*Niger. J. Physiol. Sci. 28(December 2013) 127–133*
www.njps.com.ng

Niger. J. Physiol. Sci.

# Interactions of PPAR α and GLUT4 in DOCA/salt-induced renal injury in mice

**Ighodaro Igbe[1*], Eric Kelly Omogbai[1] and Adebayo Oyekan[2]**

*[1]Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Benin, Nigeria. [2]Center for Cardiovascular Diseases, College of Pharmacy and Health Sciences, Texas Southern University, Houston, Texas*

**Summary:** Diminished insulin sensitivity is a characteristic feature of various pathological conditions such as hypertension and activation of peroxisome proliferator activated receptor α (PPARα) has been shown to enhance insulin resistance and reduce capacity for glucose uptake in muscles. The present study was designed to evaluate the interactions of PPARα and GLUT4 in a model of hypertensive renal injury by studying deoxycorticosterone acetate (DOCA)-salt induced hypertension in wild-type (WT) and PPARα knockout (KO) mice. PPARα WT and KO mice were uninephrectomized (UNx) and implanted subcutaneously DOCA and drank 1% sodium chloride/1% potassium chloride with or without a GLUT4 antagonist, indinavir (20 mg/kg/day, s.c) or PPARα ligand, fenofibrate (100 mg/kg/day, orally). DOCA/salt treatment increased urinary sodium excretion and urine volume ($p<0.05$) in PPARα KO mice compared to WT littermates. Indinavir increased proteinuria ($p<0.01$) in DOCA/salt-treated PPARα KO mice compared to WT littermates but did not affect heart and kidney weight index in DOCA/salt KO or WT-treated mice. Urinary sodium excretion ($U_{Na}V$) and urine volume (UV) were increased by indinavir ($p<0.01$) and fenofibrate ($p<0.05$) in DOCA/salt-treated PPARα KO mice compared to WT mice. Urinary nitric oxide was greater in both fenofibrate ($p<0.05$) and indinavir-treated WT mice ($p<0.05$) compared to KO mice. These data suggest that in hypertensive nephropathy, GLUT4 probably exerts a renoprotective role that was enhanced with the activation of PPARα receptors by a mechanism that may be related to increased nitric oxide production.

**Keywords***:* DOCA, Indinavir, Fenofibrate, PPARα, Nephropathy, GLUT4



**Address for correspondence:* igbe.ighodaro@uniben.edu

*Manuscript Accepted: October, 2013*

## INTRODUCTION

Peroxisome proliferator-activated receptors (PPARs) are ligand-activated transcription factors belonging to the nuclear receptor superfamily. The family also includes the steroid and thyroid hormone receptors and they have been widely reported to regulate lipid and glucose metabolism and to modulate gene expression in virtually every cell in the body (Ziouzenkova et al. 2004; Marx et al, 2004). PPARα is found in tissues (such as liver, kidney, heart, and muscle) where fatty acid catabolism is important where they regulate genes that are involved in lipid and lipoprotein metabolism (Ziouzenkova et al. 2004; Marx et al, 2004). PPARα is activated by natural ligands including polyunsaturated fatty acids (PUFAs) such as docosahexanoic acid (DHA) and eicosapentaenoic acid (EPA), oxidized phospholipids, lipoprotein lipolytic products and by synthetic ligands including fibrates, such as fenofibrate and gemfibrozil (Finck et al, 2005).

Activation of PPARα represses glucose transporter 4 (GLUT4) gene transcription, a key component of the insulin-responsive glucose uptake pathway. Earlier report has shown that subchronic inhibition of GLUT4 has been shown to alter angiotensin II-induced changes in systemic and renal haemodynamics by attenuating angiotensin-induced increase in medullary blood flow and glomerular filteration rate (Igbe et al, 2013). Chronic activation of PPARα in muscle has been shown to reduce capacity for glucose uptake and insulin resistance (Finck et al, 2005) and overexpression of PPARα in muscle was sufficient to cause glucose intolerance and insulin resistance in the context of a "lean" phenotype. Moreover, PPARα activation reduced agonist-stimulated endothelin-1 (ET-1) expression and production (Irukayama-Tomobe et al. 2004). In addition, PPARα activators were also suggested to regulate vascular tone by stimulating NO-dependent vasodilation and by inhibiting ET-1-induced vasoconstriction. Concomitantly, endothelial

dysfunction, typically found under AII infusion was prevented by DHA (Diep et al, 2002a). Although these effects cannot be conclusively and unambiguously shown to be unrelated to blood pressure reduction, in some models such as the deoxycorticosterone acetate (DOCA)-salt hypertensive rat, they have occurred in response to PPARα activators in the absence of significant blood pressure reduction (Iglarz et al, 2003). In the present study, we determined the interactions of PPARα and GLUT4 in a model of hypertension accompanied by renal injury by studying DOCA/salt- induced hypertension in wild-type and PPARα knockout mice using indinavir, a GLUT4 antagonist, which induces insulin resistance by inhibiting insulin stimulated glucose uptake.

## MATERIALS AND METHODS

### Drugs and chemicals

Fenofibrate (Sigma-Aldrich, St. Louis, MO) was suspended in 2 % carboxymethycellulose sodium salt (CMC) (Sigma-Aldrich, St. Louis, MO), Indinavir (Crixivan, Merck & Co. USA) dissolved in mineral oil, NaCl, lidocaine and ketamine (Sigma-Aldrich, St. Louis, MO), KCl (Fischer Scientific, USA), and deoxycorticosterone acetate (DOCA) pellets were used as 25mg slow release pellets prepared in our laboratories.

### Animals

PPARα wild type knockout male mice (Jackson Laboratories, MN) weighing between 25 and 30 g were maintained on standard mouse diet (Purina Chow; Purina, St Louis, MO) and allowed *ad libitum* access to water and food until the beginning of the experiments. The study was approved by the Institutional Animal Ethical Committee, and experiments were conducted as per the guidelines of CPCSEA

### Surgical preparation (Uni-nephrectomy and DOCA implantation)

Animals were anesthetised with ketamine (150 mg/kg i.p) and the site of surgery on the left flank was shaved and the expected line of skin incision was irrigated with 0.2 % Lidocaine (total 50 µl) at two or more subcutaneous sites. A straight cut was made on the skin and the skin was separated from the subcutaneous tissue by blunt dissection. A laparatomy was done by cutting through the skin to expose the abdominal cavity. The kidney was exposed and a loop was made around the hilus using chromic catgut 4-0 (LOOK®, Angiotech, Reading, USA) taking care not to tamper with the adrenal gland. The kidney capsule was carefully teased out to free the adrenal gland, the loop was secured tightly and the kidney freed behind the tie. The abdominal muscle layer was closed with 4-0 chromic catgut while the skin layer was closed with silk 6-0 using simple interrupted suturing pattern. In sham-operated mice, the above procedure was repeated and though the kidney was located, it was not removed.

After one day of recovery, the animals were put under light anaesthesia using ketamine (100 mg/kg). An incision was made on the neck of the animals and a 25 mg DOCA pellet was subcutaneously implanted into the uninephrectomized animal using a trochar.

### Experimental groups

Animals were divided into four groups (n= 5 per group).

Group 1; Sham-operated group received tap water

Group 2; Uninephrectomized mice that received 25 mg DOCA, 1% NaCl/ 0.1% KCl solution as drinking water (Newaz et al, 2002) and 2% carboxymethylcellulose (5ml/kg)/mineral oil as vehicles for fenofibrate and indinavir respectively**.**

Group 3; Uninephrectomized mice that received 25 mg DOCA, 1% NaCl/ 0.1% KCl solution as drinking water and fenofibrate (a PPARα agonist), 100 mg/kg *p.o* (Legendre et al, 2002).

Group 4; Uninephrectomized mice that received 25 mg DOCA, 1% NaCl/ 0.1% KCl solution as drinking water and indinavir, GLUT4 antagonist, 20 mg/kg (s.c twice weekly).

All animals were treated for 21 days and were placed in metabolic cages for 24 h urine collection on days 0 and 21. After 21 days treatment, they were anesthetized; the hearts and kidneys were removed and weighed to determine the heart weight (HW) and kidney weight (KW) indices. Urinary $Na^+$ excretion ($U_{Na}V$) was determined by flame photometry (Genway FP7, Jenway Ltd, Essex, UK) while urine volume (UV) was determined by gravimetrically.

### Determination of protein.

The BIORAD assay is a colorimetric assay for protein determination. Briefly, a 1 in 5 dilution of the BIORAD reagent (Bio-rad Laboratories, Inc, Hercules, CA) was made and varying standard concentrations ( 0.125, 0.25, 0.5, 1 and 2 mg/ml) was prepared from Bovine Serum Albumin (BSA). A 1 in 5 dilution of urine samples was made and thereafter 990 µl of diluted BIORAD reagent was added to 10 µl of each urine sample and standard concentration before absorbance was determined at 595 nm with a spectrophotometer (Spectronic, Genysis 5, Spectronic Instrument Inc, Rochester, USA).

### Determination of nitric oxide (NO)

Nitric oxide (NO) was determined colorimetrically in urine samples using the Griess assay. Griess reagent was freshly prepared by mixing Solutions A and B in a 1:1 ratio. Solution A contains 1 g sulphanilamide dissolved in 5 ml phosphoric acid ($H_3PO_4$) and 95 ml distilled water while Solution B contains 100 mg N-(1-naphthyl)ethylenediamine (NEDD) dissolved in 100 ml distilled water. Standard concentrations of

$NaNO_2$ at 1, 2, 5, 10 and 20 µM/ml were prepared in distilled water for plotting of the standard curve by adding equal volumes of the Griess reagent and the varying standard concentration. For the samples, 0.5 ml Griess reagent and 0.5 ml of urine samples were mixed together and transferred into a curvette. A blank was done for each sample (0.5 ml of Griess reagent and 0.5 ml of distilled water). After measuring absorbance at 540 nm by a spectrophotometer (Spectronic, Genysis 5, Spectronic Instrument Inc, Rochester (USA) and determination of NO concentrations in the samples, the values obtained from the blank were subtracted from each sample to obtain the actual concentration of NO in the urine.

**Statistical Analysis**

All data were expressed as mean ± standard error of mean (SEM). Differences between knockout (KO) and wild type (WT) littermates were tested for statistical significance by the 2-tailed Student *t* test or a 2-way ANOVA with Bonferroni post hoc analysis using GraphPad Prism (version. 4.01). In all cases, $p<0.05$ was considered statistically significant.

## RESULTS

**Effect of inhibition of GLUT4 or PPARα activation on kidney injury in DOCA/salt-treated PPARα KO or WT mice**

Figure 1 illustrates that on Day 0, there were no differences in urinary protein excretion ($U_{prot}V$) between any of the groups in both WT and KO mice. There was also no change in urinary protein excretion between the control KO and WT mice over the entire course of study. However, proteinuria was worse in KO mice compared to WT mice that received DOCA/salt treatment. On Day 21, proteinuria increased in WT mice by 4.59 ± 1.67, 5.10 ± 0.51 and 11.1 ± 1.63 mg/24h in DOCA/vehicle, DOCA/fenofibrate and DOCA/indinavir groups, respectively. By contrast, proteinuria increased 6.90 ± 1.12, 12.07 ± 2.20 and 37.48 ± 1.04 mg/24h ($p<0.01$) in DOCA/vehicle, DOCA/fenofibrate and DOCA/indinavir in KO mice respectively. These data indicate that renal injury increased progressively in DOCA/salt treated animals as the treatment progressed and that GLUT4 appears to confer renoprotective effect.

**Effect of inhibition of GLUT4 or PPARα activation on organ weight index in DOCA/salt-treated PPARα KO or WT mice**

Figure 2 illustrate that KW index was similar in WT (0.011 ± 0.01) and KO (0.011 ± 0.01) mice in DOCA/salt treated group but greater ($p<0.05$) than the sham group (WT, 0.007 ± 0.0005; KO, 0.007 ± 0.0006). There was no significant difference in KW index between the DOCA/fenofibrate treated group (WT, 0.011 ± 0.0004; KO, 0.009 ± 0.0004) and DOCA/indinavir treated group (0.01 ± 0.0003; KO, 0.01 ± 0.0008). By contrast, there were no significant differences in the HW index between the controls and any of the treated groups as shown in fig 3. These data indicate interaction of GLUT4 and PPARα did not affect DOCA/salt-induced organ enlargement.

**Effect of inhibition of GLUT4 or PPARα activation on urinary Na excretion in DOCA/salt-treated PPARα KO or WT mice**

Figure 4 illustrates that there was no significant change in the urinary Na excretion ($U_{Na}V$) between control KO and WT mice over the entire course of study. On Day 0, in KO mice, there was a significant increase in urinary Na excretion between DOCA/vehicle group ($p<0.05$) and DOCA/fenofibrate group ($p<0.05$) compared to WT mice. On Day 21, there was a significant increase in Na excretion in DOCA/vehicle group ($p<0.05$) and DOCA/indinavir group ($p<0.01$) in the KO mice when compared to WT mice. These data indicate that GLUT4 may be involved in enhancing PPARα induced-antinatriuretic effect.

**Effect of inhibition of GLUT4 or PPARα activation on urine output in DOCA/salt-treated PPARα KO or WT mice**

Figure 5 illustrates that there was no significant change in baseline (Day 0) urine volume in any of the groups compared to the control in both WT and KO mice however on day 21, urine volume increased significantly in DOCA/vehicle ($p<0.05$) and DOCA/indinavir KO mice ($p<0.05$) when compared to WT mice.

**Effect of inhibition of GLUT4 or PPARα activation on urinary NO excretion in DOCA/salt-treated PPARα KO or WT mice**

There was no significant difference in baseline $U_{NOX}V$ (Day 0) in any of the treated WT and KO mice (figure 6). On Day 21, urinary NO excretion was greater in both fenofibrate ($p<0.05$) and DOCA/indinavir-treated WT mice ($p<0.05$) compared to KO mice. These results suggest that PPARα activation stimulates NO production and this effect may be enhanced when GLUT4 activity is diminished.

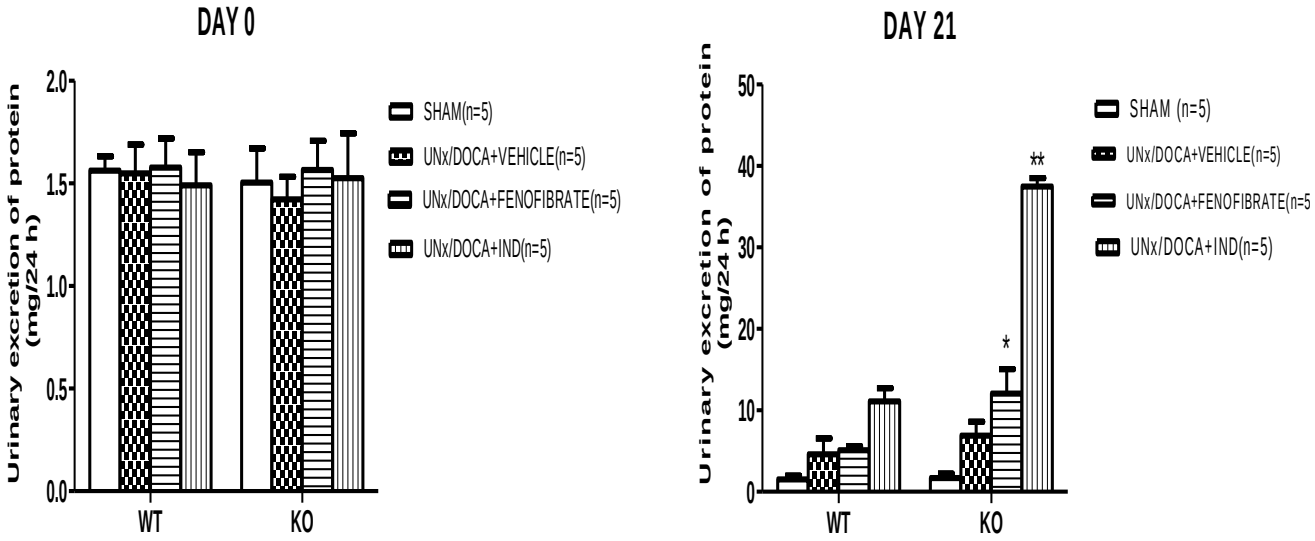


Fig. 1 Urinary protein excretion in uninephrectomised PPARα KO or WT mice treated with DOCA/1% NaCl/0.1% KCl and fenofibrate (100 mg/kg, p.o) or indinavir (20 mg/kg, s.c). ($^{*}p<0.05$, $^{**}p<0.01$ vs WT mice). Vehicle was 2% carboxymethylcellulose/mineral oil for fenofibrate and indinavir respectively. Abbreviations: UNx, uninephrectomised; KO, knockout; WT, wildtype; IND, indinavir; DOCA, deoxycorticosterone acetate; PPAR, peroxisome proliferator activated-receptor.

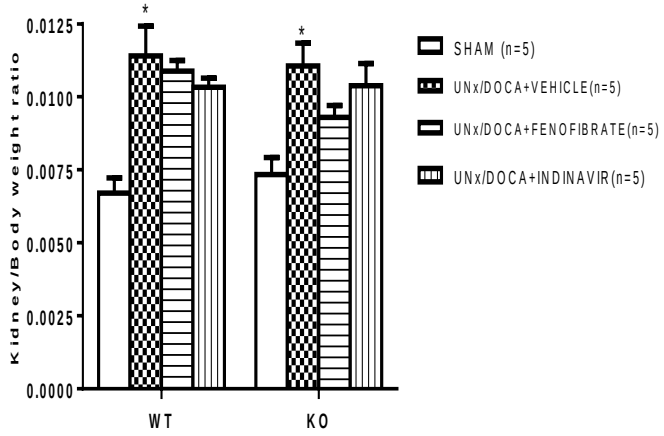


Fig. 2 Kidney weight index in uninephrectomised PPARα KO or WT mice treated with DOCA/1% NaCl/0.1% KCl and fenofibrate (100 mg/kg, p.o) or indinavir (20 mg/kg, s.c) for 21 days. ($^{*}p<0.05$ vs sham WT mice). Vehicle was 2% carboxymethylcellulose/mineral oil for fenofibrate and indinavir respectively. Abbreviations: UNx, uninephrectomised; KO, knockout; WT, wildtype; IND, indinavir; DOCA, deoxycorticosterone acetate; PPAR, peroxisome proliferator activated receptor.

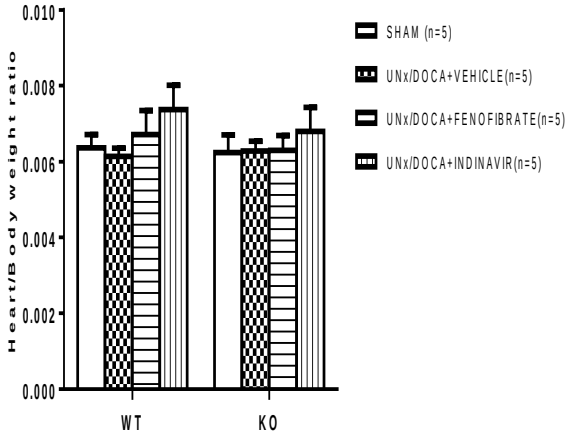


Fig. 3 Heart weight index in uninephrectomised PPARα KO or WT mice treated with DOCA/1% NaCl/0.1% KCl and fenofibrate (100 mg/kg, p.o) or indinavir (20 mg/kg, s.c) for 21 days. ($^{*}p<0.05$ vs sham WT mice). Vehicle was 2% carboxymethylcellulose/mineral oil for fenofibrate and indinavir respectively. Abbreviations: UNx, uninephrectomised; KO, knockout; WT, wildtype; IND, indinavir; DOCA, deoxycorticosterone acetate; PPAR, peroxisome proliferator activated receptor.

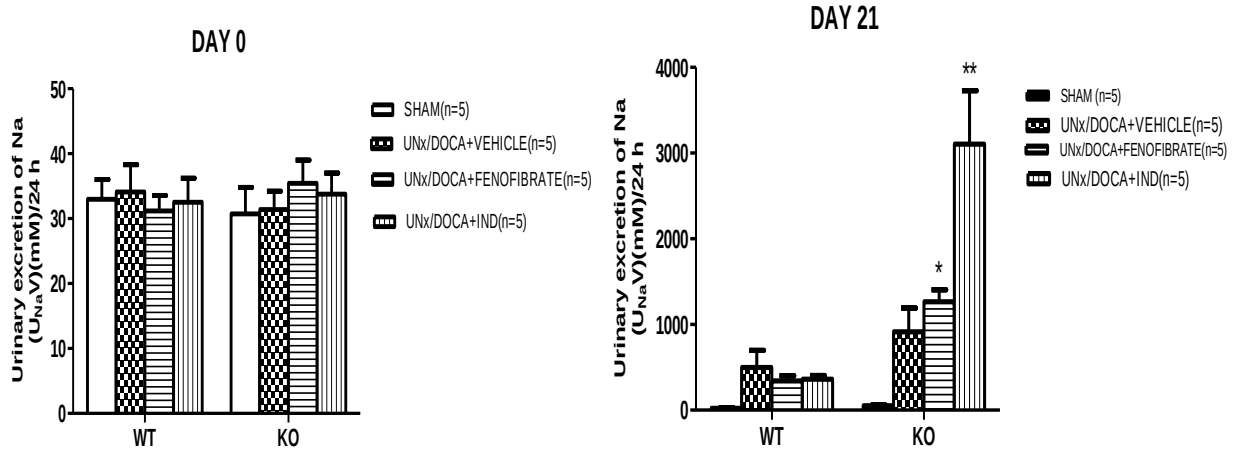


Fig. 4 Urinary Na excretion in uninephrectomised PPARα KO or WT mice treated with DOCA/ 1% NaCl/0.1 % KCl and fenofibrate (100 mg/kg, p.o) or indinavir (20 mg/kg, s.c). ($^{*}p<0.05$, $^{**}p<0.01$ vs WT mice). Vehicle was 2% carboxymethylcellulose/mineral oil for fenofibrate and indinavir respectively. Abbreviations: UNx, uninephrectomised; KO, knockout; WT, wildtype; IND, indinavir; DOCA, deoxycorticosterone acetate; PPAR, peroxisome proliferator activated receptor.

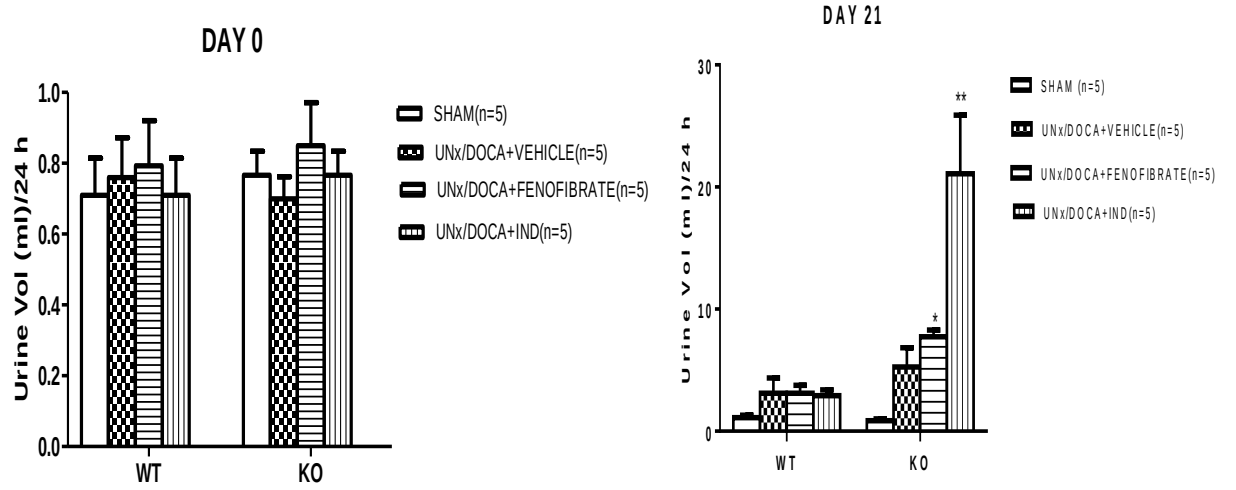


Fig. 5 Urine volume in uninephrectomised PPARα KO or WT mice treated with DOCA/1% NaCl/0.1% KCl and fenofibrate (100 mg/kg, p.o) or indinavir (20 mg/kg, s.c). ($^{*}p<0.05$ $^{**}p<0.01$ vs WT mice). Vehicle was 2% carboxymethylcellulose/mineral oil for fenofibrate and indinavir respectively. Abbreviations: UNx, uninephrectomised; KO, knockout; WT, wildtype; IND, indinavir; DOCA, deoxycorticosterone acetate; PPAR, peroxisome proliferator activated receptor.

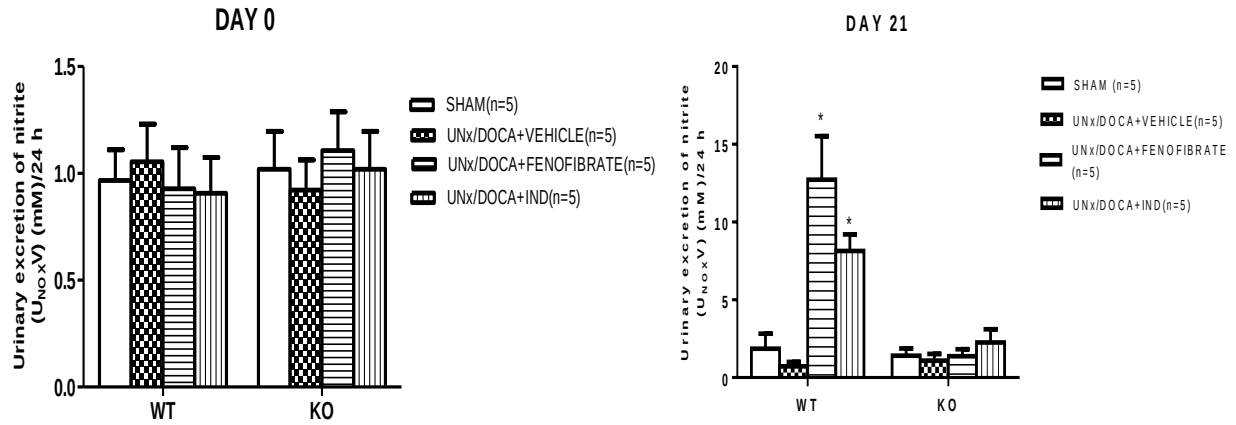


Fig. 6 Urinary NO excretion in uninephrectomised PPAR-α KO or WT mice treated with DOCA/1% NaCl/0.1% KCl and fenofibrate (100 mg/kg, p.o) or indinavir (20 mg/kg, s.c). ($^{*}p<0.05$ vs KO mice). Vehicle was 2% carboxymethylcellulose/mineral oil for fenofibrate and indinavir respectively. Abbreviations: UNx, uninephrectomised; KO, knockout; WT, wildtype; IND, indinavir; DOCA, deoxycorticosterone acetate; PPAR, peroxisome proliferator activated receptor.

## DISCUSSION

In the cardiac-specific PPARα-overexpressing mouse, the expression of genes involved in cardiac fatty acid uptake and oxidation was increased, whereas the expression of genes involved in glucose transport (GLUT4) and utilization was repressed (Finck et al, 2002; Huss and Kelly, 2004). Although, PPARα activation was shown to lower AII-induced elevated blood pressure (Diep et al, 2002a), in some models such as the DOCA-salt hypertensive rat, protection from hypertensive injury occurred in response to PPARα activation in the absence of significant BP lowering (Hou et al, 2010). In the DOCA/salt model of hypertension, the combination of DOCA/salt with unilateral nephrectomy results in hypertension, renal hypertrophy, and nephrosclerosis (Sellye, 1942; Newaz et al, 2002). These previous studies demonstrated both hypertension and nephropathy using this model and that the time course of the nephropathy used in the present study is consistent with the previous studies. Nephropathy, as evaluated by proteinuria, was greater in DOCA/salt-treated PPARα KO mice compared with DOCA/salt-treated WT mice, suggesting that PPARα may be function as a renoprotective molecule. These data are consistent with observations in other studies in which the course of hypertension and the attendant renal injury was exacerbated in DOCA/salt hypertensive PPARα KO mice (Newaz et al, 2002). Fenofibrate, a PPARα activator prevented renal injury as manifested by the decreased proteinuria in DOCA/salt-treated WT mice compared to their KO littermates over the course of the study. This observation was congruent to the known renoprotective effects of PPARα (Hou et al, 2010; Tanaka et al, 2011). In this study, on day 21, GLUT4 inhibition worsened renal injury as seen in the increased proteinuria in the PPARα KO mice compared to its WT littermates, suggesting that GLUT4 renoprotective role may be due to the presence of PPARα receptors. It is known that both PPARα and GLUT4 stimulation exert renoprotective effects independently through increased nitric oxide production (Goya et al, 2004; Yakubu et al, 2010). There were no differences in body weight during the administration period between the groups (results not shown). Indinavir did not affect kidney weight index between (DOCA/salt indinavir-treated) WT and KO mice suggesting that though GLUT4 appears to prevent worsening of renal injury in the presence of PPARα, but it does not prevent renal hypertrophy. High oxidative stress and inflammation are major mediators of progressive renal injury and PPARα activators have been shown to alleviate renal injury via their anti-inflammatory action (Delerive et al, 2001; Marx et al, 2004). Based on the prominent distribution of PPARα in the kidney, PPARα is likely to affect renal transport (Braissant et al, 1996; Guan et al, 1997; Yang et al, 1999). In our experiments, we sought to evaluate the interactions between PPARα and GLUT4 in the kidney in response to manipulations that impact renal injury. Comparing $U_{Na}V$ was higher in DOCA/salt treated KO mice implying that PPARα play a role in enhancement of Na reabsorption in the nephron. The premise was that in the absence of the PPARα gene, the loss of its renal protective effect will render the kidneys more injury prone and interfere with the normal reabsorptive capacity of the kidney leading to increased $Na^+$ excretion (Seylle, 1942; Newaz et al, 2004). Our data are incongruent with previous reports in the rat showing that PPARα ligands inhibited $Na^+$ transport in the proximal tubule of the rat (Newaz et al, 2004) and that clofibrate improved pressure natriuresis in Dahl salt-sensitive rats (Allonso-Galicia et al, 1998). It appears that this is a species-dependent effect. On Day 21, fenofibrate group produced a significant increase in $U_{Na}V$ in KO mice when compared to its WT littermates suggesting that fenofibrate may be having antinatriuretic effect. In the indinavir treated KO mice, there was a significant increase in $U_{Na}V$ compared to the WT mice. This increase in $U_{Na}V$ in indinavir-treated KO mice was greater that of the DOCA/salt and fenofibrate treated KO mice suggesting that GLUT4 may be enhancing the role of PPARα in increasing sodium reabsorption in the renal tubule. The increase in urine volume in indinavir-treated group compared to fenofibrate and DOCA/salt groups correlates with the resultant increase in urinary sodium excretion in KO mice compared to its WT littermates, thus suggesting that GLUT4 activation will enhance sodium reabsorption and this effect may be enhanced in the presence of PPARα receptors.

PPARα modulates nitric oxide synthase (NOS)-induced NO production. PPARα ligands enhance NOS expression and NO release, implying a vasculoprotective effect (Goya et al, 2004). Thus we evaluated the role of GLUT4 in modulating PPARα-mediated production of nitric oxide in DOCA/salt treated WT and KO mice. On Day 21, $U_{NOX}V$ was increased significantly in fenofibrate treated WT mice suggesting that activation of PPARα will enhance NO production. Indinavir-treated group showed increased $U_{NOX}V$ in WT mice, suggesting that GLUT4 maybe enhancing the PPARα receptor induced increase in NO production. In conclusion, GLUT4 exerts a renoprotective role and this effect is enhanced in the presence of PPARα receptors and this renoprotective effect seems to be related to in nitric oxide production.

**Acknowledgement**
This study was supported by National Institutes of Health grant HL03674. The facilities of the RCMI program at Texas Southern University were used for this study.

## REFERENCES

Alonso-Galicia, M., B. Frohlich, and R.J. Roman. (1998). Induction of P4504A activity improves pressure natriuresis in Dahl S rats. *Hypertension* 31 (1); 232–236

Braissant, O., F. Foufelle, C. Scotto, M. Dauça, and W. Wahli. (1996). Differential expression of peroxisome proliferator-activated receptors (PPARs): tissue distribution of PPAR-*α*, -*β*, and -*γ* in the adult rat. *Endocrinology* 137(1):354–366.

Diep, Q.N., F. Amiri, R.M. Touyz, J.S. Cohn, D. Endemann, and E.L. Schiffrin. (2002). PPARα activator effects on Ang II-induced vascular oxidative stress and inflammation. *Hypertension* 40:866–871

Finck, B.N., C. Carlos Bernal-Mizrachi, D.H. Han, T. Coleman, N. Sambandam, L.L, LaRiviere, J.O. Holloszy, C.F. Semenkovich, and D.P. Kelly. (2005). A potential link between muscle peroxisome proliferator-activated receptor-α signalling and obesity-related diabetes. *Cell metabolism* 1(2): 133-144

Finck, B.N., J.J Lehman, T.C. Leone, M.J. Welch, M.J. Bennett, A. Kovacs, X. Han, R.W. Gross, R. Kozak, G.D. Lopaschuk, and D.P. Kelly. The cardiac phenotype induced by PPARα overexpression mimics that caused by diabetes mellitus. *J. Clin. Invest.* 109: 121–130.

Goya, K., S. Sumitani, X. Xu, T. Kitamura, H. Yamamoto, S. Kurebayashi, H. Saito, H. Kouhara, S. Kasayama, and I. Kawase. (2004). Peroxisome proliferator activated receptor alpha agonists increase nitric oxide synthase expression in vascular endothelial cells. *Arterioscler. Thromb. Vasc. Biol*. 24 (4): 658–663

Guan, Y., Y. Zhang, L. Davis, and M.D. Breyer. (1997). Expression of peroxisome proliferator activated receptors in urinary tract of rabbits and humans. *Am. J. Physiol.* 273 (6): F1013–F1022.

Hou, X., Y.H. Shen, C. Li, F. Wang, C. Zhang, P. Bu, and Y. Zhang. (2010). PPAR alpha agonist fenofibrate protects the kidney from hypertensive injury in spontaneously hypertensive rats via inhibition of oxidative stress and MAPK activity. *Biochem Biophys Res Commun*.394(3):653-659

Huss, J.M., and D.P. Kelly. (2004). Nuclear receptor signaling and cardiac energetics. *Circ Res* 95:568–578.

Igbe, I., E.K.I., Omogbai, and A.O. Oyekan. (2013). Role of GLUT4 on angiotensin 2-induced systemic and renal hemodynamics *J Exp Pharmacol* 5:1 – 13.

Iglarz, M., R.M. Touyz, F. Amiri, M.F. Lavoie, Q.N. Diep, and E.L. Schiffrin. (2003). Effect of peroxisome proliferator-activated receptor-α and-γ activators on vascular remodeling in endothelin-dependent hypertension. *Arterioscler Thromb Vasc Biol* 23: 45–51.

Irukayama-Tomobe, Y., T. Miyauchi, Y. Kasuya, S. Sakai, K. Goto, and I. Yamaguchi. (2004). Activation of Peroxisome Proliferator-activated Receptor-alpha Decreases Endothelin-1-induced p38 Mitogen-activated Protein Kinase Activation in Cardiomyocytes. *J. Cardiovasc Pharmacol* 44:S358–S361.

Legendre C., E Causé, E. Chaput, R. Salvayre, T. Pineau, and A.D. Edgar. (2002). Fenofibrate induces a selective increase of protein-bound homocysteine in rodents: a PPARα-mediated effect. *Biochem. Biophys. Res. Comm.* 295(5):1052-1056

Marx, N., H. Duez, J.C. Fruchart, and B. Staels. (2004). Peroxisome Proliferator-Activated Receptors and Atherogenesis: Regulators of Gene Expression in Vascular Cells. *Circ. Res.* 94:1168–1178

Newaz, M.A., K. Ranganna, and A.O. Oyekan. (2004). Relationship between PPARα activation and NO on proximal tubular Na+ transport in the rat. *BMC Pharmacology*. 6: 1-9

Newaz, M.A., Z. Yousefipour, and A.O. Oyekan. (2002). Effects of Pergolide on Blood Pressure and Tissue Injury in DOCA-salt Hypertension. *Blood pressure* 11: 110-115

Delerive, P., J.C. Fruchart, and B. Staels. (2001). Peroxisome proliferator-activated receptors in inflammation control. *J. Endocrinol.* 169: 453-459

Sellye, H. (1942). Production of nephrosclerosis by overdosage with deoxycorticosterone acetate. *Can. Med. Assoc. J.* 47: 515–519.

Tanaka. Y., S. Kume, S. Araki, K. Isshiki, M. Chin-Kanasaki, M. Sakaguchi, T. Sugimoto, D. Koya, M. Haneda, A. Kashiwagi, H. Maegawa, and T. Uzu. (2011). Fenofibrate, a PPARα agonist, has renoprotective effects in mice by enhancing renal lipolysis. *Kidney Int*. 29, (8):871-82.

Yakubu, M.A., R.H. Nsaif, and A.O. Oyekan. (2010). Regulation of cerebrovascular endothelial peroxisome proliferator activator receptor alpha expression and nitric oxide production by clofibrate. *Bratisl Lek Listy*.111(5):258-64.

Yang, T., D.E. Michele, and J. Park. (1999). Expression of peroxisomal proliferator-activated receptors and retinoid X receptors in the kidney. *Am. J. Physiol.* 277 (6): F966–F973.

Ziouzenkova, O., and J. Plutzky. (2004). Lipolytic PPAR activation: new insights into the intersection of triglycerides and inflammation? *Curr. Opin. Clin. Nutr. Metab. Care* 7 (4): 369–75.