# Effect of Ability of Vegetable Materials Solutions and Environmental Conditions on Infertilization of Aedes AEGYPTI Mosquito Using Multivariate Adaptive Regression SPLINES (Mars)

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### Abstract:

**Background**: Dengue hemorrhagic fever (DHF) is a disease caused by an Arbovirus (dengue) virus which is transmitted by the Aedes aegypti mosquito as the main vector and Aedes albopictus as a secondary vector for dengue fever in tropical areas, including Indonesia. Control of vector-borne diseases currently still relies on the use of insecticides. The use of this insecticide has several weaknesses including the emergence of populations that are resistant to insecticides, environmental contamination and killing organisms.

**Materials and Methods**: This study aims to determine the effect of the ability of vegetable ingredients with a solution of pineapple (ananas comosus), bromelin, and sugar on the fertilization of the Aedes aegypti mosquito. This research is experimental with two treatments, namely the type of solution and the concentration of the solution. The samples used were pre-adult mosquitoes. The analysis technique used is Two-Way Analysis of Variance and MARS.

**Results:** The results showed that there were differences in the number of Aedes aegypty eggs produced after consuming a solution of pineapple (ananas comosus), bromelain, and sugar in the fertilization of Aedes aegypti mosquitoes, and sugar solution, as well as the ability to hatch eggs and the longevity of the mosquitoes, although with variations in concentration. no different, but the solution of pineapple (ananas comosus), bromelin, and sugar in the fertilization of Aedes aegypti mosquitoes can at least suppress the number of mosquito populations.

**Conclusion:** The ability of vegetable ingredients in a solution of Bromelain and Sugar with a solution concentration of above 1 percent succeeded in reducing the number of mosquito populations, so that it had an impact on reducing the morbidity rate by these mosquitoes, namely DHF.

Key Words: infertility, Aedes aegypti, bromelain enzyme extract, pineapple, MARS

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### I. Introduction

Dengue fever is a serious public health threat and an economic burden throughout the tropics [1]. The socio-economic and social impact of dengue is also very large because of this disease, and there is a tendency for an epidemic to explode that can paralyze the community and even the country [2]. Dengue infection in Indonesia causes an economic burden of more than 300 million US dollars per year or equivalent to Rp. 3.9 trillion [3]. The burdens and losses are related to medical expenses, the loss of patient productive time, and family productive time.

Control of vector-borne diseases currently still relies on the use of insecticides. The use of this insecticide has several weaknesses including the emergence of populations that are resistant to insecticides, environmental contamination and killing non-target organisms. However, the role of chemical insecticides has a very vital role, especially when there is an increase in cases or extraordinary occurrences of vector-borne diseases [4]. In the Regulation of the Minister of Health of the Republic of Indonesia Number 50 of 2017 concerning Environmental Health Quality Standards and Health Requirements for Disease-Carrying Vectors and Animals and their Control, it is stated that vector-borne diseases and disease-carrying animals are still a public health problem, both endemic and as new diseases that cause disease. has the potential to cause outbreaks so that it is mandated, among others, to develop vector control technology and disease-carrying animals [5]. The

negative impact of using insecticides has triggered the development of new methods, one of which is the method of genetically modified mosquitoes (GMM). The strategy currently being developed in GMM is the release of sterile male insects that have been exposed to radiation into the population and the integration of the dominant lethal gene under the influence of a specific promoter in immature male insects [6], and this GMM technology can be applied and adapted to other vector control methods. otherwise it can be integrated in an integrated vector control program. The sterile insect technique has been widely used and successfully controlled several types of pest species, including the eradication of the screwworm fly in the New World, Cochliomyia hominivorax in the United States, Mexico and throughout Central America, the control of the Mediterranean fruit fly Ceratitis capitata and the eradication of the tsetse fly. Glossina austeni on the island of Zanzibar. South Africa has now successfully used TSM to control the Mediterranean fruit fly (Medfly) in the Hex River basin [7].

Insect sterilization process can be done by using gamma rays and chemicals (chemosterilan). The use of chemosteril can cause insects to not be able to lay eggs, the eggs produced do not hatch, larvae cannot become pupae or pupa development is not perfect. Insect sterilization process using cemosteril is currently not recommended because it pollutes the environment and is carcinogenic [8]. Vegetable materials are found in nature and are usually used incidentally by humans. There are several vegetable ingredients that also contain active ingredients such as pineapple (Ananas Comosus) containing the enzyme bromelain. Bromelain enzyme that can stimulate the production of prostaglandins. Increased levels of prostaglandins cause stimulation of uterine contractions. Prostaglandins not only affect the contraction of the pregnant uterus, but also have an effect on uterine contractions in the non-pregnant state. In addition, pineapple also contains serotonin. Serotonin is a neurotransmitter in the central nervous system. In the uterus, serotonin can also play a role in stimulating uterine contractions [9]. Eggs that develop in the ovaries of the female mosquito. The internal reproductive system of the female mosquito consists of a pair of ovaries, a duct system that acts as a place for the release of eggs, and associated glands. [10].

Several studies on DHF, among others, [11], predicting larval density in the rainy and dry seasons based on health services, behavior, environmental conditions, and breeding sites in Banjarbaru City using Partial Least Square (PLS), [12], conducted Structural Equation Modeling Based on the Variance Density Index of Larvae in the Rainy Season in Banjarbaru City, and [13], predicting the incidence of Dengue Hemorrhagic Fever based on climatic factors with Multivariate Adaptive Regression Spline Longitudinal.

Pineapple fruit which contains enzymes bromelain and serotonin besides being antiseptic can also affect the occurrence of uterine contractions in mice, so it is assumed that this can also affect the reproductive system of insects, especially mosquitoes which are also complete. The scope of this research is about the effect of pineapple fruit solution contained in pineapple (Ananas comosus) on infertility in Aedes aegypti mosquitoes. This study aims to determine the effect of vegetable ingredients with pineapple (Ananas comosus) solution, Bromelain Enzyme Extract, and Sugar in the fertilization of Aedes aegypti mosquitoes. The benefit of this research is that by obtaining the ability of bromelain solution and extract from pineapple as a vegetable material for the fertilization of Aedes aegypti mosquitoes, it is hoped that mosquito population control efforts can be useful for the government and the community because it can be used to make policies in the health sector, especially in efforts to prevent dengue.

## **II. Material And Methods**

The population was all pre-adult stage mosquitoes (aged from breeding in a special room at the Department of Environmental Health Poltekkes Kemenkes Banjarmasin in Banjarbaru. The sample in this study was 210 pairs of pre-adult mosquitoes for all treatments (10 pairs/treatment) on a laboratory scale, and 20 on a field scale (Steel & Torrie (1995) [14]. Research variables consist of independent variables and dependent variables. There are two independent variables, namely the type of solution used as food for immature mosquitoes (pineapple fruit solution, bromelain extract solution, aqueous solution). sugar) (X1) and concentration of solution (0%, 1%, 2%, 4%, 6%, 8%, 10%) (X2) Dependent variable is Infertility (Y) i.e. Number of eggs and hatched eggs (ratio fruit/tail and % of hatched eggs). The conceptual framework of the research is as follows:



Figure 1. Conceptual Framework

The research steps are as follows:

- 1. Preparation of the research room, the research room is prepared separately from other rooms in the entomology laboratory, conditions are expected so that the research room is free from everything that might interfere with the research process such as chemicals and others.
- 2. Training of research assistants: The training was conducted to ask the perceptions of researchers, data collectors and research assistants. Equalization of perceptions about larva surveys, breeding mosquitoes, observing mosquito cycles, and collecting data on environmental conditions (data on water temperature, water pH, air temperature, humidity, number of mosquito eggs and hatching mosquito eggs) data recapitulation table attached.
- 3. Making pineapple bromelain extract by maceration method (soaking) and pineapple solution by blending ripe pineapple with a solution concentration of (0%, 1, 2%, 4%, 6%, 8%, 10%).
- 4. Rearing, which is the maintenance of mosquitoes in one cycle from mosquito larvae to eggs in a special place/space.

#### The analysis steps are as follows:

The initial stage is to determine the maximum number of basis functions (BF), maximum interaction (MI), and minimum number of observations between knots or minimum observations (MO). The general basis function used is two to four times the number of predictor variables. The predictor variables used in this study were two variables so that the basis function used varied for each predictor variable. For the 2 base function predictor variables, namely 4, 6, 8. The maximum interaction (MI) used is 1, 2, and the number of MO used is 0, 1, 2, 3 and 5. The stages of forming the MARS model itself are carried out by trying all a predefined combination of BF, MI, and MO values. From each of these combinations, a GCV value will be generated and it can be determined which predictor variables are included in the model.

### **III. Results And Discussion**

Banjarbaru City has an area of  $371.30 \text{ km}^2$  (37,130 ha). The entire area of Banjarbaru City is part of the urban area which is divided into 5 sub-districts and 20 urban villages. Banjarbaru City is located at coordinates  $3^025'40"$  to  $3^028'37"$  south latitude and  $114^041'22"$  to  $114^054'25"$  east longitude. Located at an altitude of 66 feet above sea level with a relatively flat area. The city of Banjarbaru also recognizes two seasons, namely dry and rainy seasons. This situation is closely related to the wind currents blowing in Indonesia. From June to September the wind flows from Australia and does not contain much water vapor, resulting in a dry season in Indonesia. On the other hand, from December to March, the wind currents contain a lot of water vapor from Asia and the Pacific Ocean after passing through several oceans, and in these months the rainy season usually occurs, the situation changes in April-May and October-November. In 2018 the air temperature ranged from 20.2°C to  $36.60^{\circ}$ C and humidity ranged from 76% -88%. In the last 5 years, rainfall in 2018 was the highest at 434.5 mm and the lowest at 76.0 mm with an average of 209.7 mm (BPS Kota Banjarbaru, 2019) [15].

umber of Aedes aegypti Mosquito Eggs									
Table 1. Number of Eggs in Aedes aegypti Infertilization Research									
	Concentration of Pineapple (Ananas comosus) fruit							Means	
	solution								
Solution Type	Container Color	0%	1%	2%	4%	6%	8%	10%	
Pineapple fruit	Green	994	702	514	643	475	706	636	667
solution	Red	-	-	-	-	-	-	-	0
	Yellow	-	-	-	-	-	-	-	0
Pineapple Bromelain	Green	994	1003	1198	1652	2001	1602	1351	1400

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Enzyme Extract	Red	-	-	-	-	-	27	-	27
	Yellow	-	-	-	-	-	-	-	0
Sugar solution	Green	994	1031	1235	1038	1203	552	1017	1010
	Red	-	-	-	-	358	340	163	287
	Yellow	-	74	374	29	124	149	20	128

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Based on Table 1, in the first stage experiment (Green Container Color), the pineapple fruit solution (Ananas comosus) showed the high egg-laying ability of the Aedes, sp (Aedes aegypty) mosquito, the mosquito lays eggs throughout its life or many times. Stage two (Red Container Color) and three (Yellow Container Color) pre-adult mosquitoes die before they can lay their eggs. Pre-adult mosquitoes in stages two and three live for only 3 - 9 days, although the mosquitoes are active and adaptable, they are even seen to still consume pineapple solution. Stage one Average 667 eggs per concentration with ten pairs of mosquitoes or about 53 eggs per female mosquito in her lifetime. In its lifetime, the mosquito lays eggs more than once, which is 2-6 times. Likewise, in the experiment using a solution of bromelain enzyme extract, only in stage one produced eggs with an average of 1400 eggs per pair per concentration of solution with an average of more than one egg laying. The average life span for stages two and three is 3-16 days. In general, mosquitoes can survive longer in a sugar solution, even under unfavorable conditions and can produce 1010 eggs per pair per concentration. In experiments using a sugar solution, observations can be made up to the third stage, although compared to stage one the number of eggs is less.

Table 2. Hatching Eggs in Aedes aegypti Infertilization Research									
Number of Eg	ggs that Hatch	Co	ncentrati	on of Pinea	apple (Ana	anas como	sus) fruit s	olution	Mean
Solution Type	Container Color	0%	1%	2%	4%	6%	8%	10%	
pineapple fruit	Green	188	80	136	60	40	44	125	96
solution	Red	0	0	0	0	0	0	0	0
	Yellow	0	0	0	0	0	0	0	0
Pineapple	Green	188	119	137	60	32	123	46	101
Bromelain Enzyme	Red	0	0	0	0	0	7	0	1
Extract	Yellow	0	0	0	0	0	0	0	0
sugar solution	Green	188	63	36	21	38	29	26	57
	Red	0	0	0	0	58	28	31	17
	Yellow	0	0	0	0	0	0	0	0

### Aedes aegypti Mosquito Eggs That Hatch

Based on Table 2, the Aedes aegypti eggs hatched in the experiment using a solution of pineapple (Ananas comosus) ranged from 40 to 188 eggs, meaning that the average number of eggs that hatched from all the eggs produced was 96 eggs. Aedes aegypti eggs hatched in experiments using the bromelain enzyme pineapple (Ananas comosus) ranged from 32 to 188 eggs, meaning that only 101 eggs hatched from all the eggs produced. Aedes aegypti eggs that hatched in the experiment using a sugar solution ranged from 21 to 188 eggs, meaning that only 57 eggs hatched from all the eggs produced.

Table 3. Perce	entage of hatc	hing eggs in A	Aedes aegypti	Infertilization	Research
	0	0 00	071		

Percentage									
hatch	0%	1%	2%	4%	6%	8%	10%	Mean	
	pinpineapple fruit solution	18.91	11.40	26.46	9.33	8.42	6.23	19.65	13.58
Solution Type	bromelain extract solution	18.91	11.86	11.44	3.63	1.60	7.68	3.40	6.60
1 3 50	sugar solution	18.91	6.11	2.92	2.02	3.16	5.25	2.56	3.67

Based on Table 3, the percentage of Aedes aegypti eggs that hatched in the experiment using a solution of pineapple fruit (Ananas comosus) ranged from 6.2% - 26.6.7%, meaning that the average number of eggs hatched from all eggs produced was 13.6%. The percentage of Aedes aegypti eggs that hatched in the experiment using the pineapple bromelain enzyme (Ananas comosus) ranged from 1.6% - 18.9%, meaning that only 6.6% of all eggs hatched. The percentage of Aedes aegypti eggs that hatched in the experiment using a

sugar solution ranged from 2.0% - 18.9%, meaning that the eggs hatched from all eggs produced as a whole were only 3.7%.

In this study, there were two treatments for the independent variable, namely the type of solution and the concentration of the solution, so the analysis was carried out using the Two-way Anova test. The results of the test of the difference in the percentage of hatching eggs in the Aedes aegypti mosquito infertility study in the type of solution and concentration, are presented in Table 4 below.

**Table 4**. ANOVA table on the percentage of eggs hatching in the infertility study Aedes aegypti mosquito on the type of solution and concentration

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Model	2656.469 <sup>a</sup>	9	295.163	12.576	.000
X1 (Solution Type)	266.714	2	133.357	5.682	.018
X2 (konsentrasi larutan)	487.543	6	81.257	3.462	.032
Error	281.634	12	23.469		
Total	2938.103	21			
R Squared = $.904$ (Adjusted R S	quared = .832)	-			

Based on Table 4, it shows that between concentrations there is a significant difference in the percentage of hatching eggs in the Aedes aegypti mosquito infertility study. This can be seen from the value of Sig. = 0.32 which is smaller than the value of = 0.05. Likewise, in the type of bromelain extract solution, pineapple solution, and sugar solution, there was a significant difference in the percentage of hatching eggs in the Aedes aegypti mosquito infertility study. This can be seen from the value of Sig. = 0.18 which is smaller than the value of = 0.05. The contribution of the type of solution and concentration to the percentage of hatching eggs in expressing the difference was 90.4 percent. So to determine the effect of the type of solution and the concentration of the solution on the percentage of eggs hatching in the Aedes aegypti mosquito infertility study, MARS modeling was used.

The MARS model with two predictor variables based on the values of GCV and MSE and  $O-R^2$  is not centered, can be presented in the following table.

 Table 5. MARS Model Percentage of Hatched Eggs in Infertilization Research of Aedes aegypti Mosquitoes

 Various MO based on GCV, O-R<sup>2</sup> [13]

Model	BF	MI	MO	GCV	$O-R^2$	Prediction Model
1	8	2	0	54.384	0.647	Y = 9.517
2	8	2	1	53.215	0.762	Y = 5.694 + 2.676 * BF2
						With
						BF2 = max(0, 5.000 - X2)
3	8	2	2	51.690	0.854	Y = 2.333 + 3.564 * BF2 + 2.093*BF5
						with
						BF2 = max(0, 5.000 - X2)
						BF3 = (X1 = 1)
						BF5 = max(0, X2 - 1.000) * BF3
4	8	2	3	42.449	0.908	Y = 13.939 - 2.211 * BF5
						With
						BF4 = (X1 = 2  OR  X1 = 3)
						BF5 = max(0, X2 - 1.000) * BF4
5	8	2	5	54.384	0.647	Y = 9.517
6	8	2	10	54.384	0.647	Y = 9.517

Source: Processed Data

Table 5 shows all the possible models that have been tested, the best model is obtained with the criteria of the model having the smallest GCV value, namely the 4th model with the number of BF = 8, MI = 2, and MO = 3 with a GCV value of 42,449 and an O-R<sup>2</sup> value. of 0.908. The best MARS model is:

Y = 13.939 - 2.211 \* BF5

With

BF4 = (X1 = 2 OR X1 = 3)BF5 = max(0, X2 - 1) \* BF4

The interpretation of the MARS model written in equation (1), namely the BF5 coefficient will be meaningful if the value of X2 (solution concentration) is greater than 1 percent and X1 (type of pineapple bromelain enzyme extract solution) or (type of sugar solution). This influence means that every increase in one basis function (BF5) can reduce the percentage of hatching Aedes aegypti eggs by 2.221 percent.

#### **IV.** Conclusion

Based on the results of the analysis and discussion of senile cataract cases, the following conclusions are obtained.

- 1. Environmental conditions that play a role in the research process on the fertilization of Aedes aegypti mosquitoes are air temperature and humidity, temperature and PH of hatching water.
- 2. There is a difference in the percentage of Aedes aegypti eggs that hatch in the Aedes aegypti mosquito fertilization research process using pineapple fruit (Ananas cumosus) solution, pineapple (Ananas cumosus) bromelain enzyme extract, and sugar solution.
- 3. There is a difference in the percentage of Aedes aegypti eggs that hatch in the Aedes aegypti mosquito fertilization research process using a solution concentration of 0%, 1%, 2%, 4%, 6%, 8%, and 10%
- 4. Statistically (solution concentration) was greater than 1 percent and X1 (type of pineapple bromelain enzyme extract solution) or (type of sugar solution) had a lowering effect on the percentage of hatching Aedes aegypti eggs.

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