ORIGINAL RESEARCH

Preliminary Study on the Evaluation of Skeletal Muscle Damage in Patients with Diabetes Mellitus Using Ultrasonic Shear Wave Elastography

Xue Fang, MM; Zhifen Han, MM; Yu Kang, MM; Xiaoxi Sha, MM; Yan Zhang, MD

ABSTRACT

Objectives • This study aimed to compare gastrocnemius muscle stiffness levels in subjects with and without type 2 diabetes mellitus (T2DM) using shear wave elastography (SWE).

Methods • This is a preliminary study enrolled patients with T2DM and healthy subjects at the affiliated Hospital of Chengdu University of Traditional Chinese Medicine between September 2021 and June 2022. Gastrocnemius muscle stiffness was measured using SWE.

Results • A total of 120 individuals (mean age: 52.09 ± 5.40 years, 85 males) were enrolled, including 70 patients with T2DM and 50 healthy subjects. There was no significant difference in E at neutral ankle position, plantar flexion position and E_{BMI} at neutral ankle position between T2DM patients and healthy subjects ($P > .05$). E at upright position (43.89 ± 14.93 vs. 51.71 ± 9.48, $P = 0.001$), E_{BMI} at plantar flexion position (1.17 (0.82-1.29) vs. 1.55 (1.21-1.84), $P < .001$) and upright position (1.72 (1.23-2.16) vs. 2.10 (1.88-2.29), $P < .001$) of the T2DM patients were significantly lower than those of healthy subjects. In T2DM patients, E at upright position was negatively correlated with the disease course (r=-0.645, $P < .001$), Hemoglobin A1c (HbA_{1c}) concentration (r=-0.741, $P < .001$), and advanced glycation end-product (AGEs) (r=-0.675, $P < .001$) but not with age ((r=-0.116, $P = .351$).

Conclusion • SWE results found that active muscle stiffness was significantly lower in T2DM patients compared to healthy controls, suggesting that evaluation of active muscle stiffness using SWE may be valuable in T2DM patients to prevent gastrocnemius muscle damage. (*Altern Ther Health Med.* 2024;30(1):314-317).

Xue Fang, MM, Attending doctor; School of Medical and Life Sciences, Chengdu University of Traditional Chinese Medicine, Chengdu, Sichuan, China. **Zhifen Han,** MM, Attending doctor; **Yu Kang,** MM, Chief physician; **Xiaoxi Sha,** MM, Attending doctor; **Yan Zhang,** MD, Associate chief physician; Department of Ultrasound Medicine, Hospital of Chengdu University of Traditional Chinese Medicine, Chengdu, Sichuan, China

Corresponding author: Yu Kang, MM
E-mail: zyfycsyxkang@cdutcm.edu.cn

INTRODUCTION

Diabetic myopathy is a common complication of type 2 diabetes mellitus (T2DM) defined as the failure to preserve muscle mass and function,[1,2] which can lead to pain and swelling of the affected extremity, ambulatory difficulty, and elevated healthcare costs.[3,4] As skeletal muscle plays a key role in glucose homeostasis and locomotion, diabetic myopathy is believed to contribute to the progression of other diabetes complications and comorbidities, including uncontrolled hyperglycemia, sedentarism, and obesity.[2,4] Early detection and treatment of diabetic myopathy are important in improving patient outcomes. Existing evaluation methods for diabetes myopathy have notable limitations, including invasiveness of muscle biopsy, poor sensitivity and specificity in muscle enzyme serology and high costs of magnetic resonance imaging (MRI).[5] Ultrasound shear wave elastography (SWE) is a new quantitative and non-invasive method used for assessing gastrocnemius muscle stiffness,[6-8] which is a well-known marker for muscle performance and integrity. Research has shown that the detection results of SWE on muscle stiffness are consistent with muscle pathology.[9] Compared to conventional ultrasound, the SWE technique can detect reduced muscle elasticity even when no organic changes occur in the muscle in the early stages of sarcopenia.[10,11] Therefore, SWE can potentially be an effective and non-invasive diagnostic tool for the early detection of diabetic myopathy in T2DM patients. However, there is a lack of studies investigating the use of SWE in assessing muscle stiffness in T2DM. This study aimed to compare gastrocnemius muscle stiffness levels in T2DM patients and healthy subjects using SWE.

METHODS

Study design and subjects

This is a preliminary study enrolled patients with T2DM and healthy subjects at the affiliated Hospital of Chengdu University of Traditional Chinese Medicine between September 2021 and June 2022. Inclusion criteria: 1) aged 40–60 years old. Exclusion criteria: 1) history of double lower-limb deformity and trauma surgery; 2) history of progressive muscular dystrophy, myasthenia gravis, myositis and other muscle diseases; 3) history of nervous system diseases; or 4) an occupational history of athletics and heavy physical labor. This study was approved by the ethics committee of the affiliated Hospital of Chengdu University of Traditional Chinese Medicine. All subjects signed the written informed consent.

Procedures

Subjects' information, including age, sex, height, weight, body mass index (BMI) and Hemoglobin A_{1c} (HbA_{1c}) concentration were collected. Disease course and the accumulation level of advanced glycation end-product (AGEs) were only collected in T2DM patients. Diagnosis of T2DM was made in accordance with the guidelines established by the World Health Organization.[12]

All examinations were performed by the same ultrasound physician using a high-performance ultrasound system (Resona R9/Mindray) and a linear array probe (L14-3MU) from 3 to 15 MHz in musculoskeletal mode. Results of stiffness (E: Young's modulus) are measured using kilopascals (kPa) and range from 0 to 200, which was standardized using BMI (E_{BMI}= E/BMI).

All subjects were examined in the following positions: 1) neutral ankle position: the feet were positioned shoulder-width apart outside the examination bed with the toes naturally sagging (Figure 1 a&b); 2) plantar flexion position: the feet were positioned shoulder-width apart on the examination table, with the maximum extent of ankle metatarsal flexion (Figure 1 c&d); 3) upright position: standing upright with feet shoulder-width apart (Figure 1 e&f). E and E_{BMI} of the neutral ankle position represent passive stiffness, whereas E and E_{BMI} of the plantar flexion position and upright position represent active stiffness.

The medial head of the left gastrocnemius muscle was first scanned for the longitudinal section parallel to the long axis of the muscle fiber, and the detection site with the thick muscle layer and clear muscle fiber texture was marked. Then, in the abovementioned three postures, the marked site was scanned in SWE mode with the size of the sampling frame set to 15 × 15 mm, including the muscular layer or placed in the muscular layer. Large blood vessels, nerves, and the muscular septum were avoided. After the motion stability index met the standard value, the diameter of the measuring frame was adjusted to 8 mm and the measuring frame was placed in the sampling frame with uniform color. The average E in the measuring frame was recorded as the E of the gastrocnemius muscle, and the average value of each data point for the three repeated measurements was recorded.

Figure 1. Schematic diagram of different postures and shear wave elastography. a, b: Neutral ankle position and the associated shear wave elastography; c, d: Plantar flexion position and the associated shear wave elastography; e, f: Upright position inspection and the associated shear wave elastography.

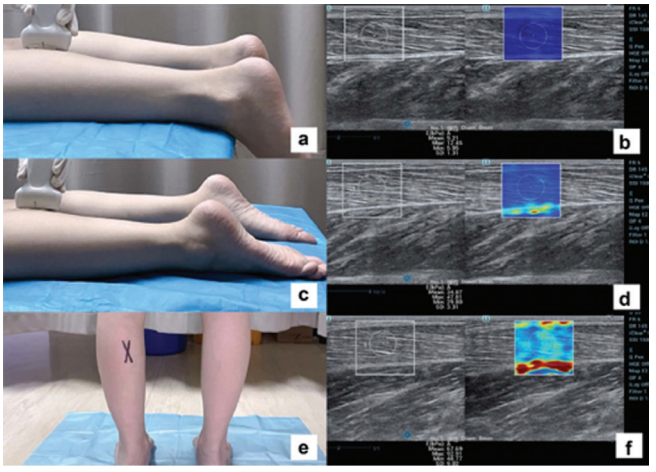


Table 1. Basic characteristics

Variables	Healthy subjects (n = 50)	T2DM patients (n = 70)	*P* value
Male, (n/%)	35, 70.00%	50, 71.43%	.865
Age (years), Mean ± SD	51.22 ± 4.94	52.79 ± 5.67	.112
Height (cm), Mean ± SD	160.8 ± 7.18	164.22 ± 5.76	.068
Weight (kg), Mean ± SD	64.80 ± 8.28	67.54 ± 9.71	.103
BMI (kg/m^2), Mean ± SD	24.31 ± 2.45	25.17 ± 3.05	.097
HbA_{1c}, Mean ± SD	4.48 ± 1.34	7.92 ± 3.76	.003
AGEs, Mean ± SD	None	85.63 ± 13.42	-

Abbreviations: BMI, body mass index; HbA_{1c}, Hemoglobin A_{1c}; AGEs, advanced glycation end-product.

Statistical analysis

Statistical analysis was performed using SPSS Statistics version 26.0 (IBM, Armonk, NY, USA). Continuous data with a normal distribution were described as means ± standard deviations and analyzed using Student's *t* test; otherwise, they were presented as medians (interquartile range, IQR) and analyzed using the Wilcoxon rank-sum test. Categorical data were described as n (%) and analyzed using the chi-square test or Fisher's exact test. Pearson's correlation test was used to analyze the correlation between E and age, disease course, HbA_{1c}, AGEs. Two-sided $P < .05$ were considered statistically significant.

RESULTS

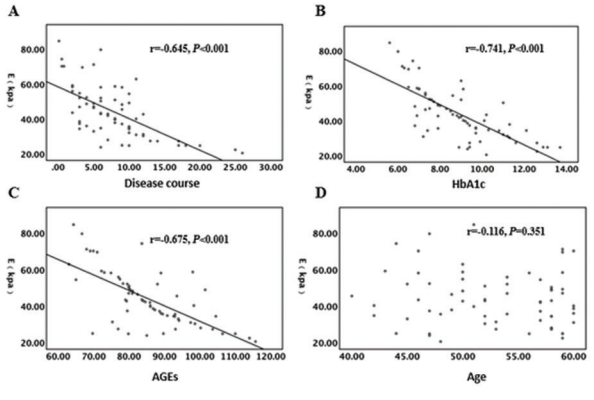
A total of 120 individuals (mean age: 52.09 ± 5.40 years, 85 males) were enrolled, including 70 patients with T2DM and 50 healthy subjects. There were no significant differences in sex, age, height, weight or BMI between T2DM patients and healthy subjects ($P > .05$). HbA_{1c} concentration was higher in T2DM patients than healthy subjects (7.92 ± 3.76 vs. 4.48 ± 1.34, $P = .003$) (Table 1). There was no significant difference in E at neutral ankle position, plantar flexion position, and E_{BMI} at neutral ankle position between T2DM patients and healthy subjects ($P > .05$). E at upright position

Table 2. Comparison of E and E_{BMI} of the gastrocnemius muscle in different positions between T2DM patients and healthy subjects

Variables		Healthy subjects (n = 50)	T2DM patients (n = 70)	*P* value
E (kPa)	Neutral ankle position	10.05 ± 2.79	10.53 ± 2.20	.285
	Plantar flexion position	30.08 (25.20- 38.36)	29.88 (24.02- 36.68)	.637
	Upright position	51.71 ± 9.48	43.89 ± 14.93	.001
E_{BMI} (kPa, kg/m²)	Neutral ankle position	0.42 (0.32-0.52)	0.41 (0.34-0.50)	.667
	Plantar flexion position	1.55 (1.21-1.84)	1.17 (0.82-1.29)	<.001
	Upright position	2.10 (1.88-2.29)	1.72 (1.23-2.16)	<.001

Note: E, Young's modulus; E_{BMI}, body mass index standardized Young's modulus (E_{BMI} =E/BMI). E and E_{BMI} of the neutral ankle position represent passive stiffness, whereas E and E_{BMI} of the plantar flexion position and upright position represent active stiffness.

Figure 2. Scatter diagram of the relationship between E of the gastrocnemius muscle at upright position and disease course, HbA_{1c}, AGEs and age in T2DM patients.



(43.89 ± 14.93 vs. 51.71 ± 9.48, P = .001), E_{BMI} at plantar flexion position (1.17 (0.82-1.29) vs. 1.55 (1.21-1.84), P < .001) and upright position (1.72 (1.23-2.16) vs. 2.10 (1.88-2.29), P < .001) of the T2DM patients were significantly lower than those of healthy subjects (P < .05) (Table 2). In T2DM patients, E at upright position was negatively correlated with the disease course (r=-0.645, P < .001), HbA_{1c} concentration (r=-0.741, P < .001), and AGEs (r=-0.675, P < .001) but not with age ((r=-0.116, P = .351) (Figure 2).

DISCUSSION

This study found that active muscle stiffness was significantly lower in T2DM patients compared to healthy controls using SWE, suggesting that evaluation of active muscle stiffness using SWE may be valuable in T2DM patients to prevent gastrocnemius muscle damage.

The study found that the active muscle stiffness was significantly lower in T2DM patients compared to healthy subjects. This finding is consistent with previous studies using SWE. Alfuraih et al. reported that decline in skeletal muscle stiffness was positively correlated with muscle weakness in patients with idiopathic inflammatory myopathies.[5,13] Chen et al. demonstrated that in elderly T2DM patients, sarcopenia patients had lower muscle stiffness than non-sarcopenia patients.[14] Alfuraih et al. reaffirmed that individuals with T2DM had an accelerated decline in muscle strength compared with healthy subjects,[15] and SWE might be a sensitive assessment tool to detect the changes. Furthermore, the reduced active muscle stiffness among T2DM patients reflects the decrease in contractile function. This condition might be due to long-term hyperglycemia which leads to the glycosylation of proteins and enzymes involved in the production of AGEs.[16] AGEs downregulate the protein kinase B signal pathway through adenylate-activating enzyme to induce myogenesis disorder or muscle atrophy.[17] This study found no significant differences in passive muscle stiffness. Gastrocnemius muscle stiffness significantly increased from passive muscle stiffness to active muscle stiffness.[18] Meanwhile, T2DM is primarily a metabolism disorder and does not cause significant muscle tone changes in the early stages. Therefore, although the standardized passive muscle stiffness was lower in T2DM patients, the difference was relatively small and insignificant. Passive muscle stiffness plays an important role in stabilizing joints, whereas active muscle stiffness is essential for dynamic stability.[19] Thus, active muscle stiffness can better represent the dynamic ability to perform activities of daily living in T2DM patients. This study demonstrated reduced active muscle stiffness in T2DM patients, and their ability to perform daily activities might be affected.

None of the T2DM patients in this study showed apparent muscle injuries or functional decline, such as myalgia and muscle weakness. However, their muscle contractile function was decreased, indicating it was still in the subclinical stage. SWE technology can detect the decrease of active hardness in skeletal muscle contraction from the changes in the contraction of muscle fiber length. It is important to consider the factors related to forced muscle contraction, such as the standardization of BMI in this study to reflect the decline of muscle function. However, there is no need to correct the measurement in the state of muscle isometric contraction, which may provide a new idea for using ultrasonic SWE technology to detect the changes in muscle contraction function in pathological conditions. Additionally, Chen et al. used SWE to assess muscle mass and function in T2DM patients and found that sarcopenia patients had less muscle stiffness.[14] However, subjects in this previous study were aged 50-80 years, which was significantly older than the studied population in this study. The present study demonstrated that SWE can be applied in younger T2DM patients to detect skeletal muscle damage in the subclinical stage.

The present study showed that the decline in the active stiffness of the gastrocnemius muscle at upright position in T2DM patients was not significantly related to age. This finding was contradictory to the finding in Alfuraih's study. [13] The discrepancy might be due to that the age range in our study was narrower (40-60 years vs. 20-94 years). The study also found that E at upright position was significantly correlated with HbA_{1c} and AGEs. Past studies showed that high HbA_{1c} concentration might lead to loss of muscle mass and strength.[20] AGEs induce skeletal muscle atrophy and insulin resistance, which can reduce the tension of muscle

contraction.[21] In clinical practice, the AGEs level is commonly used to assess the risk of complications in T2DM patients. Therefore, active muscle stiffness might also be an effective and sensitive indicator of risk assessment for T2DM patients.

However, there were still several limitations in this study. Firstly, the cut-off value of E for evaluating gastrocnemius muscle stiffness was not determined. Further research is needed to determine the cut-off value for clinical reference. Secondly, this study investigated muscle stiffness during isometric and centripetal contractions (upright position and plantar flexion position), however, other muscle contraction forms, such as isokinetic and eccentric contraction were not studied. Thirdly, this study was limited by the single-centered design. Multi-center studies with larger sample sizes were needed to furtherly confirm the findings.

In conclusion, early detection and intervention would prevent gastrocnemius muscle damage and improve patients' functions and quality of life.

CONFLICT OF INTEREST

The authors have no potential conflicts of interest to report relevant to this article.

AUTHOR'S CONTRIBUTIONS

All authors contributed to the study's conception and design. Material preparation, data collection, and analysis were performed by Xue Fang, XiaoXi Sha, ZhiFen Han and Yan Zhang. The first draft of the manuscript was written by Yu Kang and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript. Xue Fang and Zhifen Han contributed equally to this work

FUNDING/SUPPORT

This study was funded by the National Administration of Traditional Chinese Medicine (grant number: JDZX2015218).

ETHICAL STATEMENTS

This study was performed in accordance with the Helsinki Declaration of 1964 and later versions and was approved by the Hospital of Chengdu University of Traditional Chinese Medicine Ethics Committee.

REFERENCES

1. D'Souza DM, Al-Sajee D, Hawke TJ; Diabetic myopathy: impact of diabetes mellitus on skeletal muscle progenitor cells. *Front Physiol*. 2013;4(379. doi:10.3389/fphys.2013.00379.
2. Hernández-Ochoa EO, Llanos P, Lanner JT. The Underlying Mechanisms of Diabetic Myopathy. *J Diabetes Res*. 2017;2017:7485738. doi:10.1155/2017/7485738
3. Huang BK, Monu JU, Doumanian J. Diabetic myopathy: MRI patterns and current trends. *AJR Am J Roentgenol*. 2010;195(1):198-204. doi:10.2214/AJR.09.2494
4. Bassi-Dibai D, Santos-de-Araujo AD, Dibai-Filho AV, et al; Rehabilitation of Individuals With Diabetes Mellitus: Focus on Diabetic Myopathy. *Front Endocrinol*. 2022;13(869921. doi:10.3389/fendo.2022.869921.
5. Alfuraih AM, O'Connor P, Tan AL, et al. Muscle shear wave elastography in idiopathic inflammatory myopathies: a case-control study with MRI correlation. *Skeletal Radiol*. 2019;48(8):1209-1219. doi:10.1007/s00256-019-03175-3
6. Sigrist RMS, Liau J, Kaffas AE, Chammas MC, Willmann JK. Ultrasound Elastography: Review of Techniques and Clinical Applications. *Theranostics*. 2017;7(5):1303-1329. doi:10.7150/thno.18650
7. Šarabon N, Kozinc Ž, Podrekar N. Using shear-wave elastography in skeletal muscle: A repeatability and reproducibility study on biceps femoris muscle. *PLoS One*. 2019;14(8):e0222008. doi:10.1371/journal.pone.0222008
8. Wang Z, Lyu G, Zhong H, Yan L, Xu Z. Shear Wave Elastography for Detecting Calf Muscle Stiffness: An Effective Tool for Assessing Sarcopenia. *J Ultrasound Med*. 2023;42(4):891-900. doi:10.1002/jum.16082
9. Bernabei M, Lee SSM, Perreault EJ, Sandercock TG. Shear wave velocity is sensitive to changes in muscle stiffness that occur independently from changes in force. *J Appl Physiol*. 2020;128(1):8-16. doi:10.1152/japplphysiol.00112.2019
10. Wang JC, Wu WT, Chang KV, et al. Ultrasound Imaging for the Diagnosis and Evaluation of Sarcopenia: An Umbrella Review. *Life (Basel)*. 2021;12(1):9. doi:10.3390/life12010009
11. Creze M, Nordez A, Soubeyrand M, Rocher L, Maître X, Bellin MF. Shear wave sonoelastography of skeletal muscle: basic principles, biomechanical concepts, clinical applications, and future perspectives. *Skeletal Radiol*. 2018;47(4):457-471. doi:10.1007/s00256-017-2843-y
12. Alberti KG, Zimmet PZ. Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus provisional report of a WHO consultation. *Diabet Med*. 1998;15(7):539-553. doi:10.1002/(SICI)1096-9136(199807)15:7<539::AID-DIA668>3.0.CO;2-S
13. Alfuraih AM, Tan AL, O'Connor P, Emery P, Wakefield RJ. The effect of ageing on shear wave elastography muscle stiffness in adults. *Aging Clin Exp Res*. 2019;31(12):1755-1763. doi:10.1007/s40520-019-01139-0
14. Chen ZT, Jin FS, Guo LH, et al. Value of conventional ultrasound and shear wave elastography in the assessment of muscle mass and function in elderly people with type 2 diabetes. *Eur Radiol*. 2023;33(6):4007-4015. doi:10.1007/s00330-022-09382-2
15. Leenders M, Verdijk LB, van der Hoeven L, et al. Patients with type 2 diabetes show a greater decline in muscle mass, muscle strength, and functional capacity with aging. *J Am Med Dir Assoc*. 2013;14(8):585-592. doi:10.1016/j.jamda.2013.02.006
16. Mengstie MA, Chekol AE, Behaile TA, et al; Endogenous advanced glycation end products in the pathogenesis of chronic diabetic complications. *Front Mol Biosci*. 2022;9(1002710. doi:10.3389/fmolb.2022.1002710.
17. Chiu CY, Yang RS, Sheu ML, et al. Advanced glycation end-products induce skeletal muscle atrophy and dysfunction in diabetic mice via a RAGE-mediated, AMPK-down-regulated, Akt pathway. *J Pathol*. 2016;238(3):470-482. doi:10.1002/path.4674
18. Hoffman LR, Koppenhaver SL, MacDonald CW, et al; Normative Parameters of Gastrocnemius Muscle Stiffness and Associations with Patient Characteristics and Function. Normative Parameters of Gastrocnemius Muscle Stiffness and Associations with Patient Characteristics and Function. *Int J Sports Phys Ther*. 2021;16(1):41-48. doi:10.26603/001c.18803
19. Duan XH, Allen RH, Sun JQ. A stiffness-varying model of human gait. *Med Eng Phys*. 1997;19(6):518-524. doi:10.1016/S1350-4533(97)00022-2
20. Qiao YS, Chai YH, Gong HJ, et al; The Association Between Diabetes Mellitus and Risk of Sarcopenia: Accumulated Evidences From Observational Studies. *Front Endocrinol*. 2021;12(782391. doi:10.3389/fendo.2021.782391.
21. Du H, Ma Y, Wang X, et al; Advanced glycation end products induce skeletal muscle atrophy and insulin resistance via activating ROS-mediated ER stress PERK/FOXO1 signaling. *Am J Physiol-Endoc M*. 2023;324(3):E279-E287. doi: 10.1152/ajpendo.00218. 2022.