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Renometabolic disorder in experimental rat model of polycystic ovarian syndrome is reversed by acetate-mediated inhibition of pyruvate dehydrogenase kinase 4

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Abstract

Background Chronic Kidney disorders is a global public health problem, including in women with polycystic ovarian syndrome (PCOS), and is characterized by renal fibrosis, nephrotoxicity and glomerulonephritis, which increases the possibility of renal failure and organ transplant. Pyruvate dehydrogenase kinase 4 (PDK4) has been implicated in mitochondria dysfunction, contributing to metabolic dysregulation in different organs, including kidney. Studies have shown that short chain fatty acids, particularly acetate, alleviates metabolic alterations in experimental models. Hence, the present study investigated the therapeutic potential of acetate on renometabolic disorders associated with experimental PCOS model. The study in addition elucidates the probable involvement of PDK4 in PCOS-associated renometabolic disorders.

Methods Eight-week-old nulliparous female Wistar rats were randomly allotted into four groups (n = 5). Letrozole (1 mg/kg *bw*) was used to induce PCOS for 3 weeks. Thereafter, acetate (200 mg/kg *bw*) was administered for 6 weeks, uninterruptedly. Biochemical parameters from the plasma and renal tissue, as well as histology of ovaries were performed with appropriate methods.

Results Experimental PCOS rats were characterized with elevated circulating testosterone and the presence of multiple ovarian cysts. In addition, rat with PCOS also manifested insulin resistance, increased plasma urea and creatinine levels, increased renal Gamma glutamyl transferase (GGT), malondialdehyde (MDA), Nuclear factor -kappa B (NF-kB), Tumor necrosis factor -alpha (TNF-a), Transforming growth factor -beta 1 (TGF-B1), caspase-6, Histone deacetylase 2 (HDAC2), while a decrease in glucose-6 phosphate dehydrogenase (G6PD), reduced glutathione (GSH), renal nitric oxide (NO) and endothelial nitric oxide synthesis (eNOS), when compared with animals in the control group. These were associated with elevated level of PDK4 in the renal tissue. However, administration of acetate ameliorates these renal/metabolic abnormalities.

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Conclusion Altogether, the results from the present study suggests that acetate ameliorates renal dysfunction in PCOS via downregulation of PDK4.

Keywords Acetate, Apoptosis, PCOS, PDK4, Renal disorders

Introduction

Despite decades of research, polycystic ovarian syndrome (PCOS), an endocrine and metabolic disorder remains the leading cause of infertility in women of child-bearing age globally, with 6–21% of women diagnosed while others are either undiagnosed or misdiagnosed [1, 2]. The distinctive manifestation of PCOS include hyperandrogenism (as observed with the presence of hirsutism), amenorrhea/oligomenorrhea (irregularity in the menstrual cycle) and the presence of multiple cysts in the ovaries [3, 4]. Nevertheless, treatment of this condition has often been suboptimal, owing to its unknown etiology. However, several factors including sedentary lifestyle, genetic and epigenetic alterations have been reported to play critical roles in the development of PCOS and its comorbidities [5, 6]. Studies have implicated insulin resistance (IR) in the manifestation of PCOS and its attendant morbidities including obesity, cardiomyopathies and chronic renal disease [7–9]. Similarly, other investigations, including recent investigations from our laboratory have also linked PCOS with hormonal imbalance, chronic low-grade inflammation, oxidative stress, hyperinsulinemia, and hyperandrogenism, which impair follicular development and increase the risk of infertility and related comorbidities which include metabolic diseases (type 2 diabetes mellitus and endometrial cancer) [10, 11, 12, 71]. Although the pathophysiological mechanism is inconclusive.

Beyond reproductive health, PCOS often leads to metabolic and renal complications. The kidneys are bean-shaped organs in the body that are involved in glucose regulation, water balance and electrolyte balance, and can promote glucose homeostasis during prolonged starvation, among others [13]. However, studies have shown that metabolic disorders such as type 2 diabetes mellitus (T2DM), obesity, PCOS, can alter these physiological functions of the kidneys [14, 15]. Emerging evidence suggest that women with PCOS are at higher risk for chronic kidney disease (CKD) which manifest through renal fibrosis, nephrotoxicity and glomerulonephritis, and this can potentially progress to renal failure, which may necessitate organ transplantation [16, 17]. Although the mechanism linking PCOS and CKD is ill defined, hyperglycemia and insulin resistance have been reported as important players in disruption of glomerular filtration barrier, resulting from excessive accumulation of lipid in the renal tissue, inflammation, and oxidative stress which progresses to cellular apoptosis [18–20]. Studies have shown a strong correlation between CKD and metabolic abnormalities associated with obesity, diabetes mellitus (DM), and cardiovascular diseases (CVD) and PCOS [20, 21]. In addition, IR has been shown to increase renal resistive index, which causes a significant elevation in the blood pressure of PCOS women, and explains the relationship between renal dysfunction and hypertension, dyslipidemia, and cardiomyopathies [22, 23]. However, further investigation into the pathophysiological mechanisms of PCOS-associated renal disorder would provide a possible therapeutic target for prevention and management of this life-threatening condition in individuals that suffer PCOS.

Pyruvate dehydrogenase kinases (PDKs) play critical roles in the pyruvate dehydrogenase complex (PDC), inhibiting the conversion of pyruvate to acetyl coenzyme A (acetyl coA) for energy production. Among the recognized PDKs, PDK4 is highly expressed in the kidneys [24, 25]. Recent studies reported that PDK4 promoted succinate accumulation in the kidney, following renal injury and contributed to excessive production of reactive oxygen species (ROS) with subsequent renal apoptosis [26, 27]. Likewise, authors demonstrated that pharmacological inhibition of PDK4 abated kidney damage by suppressing succinate accumulation as well as excessive mitochondrial production of ROS [27, 28]. Similarly, PDK4 has been implicated as a hub for diabetic kidney disease (DKD) development, resulting from redox imbalance [29]. In addition, inhibition of PDK4 has been reported to improve nuclear factor erythroid 2 like 2 (Nrf2) as well as a spectrum of antioxidant enzymes, suggesting PDK4 as a promising therapeutic target in the management of chronic kidney disease [30].

Short chain fatty acids (SCFAs) are emerging therapeutic agents in the management of metabolic-related disorders. The main components of these SCFAs include acetate, butyrate, and propionate are synthesized from the gut microbial activities in the presence of fermented dietary fibers and have been documented to regulate metabolic health/overall wellness via epigenetic modulation [31]. Acetate, has been a major focus in biomedical research due to its abundance in circulation which makes it a potent therapeutic agent in the management of metabolic alterations. It is well-documented that these SCFAs, particularly acetate elicit their beneficial effect via inhibition of histone deacetylases (HDACs) and modulation of G-protein coupled receptors (GPCRs) 41 and 43 [32–34]. Previous studies including studies from our laboratory have shown that acetate demonstrated anti-inflammatory, anti-lipolytic, antioxidative and anti-apoptotic role

in metabolic dysfunction and related syndrome [23, 35–37]. In addition, a recent study from our laboratory demonstrated that acetate attenuates renal dysfunction in experimental PCOS model by decreasing androgen excess, apoptosis, oxidative stress, and NF-kB/NLRP3 immunoreactivity. However, the possible physiological mechanism is inconclusive [23]. Hence, the present study hypothesized that acetate would mitigate renometabolic abnormalities associated with PCOS model by suppression of PDK4.

Materials and methods

Experimental design and animal grouping

Female Wistar rats (Eight-week-old) were procured from the animal house of Afe Babalola University. The rats had free access to standard rat chow and tap water. The rats were acclimatized for two weeks and it was observed that animals had regular estrous cycles before the commencement of the experiment, which was determined through vaginal cytology. The rats were randomized into four groups with n = 5/group, namely control (CON), acetate (ACT), Letrozole-induced PCOS (LET) and LET + ACT groups. Rats were maintained in a colony under standard environmental conditions, with temperature and relative humidity of 22–26^{0}C and 50–60% respectively, and a dark/light cycle of 12-hours respectively.

Induction and confirmation of PCOS

Uninterrupted administration of letrozole (1 mg/kg; p.o., Sigma-Aldrich, St Louis, MI.) was given for three weeks to induce experimental PCOS in LET and LET + ACT groups as previously documented [38, 39]. Manifestation of PCOS was confirmed using diagnostic criteria [4] which included evaluating the estrous cycle using vagina cytology as previously described [40, 41] (supplementary Figure 1) and testosterone level. Histological evaluation of the ovaries at the end of the treatments also confirmed the presence of multiple cysts in PCOS rats.

Treatment of experimental animals

After the induction of PCOS in LET and LET + ACT groups, control and LET groups received vehicle (normal saline, per *os* (p.o.)), while ACT and LET + ACT groups received treatment with sodium acetate (200 mg/kg, p.o., Sigma-Aldrich, St Louis, MI). The treatments lasted for six weeks [42].

Collection of samples

At the end of the experiment, the rats were fasted overnight and anesthetized via injecting intraperitoneally with a single dose of sodium pentobarbital (50 mg/kg) [43, 44]. Blood sample was collected via cardiac puncture into heparinized tube and centrifuged at 704 g for 5 min at room temperature. Plasma was stored at – 80 °C until it was required for biochemical assays. Thereafter, the animals were decapitated to isolate the ovaries and kidneys. The left ovary was used for histology while the right ovary was used for biochemical analysis as previously documented [10].

Renal tissue homogenate preparation

The isolated kidneys of each rat were weighed. Thereafter 100 mg section of the renal tissue was carefully separated, minced and homogenized in 1 ml of 0.25 M sucrose/0.2 mM EDTA adjusted to pH 7.5 with Tris buffer. The tissue homogenates were centrifuged at 4 °C for 12 min at 750 g. The supernatant fluid was collected and stored at – 80 °C until it was required for biochemical assays.

Biochemical analysis

Metabolic and endocrine parameters

Fasting blood glucose level was determined with a handheld glucometer (ONETOUCH-LifeScan, Inc., Milpitas, CA, USA). Plasma insulin was determined using Rat ELISA kit purchased from Calbiotech Inc. (Cordell Ct., El Cajon, CA 92020, USA). Insulin sensitivity was determined using quantitative insulin sensitivity check index (QUICKI) as previously reported [10]. Plasma concentrations of testosterone was determined using Rat ELISA kits obtained from Calbiotech Inc. (Cordell Ct., El Cajon, CA 92020, USA) in adherence to manufacturer's procedures.

Triglyceride, oxidative stress and inflammatory markers

The concentration of triglyceride was determined in the plasma and renal tissue using standard colorimetric methods with kits obtained from Fortress Diagnostics Ltd. (Antrim, UK). The levels of MDA were determined in the renal tissue by standard non-enzymatic spectrophotometric method using assay kits procured from Randox Laboratory Ltd. (Co. Antrim, UK). Concentrations of nuclear factor kappa B (NF–kB), tumor necrosis factor alpha (TNFα), gamma glutamyl transferase (GGT), reduced glutathione (GSH), and glucose 6-phosphate dehydrogenase (G6PD) were determined in the renal tissue homogenate as previously described [10, 45, 46].

Transforming growth factor beta 1 (TGF-β1), caspase-6 and histone deacetylase 2 (HDAC2)

The level of TGF-β1, caspase-6, HDAC2 were determined in the renal tissue using the principle of sandwich immunoassay with rat ELISA kit obtained from ELK Biotechnology Co. Ltd. (1312 17th Street #692 Denver, CO 80202 USA) in adherence to the manufacturer's guidelines.

Nitric oxide (NO) and endothelial nitric oxide synthesis (eNOS)

Nitric oxide was determined in the renal tissue by standard spectrophotometric method using kits from Oxford Biomedical Research Inc., (Oxford, UK), while eNOS level was determined in the renal tissue using sandwich ELISA method with ELISA kits obtained from Elabscience Biotechnology Inc. (Wuhan, Hubei, P.R.C., China) in compliance with the manufacturer guidelines.

Pyruvate dehydrogenase kinase 4

The level of PDK4 was determined from the renal tissue using rat ELIZA kits obtained from Korain Biotech Co., Ltd (Shanghai, China) in compliance with the manufacturer's guideline.

Histological evaluation of ovaries

Ovarian tissue was evaluated using the hematoxylin and eosin (H & E) stains, a section of the ovary was fixed in 10% formol saline overnight, and thereafter was dehydrated, embedded in paraffin, and sectioned at a thickness of 5-µm [47]. The slides were prepared and examined using OPTO-Edu industrial camera light microscope and a computer (Nikon, Japan). For the stereological assessment of ovarian follicles, 10 sagittal sections of the ovaries from each animal was analyzed in a serial section. The mean total number of normal follicles and degenerated follicles were determined as reported by Malamed et al. [48].

Histological evaluation of kidneys

Hematoxylin and eosin (H & E) staining technique was used to evaluate the renal tissue. A section of the kidney was fixed in 10% formol saline overnight, which was thereafter dehydrated, and embedded in paraffin. Tissue was sectioned at a thickness of 5-µm. The slides were prepared and examined using OPTO-Edu industrial camera light microscope and a computer (Nikon, Japan) (Supplementary Figure 2). Histopathological grading of the renal tissue degeneration was performed using the method described by [49]. The tubuloglomerular degeneration score was assessed and graded according to the scale introduced, which includes grade 0 = 0%, 1 + = 1-25%, 2 + = 26-50%, 3 + = 51-75%, and 4 + = 76-100%.

Data analysis

All data from this present study were expressed as means ± SD. Shapiro-Wilk test was used to test the normality of the data. Statistical group analysis was carried out with GraphPad Prism software version 9 (Graphpad software Inc., California, USA). One-way ANOVA was used in comparing the mean values of variables among the groups. Post hoc analysis was performed using Bonferroni's test, thereafter the statistically significant difference was considered at $p < 0.05$.

Results

Glucose homeostasis in rats with PCOS is regulated by sodium acetate

A significant increase ($p < 0.05$) in fasting insulin, a decrease in QUICKI, while no significant difference in fasting glucose was observed in PCOS animals when compared with animals in the control group. However, upon administration of sodium acetate, there was a significant decrease ($p < 0.05$) in fasting insulin, an increase in QUICKI while no change in fasting glucose level in PCOS animals when compared with untreated PCOS animals (Figure 1).

Hormonal profile and ovarian morphology in rats with PCOS is restored by sodium acetate

The testosterone level of rats with PCOS was significantly increased ($p < 0.05$) when compared with the control group. In addition, the ovarian photomicrograph of rats with PCOS showed disrupted ovarian morphology when compared with the control group. Nevertheless, administration of sodium acetate significantly decreased ($p < 0.05$) the testosterone level and restored ovarian morphology in PCOS animals when compared with untreated PCOS group. Similarly, stereological evaluation of ovarian follicles showed a significant decrease in the number of normal follicles in LET group compared with control group, and this was significantly increased with sodium acetate administration. Likewise, PCOS animals showed a significant increase in the number of degenerated follicles compared to control group. Nevertheless, degenerated follicles were significantly decreased in rats that received sodium acetate treatment when compared to untreated PCOS animals (Figure 2)

Elevated levels of renal function markers in rats with PCOS is attenuated by sodium acetate

Renal urea, creatine and creatinine kinase were significantly increased ($p < 0.05$) in animals with PCOS when compared with the control group. Nonetheless, administration of acetate significantly decreased ($p < 0.05$) renal urea, creatine and creatinine kinase in animals with PCOS when compared with untreated PCOS group (Figure 3).

Increased circulating and renal lipid in rats with PCOS is reversed by sodium acetate

A significant increase ($p < 0.05$) in circulating/renal TG in rats with PCOS when compared with animals in the control group. Nevertheless, when sodium acetate was administered there was a significant decrease ($p < 0.05$)

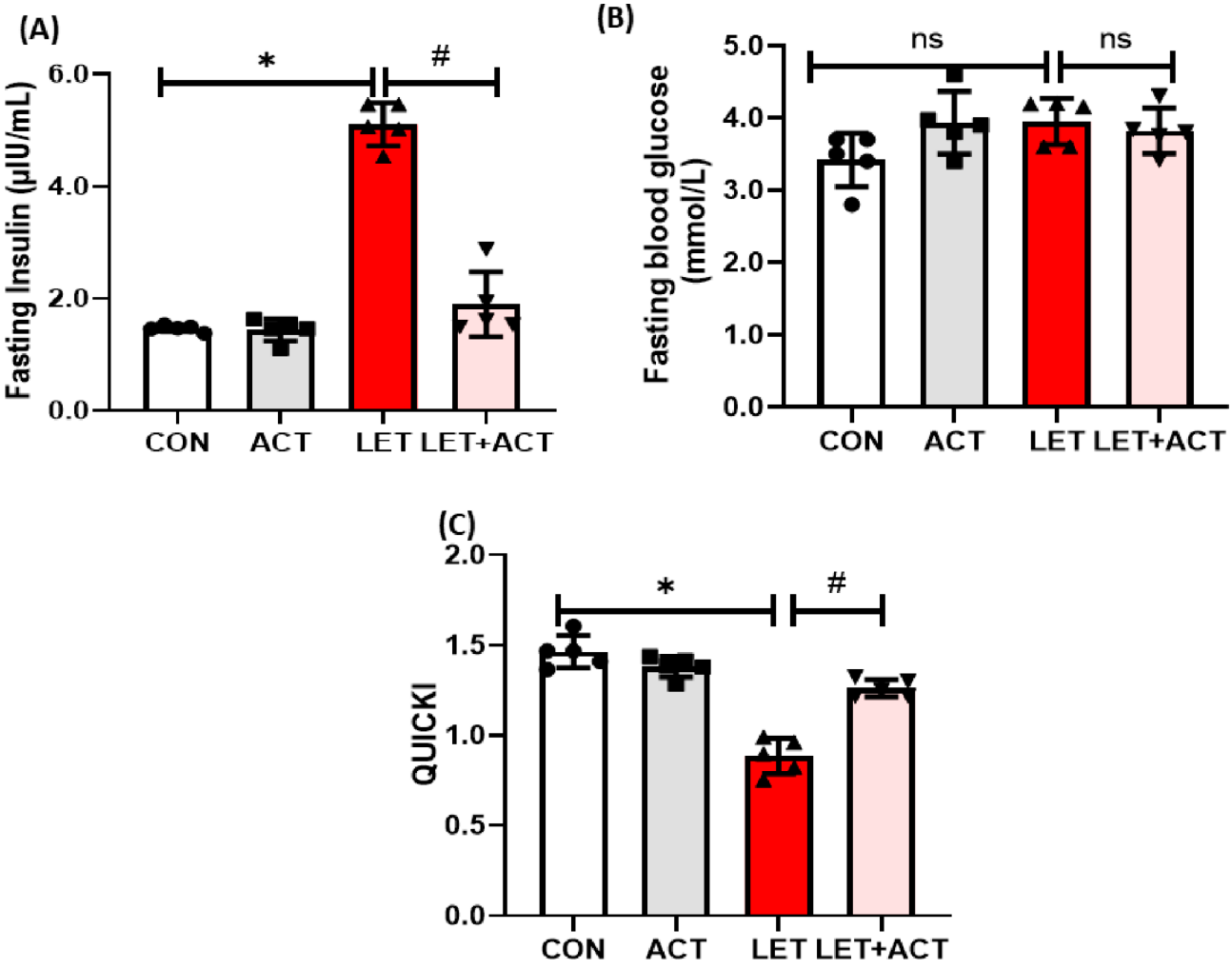

Fig. 1 Effect of acetate on fasting insulin (**a**), fasting blood glucose (**b**), QUICKI (**c**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (*p < 0.05 vs. control, #p < 0.05 vs. LET). Photomicrographs showing the ovarian histology of CON and ACT groups with normal follicle, LET group with cystic follicle and LET + ACT group with improved ovarian follicle. H & E stains. Mag. X200. Blue arrows signify oocyte (OC); Red arrow signifies cyst/ degenerated follicle. Quantitative insulin sensitivity check index (QUICKI). Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

in circulating/renal TG in PCOS animals when compared with untreated PCOS group (Figure 4).

Elevated renal inflammatory responses and oxidative stress in rats with PCOS is attenuated by sodium acetate

A significant increase ($p < 0.05$) was observed in renal NF–kB, TNF-a, MDA, and GGT, while a decrease in renal G6PD and GSH in animals with PCOS when compared with control group. However, administration of sodium acetate significantly decreased ($p < 0.05$) renal NF–kB, TNF-a, MDA, and GSH, and increased renal GGT in animals with PCOS when compared with untreated PCOS group (Figure 5)

Depletion of renal nitric oxide synthesis in rats with PCOS is alleviated by sodium acetate

Nitric oxide synthesis (NO and eNOS) in the renal tissue of rats with PCOS was significantly decreased ($p < 0.05$) when compared with animals in the control group. Nevertheless, administration of sodium acetate significantly increased ($p < 0.05$) in animals with PCOS when compared with untreated PCOS group (Figure 6).

Elevated levels of renal proangiogenic and apoptotic markers in rats with PCOS is reversed by sodium acetate

TGF-B1 and Caspase-6 levels were significantly elevated ($p < 0.05$) in rats with PCOS when compared with control group. Nonetheless, upon administration of sodium acetate there was a significant reduction ($p < 0.05$) in TGF-B1 and Caspase-6 levels in PCOS animals when compared with untreated PCOS animals (Figure 7).

Exacerbated levels of renal HDAC2 and PDK4 in rats with PCOS is abated by sodium acetate

HDAC2 and PDK4 levels were significantly elevated ($p < 0.05$) in rats with PCOS when compared with control

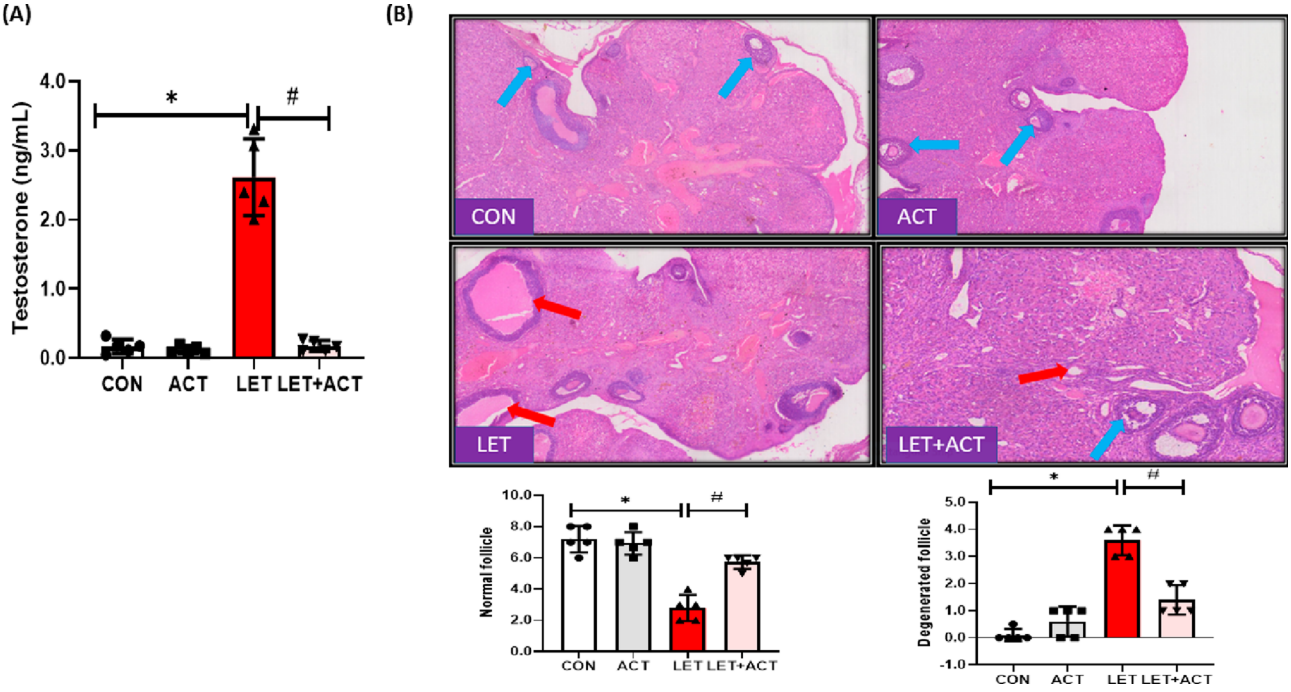

Fig. 2 Effects of acetate on plasma testosterone (**a**) ovarian morphology (**b**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (**p < 0.05 vs. control, #p < 0.05 vs. LET).* Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

group. However, upon administration of sodium acetate there was a significant reduction ($p < 0.05$) in HDAC2 and PDK4 levels in PCOS animals when compared with untreated PCOS animals (Figure 8).

Discussion

It has been extensively reported that PCOS is a critical contributor to CKD [50], necessitating the need for urgent intervention to improve the quality of life of women who are at risk or suffer PCOS. The present study investigated the beneficial effect of acetate, a short chain fatty acid on metabolic and/or renal dysfunction associated with PCOS. Interestingly, acetate was found to ameliorate renometabolic abnormalities in PCOS rat model by suppression of PDK4. Additionally, rats with PCOS showed biochemical, hormonal and histological changes, which confirmed the induction of PCOS, and these were consistent with the observation of [51]. The result also revealed renal lipid accumulation, inflammation (NF–kB, TNF-a), lipid peroxidation (MDA) and antioxidant depletion (G6PD/GSH) which resulted in renal dysfunction, characterized with elevated level of plasma urea and creatinine/creatinine kinase as well as renal GGT when compared with control animals. Likewise, renal endothelia nitric oxide synthesis was depleted, while renal TGF-B1, caspase-6, HDAC2 and PDK4 were elevated in PCOS rats when compared with control group. Nevertheless, acetate administration ameliorated these systemic and renal metabolic alterations in PCOS and these were accompanied by PDK4 reduction.

Impaired insulin sensitivity/IR is a critical feature of PCOS that is responsible for most metabolic changes in PCOS model [52, 53]. Most metabolic changes including lipolysis in the adipose tissue which promotes ectopic lipid accumulation [50, 54] as observed in the kidneys of PCOS animals with elevated level of renal TG when compared with control animals. This initiated inflammatory responses and decreased the antioxidant defense system as demonstrated in this study by elevated levels of renal NF–kB, TNF-a and MDA, and depletion in G6PD/GSH levels in animals that developed PCOS when compared with control animals. Hence, the above observations suggest that PCOS development is associated with renal inflammation/oxidative stress, which is driven by excessive intrarenal lipid accumulation.

Results from this present study also revealed that renal nitric oxide synthesis (NO and eNOS) was depleted in PCOS rats when compared with the control group. Nitric oxide (NO) and endothelial nitric oxide (eNOS) are essential biomolecules that preserve the integrity of the endothelial lining of the blood vessels supplying nutrients through the blood to vital organs, including the kidneys [55]. Studies have shown that depletion in renal NO and eNOS compromised blood flow to the kidneys, thus impairing renal function and exposing kidney to ischemia and cellular death [23, 55, 56], as validated with elevated levels of renal pro-fibrotic marker, TGF-β1, and

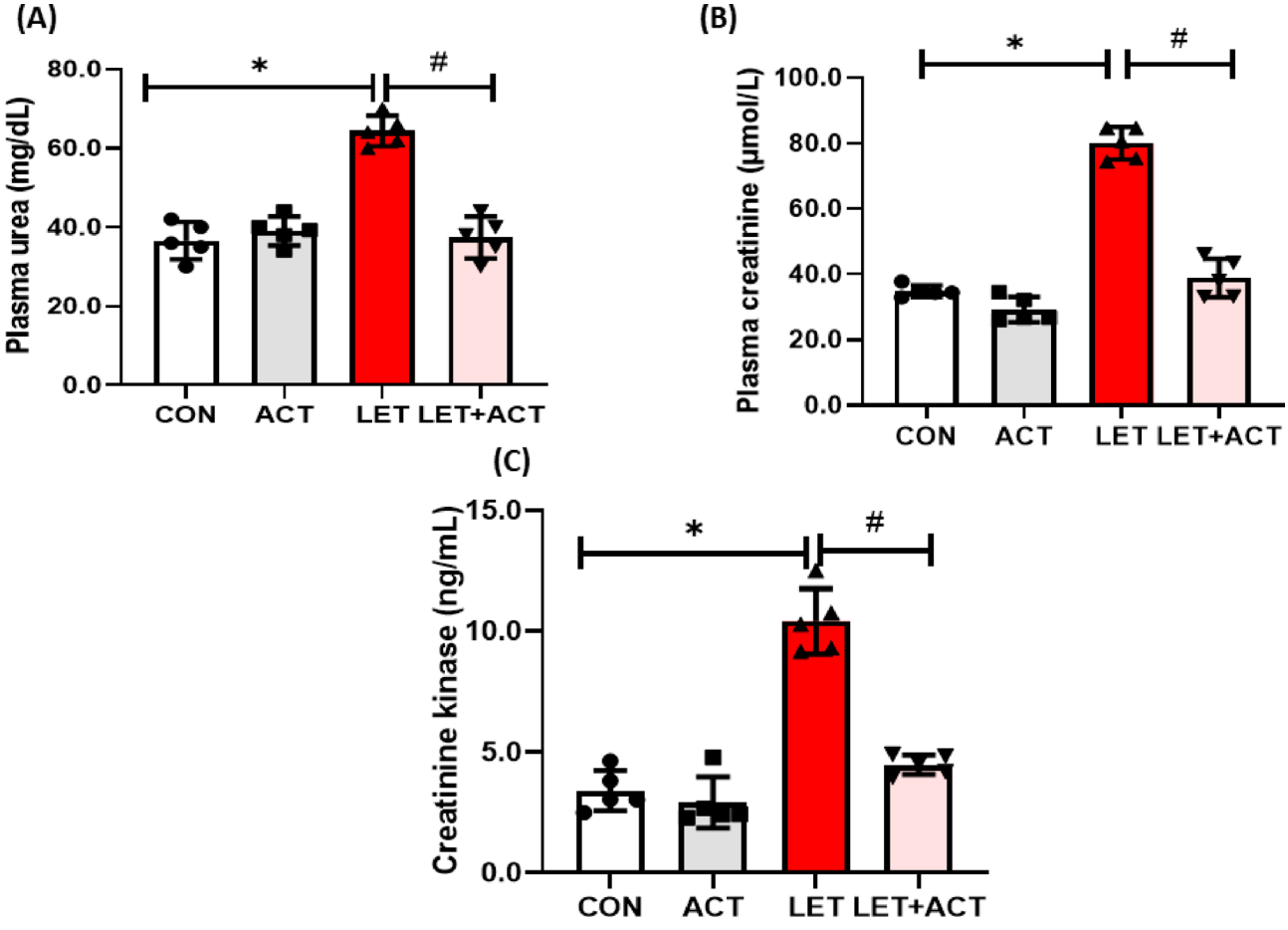

Fig. 3 Effect of acetate on urea (**a**), creatine (**b**), creatinine kinase (**c**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (*p < 0.05 vs. control, #p < 0.05 vs. LET). Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

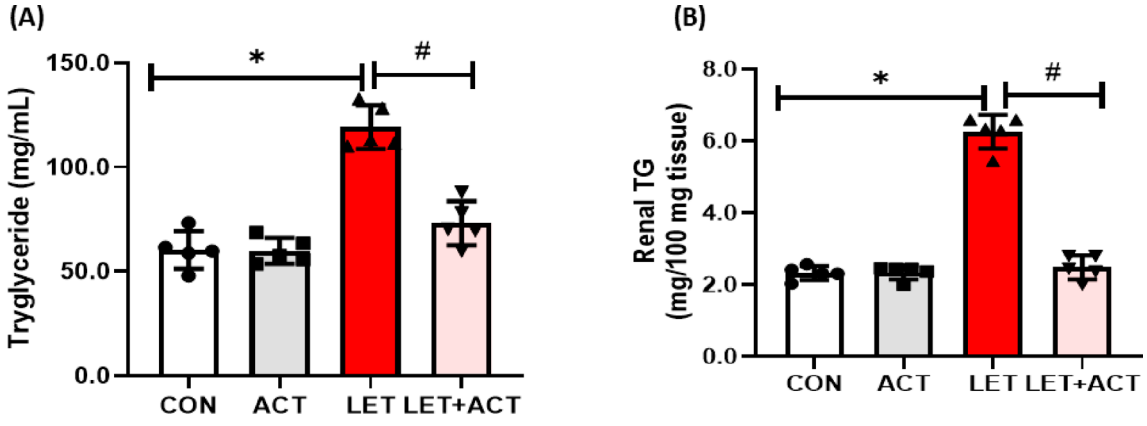

Fig. 4 Effect of acetate on plasma triglyceride (**a**), renal triglyceride (**b**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (*p < 0.05 vs. control, #p < 0.05 vs. LET). Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

apoptotic marker (caspase-6) in PCOS animals when compared with control animals. Additionally, results from the present study show elevated level of renal HDAC2 in PCOS animals when compared with control. Several studies including studies from our laboratory have reported HDAC2, as a critical epigenetic modulator in the development of pathological conditions associated with PCOS, including renometabolic disorders [18, 22, 57].

More so, the present result revealed a significant increase in renal function markers (urea, creatinine and creatinine kinase) in PCOS animals, and this is also

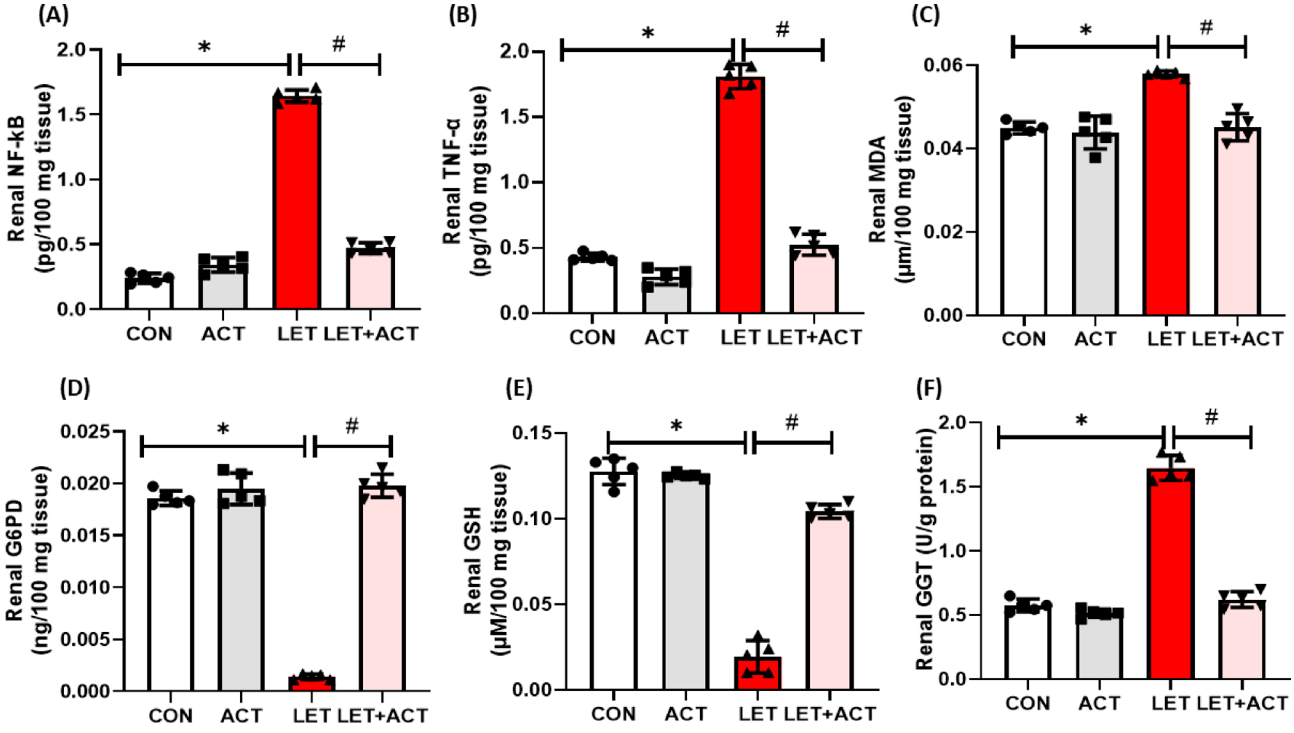

Fig. 5 Effect of acetate on NF-κB (**a**), TNF-α (**b**), MDA (**c**), G6PD (**d**), GSH (**e**), and GGT (**f**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (*p < 0.05 vs. control, #p < 0.05 vs. LET). Nuclear factor kappa B (NF-κB); Tumor necrosis factor-alpha (TNF-α); Malondialdehyde (MDA); Glucose-6-phosphate dehydrogenase (G6PD); Reduced glutathione (GSH); Gamma glutamyl transferase (GGT). Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

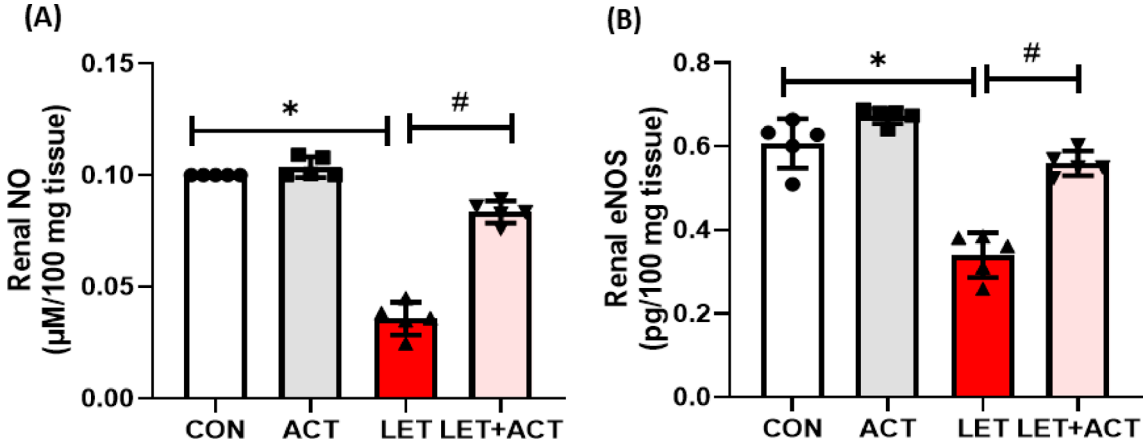

Fig. 6 Effect of acetate on renal NO (**a**), renal eNOS (**b**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (*p < 0.05 vs. control, #p < 0.05 vs. LET). Nitric oxide (NO); Endothelial nitric oxide synthase (eNOS). Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

characterized by disrupted corticomedullary architecture, comprising of disrupted glomeruli, adherence of abnormal segments of tuft to bowman's capsule and cellular infiltration as revealed in the photomicrographs of kidney histology (supplementary Figure. 2). These collectively connotes that PCOS animals elicits renal dysfunction. Notably, there was a significant increase in renal PDK4 in PCOS animal model when compared with control. This significantly corresponds to an increase in renal TG, inflammatory responses (NF–kB, TNF-a), renal lipid peroxidation (MDA) as well as a significant decrease in antioxidant system (G6PD/GSH) of the kidneys. Our present observations are in consonance with earlier observations that have implicated elevated level of PDK4 in oxidative stress-driven renal injury and diabetic kidney disease [26, 27, 29]. Similarly, Oh et al. demonstrated elevated expression of PDK4 mRNA in the kidney of mice induced with renal injury using cisplatin, and treatment with PDK4 inhibitor attenuated kidney injury with evidence of improved morphology and function

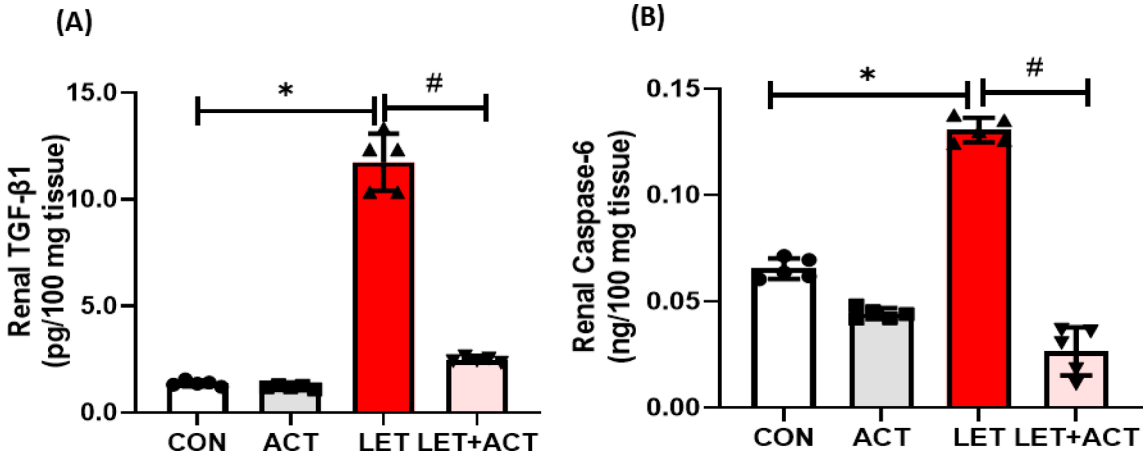

Fig. 7 Effect of acetate on renal TGF-β1 (**a**), renal caspase-6 (**b**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (*p < 0.05 vs. control, #p < 0.05 vs. LET). Transforming growth factor beta 1 (TGF-β1). Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

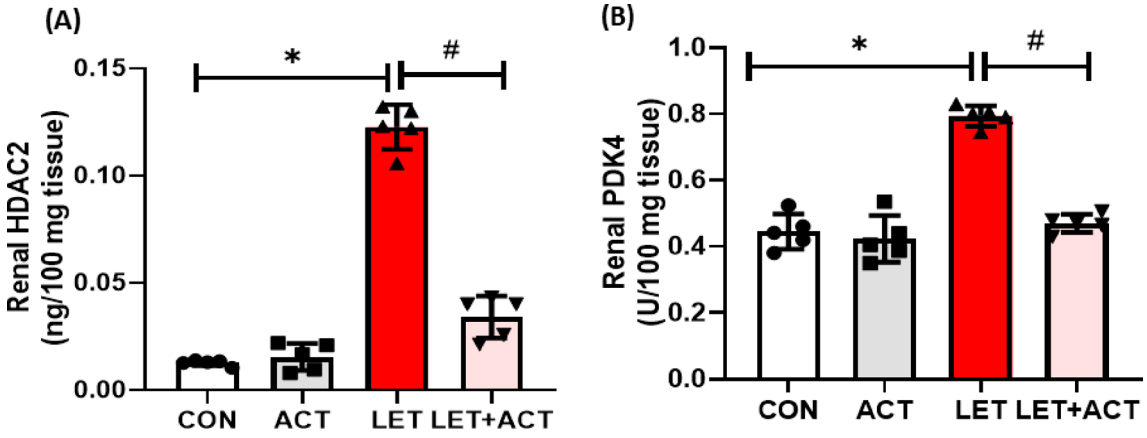

Fig. 8 Effect of acetate on renal HDAC2 (**a**), renal PDK4 (**b**) in LET-induced PCOS. Data are expressed as mean ± S.D, n = 5. (*p < 0.05 vs. control, #p < 0.05 vs. LET). Histone deacetylase 2 (HDAC2); Pyruvate dehydrogenase kinase 4 (PDK4). Control (CON); Acetate (ACT); Letrozole (LET); Letrozole + acetate (LET + ACT)

[28]. Additionally, Thome et al. reported elevated expression of PDK4 in mice induced with CKD [58]. Therefore, the present result suggests that PCOS-induced renometabolic disturbance and/or renal dysfunction is driven by elevated level of PDK4, which is associated with renal inflammation and oxidative stress. Hence, inhibition of PDK4 would possibly provide an effective therapeutic management for PCOS-induced renal dysfunction.

Interestingly, the results show that administration of SCFA, acetate reversed these endocrine and renometabolic abnormalities observed in PCOS animals when compared with untreated PCOS group. Results from this study revealed that acetate reversed altered glucose indices and hyperandrogenemia/impaired ovarian function associated with PCOS, as observed by a decrease in the circulating level of insulin, improved insulin sensitivity (QUICKI), decrease in circulating testosterone level and restoration of ovarian morphology in PCOS animals when compared with untreated PCOS animals. These findings are in conjunction with previous studies including findings from our laboratory [53, 59]. In addition, acetate attenuated elevated levels of plasma urea, plasma creatinine/creatine kinase, elevated circulating/renal triglyceride levels, elevated level of inflammatory responses and restored antioxidant levels with preserved corticomedullary architecture in PCOS animals when compared with untreated PCOS group. Moreover, acetate improved nitric oxide synthesis (NO and eNOS) in PCOS animals, demonstrating that SCFA, including acetate protects against endothelial dysfunction, ischemia and cell death in the kidneys of PCOS animals. This is similar to previous study which reported the beneficial effect of SCFAs, including acetate in metabolic-induced endothelial dysfunction [60].

Furthermore, results from this present study also revealed that acetate ameliorates excessive angiogenesis/fibrosis and apoptosis in the kidneys of PCOS animals as observed by a significant decrease in the levels of renal

pro-angiogenic/fibrotic marker (TGF-β1) and apoptotic marker (caspase-6), when compared with untreated PCOS group. Interestingly, the present results also show that acetate significantly suppressed the levels of renal HDAC2/PDK4, which were accompanied by reduction of renal inflammatory responses, oxidative stress, renal apoptosis in PCOS animals when compared with the untreated PCOS group. Thus, validating anti-fibrotic, anti-apoptotic properties of acetate [61]. Hence, the present results as detailed above suggests that PDK4 inhibition by acetate improves renometabolic disturbance in PCOS rat model. Nevertheless, the present study is not without limitations in such that the cause-effect relationship between PDK4 and other biochemical parameters were not determined and the molecular mechanism underlying the beneficial effect of acetate on renometabolic dysfunction associated with PCOS was not investigated. However, the present data provides a preclinical basis for future molecular investigation. It also adds preclinical relevance for the management of renal dysfunction in PCOS women.

Conclusion

The findings of this study provide evidence that acetate possesses significant therapeutic potential in ameliorating renometabolic disorders associated with PCOS in experimental rat model. Specifically, acetate administration reversed the key markers of renal dysfunction including elevated plasma urea, creatinine, renal oxidative stress and inflammation. Moreover, acetate mitigated the disruption in renal metabolic function, which is accompanied by suppressed level of PDK4, which is a critical regulator implicated in metabolic dysregulation. Given the growing prevalence of PCOS and its associated complications including chronic kidney disease, the result of this study demonstrates acetate as a promising candidate that offers therapeutic intervention for renometabolic disorder in PCOS by targeting PDK4. However, future studies are warranted to further explore the molecular mechanisms underlying the renoprotective effect of acetate as well as its potential clinical applicability in PCOS-related renal dysfunction.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12882-025-04157-5.

Supplementary Material 1

Author contributions

KSO and SEA conceived and designed the research. KSO, SEA and CLA conducted the experiment. KSO and SEA analysed and interpreted the data, and drafted the manuscript. CLA, AA, IOA, OEA, MBA, COA, GOO, SOO, OFA, OAA, KA, PAO, and TEA contributed reagents to the project. KSO, SEA, CLA, AA, IOA, OEA, MBA, COA, GOO, SOO, OFA, OAA, KA, PAO, and TEA read, revised and approved the final manuscript for submission.

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Data availability

Data will be made available from the corresponding author upon request.

Declarations

Ethical approval and consent to participate

This research was performed in adherence to guidelines from the National Institutes of Health Guide for the Care and maintenance of Laboratory Animals, and the protocol was approved by the Ethical Review Board of Afe Babalola University, Nigeria, with the protocol number ABUADERC/10/2023.

Consent for publication

Not applicable.

Clinical trial number

Not applicable.

Competing interests

The authors declare no competing interest.

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