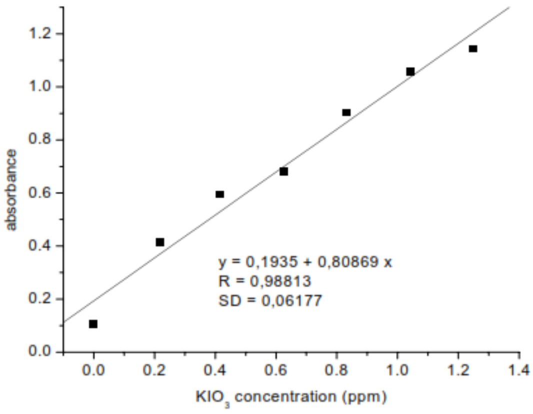

Fig. 3 Calibration Curve of Iodate

Based on Fig. 3, there is linear relationship between iodate concentration and absorbance value. This fact validated with correlation coefficient of closing one calibration curve.

The result of iodate determination in seawater and seaweed (*Eucheuma cottonii*) using spectrophotometric method plotted to calibration curve is presented in Table I. Iodate concentration in the samples below is average concentration for 4 times repetition. Based on Table I, bioconcentration (enrichment) factor of iodate in seaweed (*Eucheuma cottonii*) from the three samples (cluster) is different; in Coastal area of Ambon Island, Western Seram and Southeast Maluku respectively are 23.78, 23.28 and 27.26. Average concentration in seaweed on table is average concentration based on dry weight of the samples. It means that seaweed (*Eucheuma cottonii*) can be recommended as alternative food source of iodine to prevent IDD.

TABLE I
CONCENTRATION OF IODINE SPECIES IN SAMPLES

No	Sample and Location	Average Concentration
1	Seawater (Ambon - Hutumuri)	0.2655 mg/L
2	Seawater (Western Seram - Pelita Jaya)	0.2719 mg/L
3	Seawater (Southeast Maluku - Kei Kecil)	0.1760 mg/L
4	*Euchema cottoni* (Ambon - Hutumuri)	6.3122 mg/kg
5	*Euchema cottoni* (Western Seram - Pelita Jaya)	6.3293 mg/kg
6	*Euchema cottoni* (Southeast Maluku - Taar)	6.2333 mg/kg
7	*Euchema cottoni* (Southeast Maluku - Ohoidertawun)	3.7406 mg/kg
8	*Euchema cottoni* (Southeast Maluku - Wab)	4.4207 mg/kg

The results show that the lowest iodide content in *Eucheuma cottonii* is found in Southeast Maluku marine (Ohoidertawun and Wab); while iodine contents in marines of Ambon Island, West Seram and Southeast Maluku (Taar) are almost the same. The difference of iodide content in seaweed (*Eucheuma cottoni*) is caused by the different marine conditions for cultivation. Marine condition and the food consumed by eucheuma cottonii affect the amount of iodine absorbed in seaweed. So, the circulation of sea water (include water flow) in marine has impact to the lowest and highest nutrition, including the content of iodine in seaweed. Salinity has an effect on osmoregulation process in seaweed. The process causes iodine accumulation and the other compounds in sea into seaweed.

Fig. 4 Seaweed (*Eucheuma cottonii*)

Maluku province has an area 705.645 km^2 with 90% (658.294 km^2) marine. The total area of seaweed cultivation in Maluku is 266,17 km^2 and now the area used is 102,59 km^2. Some area used for seaweed cultivation is in marine of Ambon Island, West Seram and Kei Island. Maluku marine is very potential cultivation area for seaweed, when *Eucheuma cottonii* is the most widely cultivated type of seaweed because of the easy treatment and short harvest time.

The extents of seaweed cultivation area in 2015 is estimated at 90,000 ha or 540,000 tons in the Maluku province. To achieve the production targets, government has development strategy done in 2 stages. In the first stage, the government finances the minimum production of seaweed. So the production of seaweed can meet the needs required by society.

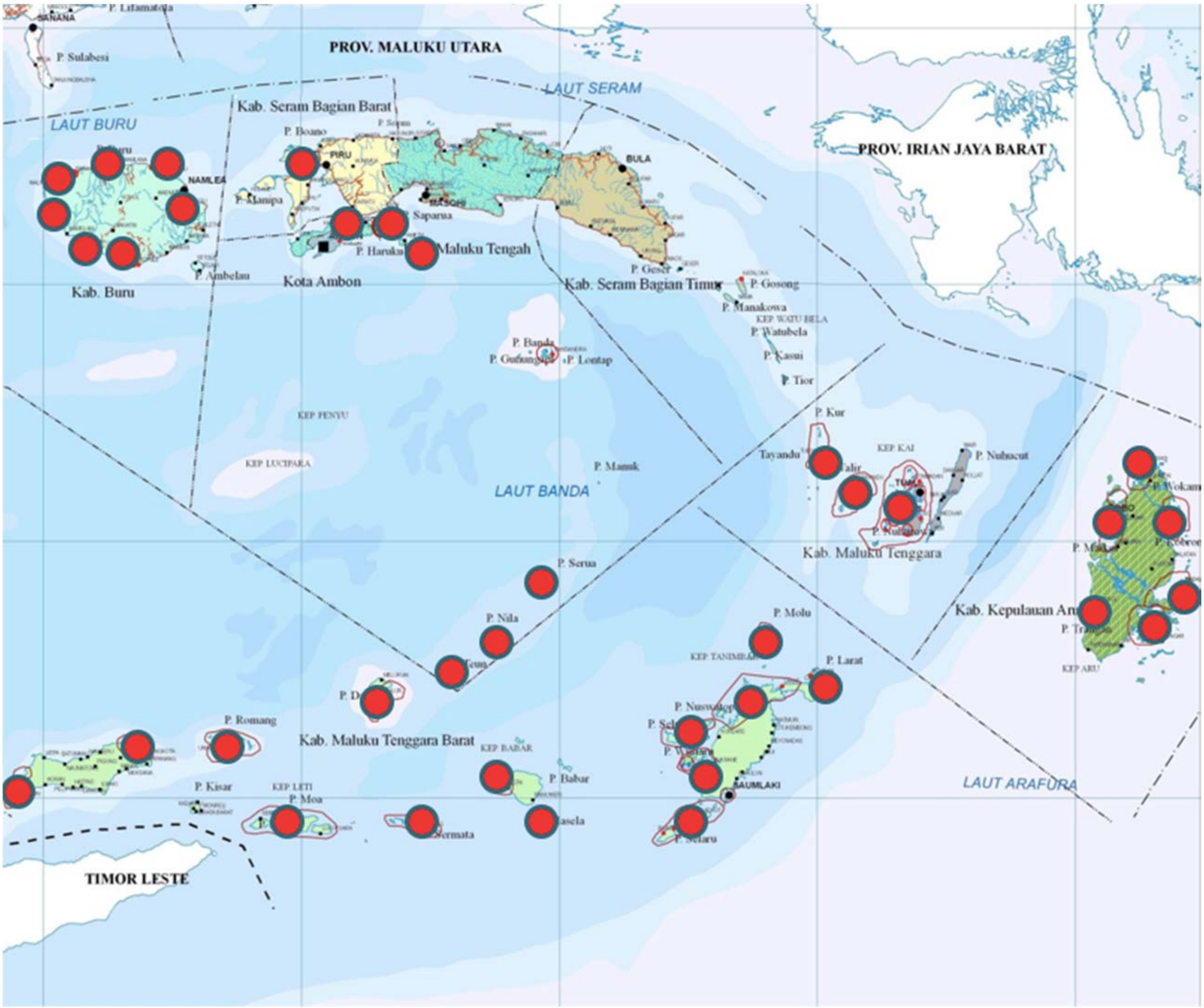

Fig. 5 Location (Cluster) of seaweed cultivation

Cluster development and its processing centers was done gradually starting from West Southeast Maluku Cluster, Seram Cluster, Aru Islands Cluster, Southeast Maluku Cluster, Southwest Maluku Cluster and Buru Island Cluster. Southeast Maluku Cluster was the most potential cluster producer of seaweed (*Eucheuma cottonii*) that is strongly supported by the aquatic resources, including the content of iodium there.

Congenital hypothyroid (a disease caused by thyroid disorders) is one of the diseases that are not infectious but causes growth disorders, development and mental retardation. Diseases caused by thyroid disorders can be detected 3 days after the baby is born. The first treatment for babies who have congenital hypothyroidism is giving thyroxine hormone in tablet form. Deficiencies of iodine also have an impact on caused by thyroid disorders. National suvery results of BPS 2002 showed that the prevalence of endemic in primary school students is 9.8%. This is considered a public health problem [4]. To overcome this problem, the government has been intensively applying the salt iodization programs, lipiodol injection and recommending to consume the sea food containing iodine.

The content of iodine in seaweed can be one alternative to prevent iodine deficiency which can cause hypothyroidism.

IV. Conclusion

Bioconcentration factor of iodate in *Eucheuma cottonii* from the three samples (cluster) is different; in Coastal area of Ambon Island, Western Seram and Southeast Maluku respectively are 23.78, 23.28 and 27.26. Based on this fact, *Eucheuma cottonii* can be recommended as food alternative which are good source of iodine.

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