Effect of *Arthrospira maxima* Setchell et Gardner and *Chlorella vulgaris* Beijerinck on Eritrhocyte and Thrombocyte Profile of Hyperglycemia Wistar Rats (*Rattus norvegicus* Berkenhout, 1769)

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Keywords: Arthrospira maxima, Chlorella vulgaris, Erithrocyte profile, Thrombocyte profile, Hyperglycemia

Abstract: Community diet patterns that tend to be unhealthy with the consumption of fast food can cause metabolic disorders or metabolic syndromes such as diabetes mellitus. Diabetes mellitus is characterized by a chronic hyperglycemia condition that is an increase in blood glucose levels and keto-acidosis. Handling of diabetes mellitus patients using synthetic chemical drugs has dangerous side effects. The aims of this study examines the effect of microalgae that is suspected of being potential as an alternative medicine for the prevention or cure of diabetes mellitus. The purpose of this study was to determine the effect of administration of Arthrospira maxima and Chlorella vulgaris on the erythrocyte and platelet profiles of Wistar rats (Rattus norvegicus Berkenhout, 1769) hyperglycemia on the induction of DMT2. Twenty Wistar male rats were divided into 5 groups consisting of three control groups namely healthy controls, hyperglycemia, and drugs, and 2 treatment groups namely A. maxima and C. vulgaris. DMT2 induction uses a high carbohydrate diet and a single dose streptozotocin. Erythrocyte and platelet profiles were measured at D0, D15, and D30. Body weight and blood glucose levels are measured every 10 days. Data were analyzed using One-Way ANOVA. The erythrocyte profile was low in the control hyperglycemia rat after DM induction of D15. Erythrocyte profiles in hyperglycemia rat with this microalgae treatment, not significantly increased ($p>0.05$). Platelet profile in both the hyperglycemia and treatment groups was not significantly affected ($p>0.05$). Erythrocyte profiles in hyperglycemic rats have increased, platelet profile have decreased, and returned to better conditions after being given A. maxima and C. vulgaris.

1 INTRODUCTION

Healthy food is food that is good in the processing, cooking, serving, packaging and contains nutrients that are good function for the body. Foods that are publicly marketed today are foods that are ready to eat, good taste, look attractive, prices are relatively cheap, but the nutritional content is low and sometimes contains contaminants. Fast food products are increasingly becoming the menu of choice for most people rather than traditional foods. Good taste that appears because there is a high fat content. Increased fast food that increases without being balanced with adequate exercise can cause metabolic syndromes such as obesity and diabetes (Monteiro et al., 2016).

Metabolic syndrome is a set of factors consisting of hypertension, hyperglycemia, hyperlipidemia, and obesity that are closely related to the disruption of diabetes mellitus (Bonomini, Rodella, & Rezzani, 2015). Hyperglycemia is a condition of high glucose levels in the blood caused by consumption of high carbohydrates, stress conditions, and keto-acidosis (Lanywati, 2001)(Moore, 2008). Diabetes Mellitus Type 2 (DMT2) with insulin resistance has a high global pravalence including in Indonesia even reaching 90% (Anonim, 2014).

Patients with diabetes mellitus are often treated using synthetic chemical therapy that have side effects and need to avoid danger. Research on alternative therapy for the prevention or cure of diabetes mellitus is currently being done. One material that is widely studied is microalgae. Microalgae, especially cyanobacteria, are known to have therapeutic abilities (Belay, Ota, Miyakawa, & Shimamatsu, 1993).

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Chlorella sp. is an algae that can overcome heavy metals that are diabetagenic such as mercury and cadmium. *Arthrospira maxima* also has other nutritional components such as minerals, vitamins, chlorophyll, glycolipids, phycocyanin, carotenoids, tocopherols, linolenic acids, phenolic compounds, sulfolipids, and Superoxide Dismutase (SOD) as an antioxidant that is abundant in the activity of Reactive Oxigen Species (ROS) (Cousens, 2007)(Farag, Alagawany, El-Hack, & Dhama, 2016). In addition, this study also uses metformin as an anti-hyperglycemia agent(Mantzoros, 2006).

In the condition of hyperglycemia, glucose levels in cell fluid are also high. This situation has the potential to interfere with the function of cells to organs, including blood with its components. This is presumably because erythrocytes exposed to high glucose will change their character and become easily damaged and activate platelet function. The purpose of this study was to determine the effect of administration of *Arthrospira maxima* and *Chlorella vulgaris* on the erythrocyte and platelet profiles of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia on the induction of DMT2.

2 METHODS

This research is part of Mulyati (2017) regarding physiological responses in DMT2 induced rat with high carbohydrate diet and continued with the Widiyanto (2018) research concerning the effect of administration of *Spirulina* sp. and *Chlorella* sp. to the physiological response of White Rat (*Rattus norvegicus* Berkenhout, 1769) on the induction of DMT2. The study was conducted at the UGM Unit IV Integrated Research and Testing Laboratory (LPPT) as a place for acclimation, maintenance, treatment, and testing of blood glucose levels on animal models. This study is a following by study of hyperglycemia rat research with induction of high carbohydrate diet and low dose STZ that took place in May to October 2017 with an ethical clereance certificate numbered: 00097/04 / LPPT / VIII / 2017. Then followed by this study which began from 11 December 2017 to 11 January 2018 after obtaining approval from the Animal Ethics Commission Team with certificate number: 00167/04 / LPPT / I / 2018.

Test animals in this study were white rats (*Rattus norvegicus*, Bekenhout 1769) male Wistar strain obtained from LPPT UGM Unit IV. Rats aged 19 weeks weighing 300-400 grams after being induced by DMT2 and high carbohydrate diets. Twenty white rats were acclimated for 7 days and grouped randomly. Each cage consist of 4 rat.

Animal models are given commercial standard feed and given reverse osmosis (RO) drinking water. There are two stages of treatment in this study. The first stage is the induction of DMT2 by administering high carbohydrate diet and followed by injection of 30mg/kg.bw low dose streptozotosin (STZ) to obtain animal models of hyperglycemia for 45 days. The study continued with the induction of hyperglycemia using low-dose STZ booster on intraperitonially for 10 days before the treatment of microalgae and diabetes/metformin drugs.

The dosage given for each microalgae is 2500 mg/kg.bw dissolved in 12 ml of distilled water and metformin 10 mg/kg.bw dissolved in 4 ml of distilled water. Body weight and glucose levels are measured every 10 days. Hematology profiles are measured at D0, D15, and D30 and tested using a hematology analyzer. The results of the data were analyzed using One-Way ANOVA on SPSS 16.0.

Table 1: Grouping animal model

No.	Groups	Explanation
1.	Negative Control (NC)	Without treatment
2.	Positive Control (PC)	Hiperglicemic Control
3	Treatment 1 (M)	Metformin 100 mg/kg.bw in doses after hiperglicemia
4.	Treatment 2 (AMX)	*Arthrospira maxima* 2500mg/kg.bw in doses after hiperglicemia
5.	Treatment 3 (CVL)	*Chlorella vulgaris* 2500mg/kg.bw in doses after hiperglicemia

3 RESULTS AND DISCUSSION

The results of this study present the effect of consumption of *Arthrospira maxima* and *Chlorella vulgaris* at a dose of 2500 mg/kg.bw on erythrocyte profiles consisting of RBC, Hgb, HCT, MCV, MCH, MCHC, and platelet profiles consisting of Platelets, MPV, PDW, P-LCR, and PCT along with supporting data in the form of body weight and blood glucose. The results are presented in tabular and figure form as follows:

Rat body weight is secondary data that is used as supporting data for blood chemical levels that have been tested. While the data of blood glucose levels as a mark to determine the success of induction of hyperglycemia. The mean of the data can be presented in the form of a continue graph which is presented as figure 1, table 2 and table 3.Negative or healthy control group increased not significant in body weight that showed to be stable. This is due to the fact that rats are given feed and drink in an adlibitum routinely so that they are growth and development. Healthy control have undisturbed metabolism. In the group of treated rats (positive control, *A. Maxima*, *C. Vulgaris*, and Metformin) weight low increased not significant. This can be due to the induction of STZ given which makes high glucose levels.

Blood glucose levels in hyperglycemia group has higher blood glucose levels compared to negative control group and shows the success of STZ induction. Rats glucose levels increase after 3 days induced hyperglicemia and decrease after 10 days induced hyperglicemia. Decrease in all groups occurred insignificantly. According to Yosti's research (2017), *Chlorella vulgaris* can reduce blood glucose levels at a dose of 4 mg/20 g.bw. *A. Maxima* was proven to have a protein of 65-70% and high fiber, vitamins and minerals. The beneficial effects of spirulina are for the treatment of malnutrition, obesity, and diabetes mellitus which have been reported by (Ambrosi, Reinehr, Bertolin, Costa, & Colla, 2005).

Table 2: Body weight of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Body Weight (gram)			
	H 0	H 10	H 20	H 30
NC	358.00 ± 34.52^{b,x}	370.75 ± 38.95^{b,x}	376.20 ± 44.77^{b,x}	385.60 ± 52.93^{b,x}
PC	276.25 ± 25.66^{a,x}	289.43 ± 27.43^{a,x}	290.15 ± 23.31^{a,x}	296.17 ± 28.13^{a,x}
M	288.00 ± 30.31^{a,x}	294.13 ± 41.21^{a,x}	299.40 ± 42.34^{a,x}	308.43 ± 43.58^{a,x}
AMX	300.15 ± 28.67^{a,x}	304.30 ± 28.89^{a,x}	299.13 ± 28.39^{a,x}	302.03 ± 30.75^{a,x}
CVL	295.10 ± 41.73^{a,x}	299.35 ± 44.11^{a,x}	297.93 ± 43.24^{a,x}	300.95 ± 48.43^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 3. Blood glucose of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Blood Glucose Level (mg/dL)			
	H -7	H 0	H 15	H 30
NC	81 ± 12.69^{a,x}	87 ± 4.08^{a,x}	106 ± 10.60^{a,x}	96 ± 8.77^{a,xy}
PC	250 ± 99.95^{c,x}	296 ± 55.09^{b,x}	184 ± 104.98^{a,x}	186 ± 75.26^{a,x}
M	237 ± 105.75^{bc,x}	292 ± 134.32^{b,x}	193 ± 82.10^{a,x}	223 ± 127.06^{a,x}
AMX	190 ± 14.39^{bc,x}	210 ± 79.19^{ab,x}	175 ± 62.95^{a,x}	187 ± 110.62^{a,x}
CVL	139 ± 33.64^{ab,x}	277 ± 185.48^{b,x}	142 ± 62.89^{a,x}	153 ± 68.16^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

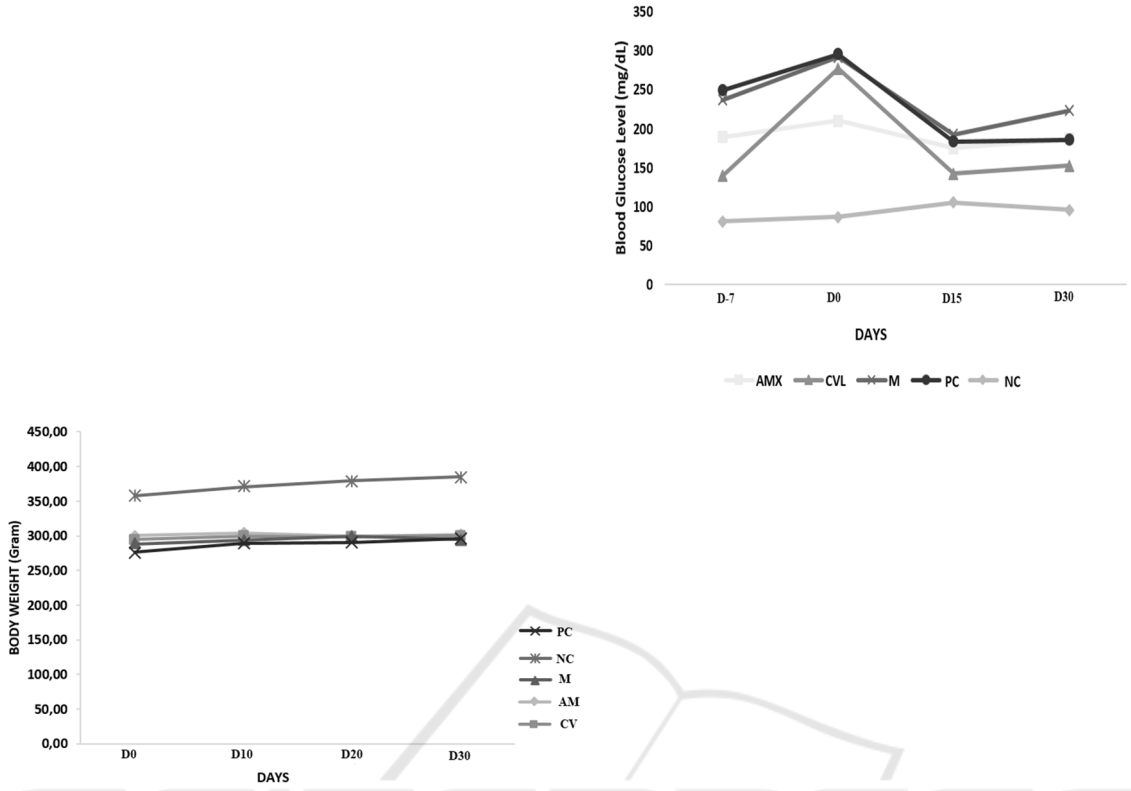

Figure 1: Body weight (a) and Blood Glucose Levels(b) of Wistar Rats (*Rattus norvegicus* Berkenhout,1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Table 4. Red blood cells of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	7.93 ± 0.2 ^{b,x}	7.97 ± 0.31 ^{a,x}	7.74 ± 0.32 ^{a,x}
Hiperglicemia control	7.76 ± 0.25 ^{b,x}	8.06 ± 0.54 ^{a,x}	8.03 ± 0.35 ^{a,x}
Metformin	7.32 ± 0.66 ^{a,x}	8.08 ± 0.44 ^{a,y}	8.22 ± 0.12 ^{a,y}
Arthrospira maxima	7.91 ± 0.33 ^{b,x}	7.89 ± 0.18 ^{a,x}	8.12 ± 0.19 ^{a,x}
Chlorella vulgaris	7.68 ± 0.17 ^{b,x}	7.54 ± 0.43 ^{a,x}	8.04 ± 0.32 ^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 5: Hematocrite levels of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy Control	43.93 ± 0.90 ^{a,x}	43.30 ± 2.46 ^{a,x}	43.18 ± 1.05 ^{a,x}
Hiperglicemia Control	42.63 ± 2.22 ^{a,x}	45.23 ± 2.61 ^{a,x}	44.13 ± 2.34 ^{a,x}
Metformin	42.90 ± 2.47 ^{a,x}	45.37 ± 2.68 ^{a,x}	47.45 ± 1.47 ^{a,x}
Arthrospira maxima	42.55 ± 3.91 ^{a,x}	44.80 ± 0.82 ^{a,x}	45.80 ± 1.27 ^{b,x}
Chlorella vulgaris	42.78 ± 0.75 ^{a,x}	42.83 ± 2.07 ^{a,x}	44.08 ± 1.77 ^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 6. Hemoglobine levels of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	14.98 ± 0.47 ^{a,x}	14.53 ± 0.40 ^{a,x}	14.70 ± 0.32 ^{a,x}
Hiperglicemia control	14.25 ± 1.01 ^{a,x}	15.40 ± 0.62 ^{a,x}	14.68 ± 0.66 ^{a,x}
Kontrol obat	14.50 ± 0.82 ^{a,x}	15.48 ± 0.98 ^{a,xy}	15.85 0.5 ^{b,y}
Athrospira maxima	14.25 ± 1.51 ^{a,x}	15.23 ± 0.46 ^{a,x}	15.63 0.40 ^{b,x}
Chlorella vulgaris	14.30 ± 0.41 ^{a,x}	14.45 ± 0.45 ^{a,x}	14.50 ± 0.82 ^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

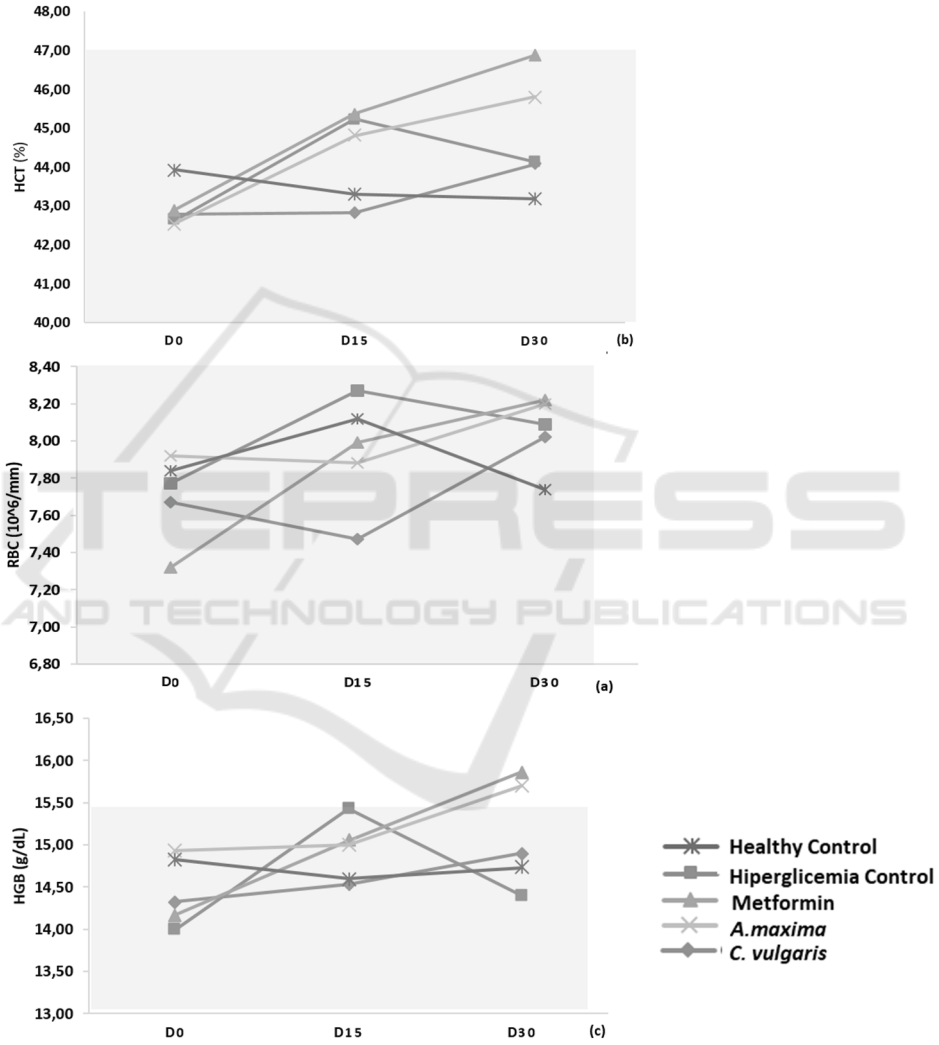

Figure 2: Red blood cells (a), Hematocrit levels (b), and Hemoglobin levels (c) Wistar rats (*Rattus norvegicus*, Berkenhout 1769) hyperglycemia with the treatment of *Chlorella vulgaris* and *Arthrospira maxima* for 30 days. Note: the blue box in Fig.2 shows the normal region of that level.

Based on reference data (Syariatin & Mulyati, 2017) a normal RBC range of 6.03-8.4 x 106 cells/uL is obtained. Figure 2a shows that hiperglicemia rats group have increased but are still in the normal range. Other groups experienced fluctuations in RBC values but were still in the normal range and no significant difference ($p>0.05$) on the average number of red blood cells affected by the factors and overall time. Groups of rats treated with metformin had significant differences in H0, H15, and H30 ($p<0.05$). This

shows that the administration of metformin 10 mg/kg.bw. can increase the RBC value and return as normal. Administration of *C. vulgaris* and *A. Maxima* at a dose of 2500 mg/kg.bw for 30 days was classified as being able to increase the total number of erythrocytes even though it was not significant ($p<0.05$).

Based on data (Syariatin & Mulyati, 2017) a normal range of hematocrit values was obtained between 34.6-47%. Fig. 2b shows that in all groups fluctuations in hematocrit values occurred. According to Emami's research shows that PVC in diabetics has decreased when compared to non-diabetics. The decrease in PCV in diabetics is possible due to dehydration and protein accumulation (Emami & Olfati, 2017). Figure 14 shows that in D0 the hyperglycemia induced rat group was not significantly different from the negative / healthy control group. In D15 the DM control mice experienced a significant increase and decreased after H15. This increase can be caused by correlating with the RBC value which shows the red blood cell production is still experiencing. But after that the HCT decreased due to the rat successfully inducing hyperglycemia and not receiving antidiabetic treatment. The possibility of rats dehydrated and characterized by a condition that is thicker blood. Decreased HCT value correlates with RBC value which also decreases after D15, this is due to the kidney failing to produce erythropoietin, and an increase in non-enzymatic reaction to glycosylation of RBC membrane protein, and both of these are also considered to be anemic factors (Emami & Olfati, 2017).

Based on data (Syariatin & Mulyati, 2017), the normal Hb range is between: 11.5-15.4 g/dL. These results indicate that Hb levels are positively correlated with RBC values and HCT values. Hyperglycemia group of rats, on D0-D15 has increased, this is possible because hyperglycemia rats are still producing erythropotein. Whereas before D15, rats in the DM group experienced decreased Hb levels. This is because hyperglycemic mouse hemoglobin is bound to high blood sugar levels in the blood stream (glycated). Hyperglicemia conditions can lyse blood cells and reduce hemoglobin levels in erythrocytes due to Reactive Oxygen Species (ROS) which cause oxidative stress. The persistent condition of hemoglobin and high glycosylation is a result of hyperglycemia associated with structural and functional changes in hemoglobin molecules (Hb), osmotic disorders, and cytoplasmic viscosity in cells(Alamri et al., 2019). Part of the glycated hemoglobin is the heme protein associated with the duration and level of hyperglycemia.

Table 7: Mean corpuscular Hemoglobin*e* levels of Wistar Rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	18.88 ± 0.66 ^{a,x}	18.86 ± 0.96 ^{a,x}	19.00 ± 0.43 ^{a,x}
Hiperglicemia control	18.50 ± 0.88 ^{a,x}	21.65 ± 1.00 ^{a,x}	18.28 ± 1.00 ^{a,x}
Metformin	18.95 ± 1.13 ^{a,x}	18.38 ± 1.28 ^{a,x}	19.03 ± 0.87 ^{a,x}
A. maxima	18.68 ± 1.16 ^{a,x}	19.30 ± 0.93 ^{a,x}	19.30 ± 1.18 ^{a,x}
C. vulgaris	18.65 ± 0.71 ^{a,x}	19.20 ± 0.80 ^{a,x}	18.05 ± 1.39 ^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 8: Mean corpuscular Hemoglobine concentration levels of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	34.08 ± 0.74 ^{a,x}	33.58 ± 0.99 ^{a,x}	34.05 ± 0.29 ^{a,x}
Hiperglicemia control	33.40 ± 0.63 ^{a,x}	34.10 ± 0.91^{a,x}	33.28 ± 0.90 ^{a,x}
Metformin	33.80 ± 0.59 ^{a,x}	32.30 ± 3.01 ^{a,x}	33.40 ± 0.42 ^{a,x}
A. maxima	33.45 ± 0.99 ^{a,x}	34.00 ± 0.50 ^{a,x}	34.15 ± 1.05 ^{a,x}
C. vulgaris	33.43 ± 0.86 ^{a,x}	33.78 ± 0.78 ^{a,x}	32.90 ± 1.01 ^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 9: *Mean Corpuscular Volume* Levels of Wistar Rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	55.40 ± 1.49 ^{a,x}	56.09 ± 1.28 ^{a,x}	55.80 ± 0.97 ^{a,x}
Hiperglicemia control	54.93 ± 1.70 ^{a,x}	56.20 ± 1.96 ^{a,x}	54.98 ± 1.98 ^{a,x}
Metformin	56.05 ± 2.10 ^{a,x}	57.00 ± 2.45 ^{a,x}	56.93 ± 1.56 ^{a,x}
A. maxima	55.78 ± 1.76 ^{a,x}	56.84 ± 2.16 ^{a,x}	56.45 ± 2.25 ^{a,x}
C. vulgaris	55.68 ± 0.80 ^{a,x}	56.83 ± 1.88 ^{a,x}	54.88 ± 2.59 ^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

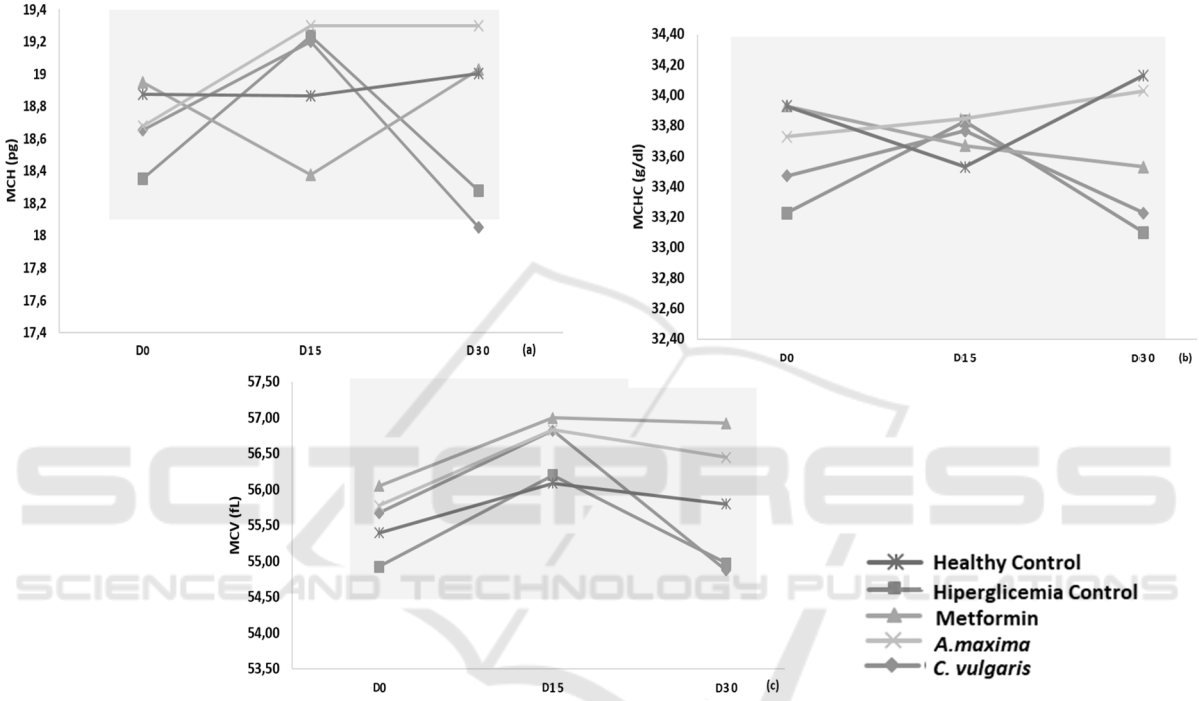

Figure 3: MCH (a), MCHC (b), and MCV levels (c) Wistar rats (*Rattus norvegicus*, Berkenhout 1769) hyperglycemia with the treatment of *Chlorella vulgaris* and *Arthrospira maxima* for 30 days. Note: the blue box in Figure 3 shows the normal region of every level.

Based on the results of the data (Syariatin & Mulyati, 2017) the normal range of MCH values is between 18.1-20.20 pg. All groups experienced fluctuations and were still in the normal range. Fig.3 shows that fluctuations occur but are still in the normal range ($p>0.05$). In the hyperglycemia group MCH values increased before D15 and decreased after D15. In the metformin group, MCH decreased before D15, due to the induction of hyperglycemia and increased after D15 which showed that metformin could control the effects of hyperglycemia thereby increasing hemoglobin levels. Indicated by the value of MCH on D30, where the value of MCH *A. maxima* has more potential to increase the value of MCH in the blood.

Based on research (Syariatin & Mulyati, 2017) the normal MCHC value ranges between 31.22-36.0%. Figure 4b shows that all groups are still in the normal range. In the DM control group, there was an increase in the value of MCHC before D15 for 25 days of STZ re-injection, this increase was possible because the mice experienced still being able to do erythropoetin production and again decreased after D15 because the rats became more chronic. In the treatment of microalgae, the results shown in graph *A. Maxima* show results that are more efficient than *C. vulgaris*. This can be seen in *A. maxima* increasing continuously from H0 to H30 and at H30 approaching the MCHC value in the healthy control group.

Based on these data the normal range of MCV values is obtained between 53.27-58.5 fL. At the D0 the MCV values of all groups increased and decreased after D15. Increases and decreases in MCV values are still in the normal range. This shows that there is no significant effect on changes in the value of MCV after microalgae treatment. Based on Figure 3 and Table 9 group *A. maxima* shows a graph that is close to the value of the metformin treatment group. This shows that A. maxima is far more potential to increase the value of MCV than C. vulgaris. Based on these results, based on the value of MCH, MCHC, and MCV are positively correlated. *A. maxima* has more potential to improve hematological parameters and has higher Fe levels than *C. vulgaris*.

Table 10: Platelets total levels of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	947.5 ± 128.26 ^{a,x}	952 ± 102,94 ^{a,x}	913.5 ± 116.866 ^{a,x}
Hiperglicemia control	874.25 ± 158.95 ^{a,x}	1031.75 ± 72.43 ^{a,x}	1035.75 ± 150.59 ^{a,x}
Metformin	812.75 ± 137.70 ^{a,x}	1207.25 ± 525.38 ^{a,x}	906.25 ± 102.48 ^{a,x}
A. maxima	804.25 ± 154.50 ^{a,x}	991 ± 100.191 ^{a,x}	878.5 ± 170.814 ^{a,x}
C. vulgaris	924 ± 92.34 ^{a,x}	1016.25 ± 86.55 ^{a,x}	849 ± 166.163 ^{a,x}

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 11: Platelet Distribution Weidth (PDW) levels of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	7.32 ± 0.53 $^{a.x}$	7.27 ± 0.51 $^{a.x}$	7.35 ± 0.57 $^{a.x}$
Hiperglicemia control	7.62 ± 0.43 $^{a.b,c,x}$	7.52 ± 0.50 $^{a.x}$	7.90 ± 0.92 $^{a.x}$
Metformin	8.03 ± 0.29 $^{a.b,x}$	8.28 ± 0.57 ^{b,x}	7.825 ± 0.35 $^{a.x}$
A. maxima	8.075 ± 0.39 $^{c.x}$	7.73 ± 0.49 $^{a.b,x}$	7.525 ± 0.40 $^{a.x}$
C. vulgaris	7.43 ± 0.25 $^{a.b,x}$	7.30 ± 0.14 $^{a.x}$	7.825 ± 0.09 $^{a.y}$

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 12: Mean Platelet Volume (MPV) levels of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	6.5 ± 0.24 $^{a.x}$	6.52 ± 0.22 $^{a.x}$	6.62 ± 0.41 $^{a.x}$
Hiperglicemia control	6.7 ± 0.22 $^{a.b,x}$	6.62 ± 0.32 $^{a.x}$	6.93 ± 0.57 $^{a.x}$
Metformin	7.07 ± 0.28 ^{b,c,x}	7.05 ± 0.34 ^{b,x}	6.83 ± 0.26 $^{a.x}$
A. maxima	7.47 ± 0.49 $^{c.y}$	6.8 ± 0.25 $^{a.b,x}$	6.68 ± 0.22 $^{a.x}$
C. vulgaris	6.65 ± 0.17 $^{a.b,x,y}$	6.55 ± 0.13 $^{a.x}$	6.85 ± 0.13 $^{a.y}$

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 13: PLCR Levels of Wistar Rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	4.25 ± 0.49 $^{a.x}$	4.57 ± 0.97 $^{a.x}$	4.85 ± 1.55 $^{a.x}$
Hiperglicemia control	5.23 ± 0.58 $^{a.x}$	4.67 ± 1.12 $^{a.x}$	5.82 ± 2.59 $^{a.x}$
Metformin	6.97 ± 1.03 $^{b.x}$	6.95 ± 1.57 $^{b.x}$	5.55 ± 1.34 $^{a.x}$
A. maxima	7.35 ± 1.96 $^{b.x}$	5.55 ± 1.05 $^{a.b.x}$	5.25 ± 0.78 $^{a.x}$
C. vulgaris	4.90 ± 1.02 $^{a.x}$	4.5 ± 0.68 $^{a.x}$	5.67 ± 0.54 $^{a.x}$

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

Table 14: Plateletcrit (PCT) levels of Wistar rats (*Rattus norvegicus* Berkenhout, 1769) hyperglycemia treated with *A. maxima* and *C. vulgaris* for 30 days.

Groups	Days		
	0	15	30
Healthy control	0.50 ± 0.50 $^{a.x}$	0.62 ± 0.05 $^{a.x}$	0.60 ± 0.05 $^{a.x}$
Hiperglicemia control	0.59 ± 0.59 $^{a.x}$	0.68 ± 0.09 $^{a.x}$	0.71 ± 0.07 $^{a.x}$
Metformin	0.58 ± 0.58 $^{b.x}$	0.85 ± 0.36 ^{a,x}	0.62 ± 0.09 $^{a.x}$
A. maxima	0.57 ± 0.57 $^{b.x}$	0.67 ± 0.03 $^{a.x}$	0.61 ± 0.10 $^{a.x}$
C. vulgaris	0.46 ± 0.27 $^{a.x}$	0.67 ± 0.06 $^{a.x}$	0.58 ± 0.12 $^{a.x}$

Notation a, b, c to compare between treatment groups on the same day (column), while x, y, z to compare between days on the same treatment group (row).

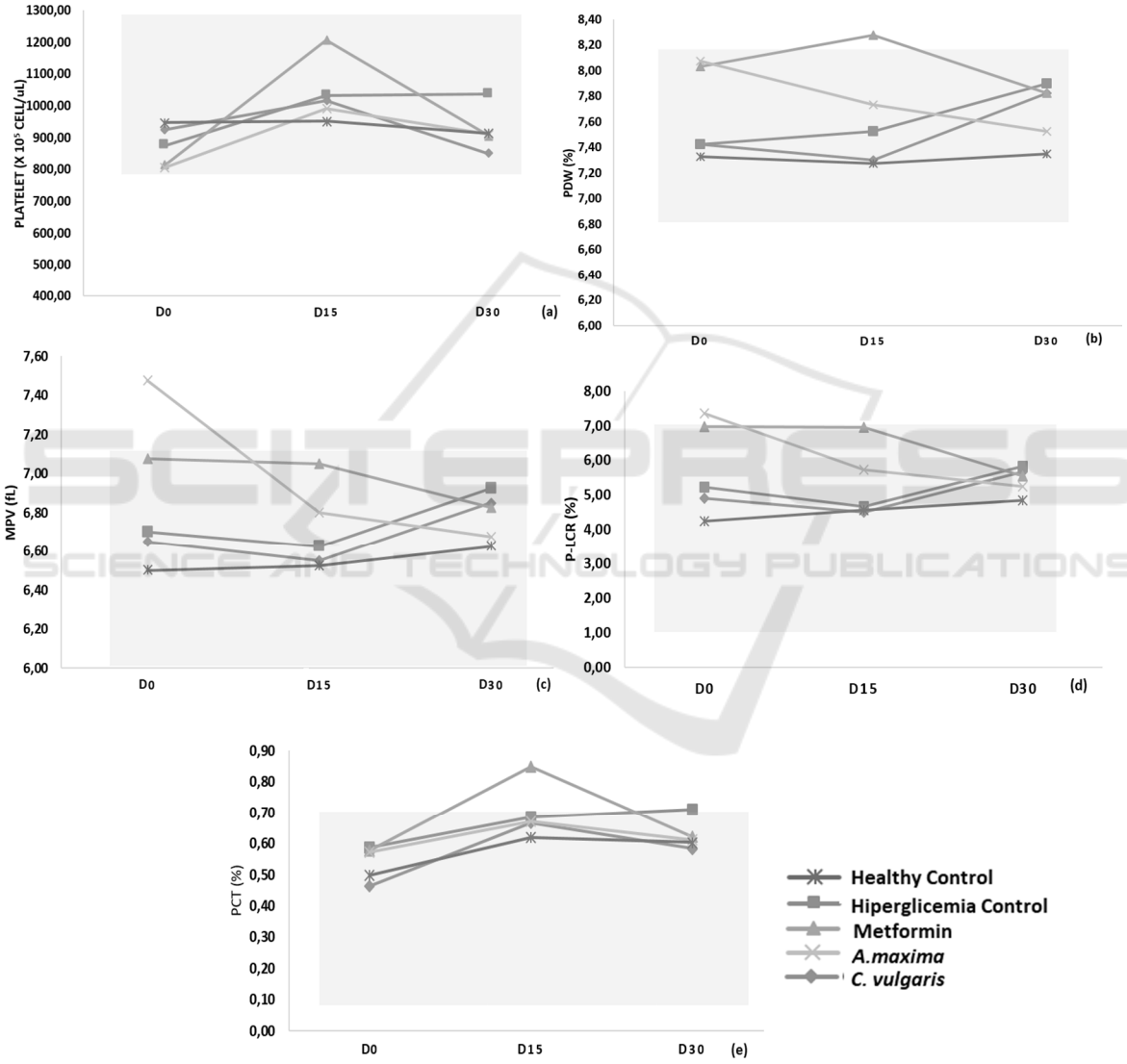

Figure 4: PLT (a), PDW (b), MPV (c), P-LCR (d), and PCT (e) Wistar rats (*Rattus norvegicus*, Berkenhout 1769) hyperglycemia with the treatment of *Chlorella vulgaris* and *Arthrospira maxima* for 30 days. Note: the blue box in Figure 4 shows the normal region of every level.

Based on these data, the normal range of PLT values can be obtained between 814-1399 x 105 cells / µL. Based on the results of this study indicate that platelet values are still in the normal range. The figure

4a shows that the hyperglycemia group has platelets that have increased continuously from D0 to D30. Moreover, it can show that the immune condition of diabetic-induced rat can still respond well. The metformin treatment showed that at D0 to D15, platelets experienced an insignificant increase. In the treatment control given microalgae, it is known that the administration of *A. maxima* and *C. vugaris* both increased in D0 to D15. After D15, the platelets both experienced a decrease, this suggests that *A. maxima* and *C. vulgaris* have the potential to reduce levels of platelet induced hyperglycemia rats.

Based on Figure 4b this study obtained a baseline ranging between 5.18-9.17%. PDW values under normal conditions indicate that platelets contained in the blood generally have the same size. While the high PDW value indicates that the platelet has a large size variation, there are disorders that affect the platelet. Based on Figure 4b it is shown that in hypergicemia control on days D0 to D30 have increased ($p>0.05$) constantly. This shows that there are variations in the form of platelets which indicate interference with the platelet. In the group of mice that received microalgae treatment and antidiabetic drugs experienced insignificant differences ($p>0.05$).

After D55, the normal range between 5.4-6.7 fL was obtained. In this study, the normal range of MPV values was obtained between 6.1-7.1 fL. In the DM group, it was shown that after D15 the MPV experienced an increase. This shows that large platelets are changing. The metformin group showed a decrease after D15. Likewise in group *A. maxima*. The *A.maxima* group had results at D30 approaching healthy control values. Group *A. maxima* is far more effective at reducing MPV values.

Normal range of P-LCR values was obtained between 2.9-6.7%. The DM control group shows that based on the difference in days, the graph shows a decrease before D15 and an increase after D15 ($p>0.05$). The metformin group decreased after D15. This shows that metformin has the ability to restore the platelet condition of hyperglycemia induced mice. In group *A. maxima* tends to have a graphic pattern that is almost the same as the metformin group. *A. maxima* group experienced a decrease from D0 to D30. An increase in P-LCR can indicate increased platelet activity due to viral marrow suppression, platelet destruction that is mediated by antigens, antibodies consumption of peripheral platelets or viral replication in platelets which then causes thrombocytopenia.

At the beginning of this study, after D55 was obtained the normal range value in the healthy control group of rat between 0.16-0.68%. In the Figures and Tables it is shown that in the hyperglycemia group the rat experienced a non-significant increase from D0 to D30 and at D30 the PCT value was higher among the other groups. In the metformin group the mice experienced an increase before D15 and decreased toward the healthy control group after D15. In groups *A. maxima* and *C. vulgaris* experienced a slight increase after D15 approached the results of metformin at D30.

4 CONCLUSIONS

This research conclude that erythrocyte profiles in hyperglycemic rats have increased, platelet profile have decreased, and returned to better conditions after being given *A. maxima* and *C. vulgaris.*

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