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by Ni Made Ayu Suardani Singapurwa

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Analysis of cyclamate sweeteners in elementary school student's drink in South Denpasar Subdistrict

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Abstract. Cyclamate is an artificial sweetener with a sweetness level of 30- 40 times greater than sucrose. This study aimed to determine the content and levels of cyclamate sweetener in unmarked beverages without labels that are prepared and sold by elementary school canteens in South Denpasar District. This research is a type of descriptive research that uses survey methods with simple random sampling techniques and experiments in the laboratory. The number of elementary schools in South Denpasar District is 72 schools. The population in this study were 145 types of unmarked beverages without labels that were prepared and sold by elementary school canteens in South Denpasar District. The number of samples sampled is 110 samples. The parameters of artificial sweeteners in this study were cyclamate tested by HPLC. Research data are presented in tables, graphs and analyzed descriptively. The results showed that of 110 samples containing 62 cyclamate (56.36%). Cyclamate levels that exceed the maximum limit requirements so that they do not meet the requirements of 44 samples (40%) and meet the requirements of 66 samples (60%), according to the maximum allowable limits refer to the Head of the Indonesian National Agency for Drug and Food Control No. 4 of 2014.

1. Introduction

School children snacks are snacks sold in the school environment and routinely consumed by most school children. Food snacks for school children play a strategic role as one of the contributing sources of nutrition for children while at school. Food snacks for school children have not all met health requirements. The results of the supervision of the quality of national school food snacks show that as many as 19.21% of school food snacks of 15,917 samples tested did not meet the requirements. The cause of samples that do not meet the requirements is due to using hazardous substances that are prohibited for food, using food additives exceeding the maximum limit, containing heavy metal contamination exceeding the maximum limit and microbiological quality that does not meet the requirements [1].

Artificial sweeteners are increasingly used as sweeteners in food and drinks. That is because artificial sweeteners have the same sweetness even more when compared to natural sweeteners. But excessive use of artificial sweeteners will cause toxic effects that are not good for health. The toxic effects caused by artificial sweeteners include headaches/migraines, dry mouth, nausea, vomiting, diarrhea, and bladder cancer [2]. The results of the Jakarta Consumer Institute's research show that 9 of 48 types of food, especially children's food, use artificial sweeteners (aspartame, saccharin, and cyclamate), which

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can negatively affect brain nerves and cancer. The use of these materials in European countries has long been banned [3].

Cyclamate is an artificial sweetener with a sweetness level of 30-40 times greater than sucrose. The carcinogenic potential of cyclamate occurs when it is converted to cyclohexylamine in the digestive tract [4]. Cyclohexylamine is toxic and is a tumor promoter [5]. Cyclamate is generally in the form of calcium, potassium, and cyclamate sodium. Cyclamate salt is white crystalline, odorless, colorless, and dissolves easily in water and ethanol. The combination of using cyclamate with saccharin is synergistic and is compatible with taste buds and as preservatives. The physical properties of cyclamate are heat-resistant, so they are often used in foods that are processed at high temperatures such as canned food [6].

Based on research results giving artificial sweeteners (aspartame, saccharin, and cyclamate) to experimental animals of mice showed that the higher the dose of artificial sweeteners given the higher the degree of damage to the liver [7]. Therefore, research was conducted on the analysis of the content of cyclamate artificial sweeteners in snacks for elementary school children in the South Denpasar District. The purpose of this study was to determine the content of cyclamate artificial sweeteners in unmarked drinks without labels prepared and sold by traders in the elementary school canteen in South Denpasar District and to determine the levels of cyclamate artificial sweeteners on unmarked beverages without labels prepared and sold by traders in the elementary school canteen in South Denpasar District are eligible or not with the maximum allowable limit.

2. Material and methods

This research was conducted in the South Denpasar Subdistrict which consisted of 8 sub-districts / villages namely Sidakarya, Sesetan, Panjer, Pedungan, Renon, Sanur, Pemogan, and Serangan with a total number of schools of 72 elementary schools. Determination of the population in this study was done by purposive sampling that is unmarked beverages without labels that are prepared and sold by traders in the primary school canteen. Bottled drinks with label brands and marketing authorization are not taken as a study population because pre-market and post-market supervision has been carried out by the competent authority. The population is 145 drinks from 72 elementary schools. The cyclamate sweetener test was carried out at the Chemical Laboratory of the Center for Drug and Food Control in Denpasar. The study was conducted by describing a phenomenon, event, event that occurred at present, by linking one variable with another variable [8]. This research uses survey and experimental methods in the laboratory.

The determination of the school canteen sampled for sampling is done at random (simple random sampling). The sampling technique was done once for each trader in the school canteen. In one school canteen which is included in the sample, criteria will be fully sampled. Based on the *Slovin* Formula [9] namely the determination of the minimum number of samples to be examined with a defined margin of error of 5% (95% confidence level), the calculation is as follows:

$$n = \frac{N}{(1 + (Nx e^2))} \tag{1}$$

Where n is the sample, N is the population and e is the margin of error (5%). With the *Slovin* formula, the minimum number of samples to be studied is 106 samples from 145 existing populations.

The cyclamate test is carried out with a qualitative test first with the precipitation method. The positive qualitative test results were continued to the level determination test (quantitative) using High-Performance Liquid Chromatography (HPLC) UV detectors. Before being used for sample testing, verification of the cyclamate sweetener method is carried out first [10]. The results of the study will refer to the Regulation of the Head of the Indonesian National Agency for Drug and Food Control No. 4 of 2014 concerning the Maximum Limit of the Use of Sweetener Additives [11].

3. Results and discussion

Based on the sampling results obtained 110 samples of drinks according to the sample criteria consisting of 51 samples (46%) colored drinks, 36 candles of ice samples (33%), 16 samples of tea drinks (14%),

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3 samples of mixed ice (3%), and miscellaneous drinks 4 samples (4%). The results of the analysis of cyclamate artificial sweeteners in the sample can be seen in Table 1 to Table 5.

Table 1. Results of cyclamate sweetener tests on color drinks.

School Code	Beverage	Level of	Maximum
	Drinks	cyclamate	limit
		(mg/kg)	10
B, D, F, G, I, Y, CC,	Chocolate	Negative	350 mg/kg
P, Q,T,V,AA1, AA2	Chocolate	Negative	
M	Chocolate	213.58	
BB	Chocolate	6440.20*)	
A, N, Y, Z,	Green	Negative	350 mg/kg
C	Green	1825.31*)	
AA	Green	223.64	
BB	Green	4852.15*)	
S	Green	399.16*)	
V	Green	326.04	
FF	Green	3435.90*)	
B, Q	Red	Negative	350 mg/kg
В	Red	585.08*)	
C	Red	98.81	
O	Red	1606.22*)	
AA	Red	368.97*)	
P	Red	668.47*)	
R	Red	3587.23*)	
FF	Red	3479.55*)	
C	Yellow	1880.61*)	350 mg/kg
N	Yellow	79.54	
M	Yellow	1402.81*)	
FF	Yellow	3496.33*)	
Q, V	White	Negative	350 mg/kg
H	White	31.86	
M	White	223.99	
H	Orange	270.24	350 mg/kg
AA	Orange	199.62	
FF	Orange	175.51	
AA	Purple	Negative	350 mg/kg
BB	Purple	4950.23*)	
CC	Purple	1181.26*)	
CC	Blue	Negative	350 mg/kg
F	Blue	216.59	
I	Blue	130.01	
Containing of Cyclama	ite	28 sample	
Does not contain Cycla	mate	23 sample	
Quantify		51 sample	
Note: *): not qualified		•	

Note: *): not qualified

In Table 1. Shows as the cyclamate sweeteners in 51 colored beverage samples. In the qualitative test results, there were 23 samples of negative colored drinks that did not contain cyclamate and 28 samples that were positive contained cyclamate. Positive samples containing cyclamate were continued with quantitative tests. In the quantitative test of 28 positive samples containing cyclamate obtained the smallest levels of 31.86 mg/kg are found in white drinks and the largest levels of 6440.20 mg/kg in chocolate drinks. In the sample of colored drinks out of a total of 51 samples tested cyclamate there

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were 16 samples (31.37%) whose levels exceeded the maximum limit requirement which is greater than 350 mg/kg. This shows 31.37% of colored drinks are not eligible. Whereas 35 samples (68.63%) were smaller than 350 mg/kg so that they met the maximum allowable limits.

Table 2. Results of cyclamate sweetener tests on ice lolly / ice stick.

School Code	Beverage Drinks	Level of cyclamate (mg/kg)	Maximum limit
			11
BB, U	Chocolate	Negative	250 mg/kg
C	Chocolate	3513.95*)	
E	Chocolate	1006.83*)	
K	Chocolate	4329.27*)	
DD	Chocolate	2298.77*)	
BB, U	Green	Negative	250 mg/kg
E	Green	1120.87*)	
G	Green	1115.57*)	
EE	Green	5017.17*)	
L, BB	Red	Negative	250 mg/kg
C	Red	2793.90*)	
G	Red	1752.50*)	
O	Red	1677.03*)	
K	Yellow	244.51	250 mg/kg
DD	Yellow	899.68*)	
EE	Yellow	2496.97*)	
L	White	Negative	250 mg/kg
O	White	177.06*)	
K	White	3150.45*)	
DD	White	1057.55*)	
L	Orange	Negative	250 mg/kg
E	Orange	2314.61*)	
E	Orange	2080.92*)	
G	Orange	81.61	
DD	Orange	2083.83*)	
EE	Orange	4221.79*)	
BB	Purple	Negative	250 mg/kg
K	Purple	5881.77*)	- 0 0
E	Blue	892.02*)	250 mg/kg
K	Blue	433.02*)	
DD	Blue	914.89*)	
Containing of Cyclan		27 sample	
Does not contain Cyc		9 sample	
Quantify		36 sample	

Note: *): not qualified

In Table 2. Shows as the cyclamate sweeteners in 36 ice wax samples. In the qualitative test results, there were 9 samples of negative ice wax that did not contain cyclamate and 27 positive samples contained cyclamate. In the quantitative test of 27 positive samples containing cyclamate, the lowest levels of 81.61 mg/kg were found in orange ice stick and the largest concentration of 6992.25 mg/kg in yellow stick ice. In the ice stick samples from a total of 36 samples, there were 25 samples (69.44%) whose levels exceeded the maximum allowable limit that is greater than 250 mg/kg, so that 69.44% ice wax did not meet the requirements. Whereas 11 samples (30.56%) levels met the maximum allowable requirements.

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Table 3. Results of cyclamate sweetener tests on tea drink.

School Code	Tea Drinks	Level of cyclamate (mg/kg)	Maximum limit
R	Tea	Negative	350 mg/kg
R	Iced Tea	Negative	
J	Tea	869.80*)	
X	Tea	Negative	
X	Iced Tea	Negative	
Y	Tea	53.35	
AA	Tea	Negative	
P	Tea	151.59	
S	Tea	227.62	
T	Tea1	Negative	
T	Tea2	Negative	
W	Tea	86.60	
GG	Iced Cup Tea	Negative	
HH	Iced Cup Tea	Negative	
II	Iced Cup Tea	Negative	
II	Cup Tea	Negative	
Containing of Cyclamate		5 sample	
Does not contain Cyclamate		11 sample	
Quantify		16 sample	

Note: *): not qualified

In Table 3. Shows as the cyclamate sweetener in 16 tea drink samples. In the qualitative test results, there were 11 samples of tea drinks that did not contain cyclamate (negative) and 5 samples that were positive contained cyclamate. In the quantitative test of 5 positive samples containing cyclamate, the smallest level was 53.35 mg/kg and the largest level was 869.80 mg/kg. In the sample of tea drinks from a total of 16 samples tested there was 1 sample (6.25%) whose levels exceeded the maximum limit requirement which is greater than 350 mg/kg. This shows 6.25% of tea drinks are not eligible. Whereas 15 samples (93.75%) met the maximum allowable limit of 350 mg/kg.

Table 4. Results of cyclamate sweetener tests on mixed fruit ice dessert.

School Code	Mixed fruit ice dessert	Level of cyclamate (mg/kg)	Maximum limit
R	Mixed fruit ice dessert	2254.19*)	350 mg/kg
V	Mixed fruit ice dessert	870.25*)	
II	Mixed fruit ice dessert	Negative	
	cup		
Containing of Cyclamate		2 sample	
Does not contain Cyclamate		1 sample	
Quantify		3 sample	

Note: *): not qualified

In Table 4. Shows as the cyclamate sweetener in 3 mixed ice samples. Based on qualitative test results obtained 1 negative sample does not contain cyclamate and 2 positive samples contain cyclamate. In quantitative tests on 2 positive samples obtained levels of 2254.19 mg/kg and 870.25 mg/kg. The results showed that 2 samples (66.67%) exceeded the maximum limit requirement, which was greater than 350 mg/kg and therefore did not meet the requirements. While 1 sample (33.33%) met the maximum allowable limit.

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Table 5. Test results on other beverages.

School Code	Beverages	Level of cyclamate (mg/kg)	Maximum limit
A	Young coconut	Negative	350 mg/kg
A	Grass jelly ice	Negative	
GG	Cup Young coconut	Negative	
П	Cup orange ice	Negative	
Containing of Cyclamate		0 sample	
Does not contain Cyclamate		4 sample	
Quantify		4 sample	

In Table 5. Shows as the cyclamate sweetener in 4 samples included in other types of drinks such as young coconut ice drinks, grass jelly ice drinks, and orange ice drinks. Based on the qualitative test results, the four samples did not contain negative cyclamate, so they met the maximum limit requirement for using cyclamate, which was a maximum of 350 mg/kg. This maximum cyclamate limit requirement refers to the Regulation of the Head of the Indonesian Center for Drug and Food Control Number 4 of 2014 concerning the maximum limit on the use of sweetener food additives.

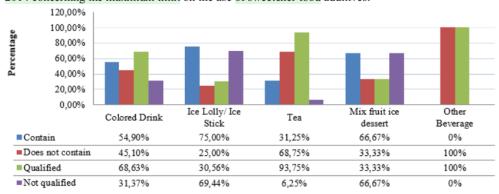


Figure 1. Cyclamate sweetener graph on hawker drinks.

Use of natural sweeteners is safe. But it has calories and can cause several adverse effects, such as cavities, increasing body weight, increasing triglycerides and diabetes. Conversely, artificial sweeteners do not have calories and do not cause health problems (do not produce cavities, no effect on blood sugar). But these sweeteners provide controversial side effects and safety. There are 5 artificial sweeteners that are permitted to use, namely accsulfame-K, aspartame, neotame, saccharin, and sucralose. But for more research and long-term surveys to be done to reduce the side effects of using artificial sweeteners [12,13].

Quantitative results show that some samples of cyclamate levels are very high and exceed the maximum allowable limit. Cyclamate metabolism in the stomach will produce cyclohexylamine compounds which are carcinogens. These compounds can cause bladder cancer and chromosomal damage. Besides cyclamate sweetener also raises many health problems, including tremors, migraine, or headaches, memory loss, confusion, insomnia, irritation, asthma, hypertension, diarrhea, abdominal pain, allergies, impotence, and sexual disorders, baldness and brain cancer. Effects arising from the use of cyclamate cannot be seen directly or in the short term because they have to wait twenty years to thirty years. However, use in children is better avoided because these sweeteners do not contain energy and nutrients. Excessive use can endanger health [14].

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The content of saccharin and cyclamate in snacks is the result of syrup containing cyclamate but still within the maximum allowable [15]. Syrup ice drinks sold by culinary traders in Malalayang beach, Manado City, obtained from 8 samples tested 6 samples contained positive cyclamate and 1 sample that exceeded the maximum allowable level of cyclamate [16]. Research on snacks has shown that in 16 samples of snacks in six traditional markets in Manado City there were 2 samples of syrup ice containing cyclamate artificial sweeteners namely red syrup ice at 931.98 mg/kg and yellow syrup ice at 848.65 mg/kg where the level exceeds the maximum allowable value [17]. The analysis of cyclamate in orange ice drinks at the University of North Sumatra obtained the results that the squeezed orange ice sold was detected to contain cyclamate artificial sweeteners with cyclamate levels exceeding the maximum allowable referring to the Regulation of the Head of the Indonesian Center for Drug and Food Control Number 4 of 2014 [18].

4. Conclusions

Laboratory test results from 110 samples containing cyclamate were 62 samples (56.36%) and those without cyclamate were 48 samples (43.64%). Samples that exceed the maximum allowable cyclamate limit were 44 samples (40%) and those who fulfill the requirements were 66 samples (60%). This requirement refers to the Regulation of the Head of the Indonesian National Agency for Drug and Food Control Number 4 of 2014, namely for drinks, tea, and mixed ice, maximum cyclamate levels of 350 mg/kg, whereas for ice stick the maximum cyclamate content is 250 mg/kg.

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