ORIGINAL ARTICLE

The Gonadal Hormone Regulates the Plasma Lactate Levels in Type 2 Diabetes Treated With and Without Metformin

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Abstract

Objective: Our previous study showed there was a gender difference in plasma lactate concentrations in subjects with type 2 diabetes. This study investigated the effect of sex hormone levels on plasma lactic acid (LA) levels in type 2 diabetes with and without metformin therapy.

Subjects and Methods: Fasting whole blood specimens of 392 type 2 diabetes patients treated with metformin ($n=199$) or not ($n=193$) were collected. LA was measured with an enzyme-electrode assay. Levels of sex hormones, including testosterone (T) and estradiol (E_2), were measured with a chemiluminescence microparticle immunoassay. Spearman's or Pearson's correlation and logistic regression analysis were performed for the factors associated with LA.

Results: The LA level in the metformin group was significantly higher than that in the non-metformin group (1.26 ± 0.43 vs. 1.14 ± 0.49 mmol/L, $P < 0.001$), and LA levels of females were significantly higher than those of males ($P < 0.001$). LA concentrations were positively correlated with E_2 level but negatively correlated with metformin and T levels ($P < 0.01$). The logistic regression analysis showed that gender, creatinine, E_2 , metformin, and T were independent factors influencing lactate levels. Analysis of subgroups demonstrated that the LA concentrations increased with the elevation of E_2 level ($P < 0.05$) but decreased with the rising of T level ($P < 0.05$).

Conclusions: Sex hormones play an important role on regulating plasma lactate levels in diabetes patients treated with metformin. E_2 up-regulates but T tend to down-regulate lactate levels.

Introduction

LACTIC ACID (LA) IS PRODUCED from the process of glucose or glycogen glycolysis. The fatal lactic acidosis is mainly due to lactic acid overproduction or underutilization. Metformin is one of the most commonly used antihyperglycemic drugs for the treatment of type 2 diabetes mellitus (T2DM). The United Kingdom Prospective Diabetes Study had shown that metformin therapy significantly reduced mortality from cardiovascular diseases and all-cause-mortality compared with sulfonylureas or insulin in T2DM patients.¹ The hypoglycemic mechanism of metformin is involved with glycolysis and lactate generation, which leads to elevating levels of blood lactate.² However, reports in recent years have indicated that the incidence of lactic acidosis induced by metformin is rare and far less than that by another biguanide drug, phenformin. Moreover, the lactic acidosis caused by

metformin mostly occurs in diabetes patients with liver and kidney dysfunction, chronic congestive heart failure, and obstructive pulmonary emphysema.³⁻⁶ Nevertheless, our previous results, by comparing serum lactate levels with and without metformin therapy in 1,024 T2DM patients, found that the incidence of hyperlactatemia was significantly increased in the metformin group, and there was a low incidence of hyperlactatemia in T2DM patients of the non-metformin group.^{7,8} Although dysfunction of the liver and kidney could affect plasma lactate concentrations, we further elucidated that plasma lactate levels in female T2DM patients were significantly higher than those in male T2DM patients, with the highest level of lactate in premenopausal women. In other words, there is a gender difference in lactate levels in diabetes.⁹ In recent decades, several animal studies have also revealed that sex hormones regulate the expression of transporter proteins (organic cation transporter 2 [OCT2],

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multidrug and toxin extrusion 1 [MATE1]) related to renal metformin transportation and excretion.^{10,11} However, to our knowledge, no studies related to the relationship between sex hormones and plasma lactate in the T2DM population had been reported. Therefore, this study investigates the potential relationship among estrogen, androgen, and plasma lactate concentrations by comparing the levels of lactate and gonadal hormone in T2DM patients with and without metformin treatment and exploring the possible effect of sex hormones on the metabolism of metformin.

Subjects and Methods

Subjects

In total, 392 patients with T2DM were recruited from the Shanghai Diabetes Center from October 2008 to January 2011, including a group receiving metformin treatment ($n=199$; male:female, 89:110) and another group without metformin treatment ($n=193$; male:female, 84:109). The inclusion criteria were as follows: (1) diagnosed as T2DM by 1999 World Health Organization criteria and American Diabetes Association standards; (2) renal function was normal or only slightly impaired, indicated by a glomerular filtration rate of >60 mL/min and creatinine (Cr) level of <130 μ mol/L; and (3) able to perform physical activity freely. Participants with the following conditions were excluded: (1) chronic liver disease, (2) congestive heart failure, (3) severe chronic obstructive pulmonary disease, (4) alcohol addiction, and (5) chronic diarrhea and innutrition state. The study was approved by the Ethics Committee of the Shanghai 6th People's Hospital. Written informed consents were obtained from all participants. The mean age was 58.24 ± 11.43 years (range, 26–77 years), and the duration after diabetes diagnosis was 7.91 ± 6.61 years (range, 15 days–38 years).

Measurement of plasma lactate concentrations

All patients were administered metformin 500 mg three times daily for at least 1 week. Fasting blood samples were collected in the morning of the third day with the patient resting. All plasma lactate assays were performed with the enzyme-electrode method (Biosen5030 Autocal glucose-lactate analyzer, EKF Diagnostics, Magdeburg, Germany); the normal range for fasting lactate in Chinese people is 0.68–1.96 mmol/L. Hyperlactacidemia was diagnosed when it exceeded 1.96 mmol/L.¹²

Determination of other biochemical indexes

Fasting plasma glucose, glycosylated hemoglobin A1c (HbA1c), indexes representing the function of liver and kidney such as Cr, urea nitrogen, and alanine aminotransferase (ALT), and sex hormones were measured on the day before lactate assay. Fasting plasma glucose was determined with the glucose oxidase method. Blood urea nitrogen, Cr, and ALT were measured on an automatic analyzer (Hitachi 7180 biochemistry automatic analyzer, Hitachi, Tokyo, Japan). HbA1c was detected with a high-performance liquid chromatography assay. Body mass index (BMI) was calculated based on the following formula: $\text{BMI} = \text{body weight (in kg)} / \text{height (in m)}^2$. Gonadal hormones, including estradiol (E_2), progesterone, testosterone (T), follicle-stimulating hormone, luteinizing hormone, and prolactin, were determined with a

chemiluminescent microparticle immunoassay (Architect i2000SR, Abbott Laboratories, Rungis, France).

Statistical analysis

All numerical values were expressed as mean $\pm$ SD. Variables that did not conform to a normal distribution were analyzed after logarithmic transform. Differences between two groups were assessed by two-tailed Student's t test. The rate of hyperlactatemia was compared with the χ^2 test. The blood lactate concentrations of two and more groups were compared by one-way analysis of variance. The Kruskal–Wallis rank sum test was used if the numerical values were in non-normal distribution. Associations between plasma lactate and other variables were assessed with Spearman's or Pearson's correlation coefficient analysis, and logistic regression analysis was performed in the normal group and the hyperlactatemia group. SPSS version 18.0 (SPSS Inc., Chicago, IL) was used for all data analyses. $P < 0.05$ was considered statistically significant.

Results

Baseline clinical characteristics of the two groups

Clinical characteristics of all T2DM patients were shown in Table 1, including the group receiving metformin treatment ($n=199$) and another group without metformin therapy ($n=193$). There was no significant difference in gender, Cr, ALT, and diabetes duration between the two groups. The metformin group had higher BMI and plasma lactate levels than the non-metformin group. The incidence of hyperlactatemia in the metformin group was higher than that in the non-metformin group (7.5% vs. 3.11%, $P=0.025$), but no lactic acidosis cases were found.

Gender differences of plasma lactate concentrations

The plasma lactate level was higher in female patients than in male patients (1.25 ± 0.46 mmol/L vs. 1.08 ± 0.36 mmol/L, $P < 0.001$) in the total group. Females with metformin administration had the highest lactate levels ($P < 0.01$) (Table 2). The levels of plasma lactate, follicle-stimulating hormone, and

TABLE 1. CLINICAL AND BIOCHEMICAL CHARACTERISTICS OF CHINESE TYPE 2 DIABETES PATIENTS TREATED WITH AND WITHOUT METFORMIN

	Metformin ($n=199$)	Non-metformin ($n=193$)	P
Age (years)	57.16 ± 10.79	59.35 ± 11.99	0.06
Diabetes duration (years)	7.78 ± 6.07	8.04 ± 7.11	0.71
FPG (mmol/L)	8.52 ± 2.73	8.36 ± 2.66	0.695
HbA1c (%)	8.79 ± 2.23	9.11 ± 2.31	0.17
BMI (kg/m^2)	26 ± 4.03	23.83 ± 3.29	<0.001
Cr ($\mu\text{mol}/\text{L}$)	65.17 ± 17.09	64.95 ± 18.15	0.81
ALT (U/L)	25.29 ± 16.25	25.93 ± 13.29	0.16
LA (mmol/L)	1.26 ± 0.43	1.08 ± 0.40	<0.001
HLA (%)	7.5%	3.13%	0.025

ALT, alanine aminotransferase; BMI, body mass index; Cr, creatinine; FPG, fasting plasma glucose; HbA1c, glycosylated hemoglobin A1c; HLA, hyperlactatemia; LA, lactic acid.

TABLE 2. GENDER DIFFERENCES OF PLASMA LACTIC ACID CONCENTRATIONS AND GONADAL HORMONE LEVELS IN CHINESE TYPE 2 DIABETES PATIENTS WITH AND WITHOUT METFORMIN

	Metformin			Non-metformin		
	Male (n=89)	Female (n=110)	P	Male (n=84)	Female (n=109)	P
Age (years)	56.37±11.83	57.81±9.86	0.35	55.22±13.23	62.62±9.86	<0.001
Diabetes duration (years)	7.99±6.75	7.61±5.51	0.68	7.72±7.78	8.34±6.59	0.24
BMI (kg/m ²)	26.09±3.95	25.92±4.11	0.76	24.00±3.30	23.69±3.32	0.52
HbA1c (%)	8.72±2.07	8.84±2.36	0.71	9.11±2.24	9.11±2.39	0.99
LA (mmol/L)	1.16±0.37	1.35±0.45	0.001	1.01±0.33	1.14±0.44	0.036
HLA (%)	4.49%	10%	0.015	1.19%	4.59%	0.18
Cr (μmol/L)	71.82±14.58	59.68±17.12	<0.001	69.32±16.25	61.63±18.94	0.001
ALT (U/L)	27.91±20.26	23.18±11.80	0.22	26.41±13.97	25.52±12.84	0.53
T (μg/L)	4.04±1.92	0.40±0.24	<0.001	3.48±1.70	2.33±1.57	<0.001
E ₂ (ng/L)	28.76±15.33	24.73±22.02	0.001	31.69±18.34	25.75±22.07	0.004
FSH (IU/L)	10.98±11.28	40.09±24.46	<0.001	10.00±10.03	48.94±28.33	<0.001
LH (IU/L)	6.53±10.22	16.92±11.49	<0.001	5.84±4.00	21.34±13.52	<0.001
PRL (μg/L)	13.34±9.97	14.06±15.90	0.85	11.99±5.39	14.57±11.33	0.38
P (μg/L)	0.40±0.33	0.75±1.77	0.96	0.64±0.57	0.77±1.37	0.58

ALT, alanine aminotransferase; BMI, body mass index; Cr, creatinine; E₂, estradiol; FSH, follicle-stimulating hormone; HbA1c, glycosylated hemoglobin A1c; HLA, hyperlactatemia; LA, lactic acid; LH, luteinizing hormone; P, progesterone; PRL, prolactin; T, testosterone.

luteinizing hormone in women were significantly higher than those in men (all $P < 0.01$). However, the levels of Cr, T, and E₂ in women were significantly lower than those in men ($P < 0.01$) (Table 2).

Gonadal hormone levels and plasma lactate concentrations in different age subgroups

Taking into account that the age of female menopause is usually between 45 to 55 years, female patients were divided into three subgroups: (1) <45 years, (2) 45–55 years, and (3) >55 years. We found that the plasma lactate levels of women <45 years old (1.72 ± 0.49 mmol/L) were higher than those of women 45–55 years old (1.31 ± 0.55 mmol/L) and of women >55 years old (1.32 ± 0.36 mmol/L) ($P < 0.05$) (Fig. 1); so was the incidence of hyperlactatemia. These data suggest

that the plasma lactate levels of menopausal women were significantly higher than those of postmenopausal women. However, in men, there was no significant difference in blood lactate levels among the three subgroups ($P > 0.05$).

Factors affecting the fasting lactate levels

In all 392 patients, the Spearman's (T, follicle-stimulating hormone, luteinizing hormone, prolactin, progesterone) and Pearson's (E₂, BMI) correlation analysis indicated that the fasting LA level was positively correlated with gender ($r = 0.193$, $P < 0.001$), Cr ($r = 0.152$, $P < 0.01$), and BMI ($r = 0.117$, $P = 0.02$) and negatively associated with T ($r = -0.187$, $P < 0.001$) and metformin ($r = -0.237$, $P < 0.001$) (Table 3). However, it had no significant correlation with age, diabetes duration, HbA1c, and E₂. In the metformin group, LA was

FIG. 1. Alteration of lactic acid levels with increasing age in female Chinese type 2 diabetes patients. Error bars indicate the 95% confidence interval. * $P < 0.05$ versus the subgroup <45 years old.

TABLE 3. CORRELATION ANALYSIS RESULTS FOR INFLUENCE FACTORS OF PLASMA LACTIC ACID CONCENTRATIONS IN CHINESE TYPE 2 DIABETES PATIENTS WITH OR WITHOUT METFORMIN THERAPY

	Total		Met		Non-Met	
	r	P	r	P	r	P
Age	0.013	0.798	-0.06	0.402	0.138	0.055
Sex	0.181	<0.01	0.23	0.001	0.163	0.024
BMI	0.117	0.02	0.061	0.394	0.051	0.488
Met	-0.237	<0.01				
E ₂	0.019	0.72	0.118	0.028	0.05	0.52
P	0.13	0.01	0.167	0.024	0.213	0.003
T	-0.187	<0.01	-0.202	0.007	-0.206	0.005
FSH	0.037	0.47	-0.033	0.645	0.119	0.102
LH	0.034	0.51	-0.027	0.708	0.129	0.075
PRL	0.048	0.36	0.101	0.166	0.01	0.894

BMI, body mass index; E₂, estradiol; FSH, follicle-stimulating hormone; LH, luteinizing hormone; Met, metformin; Non-Met, non-metformin treatment group; P, progesterone; PRL, prolactin; T, testosterone.

negatively correlated with T ($r=-0.202$, $P=0.007$), whereas it was positively correlated with E_2 ($r=0.118$, $P=0.028$) (Table 3). Furthermore, the fasting LA levels of females were positively correlated with E_2 levels in the metformin group. The logistic regression analysis indicated that factors influencing LA level were Cr ($P=0.004$), E_2 ($P=0.009$), metformin ($P=0.025$), gender ($P=0.047$), and T ($P=0.046$) (only if E_2 was removed) in all patients. The LA levels in women had no significant correlation with T and E_2 , but the LA levels in men had a negative correlation with T in the non-metformin group ($r=-0.237$, $P=0.031$).

Influence of T and E_2 levels on lactate concentrations

The patients were divided into three subgroups according to the interquartile of T levels, and the sample numbers for the three subgroups were 63, 25, and 89, respectively. The Kruskal-Wallis test indicated that the LA concentrations decreased with increasing T levels ($P=0.002$) (Fig. 2) but increased with elevating E_2 levels ($P<0.01$) (Fig. 3).

Discussion

This is the first study showing that sex hormone levels can influence the fasting LA levels in a population with T2DM. Regardless of the total group, the metformin group, or the non-metformin group, we all found that fasting LA levels were lower in men than that in women. The LA concentration in plasma was positively correlated with E_2 levels, but it was significantly negatively correlated with T levels. In addition, our preliminary results showed that the lactate concentration increased with elevation of Cr levels, and it significantly increased when Cr levels were higher than $96.5 \mu\text{mol/L}$. The LA concentrations also increased with elevation in ALT activity.^{7,9} However, there was no significant difference in liver and kidney function between men and women in the present study. Thus, the difference of lactate levels between males and females was due to the gender distinction.

FIG. 2. Plasma lactate concentrations decrease with elevation of testosterone (T) levels. Error bars indicate the 95% confidence interval. By Kruskal-Wallis test, $P=0.002$; $P<0.01$ for the T $0.5\text{--}1.0 \mu\text{g/L}$ group versus the T $<0.5 \mu\text{g/L}$ group, the T $>1.0 \mu\text{g/L}$ group versus the T $0.5\text{--}1.0 \mu\text{g/L}$ group, and the T $>1.0 \mu\text{g/L}$ group versus the T $<0.5 \mu\text{g/L}$ group.

FIG. 3. Plasma lactate concentrations increase with elevation of estradiol (E_2) levels. The regression line is drawn for women with type 2 diabetes mellitus. $P<0.001$.

Similarly, our previous study found that there was a gender difference in blood lactate levels, which decreased with the increase in age of women, and the plasma lactate levels in postmenopausal women were significantly lower than those in premenopausal women.⁹ This research was concordant with a previous related study in which lactate levels of women <45 years old were higher than those in those $45\text{--}55$ years and >55 years old.⁹ The present study further indicated that the LA concentrations increased with the elevation of E_2 levels but decreased with the increase of T levels, in the analysis of the relation between blood lactate levels and sex hormones. In other words, the gender difference in T and E_2 levels largely leads to the different plasma lactate levels. The sex hormones may affect the expression of enzyme or transporter proteins that are involved in the metabolism of LA. Singhal et al.¹³ reported that the activity of pyruvate kinase declined after gonadectomy, compared with normal rats, and the enzyme activity increased with the rise of estrogen dose; the same phenomenon was observed with phosphofructokinase. Therefore, it is possible that E_2 could increase the plasma lactate concentration by augmenting its generation. Recently, similar outcomes had been reported in some animal studies. Urakami et al.¹⁴ found that there was a gender difference in the expression of metformin-associated transporter protein (OCT2, MATE1) in mouse kidney and that this difference is regulated by different hormones. The expression of OCT2 and MATE1 is higher in male rats than in female rats.^{10,14,15} Asaka et al.¹¹ also found that androgen significantly increased the expression of OCT2 but that estrogen had the opposite effect. Therefore, we speculate that T and E_2 levels and the expression of OCT2 are closely linked. Androgen is unlikely to cause the accumulation of LA because it can increase the expression of OCT2, which will result in an increase of the proportion of metformin transported into the kidney through renal tubular epithelial cells. At the same time, androgen could increase the metformin excretion from the body through urine by high expression of MATE1. In addition, Meemam et al.^{16,17} further

reported that T significantly increased the excretion of cationic drugs by elevating the expression of OCT2 *in vivo*, whereas estrogen decreased their excretion via inhibiting OCT2 gene expression.

Our previous study showed that the gene *SLC22A2* encoding OCT2 polymorphisms may affect blood lactate levels differently based on gender, which means the *SLC22A2* gene (808G>T) variation can influence the plasma LA levels. Patients with the mutant genotype (TT) in the metformin-treated group had a higher blood lactate concentration and higher incidence of hyperlactacidemia compared with the GG genotype in the metformin-treated group and the GG or GT genotypes in the non-metformin-treated group. However, there were significant gender differences in the same genotype, and the lactate levels of women with the TT genotype were the highest in the metformin-treated group.⁸ Therefore, this study may be of great potential clinical value in the metformin administration in patients with diabetes. Because the lactate concentrations were higher in females with high E₂ levels and increased with elevation of E₂ level, the lactate concentrations should be monitored frequently in diabetes patients during metformin administration, especially in women with high estrogen levels, to avoid potential lactic acidosis, even if their liver and renal functions are in the normal range. Additionally, the logistic regression analysis indicated that the independent factors influencing LA were Cr, E₂, metformin, and gender in all patients, whereas T had obvious significance with lactate only if E₂ was removed. Therefore, we speculate that E₂ has a greater impact on blood lactate levels than T. Soric et al.¹⁸ observed a similar outcome in animals; in their study, all male rat groups receiving 17 β -E₂ treatment for 16 weeks developed more pronounced liver lactic acidosis than their matched controls, and the LA concentrations in castrated males were higher than those in intact males regardless of serum androgen levels. Some previous studies reported the capacity of estrogen to modulate the activities of several glycolytic enzymes, including phosphofructokinase, glucokinase, pyruvate kinase, and aldolase.^{13,19,20} These studies further indicated that the blood lactate levels in the non-metformin group also had a gender difference, suggesting that sex hormones may affect blood lactate levels not only by regulating the metabolism of metformin, but also by directly involvement in the process of lactate metabolism.

There are some limitations in this study. First, the blood lactate levels in the same patient before and after metformin therapy were not measured, nor was the plasma metformin concentration detected in the present study. Therefore, we could not further analyze the relationship between gonadal hormone levels and metformin metabolism. Second, the sample size of the present research was relatively small. More patients should be recruited to prove these findings. In addition, diabetes duration and age are also factors affecting blood lactate levels in diabetes patients. The diabetes duration, the metabolic rate of older individuals, heart and lung function, and other factors caused by hypoxia, which also increase blood lactate levels, should be taken into account.

In summary, metformin increases blood lactate levels. Different levels of T and E₂ are the main cause that led to different levels of blood lactate between individuals or between men and women. The LA concentrations increased with the elevation of E₂ levels but decreased with the increase

of T levels. Thus, more attention should be paid to women with diabetes and high E₂ levels after metformin administration to prevent the occurrence of lactic acidosis.

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Author Disclosure Statement

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References

1. Effect of intensive blood-glucose control with metformin on complications in overweight patients with type 2 diabetes (UKPDS 34). UK Prospective Diabetes Study (UKPDS). *Lancet* 1998;352:854–865.
2. Abbasi AA, Kasmikha R, Sotingeanu DG: Metformin-induced lacticacidemia in patients with type 2 diabetes mellitus. *Endocr Pract* 2000;6:442–446.
3. Misbin RI, Green L, Stadel BV, Gueriguian JL, Gubbi A, Fleming GA: Lactic acidosis in patients with diabetes treated with metformin. *N Engl J Med* 1998;338:265–266.
4. Stang M, Wysowski DK, Butler-Jones D: Incidence of lactic acidosis in metformin users. *Diabetes Care* 1999;22:925–927.
5. Orban JC, Giunti C, Levraut J, Grimaud D, Ichai C: Metformin-associated lactic acidosis remains a serious complication of metformin therapy. *Ann Fr Anesth Reanim* 2003;22:461–465.
6. Pertek JP, Vidal S, Mariot J, Galy-Floc'h M, Azoulay E: Metformin-associated lactic acidosis precipitated by acute renal failure. *Ann Fr Anesth Reanim* 2003;22:457–460.
7. Liu F, Lu JX, Tang JL, Li L, Lu HJ, Hou XH, Jia WP, Xiang KS: Relationship of plasma creatinine and lactic acid in type 2 diabetic patients without renal dysfunction. *Chin Med J (Engl)* 2009;122:2547–2553.
8. Li Q, Liu F, Zheng TS, Tang JL, Lu HJ, Jia WP: SLC22A2 gene 808G/T variant is related to plasma lactate concentration in Chinese type 2 diabetics treated with metformin. *Acta Pharmacol Sin* 2010;31:184–190.
9. Li Q, Liu F, Tang JL, Zheng TS, Lu JX, Lu HJ, Jia WP: The gender difference of plasma lactate levels and the influence of metformin in type 2 diabetes patients [in Chinese]. *Chin J Endocrinol Metab* 2010;26:372–376.
10. Urakami Y, Okuda M, Saito H, Inui K: Hormonal regulation of organic cation transporter OCT2 expression in rat kidney. *FEBS Lett* 2000;473:173–176.
11. Asaka J, Terada T, Okuda M, Katsura T, Inui K: Androgen receptor is responsible for rat organic cation transporter 2 gene regulation but not for rOCT1 and rOCT3. *Pharm Res* 2006;23:697–704.
12. Lu HJ, Tang JL, Yuan QY, Jia WP, Xiang KS: The establishment of normal range of whole vein blood lactic acid [in Chinese]. *Shanghai Med* 2005;28:27–29.
13. Singhal RL, Valadares JR, Ling GM: Metabolic control mechanisms in mammalian systems. I. Hormonal induction of phosphofructokinase in rat uterus. *J Biol Chem* 1967;242:2593–2598.
14. Urakami Y, Nakamura N, Takahashi K, Okuda M, Saito H, Hashimoto Y, Inui K: Gender differences in expression of organic cation transporter OCT2 in rat kidney. *FEBS Lett* 1999;461:339–342.

15. Lickteig AJ, Cheng X, Augustine LM, Klaassen CD, Cherrington NJ: Tissue distribution, ontogeny and induction of the transporters Multidrug and toxin extrusion (MATE)1 and MATE2 mRNA expression levels in mice. *Life Sci* 2008;83:59–64.
16. Meetam P, Srimaroeng C, Soodvilai S, Chatsudthipong V: Regulatory role of testosterone in organic cation transport: in vivo and in vitro studies. *Biol Pharm Bull* 2009;2:982–987.
17. Meetam P, Srimaroeng C, Soodvilai S, Chatsudthipong V: Role of estrogen in renal handling of organic cation, tetraethylammonium: in vivo and in vitro studies. *Biol Pharm Bull* 2009;32:1968–1972.
18. Soric S, Belanger MP, Askin N, Wittnich C: Impact of female sex hormones on liver tissue lactic acidosis during ischemia. *Transplantation* 2007;84:763–770.
19. Moorthy K, Yadav UC, Siddiqui MR, Sharma D, Basir SF, Baquer NZ: Effect of estradiol and progesterone treatment on carbohydrate metabolizing enzymes in tissues of aging female rats. *Biogerontology* 2004;5:249–259.
20. Singhal RL, Valadares JR: Estrogenic regulation of uterine pyruvate kinase. *Am J Physiol* 1970;218:321–327.

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