

Developing animal model

by Ardiana Ekawanti

Submission date: 09-Apr-2023 05:48PM (UTC-0500)

Submission ID: 2059846966

File name: Development_of_Animal_Model_for_turnitin.docx (21.16K)

Word count: 1374

Character count: 7416

Development of Animal Model for The Study of Gout and Dyslipidemia by Diet Modification

*Ardiana Ekawanti¹, Nurhidayati¹, Novrita Padauleng¹, Niti Wedayani¹

³ ¹ Medical Faculty Mataram University, Jl. Pendidikan No. 37 Mataram, Indonesia
*Corresponding author: HP.081907070322/email: ekawantimuhaimin@gmail.com

Abstract

Degenerative diseases is tend to be rise nowadays. Hyperuricemia and dyslipidemia are condition that play an important role in pathogenesis of several degenerative diseases e.g. cardiovascular diseases such as hypertension, infarct myocard, etc. Since ethical barrier is critical issue in studying the mechanism of these disease in real patient, so that, animal laboratory model is required. This research objective was to find out the nutrition formula, route and duration of diet in order to develop animal model for hyperuricemia and hypercholesterol condition. Research method was experimental laboratory research which was used post test only control group design. Male *Rattus norvegicus* star strain were divided into four groups: group (A) group fed by standard animal diet, group (B) group was given high cholesterol and high purin diet in duration of 14 days, group (C) group was given high cholesterol and high purin diet in duration of 21 days group (D) group was given high cholesterol and high purin diet in duration of 30 days given propyl thiouracil for the hypercholesterol group. The result were; cholesterol level of group A was 183±21.5 mg/dL, group B was 176.5±39.32 mg/dL, group C was 183.7±25.9, group D was 242.17±40.2 mg/dL. Uric acid serum level for group A was 2.1±0.1 mg/dL, group B was 3.95±0.8 mg/dL, group C 3.35±0.3 mg/dL and group D was 8.3±3.4 mg/dL. Hypothesis was examined by using Kruskal Wallis test showed that there was a differences among groups significantly with p value 0.000. It was concluded that induction of hypercholesterol and hyperuricemia condition by high cholesterol and high purin diet modification was effective after 21 days of fed by oral route.

Kata kunci: hypercholesterol, hyperuricemia, induction, diet, animal model.

Introduction

INTRODUCTION

Hyperuricemia is a metabolic disorder characterized by elevated uric acid levels. Uric acid is the end product of purine metabolism. Normal levels of uric acid in humans range from 7 mg / dL in men and 6 mg / dL in women. This condition can be caused by increased xanthine oxidase activity so that increased uric acid production, decreased uric acid excretion through the kidneys (Edwards, 2009), or increased purine intake,

The prevalence of hyperuricemia is rapidly increasing worldwide in recent decades (Edwards, 2009). NHANES survey results in the United States in 2007-2008, the prevalence of men who have uric acid levels 6.14 mg / dl reached 21.1%, 5.9% of whom suffer from gout, whereas in women, the prevalence reached 4.87 % percent and 2% of the population experienced gout (Davide, et al, 2013). Hyperuricemia has several

manifestations, ranging from asymptomatic, inflammation of the joints called gout arthritis and has also been shown to have a negative impact on cardiovascular events, such as atherosclerosis, hypertension, coronary heart disease and kidney disorders including renal failure, and also associated with metabolic syndrome (Edwards, 2009)

The condition of hyperuricemia and abnormalities in the aorta induced by diets high in cholesterol and purine.

There was an ethical barrier to study the mechanism of the diseases directly in human, so that developed animal model is needed. Animal model of hypercholesterol condition and hyperuricemia using local food stuff did not conducted yet. Therefore, the aim of this research was to find out the effective composition, route and duration of diet inducing hypercholesterol and hyperuricemia.

MATERIAL AND METHOD

Method

This research is an experimental research with complete randomized design (post test only design). This study used *Rattus norvegicus* experimental animals induced with a high cholesterol and high purine diet and in combination with oral propylthiouracil (PTU) with a dose of 200 mg/kgBW rat. Male *Rattus norvegicus* wistar strain were divided into four groups: group (A) group fed by standard animal diet, group (B) group was given high cholesterol and high purin diet in duration of 14 days, group (C) group was given high cholesterol and high purin diet in duration of 21 days group (D) group was given high cholesterol and high purin diet in duration of 30 days or given propyl thiouracil for the hypercholesterol group. At the end of treatment, blood sample were taken from the tail and assessed for cholesterol and uric acid serum.

The independent variables were diet and duration of treatment, while dependent variables were value of cholesterol and uric acid level. The formula for calculating large experimental samples from Federer (David, 2008) and minimal samples needed were six rats.

Induction of Hypercholesterol and Hyperuricemia

Animal model of hyperuricemia is male white rats Wistar strain which is stated hyperuricemia after induced rats that are high purine. High purine diet as well as high cholesterol consisted of 1000 gr standard feed mixed with 50 of quail egg yolk, 250 gr of chicken liver, 250 gr flour of *Gnatum gnemon* and 250 gr sardines and were formed as pellets. Diet administered for the longest duration 30 days orally. For the group D, on 21th day, given 200 mg/kgBW of propylthiouracil by the route of oral injection.

Blood Investigation

Examination of Uric Acid and Cholesterol Level

The level of uric acid and level of cholesterol measured from capillary blood sample . The method used for this measurement was dipstick examination. At the end of treatment or each groups, serum uric acid and serum cholesterol levels were measured from capillary blood samples (animal tail blood sample). Animals were stated as hyperuricemia if uric acid levels > 3 mg/dL, and hypercholesterolemia if cholesterol level was more than 200 mg/dL.

RESULT

Mean of uric acid and cholesterol of each groups were shown in table 1.

Table 1. Cholesterol and Uric Acid Levels

Parameter	Group			
	A	B	C	D
Asam urat serum (mg/dL)				
• Mean±SD	2.08±0.13	3.95±0.79	3.35±0.29	8.33±3.37
• Median	2.00±0.13	3.65±0.79	3.30±0.29	9.30±3.37
• Minimum	2.0	3.3	3.0	3.7
• Maximum	2.3	5.1	3.9	12.8
Cholesterol (mg/dL)				
• Mean±SD	183±21.55	176.5±39.32	183.67±25.94	242.17±40.17
• Median	184.5±21.55	186±39.32	181.00±25.94	248.50±40.17
• Minimum	153	101	151	181
• Maximum	210	213	229	291

Cholesterol level showed that all groups were within normal value, except for group D had hypercholesterol condition that cholesterol level >200 mg/dL (mean 242.17±40.17 mg/dL), and the lowest value was found in group B (176.5±39.32 mg/dL).

Uric acid level were increased in group B, C and D, while group A had normal uric acid level. The lowest value was in group A (2.08±0.13 mg/dL) and the highest value was in group D (8.33±3.37 mg/dL).

Since the data were not normal in distribution so that hypothesis was tested by using Kruskal Wallis test. Uric acid level were significantly different among group ($p = 0.000$) and cholesterol level were not different.

DISCUSSION

Developing animal model using endemic food very important in studying disease mechanism, but the obstacle in providing chemical used in inducing the specific condition, for example potassium oxonate.

Diet component used in this study were Gnatum gnemon flour contain abundant in protein (19.0g/100g), adequate amounts of essential amino acids, fatty acids and minerals. While quail

egg nutrients composition was Crude Protein : $13.30 \pm 0.08\%$ of 100 gr of eggs and 1993 kcal. Liver of chicken contain 1500 kcal of energy and 25 gr/100 gr of protein. Food stuff rich in protein also rich in purin. High calorie diet provided precursor of cholesterol production.

This diet modification could increased lipid and uric acid value of rat within 3 weeks, while Liu (2016) has found out that increased of fat (314 mg/dL) and UA (3 mmol/L) found in 8th weeks after giving high lipid and adenine diet combined with potassium oxonate .

From the result presented that cholesterol level in duration of 14th day was decreased, it may caused by lower consumed of pellet, but the limitation of this research that the amount of pellet consumed did not measured. The diet combination used in this study has potency in increasing uric acid but not cholesterol level.

CONCLUSION

The diet combination of quail egg, Gnatum gnemon flour, liver of chicken and sardines which given in 30 days has potency to generate hyperuricemia but not hypercholesterolemia.

ACKNOWLEDGMENT

We acknowledge Kemenristekdikti that had funded this study and also thank to laboratorium Hepatika for facilitating this research in assessing uric acid and cholesterol value.

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