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Refill Drinking Water Quality Analysis Based on Microbiological and Inorganic Chemicals Component Test in Kopelma Darussalam Banda Aceh

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Abstract:
Background*: This research is entitled "Analysis of the Quality of Refill Drinking Water Based on Microbiological Tests and Inorganic Chemical Content in the Kopelma Darussalam Banda Aceh Area". The purpose of this study was to determine the difference in the amount of microbiological content (Escherichia coli and Coliform) and the difference in the amount of inorganic chemical content (Cadmium (Cd), Chromium (Cr), and Cyanide (CN)) contained in raw water, drinking water refill with RO and Non RO in the Kopelma Darussalam Region.*
Materials and Methods*: The research method used is a descriptive method which aims to obtain information about the contamination of Escherichia coli and Coliform bacteria in refill drinking water using the Most Probable Number (MPN) series 5-1-1 method, and test the content of inorganic chemicals. in the form of Cadmium (Cd), Chromium (Cr) and Cyanide (CN) using Atomic Absorption Spectrophotometer by measuring the absorbance of water samples at certain wavelengths. This research approach is a quantitative approach. The parameters in this study were the number of Escherichia coli and Coliform bacteria and inorganic chemicals such as Cadmium (Cd), Chromium (Cr) and Cyanide (CN) contained in RO and Non RO refill water in Kopelma Darussalam. Data analysis was performed using ANOVA (Analysis of Variance) followed by Duncan's test at α 5%. The research was carried out from July 2020 to August 2020.*
Results *: The results of this study indicate that there is a significant difference in the amount of Escherichia coli and Coliform bacteria in RO and Non RO refill drinking water, while for raw water there is no significant difference with drinking water. RO and Non RO refills. There were no significant differences in the content of Cadmium (Cd), Chromium (Cr) and Cyanide (CN) in raw water, RO and Non RO refill drinking water.*
Conclusion *: The conclusion of the study shows that based on microbiological parameters refill drinking water in the Kopelma Darussalam area does not meet the standards of the Indonesian Minister of Health Regulation Number 492 / MENKES / PER / IV / 2010, while for the content of inorganic chemicals (Cadmium (Cd), Chromium (Cr)) and Cyanide (CN)) have met the standards.*
Keywords *: Bacteria; Inorganic Chemicals; Refill Drinking Water.*

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I. Introduction

Refill drinking water is water that has gone through various purification processes. This water has gone through several stages of filtration, either by means of ultraviolet irradiation, ozonation, or both, which aim to get clean water and suitable for use as drinking water [5]. Consumption of unhygienic drinking water can cause various digestive tract diseases. Efforts and efforts to reduce the incidence of disease include paying attention to the quality of drinking water consumed every day [11]. For this prevention, the state participates in regulating regulations and rules regarding raw water and the quality of drinking water for its citizens.

The Regulation of the Minister of Health (Permenkes) of the Republic of Indonesia Number 492/MENKES/PER/IV/2010 has stipulated requirements for the quality of drinking water based on several parameters such as microbiological parameters, inorganic chemistry, physical and chemical parameters. Drinking water parameters that are directly related to health include microbiological parameters and inorganic chemicals. For microbiological parameters, the total content of *Escherichia colibacteria* and *Coliform* allowable is 0/100 ml water [9]. For inorganic chemical parameters in the form of Cadmium (Cd), which is allowed is 0.003 mg / liter [4], allowed chromium (Cr) is 0.05 mg / liter [21] and allowed cyanide (CN) is 0.07 mg / L [17].

The process carried out to obtain good drinking water quality based on this Regulation of the Minister of Health can be carried out by processing raw water sources into clean water. The steps taken include storing

water, oxidation or the process of adding oxygen to the water so that the levels of heavy metals and other chemical substances contained in the water are easily decomposed, the stages of deposition or coagulation, filtration and disinfection. Currently, refill drinking water depot businesses have mushroomed that offer better quality drinking water because it reprocesses clean raw water by going through several stages such as filtration and sterilization.

Refill drinking water has become one of the alternatives most frequently used by urban communities, especially Banda Aceh. Syiah Kuala is one of the most populated sub-districts in Banda Aceh, and this sub-district is mostly inhabited by students and university students because in this area there are two major universities, namely Syiah Kuala University and UIN Ar-raniry. Given the density of the population in this sub-district, the need for clean water has increased, so that the refill drinking water depot business is increasingly in demand. Based on the results of Rizki's research [18], there are still *Escherichia colibacteria* and *Coliform* in refill drinking water in areas that have been monitored and areas that have not been monitored. The same research conducted by Natalia [14] in Blora Regency, Semarang, still contains bacteria *Coliform* in refill drinking water that has passed the filtering process, as well as Muzajjanah's research [13] states that refill drinking water is in Jagakarsa District, South Jakarta, there is still contamination *Escherichia coli* and *Coliform.* Apart from being contaminated by bacteria, refill drinking water can also be contaminated by inorganic chemicals. Research conducted by Tanty [23] drinking water refill around the Syahdan campus and Bina Nusantara Bogor Orchid was contaminated by Cadmium (Cd), Chromium (Cr) and Cyanide (CN).

According to Walangitan [24], contamination of refill drinking water by bacteria or inorganic chemicals can be caused by several factors, one of which is because the raw water is contaminated. The Krueng Aceh River is the source of water used by PDAM Tirta Daroy to be distributed to the community after carrying out several treatments into clean water. The clean water is used as raw material for depot entrepreneurs in the Syiah Kuala sub-district. Based on Hadi's research [2], the mouth of the river in Krueng Aceh is polluted by several heavy metals, one of which is Cadmium (Cd). The source of Cadmium (Cd) pollution comes from fishing boat activities. Fishermen coat the boats with Cd metal to prevent corrosion, but at certain times the metal will dissolve in the water [3]. Suhendrayatna [22] stated that along the Krueng Aceh Watershed (DAS) which is the main source of raw water for PDAM, there are many community activities including industry, power plant activities, hotels, markets, densely populated housing, and home industries. produces domestic waste which is directly disposed of through the Krueng Aceh watershed. Chromium contamination in the Krueng Aceh river, which becomes raw water for processing refill drinking water in the cities of Banda Aceh and Aceh Besar, can come from human activities in the form of industrial activities, mining, burning fuel, and other domestic activities. One of the biggest contributors to heavy metal is household waste. In the Krueng Aceh river flow, which becomes raw water at several depots in the cities of Banda Aceh and Aceh Besar, there are several points that have been contaminated by cyanide, including the Javanese village which is the final disposal site (TPA) in the city of Banda Aceh, the pollution comes from the flow of leachate ponds located on the banks of the Krueng Aceh river [10].

In line with the community's need for drinking water that is free from bacterial contamination and inorganic chemicals, there has been a proliferation of refill drinking water depots that offer quality, cheap and suitable drinking water because they have gone through several stages of filtration and sterilization. The price of refill drinking water is cheaper, but not all refill drinking water depots are guaranteed product safety [8].

The results of observations made at several refill drinking water depots in Syiah Kuala District, there were several refill drinking water depots that did not meet the technical and trade requirements stipulated in the Decree of the Minister of Industry and Trade of the Republic of Indonesia Number 651 / MPP / KEP / 10 / 2004, among others, did not have a business license, clean water supply letter from PDAM, did not carry out regular equipment maintenance and an unhygienic filling process that could contaminate refilled drinking water.

Based on these problems, it is necessary to carry out an analysis to determine the quality of refill drinking water based on the test for the content of bacteria and inorganic chemicals in the Kopelma Darussalam area.

II. Chemicals and Method

The materials used in this study were LB (*Lactose Broth*) Media, BGLB (Media Brilliant *Green Lactose Bile Broth*), EMB (*Eosin Methylene Blue*), Aquades, Refill drinking water samples (RO and Non RO), Crystal violet, Lugol, Alcohol 96%, Safranin and Oil immersion.

Research Location : Microbiological content tests (*Escherichia coli* and *Coliform*) were conducted at the Microbiology Laboratory of the Faculty of Medicine, Syiah Kuala University. The content of inorganic chemicals (Chromium and Cadmium) is carried out at the Faculty of Mathematics and Natural Sciences (MIPA), Universitas Syiah Kuala. The cyanide content test was carried out at the Aceh Food and Industry Standardization Center (BARISTAN).

Research Time : The research activity was carried out from July 2020 to August 2020.

Research Parameters : The parameters measured in this study were the amount of *Escherichia coli* bacteria and *Coliform* content and the content of inorganic chemicals (Cadmium (Cd), Chromium (Cr) and Cyanide (CN)) in refill drinking water.

Population and Sample : The population in this study were 5 refill drinking water depots in the Kopelma Darussalam area. Samples in this study were taken 3 depots with the type of raw water repetition, drinking water refill with RO and Non RO. This sampling was taken based on population density, the location of the water depot on the main road in the Kopelma Darussalam area and the availability of RO and Non RO types of refill drinking water.

Research Tools : The tools used in this study are test tubes, petri dishes, durham tubes, incubators, autoclaves, sterile specimen bottles, ose needles, microscopes, test tube racks, measuring pipettes, glass objects, benzene lamps, clocks, aluminum. foil and Atomic Absorption Spectrophotometer.

Research Materials : The materials used in this study were LB (*Lactose Broth*) Media, BGLB (Media Brilliant *Green Lactose Bile Broth*), EMB (*Eosin Methylene Blue*), Aquades, refill drinking water samples, Crystal violet, Lugol, Alcohol 96%, Safranin and Immersion Oil.

Research Methods and Design: The research method used is a descriptive method. The approach used is a quantitative approach.

Research Procedure:

The research procedure consisted of sterilization of tools, sampling and water examination using the MPN method for bacteria content (*Escherichia coli* and *Coliform*) and Atomic Absorption Spectrophotometer for the content of inorganic chemicals (Cadmium (Cd), Chromium (Cr) and Cyanide (CN)).

Sterilization of plastic media and tools was carried out using an autoclave at a temperature of 121^0C within 30 minutes with a pressure of 1 atm. The glass appliance is washed and dried, then wrapped in aluminum foil and put in an oven at 160°C for 2 hours.

The water collection procedure is carried out in the Kopelma Darussalam area, the depot officers first clean the gallons with an automatic brushing machine and rinse them using raw water, each gallon is filled with raw, RO and non-RO water, then the gallons are taken to the laboratory for testing.

Test the content of microbiological *(Escherichia coli* and *Coliform)* was performed using MPN method 5-1-1 series consisting of three phases, namely Test Estimation *(Presumptive Test),* a confirmation test *(Test*Convirmative) and Completeness test (*Complete Test*). The working steps for the prediction test are: 7 test tubes are used, each tube is filled with 10 ml of LB, each tube has been inserted into the durham tube in an inverted position. The test tubes were divided into 3 groups. The first five series were filled with 10 ml of water samples, one tube of the second group was inserted with 1.0 ml of water sample, and one tube of the third group was inserted with 0.1 ml of water sample. Incubated for 2 x 24 hours at 37^0C. Observed for the first 24 hours and the second 24 hours, if gas forms in the tube after 24 hours, the prediction test is positive, if no gas is formed then the incubation is continued for the second 24 hours, and if after the second 24 hours, no gas was formed, so the prediction test was declared negative.

Confirmation test procedure, 1 loop of positive LB tube was taken and then inoculated on a test tube containing 10 ml of BGLB which had been equipped with a durham tube according to the respective series. Incubated for 1 x 24 hours at a temperature of 44 ^{0}C. The number of tubes that formed gas was observed, recorded and referenced to the table MPN series 5-1-1. Finally, for the completeness test, from a positive BGLB tube, 1 loop was taken and implanted in the EMB medium aseptically. Incubated for 1 x 24 hours at 37^0C. Growing colonies were observed, if the growing bacterial colonies were red with a metallic sheen, then the colony was *Escherichia coli* and if the growing colony was slimy pink then the colony was *Coliform*. To ensure that the colony is *Escherichia coli, it is* done with gram staining.

Test the content of inorganic chemicals (cadmium, chromium and cyanide) using an atomic absorption spectrophotometer using a specific wavelength and absorbance value.

Data Analysis :

Bacterial content data were analyzed manually using the ANOVA (*Analysis of Variance*) test. To accept or reject the hypothesis the test level is used ($\alpha = 0.05$) provided that if $F_{count} \geq F_{table}$, then if there is a significant difference between treatments then the alternative hypothesis (Ha) is accepted. Conversely, if the value of $F_{count} < F_{table}$, there is no significant difference between treatments and the alternative hypothesis (H_a) is rejected. Furthermore, if there is a significant difference, then the further test used is the Duncan test. Whereas for inorganic chemicals, the ANOVA test was not carried out, because the data obtained from the research results were not different in each parameter. Data on inorganic chemical substances were directly analyzed for their feasibility in accordance with the Regulation of the Minister of Health of the Republic of Indonesia Number 492/MENKES/PER/IV/2010.

III. Results

The results of the MPN test for raw water, RO and Non RO refill drinking water carried out at 3 drinking water depots. Refill in the Kopelma Darussalam area still does not meet the standard requirements for drinking water quality testing according to the Decree of the Minister of Health of the Republic of Indonesia Number 492/MENKES/PER/IV/2010.

Tabel 1 : MPN Test Results for Raw Water, RO and Non-RO Refill Drinking Water

NO	Sample	*Coliform* / 100 ml	Average
1	A1	9	17,66
2	A2	27	
3	A3	17	
4	B1	2	3,66
5	B2	9	
6	B3	0	
7	C1	265	184
8	C2	265	
9	C3	22	

Information: A1 = Sample raw water at depot 1
A2 = Raw water sample at depot 2
A3 = Sample of raw water at the depot 3
B1 = Sample RO refill drinking water at depot 1
B2 = Sample RO refill drinking water at depot 2
B3 = Sample RO refill drinking water at depot 3
C1 = Non RO refill drinking water sample at depot 1
C2 = Non RO refill drinking water sample at depot 2
C3 = Non RO refill drinking water sample at depot 3

Based on Table 1, the estimated average value of the highest number of bacteria is found in non-RO refilled drinking water. Aprilia [1], stated that the presence of *Escherichia coli* bacteria in refill drinking water indicates that there is microbial contamination in drinking water. This is not in accordance with the purpose of the refill drinking water depot, namely to remove bacteria content by carrying out several stages of filtration, According to Sampulawa & Tumanan [20] one of the causes of the presence of bacteria in refilled drinking water is due to the lack of attention to the drinking water production process according to the Decree. The Minister of Industry and Trade of the Republic of Indonesia Number 651/MPP/kep/10/2004 concerning the technical requirements of drinking water depots and their trade. The observed bacterial contamination can be seen in table 2.

Tabel 2 : Bacterial Contamination in Raw Water, Drinking Water Refill RO and Non RO

NO	Sample	*Coliform*	*Escherichia coli*
1	A1	✓	✓
2	A2	✓	✓
3	A3	✓	-
4	B1	✓	-
5	B2	✓	-
6	B3	-	-
7	C1	✓	✓
8	C2	✓	-
9	C3	✓	✓

Water containing *Coliform* and *Escherichia coli bacteria* can be seen based on the colony color that grows on the EMB medium. According to Sunarti [16] "the colonies that grow metallic green are *Escherichia coli bacteria*, while according to Mudatsir [12]" the bright red and metallic colonies are *Escherichia coli* colonies while the dark red and slimy colonies are thegroup *Coliform"*.

To compare the estimated amount of bacterial contamination using the MPN method in refill drinking water between the three depots, the Variance Analysis (ANAVA) test was carried out, the results obtained were that there was a significant difference between F_{count} (8.446) $\geq F_{table}$ (5.14), so it is necessary toout further tests with using the Duncan test. Based on the results of the Duncan Test, there is a significant difference in the amount ofcontent *Coliform* in Non RO refilled drinking water with RO refilled drinking water. Meanwhile, thecontent *Coliform* in raw water and RO did not differ significantly.

Tabel 3 : Content of Cadmium (Cd), Chromium (Cr) and Cyanide (CN) in Raw Water, RO and Non RO Refill drinking water

NO	Sample	Cadmium (Cd)	Chromium (Cr)	Cyanide (CN)
1.	A1	<0,002	0.0069	0,002
2.	A2	<0,002	<0.002	0,001
3.	A3	<0,002	<0.002	0,006
4.	B1	<0,002	<0.002	0,001
5.	B2	<0,002	0.0050	0,001
6.	B3	<0,002	<0.002	0,001
7.	C1	<0,002	<0.002	0,002
8.	C2	<0,002	<0.002	0,001
9.	C3	<0,002	<0.002	0,001

Based on the results of research, the content of inorganic chemicals in all depots has met the standards of the Minister of Health of the Republic of Indonesia Regulation No.492/2010. This is probably because the amount of contamination from the content of Cadmium (Cd), Chromium (Cr) and Cyanide (CN) in the Krueng Aceh river, which is the main source of raw water for all the depots studied, is still lightly polluted.

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IV. Discussion

Based on research conducted by Hadi [2], the mouth of the Krueng Aceh river was contaminated with heavy metal cadmium (Cd) with a value of 0.0137-0.0168 mg / L with an average value of 0.015 mg / L. Meanwhile, the research conducted by Irhamni [6], who tested the chromium content of leachate water on the banks of the Krueng Aceh river in Kampung Jawa, had the amount of chromium (Cr) contamination of 0.0502 mg / L and could directly affect the quality of river water. Based on the monitoring carried out by Bapedal from August to October 2014, the value for each location (Segment) is as follows: Lambaro bridge segment value -17 (moderate polluted), Pango bridge segment value -17 (moderate polluted), Surabaya bridge segment value -43 (heavily polluted), Pante Pirak bridge segment value -30 (moderate polluted). The Peunayong bridge segment has a value of -42 (heavily polluted) and the Gampong Java segment has a value of -36 (heavily polluted).

The process of obtaining clean water that will be distributed to the community, both for daily needs and as raw material for processing refill drinking water, PDAM Tirta Daroy carries out several water treatment processes in general consisting of an intake, distribution tower, sedimentation / clarifier , pulsator (flocculation / sedimentation), sand filter, and reservoir [7]. Raw water taken from the Krueng Aceh river enters through the intake mouth where at this intake there is a *screen* (filter) which functions to filter coarse waste, then enters a temporary storage container called a distribution tower, then the sedimentation / clarifier process is carried out using chemicals such as alum and chlorine water. then proceed with the filtering process using silica sand which is called the sand filter process which functions to filter out mud and other inorganic chemicals. Even though the Krueng Aceh river is indicated to be contaminated by several inorganic chemicals, the presence of several treatments by PDAM Tirta Daroy allows it to reduce the amount of contamination levels in water that has been treated. Besides that, filtering tools such as sand filters and carbon filters that are exposed to the three refill drinking water depots are still functioning properly, so they can reduce the amount of inorganic chemical content. Saleh [19] stated that the completeness and quality of the equipment greatly affects the quality of refill drinking water such as filtering (sand filter, carbon filter and micro filter).

Analysis of microbiological content, for the average refill drinking water in the Kopelma Darussalam area has not met the standards, this is in line with the results of direct observation and the depot hygiene sanitation implementation questionnaire distributed to the three depots studied still does not meet the requirements according to the Decree of the Minister of Industry and RI Trade No. 651 / MPP / Kep / 10/2004 concerning Technical Requirements for Drinking Water Depots and Their Trading.

Decree of the Minister of Industry and Trade of the Republic of Indonesia No. 651 / MPP / Kep / 10/2004 has stipulated several requirements such as business requirements, raw water, processing processes, machines / equipment and drinking water quality, containers, supervision, and reports. The processing, machinery / equipment and quality of drinking water consist of: depot design and construction, raw materials, machinery and equipment, production processes, maintenance of production facilities and sanitation programs, drinking water products, employees and storage of raw water and sales. Meanwhile, the report is that the depot is obliged to report the laboratory analyst and submit the results of the supervision carried out by the competent authority.

The results of observations made at the three refill drinking water depots in the Kopelma Darussalam Banda Aceh area, the three depots still did not have a business license and the supervision carried out was not in accordance with the Decree of the Minister of Health No. 907 / Menkes / SK / VII / 2002 which required that supervision drinking water depots are regularly conducted at least 2 times a year. Supervision is carried out by Sanitation from association organizations or health workers who handle Food and Beverage Sanitation Hygiene

(HSMM) in the City / Regency or KKP assisted by Sanitarian Puskesmas. Yadav [25] stated that periodic monitoring of drinking water quality is carried out to determine whether the drinking water source is contaminated by pathogens.

The design and construction of the depot on average at the three depots did not meet the requirements, especially for spatial planning at depots one and three did not meet the requirements because the raw water storage space was too narrow and dirty, difficult to reach and dark. For building construction, floors, walls, ceilings and doors meet the requirements. As for the location, the three depots still did not meet the standards, because the three depots were located on the edge of the main road, thus allowing pollution through the air. In addition, the third depot has a water drainage system that is not good. According to Walangitan [24] DAMIU entrepreneurs and managers must maintain production facilities and sanitation programs to avoid contamination of drinking water by bacteria *Coliform*, that is, by means of buildings and parts that must be maintained, sanitized periodically, preventing entry of rodents, insects, other small animals into in buildings and filling stations.

Access and sanitation facilities at the three depots are still not up to standard, because they do not have hand washing containers, do not provide hand soap, do not have toilets and trash cans. One of the transmission of bacteria *Escherichia coli* can be caused by individuals who do not maintain cleanliness. Washing your hands with soap is one of the most effective ways to reduce the possibility of bacterial contamination. Based on the Decree of the Minister of Trade of the Republic of Indonesia Number 651/MPP/kep/10/2004, it is stated that employees related to production must be in good health, free from wounds, skin diseases or other things that are suspected of causing contamination of drinking water. Employees are required to wear appropriate headgear and shoes. Employees must wash their hands before refilling water. Employees are also not allowed to eat, smoke, spit or take any other action while doing work that can contaminate drinking water.

The water treatment facilities at the three depots have not replaced the filters routinely, this can cause the bacteria contained in the raw water to not filter properly, so that the refill drinking water still contains bacteria. Based on research conducted by Pradana and Bowo (2013), one of the reasons for the quality of refill drinking water is still not fulfilling the requirements due to the lack of awareness of depot entrepreneurs in maintaining drinking water production equipment routinely. The same research was conducted by Marpaung and Bowo (2013) which stated that one of the causes of bacterial contamination in refill drinking water could be caused by contamination of equipment and maintenance of processing equipment.

The customer service at the three depots also does not meet the requirements, where the gallon washing process does not use filtered water, but raw water. This causes the gallon to be filled with water not in a clean state, allowing it to be contaminated by bacteria found in raw water, or bacteria that adhere to the gallon because it is not washed clean. According to Nuria [15], the presence of microbial contamination in refilled drinking water can be caused by several factors, one of which is due to contamination from unsterilized gallons.

The raw water at the three depots does not have a clean water supply letter from the PDAM, does not pass a raw water quality test, and does not take raw water samples periodically. Raw water can also affect the quality of refilled drinking water. Decree of the Minister of Industry and Trade Number 651/MPP/kep/10/2004 regarding technical requirements for drinking water depots and their trade, raw water sources must be protected from chemical and microbiological contamination which is damaging or disturbing to health. The Regulation of the Minister of Health of the Republic of Indonesia Number 416/MENKES/PER/IX/1990 requires a maximum threshold for the content of *Escherichia coli* bacteria in clean water that is not 50/100 ml of sample piping, while *Escherichia coli* in piped water is 10/100 ml samples. According to Rizki [18] The more *Escherichia coli* bacteria are contaminated in raw water, the more difficult it will be to eliminate them if the drinking water treatment procedure does not meet the required standards.

Based on the results of observations carried out, the contamination of *Coliform* and *Escherichia coli* bacteria in RO and Non RO refilled drinking water may be caused by several things, including the lack of regular supervision, lack of knowledge of refill drinking water depot employees, lack of sanitation facilities, procedures. Unhygienic water treatment, contamination of water as raw material, lack of awareness of drinking water depot owners in maintaining the quality of drinking water without paying attention to regular maintenance of equipment.

This research is in line with the Sampulawa and Tumanan theory [20], which is one of the reasons for the presence of bacteria in refill drinking water due to the lack of attention to the drinking water production process according to the Decree of the Minister of Industry and Trade of the Republic of Indonesia Number 651 / MPP / kep / I0 / 2004.

V. Conclusion

There is a difference in the amount of *Escherichia coli* and *Coliform* in raw water, RO and non-RO refill drinking water. Refillable drinking water in the Kopelma Darussalam Banda Aceh area does not meet the requirements based on microbiological parameters according to the Minister of Health Regulation No.

492/2010. There is no difference in the amount of Cadmium (Cd), Chromium (Cr) or Cyanide (CN) in raw water RO and Non RO refills in Kopelma Darussalam Banda Aceh area. Refill drinking water in the Kopelma Darussalam Banda Aceh area has met the requirements based on parameters.

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