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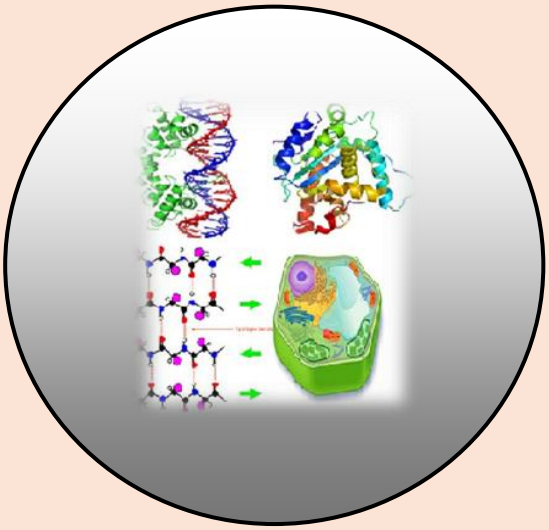
ISSN 2319-3077 Online/Electronic
ISSN 0970-4973 Print

Index Copernicus International Value
IC Value of Journal 82.43 Poland, Europe (2016)
Journal Impact Factor: 4.275
Global Impact factor of Journal: 0.876
Scientific Journals Impact Factor: 3.285
InfoBase Impact Factor: 3.66

J. Biol. Chem. Research
Volume 37 (2), Part A, 2020 Pages No. 21-27



Journal of Biological and Chemical Research

An International Peer Reviewed / Referred Journal of Life Sciences and Chemistry

Indexed, Abstracted and Cited in various International and National Scientific Databases

Published by Society for Advancement of Sciences®

J. Biol. Chem. Research. Vol. 37, No. 2, Part A: 21-27, 2020
(An International Peer Reviewed / Refereed Journal of Life Sciences and Chemistry)
Ms 37/02/1083/2020

ISSN 2319-3077 (Online/Electronic)
ISSN 0970-4973 (Print)

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RESEARCH PAPER

Received: 12/05/2020 Revised: 29/06/2020 Accepted: 30/06/2020

Analysis of Aspartame Sweeteners in Elementary School Student's Drink in South Denpasar Subdistrict

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ABSTRACT

Artificial sweeteners are substances that can cause sweetness or can help sharpen the acceptance of the sweet taste, but the calories produced are far lower than sugar. The uses of artificial sweeteners need to be aware because excessive doses can cause side effects that are detrimental to human health. This study aims to determine the content and levels of artificial sweeteners 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 and experiments in the laboratory. The number of elementary schools in South Denpasar District is 72 schools. The populations in this study were 145 types of unmarked beverages without labels prepared and sold by the elementary school canteen in South Denpasar District. The number of samples sampled was 110. The parameters of artificial sweeteners in this study were aspartame tested by HPLC. Research data are presented in tables, graphs and analyzed descriptively. Of the 110 test samples containing 35.45% aspartame. Aspartame levels of all samples meet the requirements by the maximum allowable limits referring to the Regulation of the Head of the Republic of Indonesia Drug and Food Supervisory Agency No. 4 of 2014.

Keywords: Artificial Sweeteners, Aspartame and Hawker Food.

INTRODUCTION

Food is a source of energy and various nutrients to support human life, but food can also be a source of health disturbance if the food consumed is not safe. The contribution of snacks to nutritional fulfillment is also reported to be quite important. Based on the results of the Monitoring and Verification Profile of School Children Snack Food Safety. Nationally conducted by the Food and Drug Supervisory Agency in 2008 it is known that the total school food consumption of snacks for students is 239.87 g / capita/day containing 384 kcal and 9.4 grams of protein. The survey results also showed that snack food contributed 3.1% of energy and 27.4% of protein from students' daily food consumption (Indonesian Drug and Food Control Agency, 2013). Food safety problems that are often encountered in the school environment are caused by processed food products in the school environment that is contaminated with hazardous materials (microbiological and chemical hazards), fast food in the school environment does not meet hygiene requirements, and problematic food donations (Ministry of Health, 2011). The occurrence of these problems is due to procedures for handling food that ignores the principles of food safety. These errors can be found in various aspects ranging from raw materials, handling (production process, storage, and presentation) and the procedures for distribution. Also, the ignorance of consumers, in this case, school children and teachers, the level of food safety of snacks causes food safety problems (Indonesian Drug and Food Control Agency, 2016).

Chemical hazards occur in the use of hazardous materials that should not be used in food. The use of food additives that exceeds the allowable limit as in the use of artificial sweeteners. The use of these materials is still often done by small traders who do not know the dangers but prefer the cheaper price (Ministry of Health, 2011). According to Regulation of the Head of the Food and Drug Supervisory Agency Number 4 of 2014 concerning the Maximum Limit of the Use of Food Additives Sweeteners, sweeteners are food additives in the form of natural sweeteners and artificial sweeteners that give sweetness to food products.

Even if the use is permitted, artificial sweeteners such as saccharin, cyclamate, acesulfame K, aspartame and other chemicals according to the rules of use must be limited. Although artificial sweeteners are safe for consumption in small amounts, within certain limits will pose a danger to the health of humans and animals that consume them. These restrictions are known to us as Acceptable Daily Intake. ADI is the maximum amount of artificial sweeteners in mg/kg body weight that can be consumed every day for life without causing adverse health effects (Zafar, 2017; Ardalan *et al*., 2017). Aspartate acid is a trigger for neuronal cell death because it will enter calcium radicals in neurons and methanol which will disrupt the work of the sense of sight (Ashok *et al*., 2014). The calorie value resulting from consuming aspartame is zero. This is because aspartame, when consumed, will be metabolized into several amino acids and like other amino acids, each gram will produce 4 calories, but due to the very high level of sweetness, when compared to sucrose sugar we use 200 grams, for aspartame we only need 1 gram to get the same sweet taste, so the caloric value can be ignored (Yusuf and Fatimah, 2013). Aspartame is one type of synthetic additive discovered by accident by James Schulter in 1965 when he wanted to synthesize drugs for boils and ulcers. Aspartame belongs to the class of methyl ester dipeptide compounds, namely L-aspartyl-alanine-methyl ester with the formula $C_{14}H_{16}N_2O_5$ and molecular weight 294.31 and with the sweetness of 100-200 times the saccharide sugar (Cahyadi, 2008). Aspartame is a compound with the appearance of white powder, odorless, slightly soluble in water and ethanol and tastes sweet. Aspartame is not suitable for use in foods whose production process requires high temperatures, because aspartame is only stable at room temperature and also stable at pH 4-5 so that it provides a large enough acidic atmosphere when dissolved in water (Yusuf and Fatimah, 2013).

Therefore, research was conducted on the analysis of the content of artificial sweeteners of aspartame in the snacks of elementary school children in the South Denpasar District. This study is to determine the content and levels of artificial sweetener aspartame in unmarked beverages without labels that are prepared and sold by traders in the elementary school canteen in South Denpasar District, whether or not they meet the maximum allowable limits.

MATERIALS AND METHODS

The study was conducted on 145 snack drinks at 72 Elementary Schools in South Denpasar Subdistrict in 8 villages namely Sidakarya, Sesetan, Panjer, Pedungan, Renon, Sanur, Pemogan, and Serangan. Determination of the population by purposive sampling, that is, drinks without brands without labels that are prepared and sold by traders in the primary school canteen. Aspartame sweetener testing was carried out at the Chemical Laboratory of the Center for Drug and Food Control in Denpasar. The determination of the school canteen sampled for sampling is done at random (simple random sampling). Sampling was done once for each trader in the school canteen. In one school canteen, the sample included in the sample criteria was completely sampled. Based on the Slovin formula (Husein, 2005), 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:

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

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 studied is 106 samples from 145 existing populations.

Objectively observing artificial sweeteners of Aspartame was carried out by a quantitative assay test using the simultaneous method using the HPLC UV-Vis detector (National Standardization Agency, 1992). Before being used for sample testing, verification of the artificial sweetener simultaneous method is carried out first (Indonesian Drug and Food Control Agency, 2013). The results of the study 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.

RESULTS AND DISCUSSION

Table 1 shows the results of aspartame sweetener in 51 samples of colored drinks. Based on quantitative test results there were 28 samples (54.90%) that were not detected containing aspartame and 23 samples (45.10%) containing aspartame with the smallest level of 44.20 mg/kg. In samples containing aspartame not exceeding the maximum allowable limit of 600 mg/kg, referring to the Regulation of the Head of the Indonesian Food and Drug Administration No. 4 of 2014 concerning maximum limits on the use of sweetener food additives.

Table 1. Results of Aspartame Sweetener Tests on Color Drinks.

School Code	Beverage Drinks	Level of aspartame (mg/kg)	Maximum limit
B, F, G, Y, AA, BB, CC,	Chocolate	Undetected	600 mg/kg
P, Q, T, V	Chocolate	Undetected	
D	Chocolate	53.07	
I	Chocolate	64.38	
M	Chocolate	183.28	
AA	Chocolate2	201.37	
A, Z, BB	Green	Undetected	600 mg/kg
C	Green	557.06	
N	Green	57.45	
Y	Green	59.80	
AA	Green	75.82	
S	Green	134.90	
V	Green	119.97	
FF	Green	549.83	
B, B,O,P,R,FF	Red	Undetected	600 mg/kg
C	Red	471.26	
AA	Red	114.45	
Q	Red	113.97	
M, FF	Yellow	Undetected	600 mg/kg
C	Yellow	558.36	
N	Yellow	72.25	
H, M, V	White	Undetected	600 mg/kg
Q	White	60.54	
H	Orange	56.78	600 mg/kg
AA	Orange	214.34	
FF	Orange	488.91	
BB, CC	Purple	Undetected	600 mg/kg
AA	Purple	125.20	
CC	Blue	Undetected	600 mg/kg
F	Blue	44.20	
I	Blue	61.03	
Containing of Aspartame		23 sample	
Does not contain Aspartame		28 sample	
Quantify		51 sample	

Table 2. Shows the results of aspartame sweetener in 36 ice wax samples. Based on quantitative test results, there were 23 samples (63.89%) that were not detected containing aspartame and 13 samples (31.11%) containing aspartame with various levels, namely the lowest level of 63.57 mg/kg and the highest 722.36 mg/kg. Aspartame levels in the stick ice sample meet the maximum allowable limit of 1000 mg/kg, referring to the Regulation of the Head of the Indonesian Food and Drug Administration No. 4 of 2014 concerning maximum limits on the use of sweetener food additives.

Table 2. Results of Cyclamate Sweetener Tests on Ice Lolly/ Ice Stick.

School Code	Beverage Drinks	Level of aspartame (mg/kg)	Maximum limit
C, E, K, BB, DD	Chocolate	Undetected	1000 mg/kg
U	Chocolate	99.18	
E, BB	Green	Undetected	1000 mg/kg
G	Green	470.98	
U	Green	63.57	
EE	Green	684.04	
C, O, L, BB	Red	Undetected	1000 mg/kg
G	Red	564.98	
K	Yellow	Undetected	1000 mg/kg
E	Yellow	507.58	
G	Yellow	574.01	
DD	Yellow	176.98	

EE	Yellow	722.36	
O, K, L DD	White White	Undetected 64.10	1000 mg/kg
E, G, L E DD EE	Orange Orange Orange Orange	Undetected 483.06 211,81 678.01	1000 mg/kg
BB, K	Purple	Undetected	1000 mg/kg
E, K, DD	Blue	Undetected	1000 mg/kg
Containing of Aspartame Does not contain Aspartame Quantify		13 sample 23 sample 36 sample	

Table 3. Results of Cyclamate Sweetener Tests on Tea Drink.

School Code	Tea Drinks	Level of aspartame (mg/kg)	Maximum limit
R	Tea	Undetected	600 mg/kg
R	Iced Tea	Undetected	
J	Tea	Undetected	
X	Tea	167.67	
X	Iced Tea	53.78	
Y	Tea	Undetected	
AA	Tea	Undetected	
P	Tea	Undetected	
S	Tea	Undetected	
T	Tea1	Undetected	
T	Tea2	51.98	
W	Tea	Undetected	
GG	Iced Cup Tea	Undetected	
HH	Iced Cup Tea	Undetected	
II	Iced Cup Tea		
II	Cup Tea		
Containing of Aspartame Does not contain Aspartame Quantify		3 sample 13 sample 16 sample	

Table 3. Shows the results of aspartame sweetener in 16 tea drink samples. Based on quantitative test results, there were 13 samples (81.25%) that were not detected containing aspartame and 3 samples (18.75%) containing aspartame with various levels, namely the lowest level of 51.98 mg/kg and the highest level of 167.67 mg/kg. Although these 3 samples contained aspartame, the levels did not exceed the maximum allowable limit of 600 mg/kg, referring to the Regulation of the Head of the Republic of Indonesia Food and Drug Administration No. 4 of 2014 concerning maximum limits on the use of sweetener food additives.

Table 4. Shows the results of aspartame sweetener in 3 mixed ice samples. Based on the results of the three ice mix samples were not detected to contain aspartame. The maximum allowable limit for aspartame on mixed ice is 600 mg/kg, referring to the Regulation of the Head of the Indonesian Food and Drug Administration No. 4 of 2014 concerning maximum limits on the use of sweetener food additives so that the three mixed ice samples meet the requirements.

Table 4. Results of Cyclamate Sweetener Tests on Mixed fruit ice dessert.

School Code	Mixed fruit ice dessert	Level of aspartame (mg/kg)	Maximum limit
R V II	Mixed fruit ice dessert Mixed fruit ice dessert Mixed fruit ice dessert cup	Undetected Undetected Undetected	600 mg/kg
Containing Aspartame Does not contain Aspartame Quantify		0 sample 3 sample 3 sample	

Table 5. Shows the results of aspartame sweetener in 4 beverage samples included in the sample types of other drinks such as young coconut ice drinks, grass jelly ice drinks, and orange ice drinks. Based on the test results, the four samples were not detected to contain aspartame. This shows that the four samples met the maximum allowable aspartame limit for drinks which is 600 mg/kg, referring to the Regulation of the Head of the Indonesian Food and Drug Administration No. 4 of 2014 concerning maximum limits on the use of sweetener food additives. The percentage of the results of the artificial sweetener aspartame test can be seen in Figure 1.

Table 5. Test Results on other Beverages.

School Code	Beverages	Level of aspartame (mg/kg)	Maximum limit
A	Young coconut	Undetected	600 mg/kg
A	Grass jelly ice	Undetected	
GG	Cup Young coconut	Undetected	
II	Cup orange ice	Undetected	
Containing Aspartame		0 sample	
Does not contain Aspartame		4 sample	
Quantify		4 sample	

Under conditions of the same degree of pH, the stability of aspartame decreases with increasing storage temperature. The stability of aspartame increases when the pH value is increased from 2.75 to 4.57. Aspartame is most stable at pH 2.75 when stored at 40 ° C, and stable at pH 4.57 and 20 ° C (Yakici and Arici, 2013). Research on 90 soft drink samples (brands A, B, C, D, E, and F) including 15 of each brand collected from supermarkets in Ankara Province, Turkey. Research by the spectrophotometric method was used for the quantitative determination of aspartame in the sample. The mean levels (± SE) of aspartame in brand samples A, B, C, D, E and F were found to be 156.81 ± 7.29 mg / l, 208.67 ± 8.97 mg / l respectively, 236.58 ± 17.91 mg / l, 299.54 ± 26.19 mg / l, 202.39 ± 8.08 mg / l and 223.28 ± 14.08 mg / l. The data revealed that the average levels of aspartame were found in the Turkish Food Codex in all samples. However, some samples were not found according to label information (Çelik *et al*., 2014).

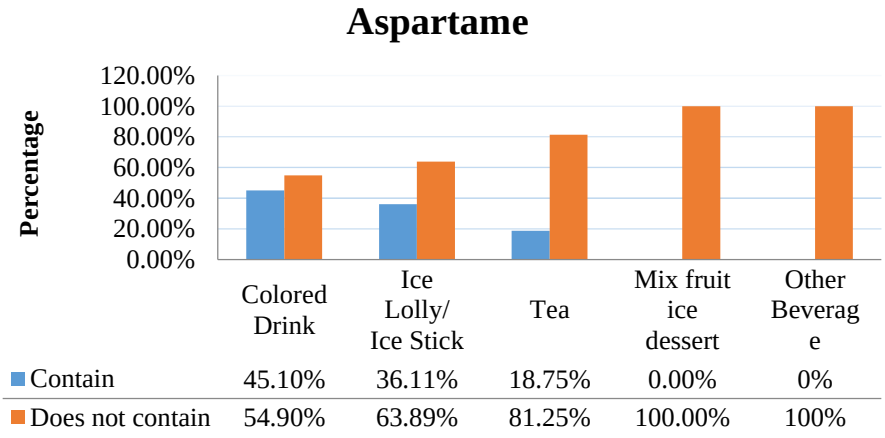

Figure 1. Test Results of Aspartame Artificial Sweeteners.

Dali *et al*. (2013) have researched artificial sweeteners of aspartame on school children's snack drinks circulating in Makassar in 7 samples containing 6 samples and 1 sample containing no aspartame. The aspartame content of the six samples is still within the maximum limit set at 600 mg/kg. Aspartame in the body is metabolized to aspartate acid, phenylalanine, and methanol that accumulates in the blood. Methanol absorption in the body is accelerated to formaldehyde (formalin), formic acid, and dicopyperazine which accumulate nucleic acids, proteins, and lipids. Some studies say the formaldehyde content can cause damage to neurons in the central nervous system, liver, lymph glands, and several other body organs. The damage is caused by the use of aspartame which exceeds the regulatory limit and is excessive for a long period (Oktavianti *et al*., 2005; Soffritti *et al*., 2005). Aspartame metabolism in the body produces as much as 40% aspartic acid, 50% phenylalanine and 10% methanol (Yusuf and Fatimah, 2013). Phenylalanine is a result of aspartame metabolism, which can be a trigger for phenylketonuria. Although in genetically consuming aspartame there is no phenylalanine.

While for patients with phenylketonuria it is forbidden to consume phenylalanine because it increases phenylalanine levels in the brain that cannot be metabolized by the body (Nantachit *et al*., 2008). Consumption of phenylalanine also makes serotonin levels decrease so that a person will experience emotional disturbances and carbohydrate syndrome. Research in various countries mentions other adverse effects of excessive use of aspartame. Diseases that arise include headaches, muscle spasms, nausea, numbness, insomnia, hearing loss, palpitations, and many others.

Aspartame can adversely affect the oxidant / antioxidant balance, induce oxidative stress, and damage the integrity of cell membranes, potentially affecting various cells and tissues and causing deregulation of cellular function, which ultimately leads to systemic inflammation (Choudhary and Pretorius, 2017). Chronic aspartame consumption and even an acceptable daily intake can cause liver cell injury (Ebraheim and Metwally, 2016). Carcinogenicity bioassay results in mice and prospective epidemiological studies, provide consistent evidence of carcinogenic potential by Aspartame (Soffritti *et al*., 2014).

CONCLUSIONS

Laboratory test results of 110 samples, containing 35.45% of aspartame and 64.54% not containing aspartame. The levels of artificial sweeteners in 110 beverage samples obtained results for aspartame levels all still meet the maximum allowable requirements. This requirement refers to the Regulation of the Head of the Republic of Indonesia Drug and Food Supervisory Agency Number 4 of 2014, namely for drinks, tea, and ice mixed with maximum aspartame levels of 1000 mg/kg.

ACKNOWLEDGMENTS

The author would like to thank those who have helped carry out this research, to the Laboratory of the Center for Drug and Food Supervision, Processing Laboratory of the University of Warmadewa, Principal of the Elementary School in South Denpasar District, traders in the school canteen, and friends who have helped carry out this research.

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