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Antihypertensive Effect of Rice Bran Derived Amino Acids Through Modulation of iNOS Expression and Renal Histopathology in DOCA Salt Induced Hypertensive Rats

Muhammad Fikri Nur[1], Devi Ramadani[1], Muh Fikry Baharudin[1], Almas Dwi Khairana[1], Saidun Fiddaroini[1], Akhmad Sabarudin[1], Anna Safitri[1], Wibi Riawan[2], Dyah Kinasih Wuragil[3], Hilman Nurmahdi[1], Aulanni'am[1*]

[1] Department of Chemistry, Faculty of Mathematics and Natural Sciences, Brawijaya University, Malang 65145, East Java, Indonesia
[2] Department of Biochemistry, Faculty of Medicine, Brawijaya University, Malang 65145, East Java, Indonesia
[3] Faculty of Veterinary Medicine, Brawijaya University, Malang 65145, East Java, Indonesia

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Corresponding Author:
Aulanniam Aulanniam
aulani@ub.ac.id

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Abstract: Hypertension is a major global health concern and a leading risk factor for cardiovascular mortality. Conventional treatment with ACE inhibitors like captopril is effective but often causes adverse effects including cough, dizziness, and renal impairment. This study evaluated the antihypertensive effects of rice bran in DOCA-salt-induced hypertensive rats. In vitro analysis revealed 45.58% soluble fiber and rich amino acid content, including 2.121 mg L-Arginine, a nitric oxide precursor promoting vasodilation. Male Wistar rats were divided into five groups: negative control, positive control, captopril treatment (5 mg/kg BW), and rice bran supplementation at 1% and 2% of 25 g feed. The 2% rice bran group significantly ($p < 0.05$) reduced renal iNOS expression by 88.32% compared to hypertensive controls, surpassing captopril's 22.96% reduction. Histopathological examination revealed notable renal protection with complete absence of fatty degeneration and minimal glomerular inflammation in the 2% rice bran group. These findings demonstrate potent antihypertensive activity through ACE inhibition, enhanced nitric oxide bioavailability, and mitigation of oxidative and inflammatory stress, establishing rice bran as a promising natural therapeutic alternative for hypertension management.

Keywords: ACE inhibitor; DOCA-salt; Histopathology; Hypertension; iNOS

Introduction

Hypertension is a significant global health problem and a primary risk factor for mortality from cardiovascular diseases in both developed and developing countries. This condition is characterized by a persistent elevation in blood pressure, defined as a systolic pressure of ≥140 mmHg and/or a diastolic pressure of ≥90 mmHg, well above the normal threshold of <120/80 mmHg (Nurmahdi et al., 2017; Whelton et al., 2018). In Indonesia, the prevalence of hypertension reached an alarming 34.1% in 2018, a concerning figure given its dangerous complications affecting vital organs such as the heart, brain, and kidneys. The escalating burden of hypertension in developing countries like Indonesia is further compounded by limited healthcare access, poor medication adherence, and the high cost of long-term pharmacological therapy, creating an urgent need for accessible and affordable therapeutic alternatives (M. J. Husain et al., 2020). Hypertension can lead to secondary conditions such as atherosclerosis, stroke, myocardial infarction, heart failure, and chronic kidney disease (Carey et al., 2018; Williams et al., 2018). Pathophysiologically, this condition is often triggered by a combination of factors, including oxidative stress, sodium retention, and systemic inflammation

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(Norlander et al., 2018). The oxidative stress theory of hypertension posits that an imbalance between reactive oxygen species (ROS) production and antioxidant defense mechanisms plays a central role in endothelial dysfunction and vascular remodeling, key pathological features of sustained hypertension (Griendling et al., 2021).

Elevated blood pressure is frequently mediated by the Renin-Angiotensin-Aldosterone System (RAAS). Within this system, the Angiotensin-Converting Enzyme (ACE) converts Angiotensin I into Angiotensin II. Angiotensin II is a potent vasoconstrictive peptide hormone that narrows blood vessels, increases vascular resistance, and stimulates the release of aldosterone, which leads to sodium and water retention, thereby increasing blood pressure (X. C. Li et al., 2017; Vn & Vn, 2021). Additionally, Angiotensin II promotes the release of Aldosterone, which in turn activates NADPH oxidase (NOX). The activation of NOX enhances the generation of free radicals, known as Reactive Oxygen Species (ROS). This overproduction of ROS disrupts the natural balance between free radicals and antioxidants in the body, ultimately causing oxidative stress (Valentini et al., 2025). Furthermore, excessive ROS production triggers the upregulation of inducible nitric oxide synthase (iNOS) in renal tissue, contributing to inflammatory responses and further exacerbating hypertensive pathology through impaired renal sodium handling and vascular tone regulation (Lee et al., 2016).

Pharmacological therapy for hypertension commonly utilizes Angiotensin-Converting Enzyme Inhibitors (ACE-Is) such as captopril. Although effective, long-term use of captopril is limited by adverse effects including cough, dizziness, and renal impairment (Dang & Vasanthan, 2019). These side effects reduce patient compliance and treatment efficacy, particularly in chronic conditions requiring lifelong medication (Burnier & Egan, 2019). Moreover, synthetic ACE inhibitors do not adequately address the oxidative stress component of hypertension, necessitating a multitarget therapeutic approach (Messerli et al., 2018). These limitations underscore the need for safer, natural antihypertensive alternatives.

The functional food paradigm suggests that dietary components with bioactive properties can serve as preventive and therapeutic agents for chronic diseases, offering a cost-effective and culturally acceptable strategy for disease management (Konstantinidi & Koutelidakis, 2019).Rice bran, an abundant yet underutilized byproduct of rice milling, offers significant potential in this regard. It contains high levels of protein (13.2–17.3%), lipids (17–22.9%), and dietary fiber (27.6–33.3%), as well as bioactive compounds such as γ-oryzanol, tocotrienols, and ferulic acid. These constituents exhibit antioxidant, anti-inflammatory, and Angiotensin-Converting Enzyme (ACE) inhibitory activities that may modulate oxidative stress and vascular tone, highlighting rice bran's promise as a functional food ingredient for hypertension prevention and management (Dang & Vasanthan, 2019).

Despite these promising properties, the specific contribution of rice bran's dietary fiber fractions (soluble versus insoluble) to its antihypertensive effects remains poorly understood. Furthermore, the molecular mechanisms underlying rice bran's effects on renal oxidative stress markers, particularly iNOS expression, have not been comprehensively investigated in hypertensive models (Henderson et al., 2012). Addressing these knowledge gaps is critical for developing evidence-based dietary interventions and optimizing rice bran's therapeutic application. This study sought to evaluate the antihypertensive potential of rice bran by analyzing its soluble and insoluble dietary fiber content, LC-MS/MS profile, and renal inducible nitric oxide synthase (iNOS) expression in a DOCA-salt-induced hypertensive rat (Rattus norvegicus) model. By elucidating these mechanisms, this research aims to provide scientific validation for rice bran as a functional food intervention, contributing to the development of accessible, evidence-based strategies for hypertension management in resource-limited settings.

Method

Preparation of Hypertensive Rats induced DOCA-salt

All animal procedures in this research were approved by the Research Ethics Committee of Brawijaya University (KEP-214-UB/2024) and are summarized in the flow diagram shown in Figure 1. Male Wistar rats (*Rattus norvegicus*) were used in this study and were divided into five groups: (1) Normotensive control, (2) Hypertensive control, (3) Captopril-treated (5 mg/kg body weight, BW), (4) Rice bran-treated (1% of 25g of food), and (5) Rice bran-treated (2% of 25g of food). The rats were acclimatized for one week before the experiment began and were provided with standard AD II chow (containing 12% water, 15% crude protein, 3-7% crude fat, 6% fiber, max. 7% ash, 0.9-1.1% calcium, and 0.6-0.9% phosphorus). Hypertension was induced over a five-week period with injections administered twice weekly (10 injections total); the first five injections contained DOCA-salt at 20 mg/kg BW, and the subsequent five injections contained 10 mg/kg BW. The DOCA-salt was dissolved in corn oil and injected subcutaneously in the cervical region. Throughout the induction period, the rats were supplied with 2% NaCl (w/v) in their drinking water. Blood pressure was measured weekly using the tail-cuff

method (CODA™ Tail-Cuff Blood Pressure System, Kent Scientific) until the end of the study at necropsy.

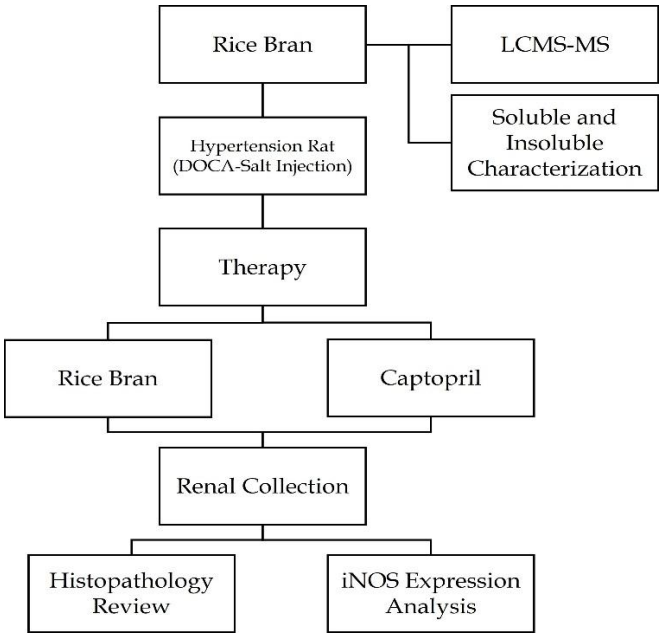

Figure 1. Research flow chart

Preparation of Rice Bran as a Therapy

The therapeutic doses of ground bran were administered in four increments, specifically 1% and 2% of the total feed provided, amounting to 25 grams per day, dissolved in 1.5 mL of drinking water. The therapy was delivered through a gastric tube once daily for a duration of 4 weeks.

Preparation of Captopril as a Therapy

The animals in the treatment group were administered captopril at a therapeutic dose of 5 mg/kg BW. For the positive control group, Captopril was freshly prepared by dissolving it in distilled water and was administered daily via oral gavage once daily for four weeks (28 days).

Analysis of Histopatology in Renal Tissue using Hematoxylin-Eosin Staining

Renal tissues were fixed in 10% paraformaldehyde, sectioned, and mounted on glass slides before undergoing deparaffinization and rehydration. The rehydration process involved sequential immersion in descending ethanol concentrations (100%, 95%, 90%, 80%, and 70%) for 5 minutes each, followed by a 5-minute rinse in distilled water. The sections were then stained with hematoxylin for 10 minutes to achieve distinct nuclear staining. After staining, the slides were rinsed under running tap water for 30 minutes and briefly in distilled water. Eosin staining was subsequently applied for 5 minutes, and the slides were immersed in distilled water to remove residual dye. Dehydration was then completed through graded ethanol series (80%, 90%, 95%, and 100%). Finally, tissue clearing was performed by immersing the slides in xylene for 5 minutes, followed by drying and mounting with Entellan, and covering with a glass coverslip.

Analysis of iNOS expression in Renal Tissue using Immunohistochemistry Method

Renal tissue was stored in 10% paraformaldehyde, sectioned, and mounted on glass slides prior to deparaffinization and rehydration. Slides were subsequently washed with Phosphate Buffer Saline (PBS) solution, treated with Hydrogen Peroxide (H_2O_2), and incubated for 20 minutes before being washed again with PBS. Blocking was conducted by adding 5% (w/v) Bovine Serum Albumin (BSA) to a PBS solution and incubating for 30 minutes at room temperature. Slides were washed with PBS solution and subsequently treated with iNOS antibody as the primary antibody. Slides were subsequently incubated with a biotin-labeled secondary antibody, followed by the addition of SA-HRP (Streptavidin-Horse Radish Peroxidase) and DAB (diaminobenzidine) solution. Counterstaining was conducted using Mayer Hematoxylin prior to the application of a cover glass. Slides were examined microscopically and subsequently analyzed using ImageJ software (Riyadi et al., 2020).

Result and Discussion

Characterization Soluble and Insoluble Fiber of Rice Bran

Based on the initial compositional analysis (Table 1), the rice bran sample was found to contain dietary fiber comprising 45.58% soluble fiber and 1.94% insoluble fiber. This composition has a strong theoretical basis for its role in maintaining cardiovascular health.

Table 1. Dietary Levels

Fiber Type	Levels (%)	SD
Soluble Fiber	45.58	1.190
Insoluble Fiber	1.94	0.076

Short-chain fatty acids (SCFAs), which are metabolites produced from the microbial fermentation of soluble fiber in the gut, contribute to the development of hypertension. They do so by influencing a wide range of complex biological pathways, such as activating specific receptors, modulating the immune and autonomic nervous systems, altering metabolic regulation, and affecting gene transcription (Mahalak et al., 2024; Wu et al., 2021).

Characterization of Rice Bran Amino Acids by LC-MS

The amino acid profile analysis of rice bran revealed 14 amino acids, including alanine, arginine,

aspartic acid, cysteine, glutamic acid, histidine, isoleucine, lysine HCl, methionine, phenylalanine, proline, serine, threonine, valine, tyrosine, and one non-essential amino acid, glycine (Table 2).

Table 2. LCMS Result on Rice Bran

Name	Ret. Time	m/z	Conc.	Area
L-Arginine	10.958	175.10	2.121	135.883
L-Aspartic Acid	6.672	134.00	5.429	7.396
L-Cystine	7.632	241.10	1.554	216
L-Glutamic Acid	6.511	148.20	3.519	103.255
Glysine	6.618	76.20	1.511	30.624
L-Histidine	9.843	156.00	1.761	133.248
L-Leucine	4.021	132.20	2.751	83.162
L-Lysine HCl	10.478	147.20	1.693	44.231
L-Methionine	4.653	150.20	1.854	2.256
L-Phenylalanine	3.645	166.20	2.235	129.568
L-Proline	4.613	116.00	2.346	354.996
L-Threonine	6.302	120.20	1.970	4.465
L-Valine	5.185	118.00	2.917	33.046
L-Tyrosine	5.403	182.00	2.071	10.099

Essential amino acids cannot be synthesized by the human body and therefore must be obtained through dietary sources, whereas non-essential amino acids can be synthesized endogenously (Mahalak et al., 2024). LC-MS/MS analysis revealed that rice bran possesses a rich amino acid profile with significant biochemical relevance to blood pressure regulation. Notably, L-Arginine was identified at a concentration of 2.121, serving as a direct precursor of Nitric Oxide (NO), a potent vasodilator that promotes vascular smooth muscle relaxation, enhances blood flow, and reduces blood pressure (Wu et al., 2021). Additionally, high levels of branched-chain amino acids (BCAAs), particularly L-Leucine (2.751) and L-Valine (2.917), contribute to improved insulin sensitivity, which is closely linked to hypertension since insulin resistance can increase renal sodium and water retention, elevating blood pressure (G. Li et al., 2024).

The amino acid composition also indicates other physiological benefits. L-Phenylalanine (2.235) and L-Tyrosine (2.071) act as precursors of neurotransmitters such as dopamine, which modulates vascular tone through central mechanisms. Moreover, L-Methionine (1.854) serves as a precursor to glutathione, a key antioxidant that protects endothelial function and supports NO production by reducing oxidative stress (Bryan, 2022). Collectively, these findings suggest that rice bran holds strong potential as a nutraceutical agent for blood pressure regulation through vasodilation, metabolic modulation, neuroregulation, and antioxidant defense mechanisms.

Histopathological Analysis of Renal Rat

The histopathological examination of renal tissues across all experimental groups (Figure 2) revealed varying degrees of structural alterations and pathological changes when compared to the normal kidney architecture. The negative control group demonstrated preserved renal histology with clearly identifiable cortical and medullary regions, providing a baseline for comparison with treatment groups that exhibited various pathological manifestations.

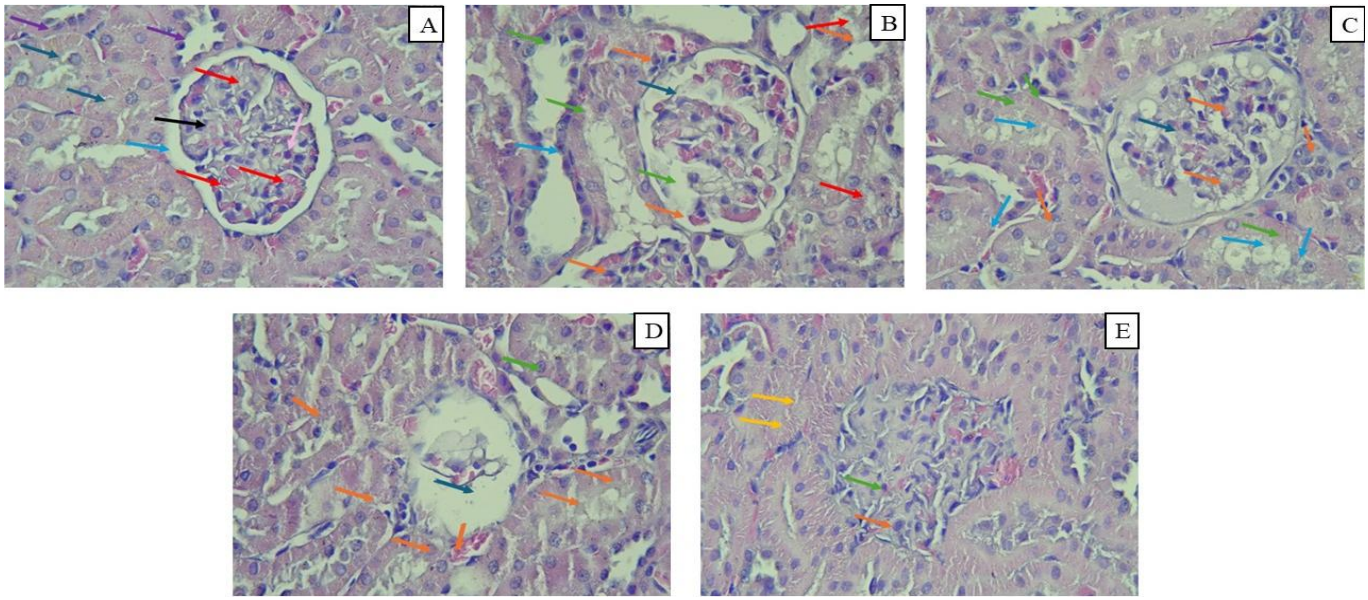

Figure 2. Renal histopathology of DOCA-salt–induced hypertensive rats at 400× magnification. (A) Negative control group; (B) Positive hypertensive control; (C) Captopril therapy group (5 mg/kg BW); (D) Rice bran therapy group (1% of 25 g total feed); (E) Rice bran therapy group (2% of 25 g total feed). Arrow colors: Black – Fatty degeneration; Blue – Karyolysis; Red – Karyopyknosis; Purple – Karyorrhexis; Yellow – Glomerular necrosis; Orange – Lymphocyte infiltration; Green – PMN cells; and Pink – Vascular congestion

In normal renal architecture, as observed in the negative control group, the kidney parenchyma maintains distinct cortical and medullary compartments. The cortex contains Bowman's capsules, glomeruli, proximal convoluted tubules, and distal convoluted tubules, while the medulla comprises collecting tubules converging toward the renal pelvis. Tubular identification is facilitated by the presence or absence of brush borders, with proximal convoluted tubules exhibiting prominent brush borders and distal convoluted tubules displaying clear lumens due to the absence of this feature (Haraldsson et al., 2008) . The glomerular structure consists of capillary tufts, basement membranes, podocytes, and resident mesangial cells, all maintaining their normal morphological characteristics in the absence of pathological insult (S. Husain, 2024; Pollak et al., 2014).

The pathological alterations observed across the experimental groups included fatty degeneration, necrosis, inflammation, and vascular congestion, though the severity and distribution of these changes varied considerably. Fatty degeneration was prominently observed in the positive, captopril, and 1% rice bran therapy groups, manifesting primarily in cortical tubular areas. This degenerative change was characterized by the accumulation of lipid droplets in non-lipid cells, creating the distinctive "signet ring cell" appearance where water-insoluble lipid droplets displaced the nucleus to the cellular periphery. During the H&E staining process, these lipid droplets dissolved in ethanol, leaving empty vacuolar spaces within the affected cells (Ivanovska et al., 2023). Notably, the 2% rice bran therapy group did not exhibit fatty degeneration, suggesting a potential protective mechanism against lipid accumulation in this treatment group.

Necrosis represented one of the most significant pathological findings across all treatment groups, affecting multiple renal compartments including glomeruli and various tubular segments. The positive control group demonstrated the most extensive necrotic changes, affecting glomeruli, proximal convoluted tubules, distal convoluted tubules, and collecting tubules. Glomerular necrosis manifested as depletion of podocytes and capillary endothelial cells, compromising the filtration apparatus. Tubular necrosis presented with characteristic nuclear alterations including karyopyknosis, where nuclei appeared condensed and shrunken; karyorrhexis, characterized by nuclear fragmentation; and karyolysis, indicating nuclear dispersion or complete disappearance. Additionally, epithelial detachment from the basement membrane into the tubular lumen was observed, indicating severe cellular injury (Daehn & Duffield, 2021). The captopril group exhibited similar necrotic patterns affecting glomeruli, proximal convoluted tubules, and distal convoluted tubules. The 1% rice bran therapy group showed extensive necrosis involving glomeruli, collecting tubules, and both convoluted tubule types, with glomerular depletion and multiple necrotic figures in tubular epithelium. The 2% rice bran therapy group demonstrated necrosis primarily in collecting tubules and both types of convoluted tubules, with all characteristic necrotic features present.

Inflammatory infiltration was a consistent finding across all groups subjected to pathological induction, though the distribution and intensity varied. The positive control group exhibited widespread inflammation extending from the glomerular regions to the tubular interstitium, with lymphocytes predominating the inflammatory cell population, though polymorphonuclear cells (PMN) were also identified in several areas (Altaleb & Rajab, 2021). The captopril group demonstrated a similar inflammatory pattern with glomerular and interstitial involvement, predominantly composed of lymphocytes with occasional PMN cells. In contrast, the 1% rice bran therapy group showed inflammatory infiltration primarily localized to the tubular interstitium rather than extensive glomerular involvement, suggesting a more restricted inflammatory response. The 2% rice bran therapy group presented an interesting pattern where glomerular inflammation was minimal while the majority of inflammatory infiltrate concentrated in the tubular interstitium, with lymphocytes predominating and PMN cells present in some areas. This varied distribution of inflammatory cells across treatment groups indicates different degrees and patterns of immune response to the induced renal injury (Vernier et al., 2023).

Vascular congestion was observed across multiple treatment groups, though with varying severity. Congestion manifested as the accumulation of blood within vessel lumens, with affected vessels appearing completely filled with erythrocytes. Under H&E staining, erythrocytes were readily identifiable due to their distinctly red appearance and high contrast with surrounding cells (Ray et al., 2019). The positive control and captopril groups demonstrated moderate to extensive vascular congestion in multiple blood vessels, indicating compromised blood flow and possible hemodynamic alterations. The 1% rice bran therapy and 2% rice bran therapy groups showed different patterns of vascular involvement, with 1% rice bran therapy group demonstrating minimal vascular congestion, while 2% rice bran therapy group exhibited more pronounced congestion in several blood vessels, suggesting differential hemodynamic responses or vascular injury patterns between these treatment groups.

The pathological diagnoses derived from histopathological examination reflected the complexity and severity of renal injury across treatment groups. The positive control and captopril groups both exhibited tubular necrosis, lymphocytic glomerulonephritis, and lymphocytic interstitial nephritis, indicating multifocal renal involvement affecting both glomerular and tubulointerstitial compartments (Joyce et al., 2017). The 1% rice bran therapy group was diagnosed with tubular necrosis and lymphocytic interstitial nephritis, with minimal glomerular involvement, suggesting a more tubulointerstitial pattern of injury. The 2% rice bran therapy group demonstrated tubular necrosis, lymphocytic glomerulonephritis, and lymphocytic interstitial nephritis, though glomerular inflammation was notably minimal compared to interstitial involvement (Martinez Valenzuela et al., 2020).

The comparative analysis of histopathological findings across all groups reveals important patterns regarding the nature and distribution of renal injury. The preservation of normal architecture in the negative control group confirmed the validity of the experimental model and staining procedures. The extensive pathological changes in the positive control group, affecting multiple parameters with considerable severity, established the baseline for injury assessment (Cohen & Schrier, 1999). The captopril group demonstrated similar pathological patterns to the positive control, suggesting limited protective efficacy under the experimental conditions employed. The treatment groups 1% rice bran therapy and 2% rice bran therapy exhibited distinct pathological profiles, with 2% rice bran therapy notably lacking fatty degeneration entirely, which may indicate specific protective mechanisms against lipid accumulation, possibly through different metabolic pathways or cellular stress responses. However, the 2% rice bran therapy group's vascular congestion pattern suggests potential hemodynamic alterations or vascular compromise that differed from other treatment groups.

The predominance of lymphocytic infiltration across all affected groups indicates a chronic inflammatory response rather than acute injury, as polymorphonuclear cells were present but not predominant (Poitou-Verkinder et al., 2015). The presence of both glomerular and tubulointerstitial inflammation in most groups suggests systemic renal injury affecting multiple compartments, though the 1% rice bran therapy group's primarily tubulointerstitial inflammation pattern indicates possible compartment-specific protective effects or injury mechanisms. The varied severity of necrosis across tubular segments and glomeruli reflects differential susceptibility of renal structures to the induced injury, with the positive control group showing the most extensive necrotic changes across all renal compartments (Hanif et al., 2025).

The histopathological evidence collectively demonstrates that all groups subjected to pathological induction experienced significant renal damage, primarily manifesting as tubular necrosis, inflammatory responses, and varying degrees of glomerular involvement (Sun et al., 2020). The differential patterns of fatty degeneration, inflammatory distribution, and vascular congestion among treatment groups suggest distinct pathophysiological mechanisms or protective effects that warrant further investigation (Zhang et al., 2021). The structural integrity and functional capacity of the kidneys were clearly compromised across treatment groups, as evidenced by the presence of necrotic cells, inflammatory infiltrates, and epithelial detachment, all of which would significantly impact renal filtration, reabsorption, and overall homeostatic functions (Xu et al., 2025).

iNOS Expression in Rat Renal Tissue

The results demonstrated that treatment with rice bran significantly reduced iNOS expression ($p < 0.05$) in the cardiac tissue of rats (Figure 3). The ACE-inhibitory compounds present in the rice bran suppress ACE enzyme activity, preventing the conversion of angiotensin I into angiotensin II. Consequently, this reduces the production of reactive oxygen species (ROS) by inhibiting NOX activation (Arendshorst et al., 2024). In the hypertensive control group, excessive ROS generation resulting from NOX activation triggered NFκB, leading to the upregulation of the iNOS gene. Elevated iNOS expression promotes the overproduction of nitric oxide (NO), which can react with superoxide anions (O_2^-) to form peroxynitrite ($ONOO^-$), a highly reactive and cytotoxic radical (Andrabi et al., 2023; Rashid et al., 2023). Moreover, increased iNOS activity contributes to the generation of superoxide radicals that are converted into hydrogen peroxide (H_2O_2), either spontaneously or through catalytic action by superoxide dismutase (SOD). The presence of peroxynitrite and hydrogen peroxide is known to cause oxidative damage, leading to tissue injury and organ dysfunction, including the renal (Radi, 2018).

The statistical grouping (Table 3) underscores a dose-dependent therapeutic effect: positive control (highest iNOS) > captopril > 1% rice bran > 2% rice bran ≈ negative control. The convergence of the 2% rice bran group with normotensive animals indicates near-complete reversal of DOCA-salt-induced inflammatory signaling, consistent with the observed histological preservation of renal structures and absence of fatty degeneration. Taken together, the significant differences among groups highlight that rice bran supplementation, particularly at 2%, confers robust anti-inflammatory and

renoprotective effects superior to standard ACE inhibition, positioning rice bran as a promising multi-target nutraceutical capable of mitigating RAAS-mediated oxidative stress while enhancing endogenous NO homeostasis (Pannangpetch et al., 2022).

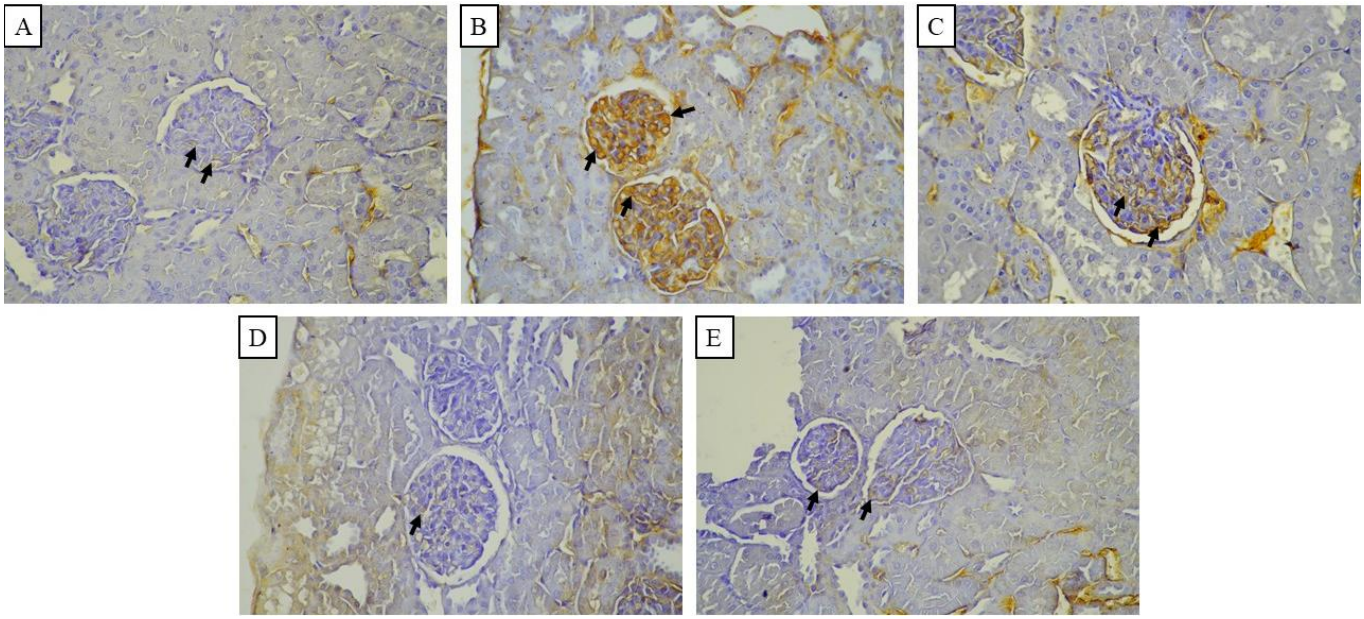

Figure 3. Renal iNOS expression in DOCA-salt–induced hypertensive rats at 400× magnification. (A) Negative control; (B) Positive hypertensive control; (C) Captopril-treated group (5 mg/kg BW); (D) Rice bran–treated group (1% of 25 g total feed); (E) Rice bran–treated group (2% of 25 g total feed).

Table 3. Levels of iNOS Expression of Each Group

Groups	Average iNOS Levels (mean±SD %area)	Increased Levels Against Negative Hypertensive Group	Decreased Levels Against Negative Hypertensive Group
Negative Hypertension	3.04 ± 0.61^{d}		
Positive Hypertension	21.04 ± 2.38^{a}	589.87	
Captopril Therapy 5 mg/kgBW	16.22 ± 2.42^{b}		22.96
Rice bran Therapy 1% of 25g of food	8.88 ± 1.53^{c}		57.82
Rice bran Therapy 2% of 25g of food	2.46 ± 0.48^{d}		88.32

Note: Superscript notation indicates a significant difference between treatment groups ($p < 0.05$).

Conclusion

This study found that rice bran, which contains 45.58% soluble dietary fiber and essential amino acids including L-Arginine (2.12), demonstrated significant antihypertensive and kidney-protective effects in DOCA-salt-induced hypertensive rats. The 2% rice bran supplementation proved more effective than the drug captopril in reducing renal iNOS expression (88.32% > 22.96% reduction), while histopathological examination showed protected kidney tissue with no fatty degeneration, minimal inflammation, and reduced vascular congestion. The researchers attribute these benefits to multiple mechanisms including ACE inhibition, improved nitric oxide availability, decreased NADPH oxidase activation, and prevention of peroxynitrite-related tissue damage, concluding that rice bran offers a safe, affordable natural alternative to synthetic medications for managing hypertension and protecting organs, though human clinical trials are needed to confirm these findings.

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Author Contributions

Muhammad Fikri Nur: investigation, interpreted the data, visualization, formal analysis, methodology, software, writing,

and editing. Devi Ramadani: formal analysis, validation, and editing. Muh Fikry Baharudin: software, review, and editing. Almas Dwi Khairana: interpreted the data, visualization, and methodology. Saidun Fiddaroini: interpreted the data, writing, editing, and formal analysis. Akhmad Sabarudin: Conceived and design the experiment, methodology, and supervision. Anna Safitri: Conceived and design the experiment, methodology, and supervision. Wibi Riawan: methodology, software, validation software and supervision. Dyah Kinasih Wuragil: Conceived and design the experiment, methodology, and supervision. Hilman Nurmahdi: methodology and validation software. Aulanniam Aulanniam: Conceived and design the experiment, resources, analyzed and interprenting the data, funding acquisition, and writing

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Conflicts of Interest
The authors declare no conflict of interest

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