BULETIN VETERINER UDAYANA

pISSN 2085-2495

eISSN 2477-2712

Received: 1 May 2026; Accepted: 27 May 2026; Published: 6 June 2026

CATALASE AND GLUTATHIONE PEROXIDASE ENZYME LEVELS IN THE SERUM OF DIABETIC RATS GIVEN A DECOCTION OF JAVANESE GINSENG (*TALINUM PANICULATUM*) LEAF SIMPLICIA

Kadar Enzim Katalase dan Glutation Peroksidase pada Serum Tikus Diabetes yang Diberi Rebusan Simplisia Daun Ginseng Jawa (*Talinum paniculatum*)

Angel Marinda[1]*, I Nyoman Suarsana[2], I Nyoman Suartha[3], I Gusti Ayu Agung Suartini[2], Anak Agung Sagung Kendran[4], I Made Kardena[5]

[1]Bachelors of Veterinary Medicine Student, Faculty of Veterinary Medicine, Udayana University, Jl. Lingkar Timur Unud, Bukit Jimbaran, Badung, Bali, 80361, Indonesia

[2]Veterinary Biochemistry Laboratory, Faculty of Veterinary Medicine, Udayana University, Jl. Lingkar Timur Unud, Bukit Jimbaran, Badung, Bali, 80361, Indonesia

[3]Veterinary Internal Medicine Laboratory, Faculty of Veterinary Medicine, Udayana University, Jl. P.B. Sudirman, Denpasar, Bali 80234, Indonesia

[4]Veterinary Clinical Pathology Laboratory, Faculty of Veterinary Medicine, Udayana University, Jl. P.B. Sudirman, Denpasar, Bali 80234, Indonesia

[5]Veterinary Pathology Laboratory, Faculty of Veterinary Medicine, Udayana University, Jl. P.B. Sudirman, Denpasar, Bali 80234, Indonesia

*Corresponding author email: angelmarindachen2004@gmail.com

How to cite: Marinda A, Suarsana IN, Suartha IN, Suartini IGA, Kendran AAS, Kardena IM. 2026. Catalase and glutathione peroxidase enzyme levels in the serum of diabetic rats given a decoction of Javanese ginseng (*Talinum paniculatum*) leaf simplicia. *Bul. Vet. Udayana.* 18(3): 545-552. DOI: https://doi.org/10.24843/bulvet.2026.v18.i03.p06

Abstract

Diabetes mellitus is a chronic metabolic disease characterized by hyperglycemia and increased oxidative stress caused by excessive Reactive Oxygen Species (ROS). Oxidative stress may affect endogenous antioxidant enzymes, including catalase (CAT) and glutathione peroxidase (GPx). This study aimed to analyze the effect of Java ginseng leaf simplicia decoction (*Talinum paniculatum*) on serum CAT and GPx levels in diabetic rats. Twenty male Sprague-Dawley rats were divided into four groups: normal control (P0), diabetic control (P1), diabetic rats treated with Java ginseng leaf decoction 50 g/100 mL (P2), and diabetic rats treated with 100 g/100 mL (P3). Diabetes was induced using a single intraperitoneal injection of streptozotocin at 40 mg/kg body weight, and treatment was administered orally for 21 days. Serum CAT and GPx levels were measured using ELISA and analyzed using ANOVA followed by Duncan's test. The results showed that CAT levels were significantly different among groups ($P<0.05$), while GPx levels were not significantly different ($P>0.05$). The 100 g/100 mL treatment group

Volume 18 No. 3: 545-552
June 2026
https://doi.org/10.24843/bulvet.2026.v18.i03.p06

showed CAT levels closer to the normal group. It can be concluded that Java ginseng leaf simplicia decoction has potential antioxidant effects in diabetic rats by improving oxidative stress conditions. Further studies are recommended using more specific extraction methods and additional oxidative stress biomarkers.

Keywords: catalase, diabetes mellitus, glutathione peroxidase, oxidative stress, *Talinum paniculatum*

Abstrak

Diabetes mellitus merupakan penyakit metabolik kronis yang ditandai dengan hiperglikemia dan peningkatan stres oksidatif akibat pembentukan *Reactive Oxygen Species* (ROS) berlebih. Kondisi tersebut dapat memengaruhi enzim antioksidan endogen seperti katalase (CAT) dan glutation peroksidase (GPx). Penelitian ini bertujuan untuk menganalisis pengaruh pemberian rebusan simplisia daun ginseng Jawa (*Talinum paniculatum*) terhadap kadar CAT dan GPx serum tikus diabetes. Sebanyak 20 ekor tikus jantan Sprague-Dawley dibagi menjadi empat kelompok, yaitu kontrol normal (P0), kontrol diabetes (P1), kelompok diabetes yang diberi rebusan 50 g/100 mL (P2), dan kelompok diabetes yang diberi rebusan 100 g/100 mL (P3). Diabetes diinduksi menggunakan streptozotocin dosis tunggal 40 mg/kg BB secara intraperitoneal, kemudian perlakuan diberikan secara oral selama 21 hari. Kadar CAT dan GPx serum diukur menggunakan metode ELISA dan dianalisis dengan ANOVA dilanjutkan uji Duncan. Hasil penelitian menunjukkan bahwa kadar CAT berbeda nyata antar kelompok perlakuan ($P<0,05$), sedangkan kadar GPx tidak berbeda nyata ($P>0,05$). Kelompok perlakuan 100 g/100 mL menunjukkan kadar CAT yang mendekati kelompok normal. Disimpulkan bahwa rebusan simplisia daun ginseng Jawa berpotensi sebagai antioksidan pada tikus diabetes melalui perbaikan kondisi stres oksidatif. Penelitian lanjutan disarankan menggunakan metode ekstraksi yang lebih spesifik dan biomarker stres oksidatif tambahan.

Keywords: diabetes mellitus, glutation peroksidase, katalase, stres oksidatif, *Talinum paniculatum*

INTRODUCTION

Diabetes mellitus (DM) is a chronic metabolic disease characterized by elevated blood glucose levels due to impaired insulin secretion, insulin resistance, or both (Qadri *et al.*, 2015). Chronic hyperglycemia can trigger increased production of Reactive Oxygen Species (ROS) through various metabolic pathways, thereby causing oxidative stress. Oxidative stress can lead to damage to pancreatic β-cells, insulin resistance, and microvascular and macrovascular complications in people with diabetes (Chen *et al.*, 2025).

The body possesses an enzymatic antioxidant defense system to neutralize ROS, including catalase (CAT) and glutathione peroxidase (GPx). Catalase functions to break down hydrogen peroxide into water and oxygen (Nandi *et al.*, 2019), while GPx reduces hydrogen peroxide and lipid hydroperoxides using reduced glutathione (Lubos *et al.*, 2011). In diabetic conditions, changes in the levels of these two enzymes may reflect an imbalance between oxidants and antioxidants. Therefore, efforts to control oxidative stress are necessary, one of which is through the use of natural ingredients with antioxidant activity.

Javanese ginseng (*Talinum paniculatum*) is an herbal plant known to contain phenolic compounds, flavonoids, saponins, and other bioactive components with potential as natural antioxidants (Lestario *et al.*, 2009). However, information regarding the effect of a decoction of Javanese ginseng leaf crude extract on the levels of catalase and glutathione peroxidase enzymes in the serum of diabetic rats remains limited. This study aims to analyze the effect of

Volume 18 No. 3: 545-552
June 2026
https://doi.org/10.24843/bulvet.2026.v18.i03.p06

administering a decoction of Javanese ginseng leaf crude extract on the levels of CAT and GPx enzymes in the serum of diabetic rats.

RESEARCH METHODS

Ethical Approval for Laboratory Animals

The use of animals in this study has been approved by the Animal Ethics Committee of the Faculty of Veterinary Medicine, Udayana University, under approval number B/179/UN14.2.9/PT.01.04/2025.

Research Subjects

The research subjects were male Sprague-Dawley strain white rats (*Rattus norvegicus*) weighing 130–140 grams. The rats were obtained from the Bio Mice and Rat Breeding and Experimentation Laboratory, Faculty of Medicine, Udayana University, Denpasar. Prior to treatment, all rats were acclimated for one week at an ambient temperature of 23–25°C and provided with pellet feed and drinking water *ad libitum*.

Research Design

This study is a laboratory experimental study using a Completely Randomized Design (CRD). After diabetes was confirmed, the test animals were randomly divided into four treatment groups, each consisting of five rats (N=20). Diabetes was induced using a single dose of 40 mg/kg body weight (BW) streptozotocin (STZ) administered intraperitoneally. Four days after induction, blood glucose levels were measured to confirm the diabetic condition using a criterion of blood glucose levels ≥200 mg/dL (Furman, 2021). Treatment began on day 5 and was administered for 21 consecutive days.

Group P0 consisted of healthy normal rats administered 1 mL of distilled water orally. Group P1 consisted of diabetic rats administered 1 mL of distilled water orally. Group P2 consisted of diabetic rats administered a decoction of Javanese ginseng leaf crude drug at a concentration of 50 g/100 mL (w/v) at a dose of 1 mL/100 g BW per day. Group P3 consisted of diabetic rats administered a decoction of Javanese ginseng leaf crude drug at a concentration of 100 g/100 mL (w/v) at a dose of 1 mL/100 g BW per day. On day 22, all rats were euthanized, and blood samples were collected.

Data Collection Method

Diabetes induction was performed using a single dose of STZ at 40 mg/kg body weight via intraperitoneal injection according to the method by Furman (2021). Blood glucose levels were measured on day 4 post-induction using a glucometer via tail vein blood. Rats were diagnosed with diabetes if their blood glucose levels were ≥200 mg/dL. Rats meeting these criteria were then randomly assigned to groups.

The treatment was administered orally via a gastric tube for 21 consecutive days. Groups P0 and P1 were each given 1 mL of distilled water per animal per day, while group P2 was administered a decoction of Javanese ginseng leaf crude drug at a concentration of 50 g/100 mL (w/v) at a dose of 1 mL/100 g BW per day, and group P3 was administered a decoction of Javanese ginseng leaf crude drug at a concentration of 100 g/100 mL (w/v) at a dose of 1 mL/100 g BW per day.

On day 22, the rats were euthanized using an intramuscular injection of ketamine-HCl at a dose of 50 mg/kg body weight. Blood was collected via the retroorbital plexus using a hematocrit pipette, yielding 1–2 mL (Amriani *et al.*, 2021). Blood samples were placed in plain vacutainer tubes, allowed to clot, and then centrifuged at 3000 rpm for 10 minutes to obtain serum. Serum

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Pendidikan Tinggi, Riset, dan Teknologi No. 158/E/KPT/2021

Volume 18 No. 3: 545-552
June 2026
https://doi.org/10.24843/bulvet.2026.v18.i03.p06

was stored at 4°C until analysis. Measurements of CAT and GPx enzyme levels were performed using the sandwich-type Enzyme-Linked Immunosorbent Assay (ELISA) method according to the protocol of the BT Laboratory kit, China, namely CAT No. E0869Ra and GPx No. E1242Ra.

Data Analysis

The data were analyzed using an Analysis of Variance (ANOVA) to determine the effect of the treatments on serum catalase and glutathione peroxidase levels. If the results showed significant differences ($P < 0.05$), the analysis was followed by Duncan's Multiple Range Test (DMRT) to determine the differences among the treatment groups.

RESULTS AND DISCUSSION

Results

The results of the analysis showed that the mean serum levels of CAT and GPx in the experimental rats varied among treatment groups (Table 1). CAT levels showed significant differences among treatment groups ($P < 0.05$), whereas GPx levels did not differ significantly ($P > 0.05$). The patterns of differences in the mean CAT and GPx levels among the treatment groups can also be seen in Graph 1 and Graph 2.

In the CAT results, the diabetic control group (P1) showed the highest mean of 56.86 ± 5.07 ng/mL, followed by group P2 at 55.95 ± 3.28 ng/mL. The normal control group (P0) had the lowest mean of 47.59 ± 6.13 ng/mL, while group P3 showed a mean of 49.05 ± 4.73 ng/mL, which was close to the normal group. Figure 1 shows a trend of decreased CAT levels in the treatment group given a decoction of Javanese ginseng leaf at a dose of 100 g/100 mL compared to the diabetic control group. These results indicate that administration of a decoction of Javanese ginseng leaf at a dose of 100 g/100 mL tends to lower CAT levels in diabetic rats.

For the GPx parameter, group P1 showed the highest mean value of 2207 ± 349.61 pg/mL, followed by group P2 at 2187 ± 349.61 pg/mL. Group P3 had a mean of 2053 ± 340.76 pg/mL, while group P0 showed the lowest value, namely 1881 ± 218.86 pg/mL. Figure 2 shows a trend toward decreased GPx levels in the treatment groups compared to the diabetes control group, although statistical analysis indicates that the differences between groups were not significant ($P > 0.05$).

Discussion

Diabetes mellitus is a metabolic disease characterized by chronic hyperglycemia. This condition can increase the formation of ROS, thereby triggering oxidative stress. Oxidative stress occurs due to an imbalance between free radical production and the body's antioxidant defense system, which can ultimately lead to cellular and tissue damage (Caturano *et al.*, 2023). The body possesses enzymatic antioxidant defense systems, including CAT and GPx, which play a role in neutralizing peroxide compounds resulting from oxidative metabolic processes (Birben *et al.*, 2012).

In this study, the untreated diabetic rat group (P1) exhibited higher levels of CAT and GPx compared to the normal group (P0). This increase is thought to be the body's adaptive response to increased oxidative stress resulting from hyperglycemia following streptozotocin (STZ) induction. STZ is known to cause damage to pancreatic β-cells, leading to hyperglycemia and increased formation of free radicals in the body (Angie *et al.*, 2025). This condition may stimulate an increase in antioxidant enzyme levels as a compensatory mechanism against oxidative stress.

Administration of the decoction of Javanese ginseng leaf crude extract to the treatment groups

Terakreditasi Nasional Sinta 4, berdasarkan Keputusan Direktur Jenderal
Pendidikan Tinggi, Riset, dan Teknologi No. 158/E/KPT/2021

Volume 18 No. 3: 545-552
June 2026
https://doi.org/10.24843/bulvet.2026.v18.i03.p06

showed a tendency to lower CAT and GPx levels compared to the untreated diabetic group. The most significant reduction was observed in group P3, which received the 100 g/100 mL concentration decoction, where CAT levels approached those of the normal group. These results indicate that a decoction of Javanese ginseng leaf crude extract has the potential to help reduce oxidative stress in diabetic conditions through its flavonoid, saponin, and phenolic compound content, which are capable of scavenging free radicals and inhibiting ROS formation (Menezes *et al.*, 2021; Rachmawan *et al.*, 2025).

The results of this study differ from those of several previous studies. Sellamuthu *et al.* (2013) reported that the administration of mangiferin to STZ-induced diabetic rats increased CAT and GPx levels in liver tissue. Sadoughi *et al.* (2020) also reported that Avicennia marina extract increased CAT and GPx levels in the liver tissue of diabetic rats. Meanwhile, in this study, there was actually a tendency for CAT and GPx levels in serum to decrease following administration of a decoction of Javanese ginseng leaf crude drug.

This difference is likely due to the type of sample used. This study utilized blood serum, whereas other studies generally use organ tissues such as the liver. Under oxidative stress conditions, cell membrane damage caused by lipid peroxidation can increase membrane permeability, allowing intracellular enzymes such as CAT and GPx to leak into the bloodstream. Consequently, enzyme levels in serum may rise as an indicator of cellular damage, while levels in tissues decrease due to depleted enzyme reserves (Tejchman *et al.*, 2021). Following antioxidant administration, cell membrane integrity may improve, leading to reduced enzyme release into the serum. Therefore, the decrease in serum CAT and GPx levels in this study may reflect improved cellular conditions, in contrast to tissue-based studies that show increased enzyme activity.

For the CAT parameter, the results of statistical tests showed significant differences among the treatment groups. This is likely because catalase is the primary enzyme responsible for rapidly breaking down hydrogen peroxide into water and oxygen, making changes in oxidative stress more easily reflected through changes in CAT levels (Nandi *et al.*, 2019). Conversely, no significant differences were found among groups for the GPx parameter. This is likely due to the more complex mechanism of GPx, which depends on the availability of reduced glutathione (GSH) as its primary cofactor. If GSH levels have not fully recovered, the GPx response may remain limited even as free radical load decreases. Additionally, GPx is a selenium-containing enzyme, so the body's selenium status can influence this enzyme's response (Brigelius-Flohé & Maiorino, 2013).

Further confirmation that the decrease in serum CAT and GPx levels is indeed associated with improvements in oxidative stress and tissue damage requires the assessment of additional parameters. Measurement of malondialdehyde (MDA) levels is important as an indicator of lipid peroxidation. Assessment of reduced glutathione (GSH) levels is also necessary to evaluate the availability of the primary cofactor for GPx. In addition, histopathological examination of the pancreas, liver, and kidneys can reveal the extent of tissue damage and regeneration following treatment. Measurements of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) can also serve as indicators of cellular damage. With these additional parameters, the mechanism of action of the decoction of Javanese ginseng leaves can be explained more comprehensively.

CONCLUSIONS AND SUGGESTIONS

Conclusion

Administration of a decoction of Javanese ginseng leaves (Talinum paniculatum) affected

Terakreditasi Nasional Sinta 4, berdasarkan Keputusan Direktur Jenderal
Pendidikan Tinggi, Riset, dan Teknologi No. 158/E/KPT/2021

Volume 18 No. 3: 545-552
June 2026
https://doi.org/10.24843/bulvet.2026.v18.i03.p06

serum CAT levels in diabetic rats; a concentration of 100 g/100 mL (w/v) yielded the best results, as it reduced CAT levels to near-normal levels. Administration of the decoction also tended to lower GPx levels, although the difference was not statistically significant.

Suggestions

Further research is recommended using a more standardized extraction method and identifying the active compounds in Javanese ginseng leaves. Additionally, measurements of additional parameters such as MDA, reduced GSH, ALT, AST, and histopathological examination of tissues should be conducted to confirm the mechanism of action and the antioxidant effects of the decoction of Javanese ginseng leaves more comprehensively.

ACKNOWLEDGEMENT

The author would like to express gratitude to the Faculty of Veterinary Medicine at Udayana University, the Bio Mice and Rat Breeding and Experimentation Laboratory, and the Biochemistry Laboratory of the Faculty of Medicine at Udayana University for providing facilities and support throughout the course of this research. The author would also like to thank the academic advisor and all other parties who contributed to the successful completion of this research.

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Volume 18 No. 3: 545-552
June 2026
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Table

Table 1. Mean Levels of Catalase and Glutathione Peroxidase Enzymes in the Serum of Experimental Rats During 21 Days of Treatment

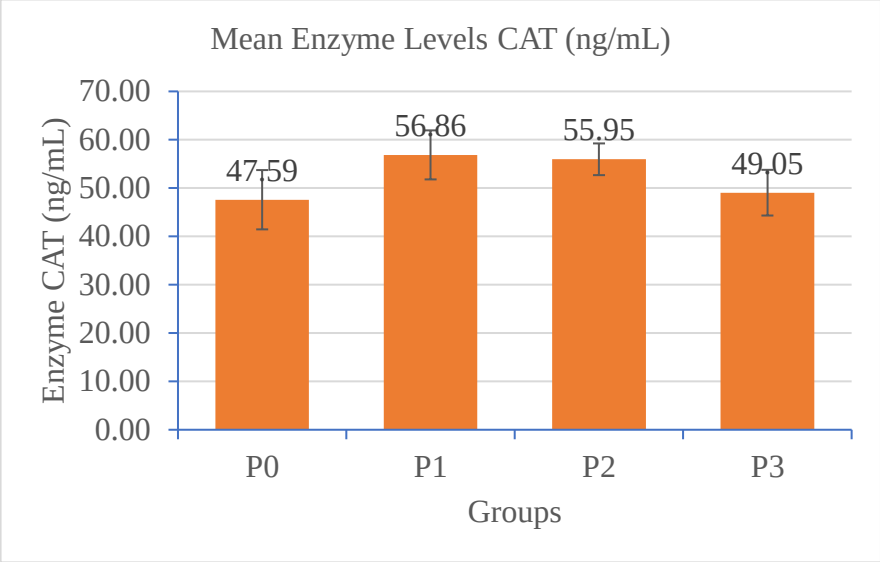
Treatment Groups	Serum CAT (ng/mL) Level	Serum GPx (pg/mL) Level
P0	47.59 ± 6.13^{a}	1881 ± 218.86^{a}
P1	56.86 ± 5.07^{b}	2207 ± 349.61^{a}
P2	55.95 ± 3.28^{b}	2187 ± 349.61^{a}
P3	49.05 ± 4.73^{a}	2053 ± 340.76^{a}

Note: Identical letters following numbers in the same column indicate no significant difference ($P > 0.05$), while different letters indicate a significant difference ($P < 0.05$); P0 (normal control group); P1 (DM group); P2 and P3 (DM groups administered decoctions of Javanese ginseng leaves at 50 g/100 mL (w/v) and 100 g/100 mL (w/v), respectively, at a dose of 1 mL/100 g body weight orally.

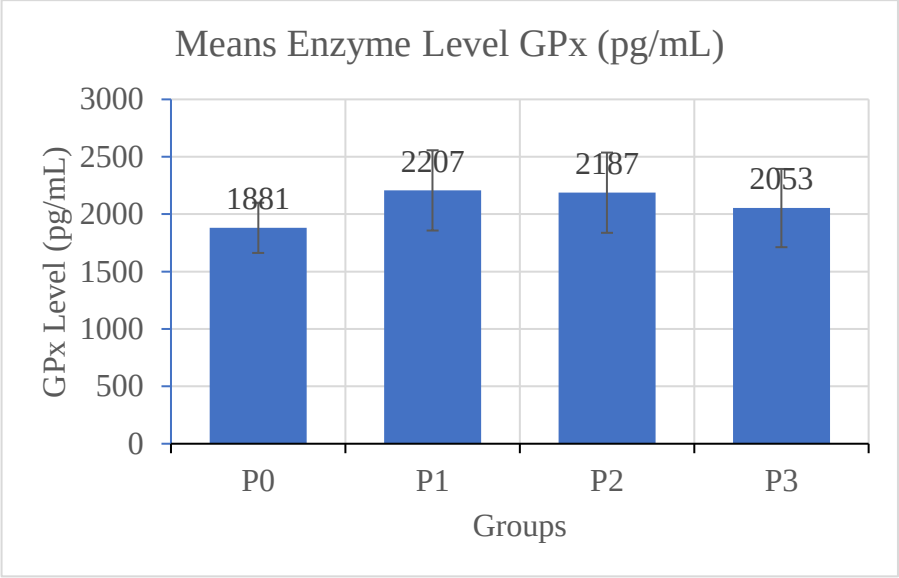
Terakreditasi Nasional Sinta 4, berdasarkan Keputusan Direktur Jenderal
Pendidikan Tinggi, Riset, dan Teknologi No. 158/E/KPT/2021

Volume 18 No. 3: 545-552
June 2026
https://doi.org/10.24843/bulvet.2026.v18.i03.p06

Graph



Graph 1. Graph of the Average Serum Catalase Enzyme Levels in Rats in Each Group. P0 = normal control group; P1 = diabetes control group; P2 = diabetes group administered a decoction of Javanese ginseng leaves at 50 g/100 mL (w/v); P3 = diabetes group administered a decoction of Javanese ginseng leaves at 100 g/100 mL (w/v)



Graph 2. Graph of the Average Serum Glutathione Peroxidase Enzyme Levels in Rats in Each Group. P0 = normal control group; P1 = diabetes control group; P2 = diabetes group administered a decoction of Javanese ginseng leaves at 50 g/100 mL (w/v); P3 = diabetes group administered a decoction of Javanese ginseng leaves at 100 g/100 mL (w/v)