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Glycemic control and electrolyte changes in diabetic rats treated with pioglitazone

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ABSTRACT

This study evaluated the effect of pioglitazone on glucose and serum electrolytes in normal and diabetic rats. Fifteen adult wister rats were divided into 3groups (n=5). Group 1 and 2 served as the normal control and diabetic untreated group respectively and each received 0.3ml distilled water. Group 3 served as diabetic treated with 4mg/kg pioglitazone. This study lasted for twelve days after which blood samples were collected under mild anesthesia through retro-orbital sinus and centrifuge to obtain serum for analysis. The weight and fasting blood glucose level of rats were determined using the weighing scale and glucose oxidase method respectively. The serum electrolytes (Na^+, K^+, and Ca^{2+}) were determined using the flame photometry method. Results showed significant increase in ($p<0.05$) in weight of pioglitazone treated group when compared with the normal control and diabetic untreated group. The average weight gain in pioglitazone treated group (29.80%) was significantly higher when compared with previous studies (5%). There was significant decrease ($p<0.05$) in fasting blood glucose level in pioglitazone treated group when compared with diabetic untreated group. Na^+ concentration was significantly higher ($p<0.05$) in pioglitazone treated group when compared with diabetic untreated group. A significant increase ($p<0.05$) in Ca^{2+} concentration was observed in pioglitazone treated group when compared with normal control and diabetic untreated groups. In conclusion, oral administration of pioglitazone caused weight gain, hypernatremia, hypercalcemia and a decrease fasting blood glucose level. Pioglitazone is therefore useful in glycemic control and maintainance of electrolyte balance in diabetic mellitus.

Keywords: Pioglitazone, glycemic control, hypernatremia, hypercalcemia, serum electrolytes.

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1.0 INTRODUCTION

Diabetes mellitus is a serious and chronic disease that occurs either when the pancreas does not produce enough insulin, or when the body cannot effectively use the insulin it produces (1). Hyperglycemia is a common effect of uncontrolled diabetes and may over time, lead to serious damage to the heart, blood vessels, eyes, kidneys and nerves. Recently, more than 400 million people have been diagnosed with diabetes and this value has been projected to increase in the nearest future (1). Therefore, the need to provide immediate treatment against diabetes cannot be overemphasized. Meanwhile, most types of diabetes mellitus cause electrolyte derangement which is often associated with increase morbidity and mortality in the affected population (2).

Hyponatremia is a decrease in serum Na^+ concentration which is often observed in uncontrolled hyperglycemia and it may results in cognitive impairment, osteoporosis and fractures (3). Since, glucose is an osmotically active substance, the hyperglycemia which occur in diabetes increases serum osmolality, resulting in movement of water out of the cells and subsequently causing a reduction of serum sodium concentration (Na^+) by dilution (dilutional hyponatremia). In diabetes mellitus, hypokalemia may also result due to redistribution of potassium K^+ from the extracellular to the intracellular fluid compartment, gastrointestinal loss of K^+ due to malabsorption syndromes (diabetic-induced motility disorders, bacterial overgrowth, chronic diarrhea states) and renal loss of K^+ due to osmotic dieresis (4). Accumulating evidences have also suggested that altered calcium homeostasis may play a role in the development of diabetes mellitus (5). Bischoff-Ferrari et al., 2006 (6) have shown that high intake of calcium, particularly from supplements result in hypercalcemia which lowers the risk of diabetes by 33% in his study population. This implies that Ca^{2+} is important in preventing the development of diabetes mellitus. The mechanism may involve a change in potential difference across voltage-gated calcium channels present on cell membrane of the pancreatic beta cells. Calcium ions then diffuse from outside the cell down their concentration gradients into the cell and therefore, cause vesicles containing insulin to move to, and fuse with, the cell surface membrane, releasing insulin by exocytosis which is utilized for glucose uptake by the cells (7).

Among the class of drugs used in the treatment of diabetes mellitus is Thiazolidinediones (TZDs) which is orally administered in diabetic patients. Pioglitazone, a thiazolidinedione class of drug is usually prescribed to manage diabetes mellitus. It is known to acts by stimulating the nuclear receptor peroxisome proliferator-activated receptor gamma (PPAR-γ) and to a lesser extent PPAR-α (8). It modulates the transcription of genes involved in the control of glucose and lipid metabolism in the muscle, adipose tissue and the liver. Pioglitazone also reduces insulin resistance in liver and peripheral tissues, decreases gluconeogenesis in the liver and reduces quantity of glucose and glycated haemoglobin in the blood stream (9).

There are controversial reports on the role of pioglitazone in the treatment of electrolyte derangements which occur in diabetes mellitus. Therefore, the purpose of this study was to ascertain the role of pioglitazone by investigating its effects on body weight changes, glucose level and serum electrolytes in diabetic wister rats.

2.0 MATERIALS AND METHODS

2.1 Animal grouping: Fifteen male Wistar rats weighing between 200-270gm were obtained from the Central Animal House, College of Medicine, University of Medical Sciences, Ondo. They were housed in well aerated cages, fed on standard rat chow and allowed free access to drinking water according to the guidelines and regulations of the National Institute of Health (10) and approved by the University of Medical Sciences Animal care committee. Pioglitazone Hydrochloride Tablets was purchased from Pharmaceutical Works Limited, Gujarat, India .The animals were divided into 5 groups of 5 rats per group. Group 1 served as control, group 2 served as diabetic untreated and group 3 was treated with 4mg/kg pioglitazone (p.o) for 12 days (11).

2.2 Induction of diabetes

Diabetes was induced in group 2 and 3 following single intra-peritoneal injection of alloxan monohydrate (Sigma Aldrich, U.S.A) at a dose of 120mg/kg using the method of Carvalho (12). After 72hours of alloxan administration, only rats with fasting blood glucose level of 250mg/kg and above were considered and selected for this study

2.3 Blood glucose and weight determination

Blood samples were obtained from the tail vein every four days (days 0 before treatment, 4, 8 and 12 post-treatment) for determination of blood glucose level. Blood glucose level was measured using the principle of glucose oxidase method as described by Trinder 1969 (13). The weights of rats in each group were determined using electronic weighing balance (WSB-8000 Omega, Taiwan) (14).

2.4 Serum electrolytes determination

After 12 days post-treatment with pioglitazone, rats were mildly exposed to sodium pentobarbital anesthesia (30mg/kg i.p) and blood samples were obtained from each rat through retro-orbital sinus for assessing the level of serum electrolytes. Serum sodium, potassium and calcium, were determined using flame photometry method as described by Hayder, 2016 (15) where compounds of metals are thermally separated into atoms at the temperature of a Bursen burner flame and some of the atoms delivered are further eager to a higher vitality level. When these energized atoms come back to the ground state, they transmit radiation, which lies principally in the visible region of the electromagnetic range

2.5 Statistical analysis

Results obtained were analyzed using ANOVA and Neuman's keul post-hoc test. Data were expressed as mean ±SEM with the level of statistical significance taken at $p<0.05$.

3.0 RESULTS

3.1 Effect of pioglitazone on body weights changes in experimental rats

Table 1 shows weight changes in the control, diabetic untreated and pioglitazone treated rats. There was significant increase ($p<0.05$) in weight at days 4, 8 and 12 in pioglitazone treated group when compare with diabetic untreated group. The percentage weight gains are 19.71%, 28.63%, and 41.32% respectively. These percentage weight gains double at day 12 (41.32%) when compared with the value at day 4 (19.71%) of pioglitazone administration. There was significant increase ($p<0.05$) in weight in pioglitazone treated group at days 4, 8 and 12 when compared with the normal control. The percentage weight gains include 16.94%, 22.76% and 26.54%.

There was no significant difference in weight of the normal control at various time intervals when compared with the initial value. There was significant decrease ($p<0.05$) in weight of diabetic untreated group at day 12 when compared with the initial value. There was significant increase ($p<0.05$) in weight at day 12 in pioglitazone treated group when compared with the initial value before treatment. At day 12, the percentage weight loss in diabetic untreated group is 26.25% while the percentage weight gain in the pioglitazone treated group is 27.97%.

3.2 Effect of pioglitazone on fasting blood glucose in experimental rats

Figure 1 showed changes in fasting blood glucose level in the control, diabetic untreated and pioglitazone treated rats. There was significant increase ($p<0.05$) in fasting blood glucose level at 72hours after diabetes induction, day 4, day 8 and day 12 in pioglitazone treated group when compared with the normal control group. However, there was significant decrease ($p<0.05$) in fasting blood glucose level at day 4, day 8 and day 12 in pioglitazone treated group when compared with the diabetic untreated group.

There was no significant difference in fasting blood glucose in diabetic untreated group at day 12 when compared with the value at 72 hours after diabetes induction. However, there was significant decrease ($p<0.05$) in fasting blood glucose level in pioglitazone treated group at day 8 and 12 when compared with the value at 72 hours after diabetes induction.

3.3 Effect of pioglitazone on sodium ion

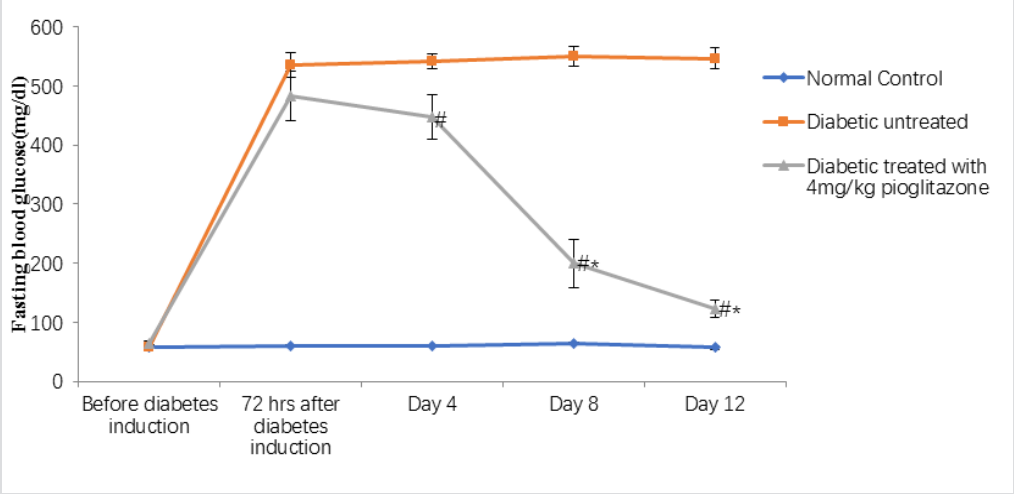

Figure 1: Effect of pioglitazone on fasting blood glucose in experimental rats. Data were expressed as Mean ± Standard Error of mean. Ash (#) indicates significant difference at $^{\#}$= P<0.05, when compared with normal control. Asterisk indicates significant difference at *= P<0.05, when compared with the diabetic untreated (n = 5).

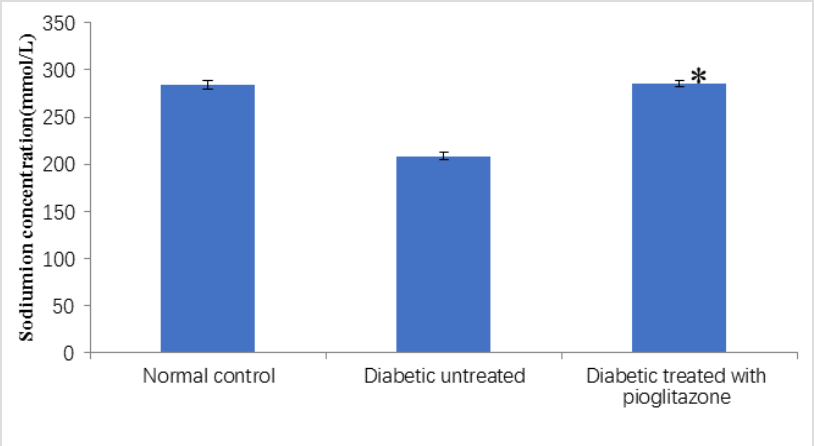

Figure 2: Effect of pioglitazone on sodium ion concentrations in experimental rats. Data were expressed as Mean ± Standard Error of mean. Asterisk indicates significant difference at *= P<0.05, when compared with the diabetic untreated (n = 5).

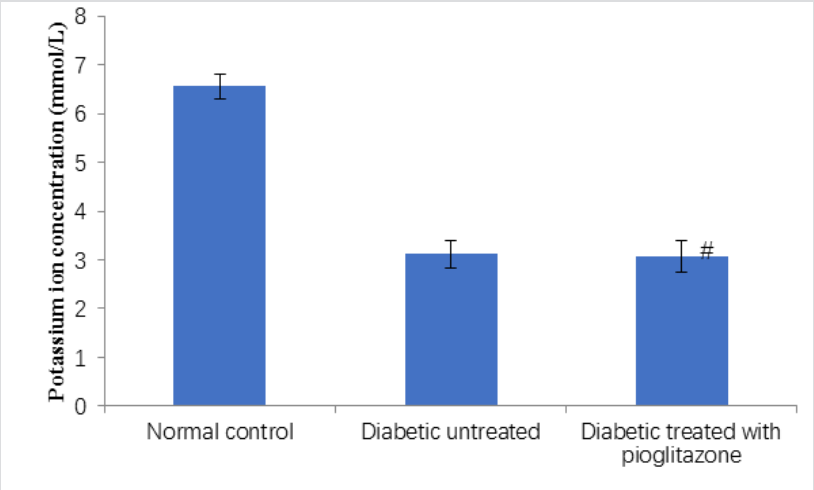

Figure 3: Effect of pioglitazone on potassium ion concentrations in experimental rats. Data were expressed as Mean ± Standard Error of mean. Ash (#) indicates significant difference at $^{\#}$= P<0.05, when compared with normal control (n = 5).

concentration in experimental animals

Figure 2 showed changes in sodium ion concentration in the control, diabetic untreated and pioglitazone treated rats. There was significant increase ($p<0.05$) in sodium ion concentration in piolitazone treated group when compared with diabetic untreated group. The percentage increase in sodium ion concentration is 36.70%. However, there was no significant difference in sodium ion concentration of pioglitazone treated group when compared to the normal control group.

3.4 Effect of pioglitazone on potassium ion concentration in experimental animals

Figure 3 showed changes in potassium ion concentration in the control, diabetic untreated and pioglitazone treated rats. There was no significant difference in potassium ion concentration in pioglitazone treated group when compared with diabetic untreated group. However, there was significant decrease ($p<0.05$) in potassium ion concentration in pioglitazone treated group when compared with the normal control with a percentage decrease of 52.42%

3.5 Effect of pioglitazone on calcium ion concentration in experimental animals

Figure 4 showed weight changes in the control, diabetic untreated and pioglitazone treated rats. There was significant increase ($p<0.05$) in calcium ion concentration in pioglitazone treated group when compared with diabetic untreated and normal control groups. The percentage increases in calcium ion concentrations are 218.18% and 174.33% respectively

4.0 Discussion and Conclusion

This study investigates the effect of pioglitazone on body weight, blood glucose level and changes in serum electrolytes in normal and diabetic wister rats. Diabetes mellitus is a metabolic disorder associated with significant weight loss (16) however, the significant weight gain observed from our study following pioglitazone administration in diabetic rats may be due the ability of pioglitazone to stimulate adipocytes differentiation and fat redistribution in the visceral and subcutaneous tissues of the body (17). This finding is consistent with the reports of Miyazaki et al., 2002 (18) who had also earlier reported weight gain in diabetic humans after pioglitazone administration. However, the average weight gain observed from this study is 29.80% which is in contrast with the findings of Miyazaki et al., 2002 (18) who reported a 5% increase in body weight after treatment with pioglitazone.

Therapies that provide strict glycemic control have remained the target in the treatment of diabetes mellitus (19). Therefore, the significant decrease in fasting blood glucose level in pioglitazone treated group observed from this study indicates that pioglitazone is acting by improving glucose tolerance. This effect of pioglitazone may be mediated by causing the regeneration of islets cells of langerhaan to produce insulin or improving the uptake of glucose in the peripheral tissues and the liver (20)

In uncontrolled diabetes mellitus, the serum concentration of Na^+ is variable reflecting the balance between hyperglycemia-induced water movement out of the cells that lower Na^+ concentration and glucosuria-induced osmotic diuresis which tend to raise Na^+ concentration (21). The significant increase in Na^+ concentration observed in pioglitazone treated group from our study may be due to its ability to stimulate glucose excretion by increase the rate of filtration and preventing the reabsorption of glucose in the renal tubular cells of the kidneys (22). Glucose being an osmotically active substance may therefore cause more water to be excreted through the kidney therefore, elevating the concentration of Na^+ in the blood serum.

Studies have shown that the incidence of hypokalemia in diabetes mellitus is higher than in general population (23). This hypokalemia may be due to the redistribution of k^+ from the extracellular to intracellular fluid compartment, gastrointestinal loss due to malabsorption syndromes or K^+ loss through osmotic diuresis (4). From this study, pioglitazone administration does not affect K^+ concentration in diabetes mellitus and this implies that pioglitazone may not be acting through the pathways that ameliorate hypokalemia.

The significant decrease ($p<0.05$) in Ca^{2+} con-

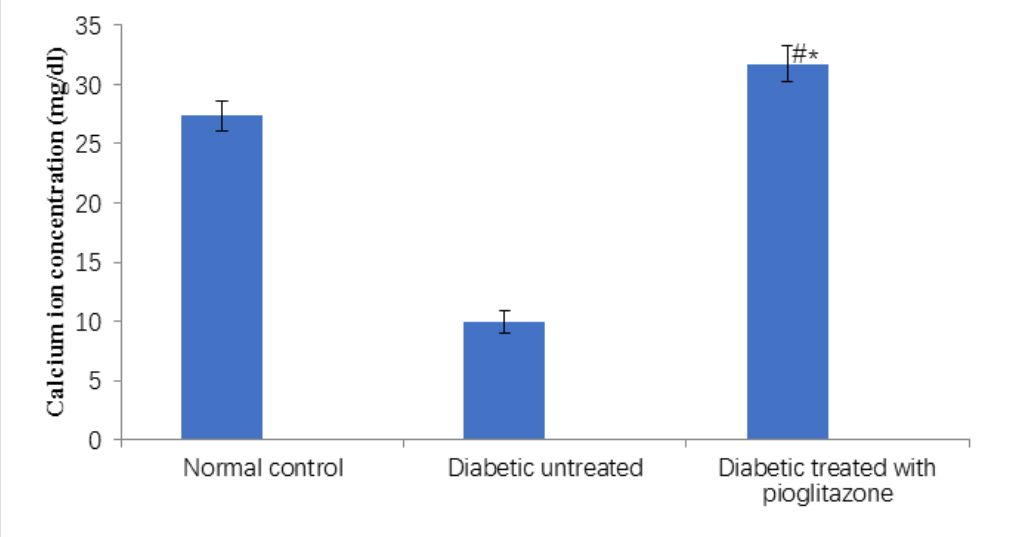

Figure 4: Effect of pioglitazone on potassium ion concentrations in experimental rats. Data were expressed as Mean ± Standard Error of mean. Ash (#) indicates significant difference at $^{\#}$ = P<0.05, when compared with normal control. Asterisk indicates significant difference at * = P<0.05, when compared with the diabetic untreated (n = 5).

Table 1: Effect of pioglitazone on body weights changes in experimental rats

Group	Initial weight before treatment (g)	Day 4 (g)	Day 8 (g)	Day 12 (g)
Normal control (0.3ml distilled water)	221.00±3.75	223.00±3.78	226.60±3.60	228.60±3.27
Diabetic untreated (0.3ml distilled water)	247.60±10.76	215.80±11.36	209.40±10.87	182.60±9.06*
Diabetic treated with pioglitazone (4mg/kg p.o)	243.80±16.26	268.80±14.31$^{\#}$	293.4±13.79$^{\#}$	311.20±10.19*$^{\#}$

Data were expressed as Mean ± Standard Error of mean. Ash (#) indicates significant difference at $^{\#}$ = P<0.05, when compared with normal control and diabetic untreated. Asterisk indicates significant difference at * = P<0.05, when compared with the initial value before treatment (n = 5).

centration in diabetic untreated rats suggests that less calcitonin and parathyroid hormone is produced from the parafollicular cell and parathyroid gland respectively. However, the significant increase ($p<0.05$) in Ca^{2+} after pioglitazone administration implies that more calcitonin was released into circulation to stimulate the activities of oesteoclasts in causing the release of Ca^{2+} from bone cells (24). Pioglitazone may also be acting by increasing the release of parathyroid hormone which inhibits Ca^{2+} excretion through the kidney hence increasing circulating Ca^{2+} (25). The secreted parathyroid hormone may also be acting by stimulating the production of 1, 25 dihydroxycolicalciferol which is necessary for intestinal absorption of Ca^{2+} (26). The Ca^{2+} concentration in pioglitazone treated group was significantly higher ($p<0.05$) than the normal control which implies that pioglitazone may be very effective in maintaining calcium homeostasis.

In conclusion, oral administration of pioglitazone causes weight gain in the diabetic rats although the percentage weight gain was higher in this study when compared with previous studies. Pioglitazone administration at 4mg/kg also causes a decrease in fasting blood glucose level which helps in ameliorating the hyperglycemia that was observed in the diabetic rats. Finally, pioglitazone causes hypernatremia and hypercalcemia which are important in maintaining electrolyte balance in the diabetic rats.

5.0 References

1. World Health Organization Guideline:sugars intake in adults and children. Geneva 2016.

2. Liamis G, Rodenburg EM, Hofman A, Zietse R, Stricker BH, Hoorn EJ. Electrolyte disorders in community Subjects: prevalence and risk factors. AM J Med, 2013; 12(6):256-263

3. Podesta MA, Faravelli I, Cucchiari D, Reggiani F, Oldani S. Neurological counterparts of hyponatremia: pathological mechanisms and clinical manifestations. Curr Neurol Neurosci Rep 2015;15: 18.

4. Yang L, Frindt G, Palmer LG. Magnesium modulates ROMK channel-mediated potassium secretion. J Am Soc Nephrol, 2010; 21: 2109-2116

5. Mathieu C, Badenhoop K. Vitamin D and type 1 diabetes mellitus: state of the art. Trends Endocrinol Metab. 2005; 16:261–266

6. Bischoff-Ferrari HA, Giovannucci E, Willett WC, Dietrich T, Dawson-Hughes B. Estimation of optimal serum concentrations of 25-hydroxyvitamin D for multiple health outcomes. Am J Clin Nutr. 2006; 84:18–28

7. Lang V, Light PE. “The molecular mechanisms and pharmacotherapy of ATP-sensitive potassium channel gene mutations underlying neonatal diabetes.”. Pharmgenomics. Pers. Med.2010; 3: 145–61.

8. Gillies PS, Dunn CJ Pioglitazone, Drugs. 2000 60(2): 333-43

9. Smith U. Pioglitazone mechanism of action Int J Clin Pract Suppl, 2001; (121) : 8-13

10. National Institute of Health (1985). Guide for the Care and Use of Laboratory Animals. NIH publication 85 – 23.

11. El Gawly HW, The effect of pioglitazone on liver of streptozotocin – induced diabetic albino wister rats. Eur Rev Med Pharmacol Sci. 2009; 13 (6):443-51

12. Carvalho EN, Carvalho NAS, Ferreira LM. Experimental model of induction of diabetes mellitus in

rats. Acta Cir Bras. 2003 (18) Special Edition. Available on URL:http://www.scielo.br/acb.

13. Trinder P. Determination of blood glucose using an oxidase –peroxidase system with a non carcinogenic chromogen. J Clin Pathol. 1969 22(2):158-61

14. Leibowitz KL, Chang GQ, Pamy PS, Hill JO, Gayles EC, Leibowitz SF, Weight gain model in prepubertal rats: prediction and phenotyping of obesity-prone animals at normal body weight International Journal of Obesity. 2007 1–12

15. Hayder Yahya Ahmed. Estimation of Serum Electrolytes for examination between Flame photometry and Ion Selective Electrodes. Int. Journal of Engineering Research and Application. 2016, 6(10), (52-54)

16. Luc Van Gaal and Andre Scheen. Weight Management in Type 2 Diabetes: Current and Emerging Approaches to Treatment Diabetes Care, 2015; 38:1161–1172.

17. Hermansen K, Mortensen LS. Bodyweight changes associated with antihyperglycaemic agents in type 2 diabetes mellitus. Drug Saf. 2007; 30:1127–1142

18. Miyazaki Y, Mahankali A, Matsuda M, Effect of

pioglitazone on abdominal fat distribution and insulin sensitivity in type 2 diabetic patients. J Clin Endocrinol Metab 2002;87: 2784–2791

19. Kahn SE, Cooper ME, Del Prato S. Pathophysiology and treatment of type 2 diabetes: perspectives on the past, present, and future. Lancet 2014; 383:1068–1083

20. Wilding J. Thiazolidinediones, insulin resistance and obesity: finding a balance. Int J Clin Pract; 2006; 60:1272–1280

21. Liamis G, Tsimihodimos V, Doumas M, Spyrou A, Bairaktari E, Elisaf M. Clinical and laboratory characteristics of hypernatraemia in an internal medicine clinic. Nephrol Dial Transplant. (2008) 23:136–143

22. Chiasson JL, Aris-Jilwan N, Bélanger R, Bertrand S, Beauregard H, Ekoé JM, Fournier H, Havrankova J. Diagnosis and treatment of diabetic ketoacidosis and the hyperglycemic hyperosmolar state. CMAJ, 2003; 168:859–866

23. Janko O, Seier J, Zazgornik J. Hypokalemia--incidence and severity in a general hospital Wien Med Wochenschr. 1992;142 (4):78-81.

24. Kameda T, Mano H, Yamada Y, Takai H, Amizuka N, Kobori M, Izumi N, Kawashima H, Ozawa H, Ikeda K, Kameda A, Hakeda Y, Kumegawa M. Calcium-sensing receptor in mature osteoclasts, which are bone resorbing cells. Biochem Biophys Res Commun.1999; 245:419-422.

25. Sammons, J., Ahmed, N., El-Sheemy, M., and Hassan, H.T “The role of BMP-6, IL-6, and BMP-4 in mesenchymal stem celldependent bone development: effects on osteoblastic differentiation induced by parathyroid hormone and vitamin D3,” Stem Cells and Development, 2004; 13(3) 273–280.

26. Wasserman, R. H., Smith, C. A., Brindak, M. E., de Talamoni, N., Fullmer, C. S., Penniston, J. T. & Kumar, R. Vitamin D and mineral deficiencies increase the plasma membrane calcium pump of chicken intestine. Gastroenterology. 1992; 102:886-894