Nigella sativa Infusion as an Antioxidant Agent Against Gentamicin-Induced Kidney Damaged in Mice

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Abstract

Background: Gentamicin is one of the most common antibiotics related to nephrotoxicity. It has been proposed that the nephrotoxicity is associated with the generation of the reactive oxygen species. Thymoquinone, an active compound of Nigella sativa, shows to have an antioxidant property. The study aims to identify the possible nephroprotective action of Nigella sativa infusion against gentamicin-induced kidney damaged in mice.

Methods: This experimental study was carried out in the Department of Cell Biology Laboratory, Universitas Padjadjaran, Bandung from 10th November 2012 to 14th December 2012. There were four groups, each consisting of 6 mice. Group I (control negative), group II (gentamicin 100 mg/kg), group III (3.9 mg Nigella sativa infusion+gentamicin 100mg/kg) and group IV (7.8 mg Nigella sativa infusion+gentamicin 100mg/kg). The kidneys were evaluated histopathologically by light microscope. The percentage average number of normal proximal tubules in group I and the percentage average number of proximal tubules damaged in group II, III and IV were measured.

Results: The results showed the percentage average number of the proximal tubules damaged in group II, III and IV were 14.53%, 7.49% and 3.94% respectively. Significant differences were observed between group II and III, group II and IV, and group III and IV.

Conclusion: Nigella sativa infusion protects against gentamicin-induced kidney damage in mice. [AMJ.2014;1(2):90–3]

Keywords: Gentamicin, kidney, Nigella sativa infusion

Infusa *Nigella sativa* sebagai Antioksidan Terhadap Kerusakan Ginjal pada Tikus yang Diinduksi Gentamisin

Abstrak

Latar Belakang: Gentamisin adalah salah satu antibiotik yang paling sering dihubungkan dengan nefrotoksisitas. Nefrotoksisitas yang terjadi berhubungan dengan pembentukan reactive oxygen species. Thymoquinone, senyawa aktif *Nigella sativa*, telah terbukti memiliki sifat antioksidan. Penelitian ini bertujuan untuk mengidentifikasi sifat nefroprotektif infusa *Nigella sativa* terhadap kerusakan ginjal pada tikus yang diinduksi gentamisin.

Metode: Penelitian eksperimental ini dilakukan di Laboratorium Departemen Biologi Sel, Fakultas Kedokteran, Universitas Padjadjaran, Bandung dari 10 November 2012 –14 Desember 2012. Ada empat kelompok masing-masing terdiri atas 6 tikus. Kelompok I (control negatif), kelompok II (gentamisin 100 mg /kg), kelompok III (3,9 mg infusa *Nigella sativa*+gentamisin 100mg/kg) dan kelompok IV (7,8 mg infusa *Nigella sativa*+gentamisin 100mg/kg). Ginjal kemudian dievaluasi secara histopatologi dengan mikroskop cahaya. Persentase rata-rata jumlah tubulus proksimal yang normal dalam kelompok I dan persentase rata-rata jumlah tubulus proksimal rusak dalam kelompok II, III dan IV diukur.

Hasil: Persentase rata-rata jumlah tubulus proksimal rusak dalam kelompok II, III dan IV adalah 14,53%, 7,49% dan 3,94%. Perbedaan signifikan yang diamati adalah pada kelompok II dengan III, kelompok II dengan IV, serta kelompok III dan IV.

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Simpulan: Infusa Nigella sativa melindungi terhadap kerusakan ginjal yang diinduksi gentamisin pada tikus. [AMJ.2014;1(2):90–3]

Kata kunci: Gentamisin, ginjal, infusa *Nigella sativa*

Introduction

Gentamicin antibiotic is widely used to treat life-threatening infections caused by negative bacteria. However, it can cause nephrotoxicity. Oxidative stress and nitrosative stress have been reported to contribute the nephrotoxicity. As a result, its clinical use is limited.1Nephrotoxicity induced by gentamicin is an essential cause of renal failure.[2] Approximately 10–15% of all cases of acute renal failure are due to the gentamicin nephrotoxicity.[3] Metabolite products of the gentamicin are excreted through kidney. It may cause damage to the renal cells and resulting in renal dysfunction.[4]

The nephrotoxicity is due to the accumulation of the drug in the renal cortex. Receptors in the proximal tubule cells are able to transport the drug into the cells through endocytosis. Accumulation of the gentamicin in the cells disrupt cellular functions that results in apoptosis and necrosis of the proximal tubule. Ultimately, acute kidney injury occurs.[5] Direct necrosis of the tubules are the characterization of the gentamicin-induced nephrotoxicity, which occur mainly in proximal tubules.[6]

Nigella sativa is one of the promising medicinal plants with many historical and religious backgrounds for curing several diseases.[7]The principle active compound of Nigella sativa is thymoquinone. Thymoquinone has most prominent activity of antioxidant. It neutralizes oxygen radicals by acting as an anion scavenger.8 This study was conducted to analyze the nephroprotective effect of Nigella sativa infusion against gentamicin-induced kidney damaged in mice.

Methods

This experiment was carried out in Animal Laboratory of Department of Cell Biology in Universitas Padjadjaran, Bandung from November 2012 to December 2012. A bottle of Nigella sativa containing powdered-capsules was purchased in Bandung Indah Plaza (BIP), Bandung. Gentamicin was obtained from the General Hospital Dr. Hasan Sadikin General Hospital Bandung. GalurBalb C strain mice (25–30g each) aged 7–8 weeks, healthy, and white male were used in this study. The mice were housed in homogenous temperature and dark-light cycle for 7 days (adaptation period) with unlimited to drink and food.

In this study, Nigella sativa infusion was made from the Nigella sativa powders. Nigella sativa powders were weighed to 3.9 mg and 7.8 mg.Then the powders were added into a container containing 100 ml of aquadest. The infusion was brought to a boil and simmered for 15 minutes until the temperature reaches 90°c. After the infusion cooled down, it was filtered using filter paper into a beaker.

Four groups of six mice each were used for the study. Group I, served as control negative, received isotonic normal saline throughout the experiment. Group II, served as control positive, received intraperitoneal injection of gentamicin 100 mg/kg/day for 8 days. Group III received gentamicin 100 mg/kg/day (intraperitoneally) for eight days and 3.9 mg of Nigella sativa infusion for ten consecutive days. Group IV received gentamicin 100 mg/kg/day (intraperitoneally) for eight days and 7.8 mg of Nigella sativa infusion for ten consecutive days.

On the 11th day, the mice were sacrificed. The kidneys of the mice were collected for histopathological examinations after sacrifice. The kidneys were fixed in 10% of formalin solution and embedded in paraffin wax. The paraffin wax was cut into 5 micrometer sections and was stained with hematoxylin and eosin. The tissues were then examined under light microscope and the number of normal proximal tubules in group I and the number of proximal tubules damaged in group II, III and IV were measured.

The data were expressed as average and percentage. The significance of differences among the four groups was assessed using Kruskal-Wallis followed by post-hoc Mann Whitney. The p value <0.05 indicated significant differences.

Results

The average number of normal proximal tubules in group control negative was 56.10