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Serum lactate levels are associated with serum alanine aminotransferase and total bilirubin levels in patients with type 2 diabetes mellitus: A cross-sectional study

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ABSTRACT

Aims: It was recently reported that lactate acts as a metabolic mediator and rises in the diabetic state, but the physiological effects are as yet poorly understood. The objective of the current study was to evaluate the significance of serum lactate elevation in type 2 diabetes mellitus (T2DM) patients.

Methods: Fasting serum lactate levels, hematological and inflammatory serum markers and anthropometric parameters, obtained employing bioelectric impedance analysis, were measured in 103 patients with T2DM.

Results: Statistically significant correlations of serum lactate levels with C-reactive peptide, insulin, aspartate aminotransferase, alanine aminotransferase (ALT), serum lipids, total bilirubin, adiponectin, homeostasis model assessment-insulin resistance, body weight, body mass index and body fat (weight or percentage of subcutaneous fat, visceral fat or total body fat), but neither fasting plasma glucose nor HbA1c, were detected. Stepwise regression analysis showed ALT to be independently positively associated with total bilirubin, while being negatively associated with serum lactate levels. Furthermore, serum lactate levels were significantly higher in patients with ALT-predominant liver dysfunction.

Abbreviations: T2DM, type 2 diabetes mellitus; MCP-1, monocyte chemotactic protein 1; AST, aspartate aminotransferase; ALT, alanine aminotransferase; γ-GTP, γ-glutamyl transpeptidase; ALP, alkaline phosphatase; LD, lactate dehydrogenase; BUN, blood urea nitrogen; Cr, creatinine; LDL, low-density lipoprotein; HDL, high-density lipoprotein; HOMA-R, homeostasis model assessment-insulin resistance; BMI, body mass index; GPR, G protein-coupled receptor; Ccr, creatinine clearance; NAFLD, non-alcoholic fatty liver disease.

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Conclusion: We found fasting serum lactate elevation in T2DM patients to be associated with the serum levels of ALT and total bilirubin independently of blood glucose control.

Trial registration: UMIN clinical trials registry (UMIN000029178).

1. Introduction

Classically, lactate has been regarded as a glycolytic waste product after exercise or as a substrate for hepatic gluconeogenesis. Recently, lactate has been drawing attention as an important intermediate metabolite exerting various effects on metabolism. Lactate is produced from glucose through glycolysis and the conversion of pyruvate by lactate dehydrogenase. Skeletal muscle and adipose tissues have been thought to play important roles in supplying lactate. Skeletal muscle is a major production site of lactate. It continuously generates and uses lactate in resting states and, furthermore, lactate production increases during exercise [1].

Adipose tissue is also a source of the lactate supply [2]. Depending on stimulation of the process by insulin and glucose uptake, adipose can convert more than 50% of the lactate metabolized from glucose [3–5]. Adipose tissue secretes various adipokines, which regulate metabolism. Lactate has been speculated to play an important role as an adipokine secreted from adipocytes, through its receptor G protein-coupled receptor 81 (GPR 81), functioning against the suppression of lipolysis in adipocytes by insulin [6,7]. In addition, lactate contributes to brain protection in hypoglycemic states, through the cellular interaction between astrocytes and neurons [8,9]. Furthermore, it has also been noted that lactate may contribute to the browning of white adipose cells *via* increased uncoupling protein 1 (UCP1) gene expressions [10]. Also, lactate reportedly exerted anti-inflammatory effects in acute hepatitis and acute pancreatitis models [11].

Type 2 diabetes mellitus (T2DM) causes vascular complications and is a major public health problem associated with lifestyle diseases such as obesity, liver steatosis, hypertension, and so on. Although serum lactate is reportedly elevated in diabetic patients [12,13], the physiological effects remain unclear. This study aimed to identify the significance of serum lactate elevation in T2DM patients, which may facilitate developing diabetes treatment strategies and new approaches to health promotion.

2. Materials and methods

2.1. Patients

The study protocol was approved by the ethics committee of Kyorin University School of Medicine (No. H28-001) and was registered with the University Hospital Medical Information Network (UMIN) Clinical Trials Registry (UMIN000029178). All procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1964, as revised in 2013. The study group consisted of 103 outpatients (Fig. 1) with T2DM (80 males and 23 females), over age 20 years. All were Japanese and were followed at the Kyorin University School of Medicine. Acute diabetic complications (diabetic ketoacidosis, diabetic coma), severe heart failure [New York Heart Association (NYHA) classification III to IV], severe renal dysfunction [creatinine clearance (Ccr) < 30 mL/min], age under 20 years, heavy alcohol consumption, viral hepatitis infection and pregnancy were exclusion criteria. Patients stopped at least moderate exercise 2 days before the blood sampling. We chose patients whose fasting plasma glucose levels above 4.0 mmol/L and also had no symptoms of hypoglycemia on the examination day to avoid lactate rise due to hypoglycemic effect. All 103 patients recruited provided fully informed consent. The characteristics of the patients are shown in Table 1.

2.2. Fasting serum lactate measurement and hematological analysis

Morning blood samples were collected from each patient after an overnight fast. The routine blood tests were performed at the general laboratory of our hospital. The serum samples were stored frozen at −80 °C until analysis. The serum lactate levels were analyzed by enzymatic methods using an automatic analyzer (BioMajesty JCA-BM9130; JEOL Ltd., Tokyo). Glucose was analyzed according to the glucose oxidase method, and HbA1c was determined by high-performance liquid chromatography, using an automatic analyzer (ADAMS Glucose GA-1171 and ADAMS A1cHA-8180V; Arkray, Tokyo). C-reactive peptide was analyzed by chemiluminescence enzyme immunoassay, using an automatic analyzer (LUMIPULSE L2400; Fujirebio, Tokyo). Serum insulin was measured by enzyme immunoassay in an automatic analyzer (A1A-2000; Tosoh, Tokyo). The 1, 5-anhydroglucitol (1, 5-AG) level

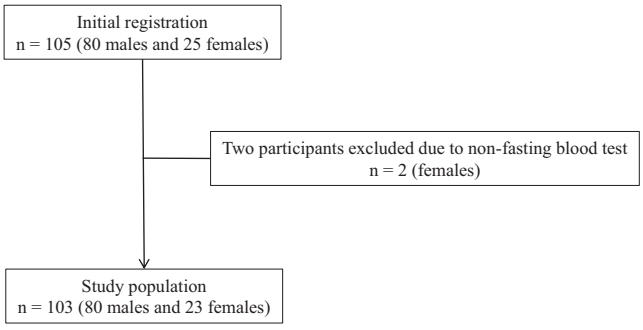


Fig. 1 – Flow chart showing inclusion and exclusion criteria. We obtained informed consent from the 105 patients enrolled in the study. However, 2 patients were excluded due to failure to fast prior to the blood tests.

Table 1 – Clinical characteristics of the T2DM patients studied.

Number (Male/Female)	103 (80/23)
Age (years)	58.6 ± 1.15
Duration of diabetes (years)	8.9 ± 0.82
Body weight (kg)	74.7 ± 1.52
Lactate (mmol/L)	1.07 ± 0.03
Fasting plasma glucose (mmol/L)	7.5 ± 0.17
HbA1c (%)	6.8 ± 0.08
HbA1c (mmol/mol)	51 ± 8
Baseline glucose-lowering therapies (n)	
MET	49
SU	17
DPP-4 inhibitor	66
TZD	7
Glinide	20
α-GI	26
SGLT-2 inhibitor	24
Insulin	16
GLP-1 receptor agonist	6

Data are expressed as means ± standard error. MET; metformin, SU; sulfonylurea, DPP-4; dipeptidyl peptidase-4, TZD; thiazolidinedione, α-GI; alpha-glucosidase inhibitor, SGLT-2; sodium glucose transporter-2, GLP-1; glucagon-like peptide-1.

was determined by the colorimetric method, ketone bodies by an enzyme cycling method, using an automatic analyzer (BioMajesty JCA-BM8060; JEOL Ltd., Tokyo). Aspartate aminotransferase (AST), alanine aminotransferase (ALT), γ-glutamyl transpeptidase (γ-GTP), alkaline phosphatase (ALP) and lactate dehydrogenase (LD) levels were determined by the enzyme reaction rate method. Blood urea nitrogen (BUN) was analyzed by ultraviolet absorption spectrophotometry. Albumin was measured by the improved bromocresol purple method. Total bilirubin was determined by the vanadate oxidation method. Triglycerides, low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol and creatinine (Cr) were measured by enzymatic methods using an automatic analyzer (LABOSPECT 008; Hitachi, Tokyo). Homeostasis model assessment-insulin resistance (HOMA-R) was calculated employing the following equation: HOMA-R = fasting plasma glucose (mg/dL) × fasting insulin (μU/mL)/405. Monocyte chemotactic protein 1 (MCP-1) and adiponectin were determined employing Enzyme-Linked Immuno Sorbent Assay (ELISA) kits (Human CCl2/MCP-1 Quantikine ELISA Kit, Human Total Adiponectin/Acrp30 Quantikine ELISA Kit; R&D Systems Inc., Minneapolis, USA) according to the manufacturer's instructions. We assessed oxidative stress using a simple method for the evaluation of reactive oxygen metabolites (ROMs). ROMs were measured using a free radical elective evaluator (FREE Carpe Diem; Diacron International) that included a spectrophotometric device reader, and measurement kits (d-ROMs test) (Wismerll Co. Ltd., Tokyo) according to the manufacturer's protocol. The results are expressed in arbitrary units (U.CARR), one unit of which corresponds to 0.8 mg/L of hydrogen peroxide.

2.3. *Anthropometric measurements*

Anthropometric parameters were measured by bioelectric impedance analysis (BIA) using a multi-frequency impedance body composition analyzer (ZEUS 9.9, Sowa medical, Fukuoka) in the fasting state. These parameters showed good correlations with the results obtained by the dual-energy X-ray absorptiometry method [14]. Body weight (kg), body fat (kg), subcutaneous fat (kg), visceral fat (kg), body fat percentage (%), and muscle mass (kg) were evaluated by BIA, while height (cm) was based on patient self-assessment. Body mass index (BMI) was calculated as weight (kg) divided by the square of the height (m^2). Obesity was defined as a BMI over 25 kg/m^2.

2.4. *Statistical analysis*

Data are expressed as means ± standard error. The Pearson test was applied to assess correlations of serum lactate levels with other hematological markers and anthropometric measurements. Stepwise regression analysis was performed to evaluate the best predictor of serum lactate levels. Normality of the data distribution was assessed by the Kolmogorov-Smirnov test. Homogeneity of variances was estimated with the F test. Mean serum lactate levels depending on liver function were compared by applying a two-sample *t* test. $P < 0.05$ was considered to indicate a statistically significant difference. Statistical analysis and data management were conducted using IBM SPSS Statistics 19 (IBM Corporation, Somers, NY).

3. Results

3.1. *Serum lactate levels correlated with hematological and anthropometric markers related to insulin resistance, but not with blood glucose control*

Table 2 shows the statistically significant correlations of serum lactate levels with hematological, inflammatory and anthropometric markers. C-reactive peptide ($r = 0.386$, $P < 0.001$), insulin ($r = 0.360$, $P = 0.001$), AST ($r = 0.292$, $P = 0.003$), ALT ($r = 0.441$, $P < 0.001$), triglycerides ($r = 0.239$, $P = 0.015$), LDL-cholesterol ($r = 0.199$, $P = 0.044$), HOMA-R ($r = 0.324$, $P = 0.003$), body weight ($r = 0.258$, $P = 0.009$), BMI ($r = 0.303$, $P = 0.002$), body fat ($r = 0.320$, $P = 0.001$), subcutaneous fat ($r = 0.296$, $P = 0.003$), visceral fat ($r = 0.335$, $P = 0.001$), and body fat percentage ($r = 0.277$, $P = 0.005$) showed positive correlations with serum lactate levels. On the other hand, HDL-cholesterol ($r = -0.221$, $P = 0.025$) and adiponectin ($r = -0.247$, $P = 0.012$) levels correlated negatively with serum lactate levels. However, while total bilirubin did not correlate significantly with serum lactate levels, the correlation coefficient suggested a relatively strong negative trend ($r = -0.191$, $P = 0.053$). There were no significant relationships of HbA1c, fasting glucose, γ-GTP, ALP, and ketone bodies with serum lactate levels.

3.2. *Stepwise regression analysis revealed ALT and total bilirubin to be associated with serum lactate levels*

In addition, we evaluated factors strongly suspected to affect serum lactate levels. We performed a stepwise regression analysis including hematological, inflammatory and

Table 2 – Correlations of serum lactate levels with hematological, inflammatory and anthropometric markers.

Marker		Lactate	
		r	*P*
HbA1c (%)	6.8 ± 0.08	0.140	0.160
HbA1c (mmol/mol)	51 ± 8	0.140	0.160
Fasting plasma glucose (mmol/L)	7.5 ± 0.17	0.182	0.066
C-reactive peptide (ng/mL)	2.38 ± 0.13	0.386	<0.001***
Insulin (μU/mL)	10.0 ± 0.86	0.360	0.001**
1.5-anhydroglucitol (μg/mL)	10.4 ± 0.75	−0.080	0.403
AST (IU/L)	23.5 ± 1.02	0.292	0.003**
ALT (IU/L)	26.5 ± 1.61	0.441	<0.001***
γ-GTP (IU/L)	48.4 ± 7.74	0.129	0.195
ALP (IU/L)	222.2 ± 6.54	0.138	0.164
LD (IU/L)	187.3 ± 3.87	−0.018	0.858
Total bilirubin (mg/dL)	0.75 ± 0.03	−0.191	0.053
BUN (mg/dL)	14.3 ± 0.44	0.124	0.213
Cr (mg/dL)	0.77 ± 0.02	0.118	0.234
Albumin (mg/dL)	4.3 ± 0.04	0.122	0.222
Triglyceride (mg/dL)	143.2 ± 8.66	0.239	0.015*
LDL-cholesterol (mg/dL)	110.8 ± 3.48	0.199	0.044*
HDL-cholesterol (mg/dL)	51.5 ± 1.51	−0.221	0.025*
MCP-1 (pg/mL)	144.1 ± 3.45	0.170	0.089
Adiponectin (ng/mL)	56.6 ± 4.58	−0.247	0.012*
Total ketone bodies (μmol/L)	197.3 ± 53.3	0.014	0.889
Acetoacetic acid (μmol/L)	50.8 ± 9.36	0.025	0.799
3-Hydroxybutyric acid (μmol/L)	146.4 ± 43.91	0.011	0.916
HOMA-R	3.5 ± 0.35	0.324	0.003**
d-ROMs test (U.CARR)	306.6 ± 5.77	0.154	0.122
Body weight (kg)	74.7 ± 1.52	0.258	0.009**
BMI (kg/m^2)	26.6 ± 0.44	0.303	0.002**
Body fat (kg)	20.9 ± 0.79	0.320	0.001**
Subcutaneous fat (kg)	17.5 ± 0.65	0.296	0.003**
Visceral fat (kg)	3.2 ± 0.17	0.335	0.001**
Body fat percentage (%)	27.4 ± 0.71	0.277	0.005**
Muscle mass (kg)	49.4 ± 0.88	0.133	0.182

Data are expressed as means ± standard error. Peason's correlation analysis.

AST; aspartate aminotransferase, ALT; alanine aminotransferase, γ-GTP; γ-glutamyl transpeptidase, ALP; alkaline phosphatase, LD; lactate dehydrogenase, BUN; blood urea nitrogen, Cr; creatinine, LDL; low-density lipoprotein, HDL; high-density lipoprotein, MCP-1; monocyte chemotactic protein 1, HOMA-R; homeostasis model assessment-insulin resistance, ROMs; reactive oxygen metabolites, BMI; body mass index.

* $P < 0.05$.
** $P < 0.01$.
*** $P < 0.001$.

anthropometric markers which had shown significant correlations with serum lactate levels on univariate analysis. These factors included C-reactive peptide, insulin, AST, ALT, triglycerides, LDL and HDL cholesterol, adiponectin, HOMA-R, body weight, BMI, body fat, subcutaneous fat, visceral fat and body fat percentage. In addition, total bilirubin, which had a relatively high correlation coefficient, 3-hydroxybutyric acid which has a structure similar to that of 2-hydroxybutyric acid and 4-hydroxybutyric acid, both of which are ligands of lactate receptor GPR81 [15], as well as HbA1c and glucose, were also included. We found only ALT (β = 4.518E−01, P = 1.32E−06) and total bilirubin (β = −2.151E−01, P = 0.0162) to be significantly associated with serum lactate levels (Table 3).

Table 3 – ALT and total bilirubin were shown to be associated with serum lactate levels by stepwise regression analysis.

Variable	Parameter estimate	Standardized estimate (β)	*P*
Intercept	8.86E+00	5.534E−17	<2E−16***
ALT	8.74E−02	4.518E−01	1.32E−06***
Total bilirubin	−2.06E+00	−2.151E−01	0.0162*

Multiple R-squared (R^2): 0.514. Adjusted R-squared (R^2): 0.433, VIF (variance inflation factor); ALT 1.003141, total bilirubin 1.007503

* $P < 0.05$.
** $P < 0.01$.
*** $P < 0.001$.

3.3. *Serum lactate was elevated in patients with ALT-predominant liver dysfunction*

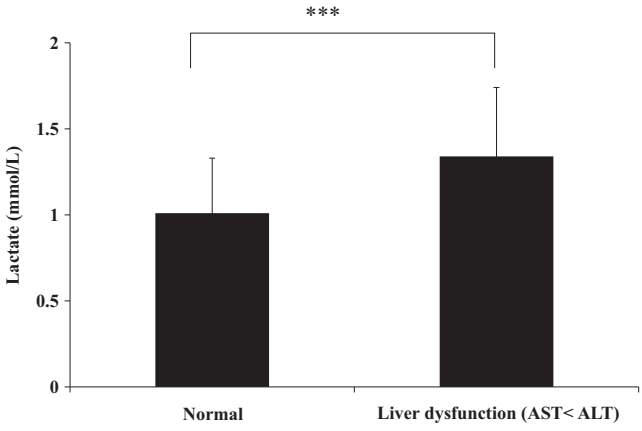
We classified patients with liver dysfunction into AST-predominant and ALT-predominant groups after defining normal and abnormal liver functions as follows: normal; AST < 30 IU/L (males and females) and ALT < 42 IU/L (males),

ALT < 23 IU/L (females). Serum lactate was significantly elevated in patients with ALT-predominant liver dysfunction as compared with those in patients with normal liver functions (n = 17 and 81, respectively, 1.34 ± 0.40 *vs* 1.01 ± 0.32 mmol/L, P < 0.001) (Fig. 2). In patients with liver dysfunction, serum lactate levels were not significantly related to a fibrosis index of 4 (FIB-4 index = (AST (IU/L) × age (years))/(plate (10^9/L) × $\sqrt{}$ ALT (IU/L)), which is a parameter of liver fibrosis (n = 22, r = −0.17, P = 0.43).

4. Discussion

The main finding of this study is that serum lactate levels are associated with indicators suggesting insulin resistance. Further analysis showed liver-related factors, specifically ALT and total bilirubin, to be the best predictors of serum lactate levels.

Fatty liver is associated with metabolic risk factors such as obesity, diabetes, and dyslipidemia [16] and has been reported as a risk factor for atherosclerosis [17,18]. It can also progress to severe related diseases such as liver steatosis, cirrhosis and hepatocellular carcinoma [19]. The gold standard for assessment of fatty liver is liver biopsy but this is an invasive procedure, and quantitative and useful biomarkers of early-stage diseases might contribute to treatment strategies aimed at suppressing the progression of such liver disorders. Lactate is a major substrate for gluconeogenesis and serum lactate elevation may serve as an indicator of impaired glucose production and lipid synthesis in states of obesity [20]. In addition, a positive correlation between serum lactate levels and lipid oxidation, suggesting that increasing gluconeogenesis from lactate is related to the promotion of lipid oxidation, has been reported in obese children [21]. Furthermore, *in vivo* magnetic resonance spectroscopy of non-alcoholic fatty liver disease (NAFLD) animal models, has suggested [1-^{13}C] lactate to be a potential biomarker of liver steatosis [22]. It has also been shown that the Warburg shift from mitochondrial respiration to cytosolic glycolysis contributes to lactate production in the early phase of liver steatosis [23]. Some previous studies have already shown that lactate is metabolized primarily by the liver [24–26] and also have suggested that liver dysfunction is associated with higher lactate levels in the acutely ill patients [27–29]. To our knowledge, however, no study has considered examined serum lactate levels in human subjects with T2DM with various serum markers and anthropometric parameters.



	n	Lactate (mmol/L)
Normal	81	1.01±0.32
Liver dysfunction (AST < ALT)	17	1.34±0.40

Fig. 2 – Comparison of serum lactate levels between patients with normal liver function and those with ALT-predominant liver dysfunction. Data are expressed as means ± standard error. ***P < 0.001. *Abbreviations:* AST; aspartate aminotransferase, ALT; alanine aminotransferase.

In our study, serum lactate levels correlated positively with ALT and negatively with total bilirubin. Furthermore, serum lactate was elevated in patients with ALT-predominant liver dysfunction not attributable to either alcohol-induced damage or viral infections. The serum lactate levels of patients with high serum total bilirubin levels (normal range: 0.4–1.5 mg/dL) were lower than those in patients with normal serum total bilirubin levels (n = 4 and 92, respectively, 0.73 ± 0.08 *vs* 1.07 ± 0.10 mmol/L, P = 0.05). However, further investigation of this issue is necessary since the number of target patients in our study was small. In recent studies, ALT elevation was found to be a risk factor related to new onset of diabetes [30], and was related to markers of inflammation and oxidative stress in NAFLD [31]. In addition, bilirubin which has antioxidant and cell-protective functions, is related to a reduced incidence of metabolic diseases and lower risk of cardiovascular disease, with an inverse correlation having been demonstrated between serum total bilirubin levels and the NAFLD incidence rate [32]. Our hypothesis, that serum lactate might serve as a biomarker of liver dysfunction, is further supported by these previously reported findings.

Lactate has an effect as a metabolic mediator suppressing lipolysis by insulin in adipose tissue. The mechanisms involve direct activation of GPR81, which is an orphan G-protein-coupled receptor. After being stimulated by lactate in adipocytes, intracellular adenylyl cyclase activity is decreased, resulting in reduced cyclic adenosine monophosphate (cAMP) production and, ultimately, the suppression of lipolysis [6,7]. GPR81 was reported to be highly expressed in adipose tissue, as well as in the brain, liver, kidney, skeletal muscle, spleen and testicles [15]. In the present study, serum lactate levels correlated negatively with the body muscle ratio (r = −0.277, P = 0.004), suggesting that lactate is a metabolite secreted from adipose tissues rather than a waste product of metabolic activities in muscles. In a past study, lactate was shown to suppress inflammatory reactions induced by Toll-like receptors and inflammasomes through activation of GPR81 in a model of acute hepatitis and acute pancreatitis [11]. Like these prior reports, our results support the hypothesis that the anti-inflammatory actions of lactate might also function in fatty liver with chronic inflammation. It has also been shown that chronic obesity due to a high fat diet can suppress GPR81 gene expression in adipose tissues [33]. It is not unreasonable to theorize that expression of

the GPR81 gene might be suppressed in fatty liver as well, possibly inducing a state of "lactate resistance" with a resultant rise in serum lactate levels. In addition, it has been found that sufficient serum lactate enhances UCP1 gene expression through mono-carboxylate transport proteins and promotes browning of white adipose tissues independently of GPR81 [10]. White adipose tissues undergoing increased fat deposition are thought to change into brown adipose tissues via the metabolism of lactate. This mechanism is regarded as a compensatory response to insulin resistance, mediated by lactate, and a similar mechanism might exist in fatty liver.

Another interesting finding of the current study is that the serum lactate level is associated with liver-related factors other than the FIB-4 index, which is an indicator of advanced hepatic fibrosis. Our results suggest serum lactate to be associated with early-stage liver dysfunction, prior to the onset of overt fibrosis.

Some biguanides can cross the mitochondrial membrane and increase L-lactate formation by inhibiting oxidative phosphorylation [34]. On the other hand, it has also been reported that levels of fasting plasma lactate in T2DM patients are similar regardless of whether or not metformin is administered [35] and that no significant differences are observed after metformin treatment [36,37]. In the current study, serum lactate levels in patients with and without metformin treatment did not differ significantly ($n = 49$ and 54, respectively, 1.11 ± 0.05 *vs* 1.04 ± 0.05 mmol/L). Some glucose-lowering therapies have been reported to be useful for fatty liver. It has suggested that the administration of pioglitazone let to metabolic and histologic improvement in subjects with nonalcoholic steatohepatitis [38]. In the current study, serum lactate levels in patients with and without thiazolidinedione treatment did not differ significantly ($n = 7$ and 96, respectively, 1.29 ± 0.26 *vs* 1.05 ± 0.03 mmol/L). It has also been shown that canagliflozin may improve liver function in T2DM with high alanine aminotransferase [39]. In the current study, serum lactate levels in patients with and without SGLT-2 inhibitor treatment did not differ significantly ($n = 24$ and 79, respectively, 1.13 ± 0.05 *vs* 1.05 ± 0.04 mmol/L). It has also been reported that liraglutide let to histological resolution of non-alcoholic steatohepatitis in T2DM [40]. In the current study, serum lactate levels in patients with and without GLP-1 receptor agonist treatment did not differ significantly ($n = 6$ and 97, respectively, 1.24 ± 0.17 *vs* 1.06 ± 0.03 mmol/L). However there were no difference in serum lactate levels depending on with or without any therapies which have been reported the utilities to fatty liver, further studies will be necessary because of the small number of patients.

In addition, some previous studies already showed that higher γ-GTP levels were associated with onset of diabetes [41–43] and γ-GTP increased in patients with nonalcoholic steatohepatitis [44,45]. However, there was no correlation between the serum lactate levels with the serum γ-GTP levels in the current study. It might be because the serum γ-GTP levels in the current study were lower than the levels which had been reported [41,44,45]. Alternatively, serum lactate may be an earlier indicator of fatty liver before γ-GTP increases.

This study has limitations. First, live imaging was not conducted to evaluate fatty liver and hepatic dysfunction because there are no highly accurate liver imaging methods for assessment of early-stage liver disorders. Second, we also did not perform invasive liver biopsies, which would have provided histological information on the relationships between fatty liver and changes in serum lactate levels. In a future study, imaging evaluations of liver dysfunction should be performed. Third, we analyzed only a small number of patients. We did not investigate non-diabetic subjects. However, according to our results, serum lactate levels are not associated with parameters of blood glucose control such as HbA1c and fasting plasma glucose. Therefore, similar results would presumably be obtained in non-diabetic patients. Fourth, we did not use a unified test meal at a day before the blood sampling and the rise of serum lactate levels due to the influence of meals could not be ignored. Finally, we had not measured other inflammatory markers or adipokines and that they might have been effected to serum lactate levels.

In conclusion, we found that fasting serum lactate elevation on T2DM was associated with serum levels of ALT and total bilirubin, independently of blood glucose control. Further clinical or basic research is warranted to elucidate the mechanism underlying the influence of rising serum lactate levels on the progression of liver dysfunction.

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Duality of interest

The authors declare that there is no duality of interest regarding the publication of this article.

Author contributions

M. I. designed the study, acquired the data, performed the experiments and analyses, and wrote the article. T. H. designed the study, acquired funding and data, performed data interpretation, and helped in drafting the article. N. M., K. K., T. M., A. K., K. T., Y. S., T. T., and T. K. were responsible for data interpretation. T. Y. contributed to the analysis. H. I. contributed to the study design and to final approval of the version to be submitted.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.diabres.2019.01.028.

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