Side effect of long term injection of high dose whitening vitamin C to plasma glucose and cholesterol level

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Abstract. The use of whitening vitamin C to make the skin brighter in the long term, however can cause side effects. The purpose of this study was to investigate the side effects of long-term high dose injections of whitening vitamin C plasma glucose and cholesterol level. This study used rats (Rattus norvegicus L.) as experimental animals. The rats randomly divided into five groups consisting of ten rats each group. Four groups were administered injections of white vitamin C and one group as control animal. The design used in this study was a complete randomized design (RAL) and the treatments were different long term of injections of high-dose whitening vitamin C, ie 30-day injections (P1), 50-day injections (P2), 70-day injections (P3), 90 days of injections (P4) and control (P0). The treatment was repeated ten times. The parameters observed were glucose and cholesterol rats plasma levels before and after high-dose whitening vitamin C injection treatment. Data was analysed by ANOVA allowed by Duncan’s Test. The results indicated that rats glucose plasma levels injected by high dose of whitening vitamin C in long-term between control and treatment showed significant differences (P = 0.005) and cholesterol plasma levels showed significant different too (P = 0.000). Glucose and cholesterol plasma levels of rats injected by high doses of whitening vitamin C in long term tend to decrease.

1. Introduction

Benefits of vitamin C to improve immune system is no doubt. High antioxidants of vitamin C can fight free radicals in the body, both coming from outside the body as well as the metabolic outcomes of our own cells. While the benefits of whitening vitamin C to tighten and brighten as result as vitamin C stimulates the formation of collagen, an extracellular protein that plays a role in toning the cells. Vitamin C inhibits the action of tyrosinase enzymes that play in inhibiting the formation of skin pigment, so that the skin becomes brighter and firmer [1, 2]. Nowadays to get skin bright and smooth by injection of whitening vitamin C has been offered by both dermatologist and beauty practitioners. The price offered for a single treatment ranges between IDR 100,000 to 300,000 for once injection. The dose of whitening vitamin C is given for once intravenous injection about 1000-4000 mg. While the recommended dose of vitamin C to maintain body health about 50-75 mg / day [3].

Vitamin C is a water-soluble vitamin, meaning that the excess of this vitamin cannot be stored by the body such as vitamin A, D, E, K that fat soluble, so that if the vitamin C are excess in the body, it will be removed from the body through the kidneys. Therefore it is advisable to consume vitamin C every day as needed [4]. Thus, the dose of whitening vitamin C given by injection is very high compared to the normal dose required. It will make the body work harder to elminate the excess of vitamin from the body [5]. Sudatri et al [6] reported that high-dose whitening vitamin C injections had...
a significant effect on the liver disfunction of rats injected by high doses of vitamin C over the long term. Research on the effects of high doses of vitamin C on the female rats reproductive system causes the length of the rat estrous cycle to be longer [6]. Injection of high doses whitening vitamin C in long-term caused increase of SGPT-SGOT level and plasma creatinine levels in mice [7].

Maintaining normal glucose levels in the blood is very important to keep the body working and functioning well. Glucose is needed by cells as fuel to generate energy. When the body is deprived of glucose, the body will not be able to work optimally, which is characterized by a state of weakness, pallor, difficulty concentrating, and so forth. High glucose levels continuously in the blood, it will also cause health problems such as diabetes, high blood pressure, kidney problems and so on. Fasting glucose levels less than 8 hours in humans is normally between 70 and 100 mg / dl. This glucose level can change after eating, sleeping or in a state of fasting. Blood glucose levels are also strongly influenced by food intake, an example of anti diabetic substances able to lower blood glucose levels, for example the leaves of breadfruit [8]. Parisa and Siamak [9] reported that consumption of vitamin C (ascorbic acid) in the long term lead to treatments rats experiencing hyperglycemia (diabetes) and also increased loss of body weight.

As well as glucose, cholesterol levels in the blood are also very important. Cholesterol is needed by the body for cell wall components and as a precursor for steroid hormones. However, high cholesterol levels in the body are intimately associated with high blood pressure, narrowing of the arteries, coronary heart disease, stroke and others. Blood cholesterol levels are strongly influenced by the diets [10]. Diets that contain trans fat, fried, organ meats will generally increase blood cholesterol levels. Diets that contain a lot of fibre and diets that contain acid will lower cholesterol levels in blood.

2. Materials and Methods

The rats randomly divided into five groups consisting of ten rats. Four groups were administered injections of whitening vitamin C and one group as control animal. The design used in this study was a complete randomized design (RAL) and the treatment were different long term of injections of high-dose white vitamin C, i.e 30-day injections (P1), 50-day injections (P2), 70-day injections (P3), 90 days of injections (P4) and control (P0). The treatment was repeated ten times.

2.1. Materials research
The material used in this study is high-dose white vitamin C (4000 mg / one injection) for humans. The experimental animals used were adult male rats aged 3-4 months with weight between 150-200 grams. The dosage used is converted from the dose used in humans to rats. The weight of an injected adult woman is estimated to be approximately 70 kg, so the dose of vitamin C given to the rats is 28 mg / once injection / rats.

2.2. Research procedure
A total of 50 male rats aged 3-4 months were measured body weight initially, testing for early glucose and cholesterol plasma level. given standard formula diet and drinking water in ad libitum and acclimatized for 4 weeks, then the test animals were divided randomly into 5 groups: P0 = control, injection of vitamin C; P1 = group of animals receiving vitamin C injections for 30 days; P2 = group of animals receiving vitamin C injections for 50 days; P3 = group of animals receiving vitamin C injections for 70 days; P4 = group of animals who received vitamin C injection for 90 days. Vitamin C injection is done intramuscularly with a 1 ml syringe. Injection is done every 2 days according to duration of treatment. The day after treatment in each group were tested for final glucose and cholesterol plasma levels.

2.2.1. Testing glucose and cholesterol levels
Blood glucose and cholesterol levels of the rats were tested with a Nesco multiple check tool. Blood glucose and cholesterol levels of rats tested were the blood glucose and cholesterol levels before the injection treatment was administered, whereas previously the rats were fasted (not fed) for approximately 8 hours. After that, blood is taken from the vein of the tail, by cutting the tip of the tail with a sterile scissors that has been smeared with alcohol, the blood coming out from the tail end is
then dropped on the sticks for testing, for further reading by the Nesco multiple check tool. The wound that arises after the cut tail is treated by betadine. Further testing is done every two weeks after treatment until the end of treatment.

2.3. Data analysis
The quantitative data obtained is statistically analysed using SPSS software and if there is any real or very real effect allowed by Duncan test. And if the data is not normally, then tested by Kruskal Wallis Test.

3. Results and Discussion

3.1. Result

3.1.1. Initial Blood Glucose Levels and Blood Glucose Final

Table 1. ANOVA assay and standard error of initial and final glucose levels of male rats injected with high doses of whitening vitamin C followed by Duncan test

| No | Treatment  | Average of initial blood glucose level (mg/dl) | Average of final blood glucose level (mg/dl) |
|----|------------|-----------------------------------------------|---------------------------------------------|
| 1  | K (Control)| 102.2 ± 8.66 a                                | 102.6 ± 5.76 a                               |
| 2  | P1 (30days)| 103.0 ± 9.93 a                                | 93.0 ± 5.54 a                                |
| 3  | P2 (50 days)| 102.2 ± 8.66 a                                | 79.8 ± 4.58 b c                             |
| 4  | P3 (70 days)| 103.0 ± 9.93 a                                | 74.0 ± 6.75 c                                |
| 5  | P4 (90 days)| 102.2 ± 8.66 a                                | 72.6 ± 5.59 c                                |

Based on blood glucose level analysed by variance analysis showed no significant difference (P = 1.00), whereas the final blood glucose level showed a significant difference (P = 0.005). There were significant differences in blood glucose levels between control rats and rats injected by high-dose whitening vitamin C for 70 days (P3) and rats injected with high-dose whitening vitamin C for 90 days (P4). There is a tendency to decrease blood plasma glucose levels after receiving high doses of whitening vitamin C in the long term.

Figure 1. Initial blood glucose and final blood glucose of male rats between controls and mice injected by high doses of whitening vitamin C in long term

3.1.2. Initial Blood Cholesterol Levels and Final Blood Cholesterol Levels
Table 2. ANOVA assay and standard error of initial and final cholesterol levels of male rats injected with high doses of whitening vitamin C followed by Duncan test

| No | Treatment        | Average Initial blood cholesterol level (mg/dl) | Average final blood cholesterol level (mg/dl) |
|----|------------------|-----------------------------------------------|---------------------------------------------|
| 1  | K (Control)      | 187.2 ± 17.89 a                              | 222.8 ± 16.92 a                             |
| 2  | P1 (30 days)     | 144.2 ± 13.26 a                              | 183.8 ± 13.69 b                             |
| 3  | P2 (50 days)     | 153.4 ± 13.65 a                              | 118.4 ± 9.08 c                              |
| 4  | P3 (70 days)     | 191.2 ± 11.05 a                              | 182.0 ± 9.03 b                              |
| 5  | P4 (90 days)     | 187.2 ± 17.89 a                              | 165.8 ± 5.23 b                              |

Initial blood cholesterol levels analysed by variance analysis showed no significant difference (P = 0.11), whereas analysis of final blood cholesterol level analysed by variance analysis showed significant difference (P = 0.000). Between control rats and all treatment rats experienced different levels of blood cholesterol. After a high dose of whitening vitamin C injection, rats blood cholesterol levels tend to decrease.

Figure 2. Initial blood cholesterol and final blood cholesterol of male rats between controls and mice injected by high doses of whitening vitamin C in long term

3.2. Discussion

Glucose levels in the blood are very volatile. Many factors that affect blood glucose levels such as; post absorption, an imbalance of hormones, genetic disorders, and wrong diet. Blood glucose levels depend on hormones released by the adrenal glands, namely adrenaline and corticosteroids. Adrenaline will spur a rise in blood glucose, whereas corticosteroids will lower it. Adrenaline that is driven continuously will result in insulin can not regulate the ideal blood glucose levels so that blood glucose levels increase. In addition, drugs or foods containing antidiabetic substances can also lower blood glucose levels such as teak leaf extract and temulawak extract [11, ginger extract [12]. Glucose levels measured are usually fasting blood glucose levels.

A decrease in blood glucose levels in rats that received an injection of high doses of whitening vitamin C may be caused by the content of ascorbic acid contained in the high-dose whitening vitamin C. Ascorbic acid is the possibility of lowering the absorption of glucose in the small intestine so that the levels of glucose in the blood plasma was decreased. A similar opinion was expressed by Zubaida & Rosdiana [13], that the apple cider vinegar and vinegar-containing phenol compound lowers blood
glucose levels. Substances that are acidic lowers blood glucose levels by reducing the activity of glucose transporter both sodium-glucose linked transporter protein 1 (SGLT 1) and glucose transporter 2 (GLUT 2). This condition causes less glucose into the bloodstream. SGLT 1 and GLUT 2 act as active transport that facilitate the absorption of glucose in the intestine. In addition, high-dose vitamin C intake causes nausea, diarrhea, heartburn and stomach cramps, hyperoxaluria, a serious health disorder when oxalic acid is excreted in the urine too much, increasing the risk of kidney stones.

The treatment of high doses of whitening vitamin C in the treated rats also decreasing total cholesterol level in blood. This decrease in cholesterol level is likely due to high level of ascorbic acid in blood of rat that are injected with high doses of whitening vitamin C in the long-term. Ascorbic acid (vitamin C) found in star fruit plays a role in lowering blood cholesterol even in small amounts. The fruit helps the process of transformation of cholesterol into bile acids in the liver, which eventually released with faeces [14]. The ascorbic acid contained in whitening vitamin C decreases cholesterol levels, through the binding mechanism of bile salt and cholesterol in the intestine, increasing the release of bile in faeces [15]. The cholesterol eventually bound and removed with faeces, it prevents the building up of cholesterol in the blood stream and blood.

4. Conclusion
Glucose levels of male rats injected with high doses of whitening vitamin C in the long-term experienced a marked decrease (P = 0.005) and cholesterol level of male rats (Rattus norvegicus L.) injected with high doses of vitamin C in the long-term tends to decrease markedly (P = 0.000).

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