DIAGNOSIS OF ARTIFICIAL HYPOGLYCEMIA DUE TO ORAL HYPOGLYCEMIC DRUGS BY HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY WITH TANDEM MASS SPECTROMETRIC DETECTION

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BACKGROUND: Artificial hypoglycemia (ArH) is a decrease of blood glucose levels less than 3 mmol/l due to the deliberate use of hypoglycemic drugs by a patient outside of medical appointments. Timely diagnosis of this kind of hypoglycemia avoids unnecessary numerous examinations and hospitalizations. However, the detection of ArH still remains an extremely difficult task for both the healthcare facility and the attending physician. The foreign literature describes cases of successful detection of deliberate intake of oral hypoglycemic drugs (OHD) using high-performance liquid chromatography with tandem mass spectrometric detection (HPLC-MS/MS). Thus, it is relevant to develop and validate a method for determining OHD using HPLC-MS/MS.
AIM: To optimize the diagnosis of ArH due to the use of OHD.
MATERIALS AND METHODS: A total of 92 patients were examined. The development of the HPLC-MS/MS method for the detection of the studied OHD (sOHD; n=1-glibenclamide, n=1-gliquidone, n=1-gliclazide, n=1-glimepiride, n=1-glipizide, n=1-nateglinide and n=1-repaglinide) in the blood was carried out in a group of patients with diabetes mellitus type 2 (n=7) who received sOHD, and a group of conditionally healthy people who did not receive any medications (n= 7). To validate the method, the determination of sOHD substances was carried out on groups of patients with hyperinsulinemic nondiabetic hypoglycemia (NDH) of unknown origin (n=11) and with insulinoma (n=67).
RESULTS: In the study of blood samples by HPLC-MS/MS in the group of patients with diabetes mellitus type 2, was confirmed in 100% of the cases the use of the drug that the patient received, in the group of conditionally healthy — sOHD were not detected. A false positive result was not obtained in any conditionally healthy and in any patient with insulinoma. ArH was diagnosed in 5 out of 11 patients in the group with hyperinsulinemic NDH of unknown origin, the method identified sOHD glibenclamide and gliclazide in the patients' blood samples. In the remaining 6 patients of this group, examinations were continued and other causes of NDH were diagnosed. The sensitivity of the method was 100% [74%; 100%], specificity — 100% [95%; 100%].
CONCLUSION: The HPLC-MS/MS method has high diagnostic accuracy in the detection and identification of sOHD (glibenclamide, gliquidone, gliclazide, glimepiride, glipizide, nateglinide and repaglinide) in blood samples of patients receiving these drugs. Currently, due to the low availability of the method, this study is advisable to use in patients with hyperinsulinemic hypoglycemia with negative results of first-line insulinoma imaging methods (computed tomography with contrast enhancement, ultrasound and magnetic resonance imaging of the abdominal cavity).

KEYWORDS: artificial hypoglycemia; sulfonylurea derivatives; glinides; substances; high-performance liquid chromatography with tandem mass spectrometric detection.

ДИАГНОСТИКА АРТИФИЦИАЛЬНОЙ ГИПОГЛИКЕМИИ ВСЛЕДСТВИЕ ПРИЕМА ПЕРОРАЛЬНЫХ САХАРОСНИЖАЮЩИХ ПРЕПАРАТОВ МЕТОДОМ ВЫСОКОЭФФЕКТИВНОЙ ЖИДКОСТНОЙ ХРОМАТОГРАФИИ С ТАНДЕМНЫМ МАСС-СПЕКТРОМЕТРИЧЕСКИМ ДЕТЕКТИРОВАНИЕМ

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ВВЕДЕНИЕ. Артифициальная гипогликемия (АрГ) — это снижение уровня глюкозы крови менее 3 ммоль/л вследствие умышленного применения сахароснижающих препаратов пациентом вне врачебных назначений. Своевременная диагностика такого рода гипогликемий позволяет избежать излишних многочисленных обследований и госпитализаций. Однако выявление АрГ до сих пор остается крайне сложной задачей как для медицинского учреждения, так и для лечащего врача. В зарубежной литературе описаны случаи успешного выявления преднамеренного приема пероральных сахароснижающих препаратов (ПССП) с помощью высокоэффективной жидкостной хроматографии с тандемным масс-спектрометрическим детектированием (ВЭЖХ-МС/МС). Таким образом, актуальна разработка и валидация метода определения ПССП с помощью ВЭЖХ-МС/МС.

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ЦЕЛЬ ИССЛЕДОВАНИЯ. Оптимизация диагностики АрГ вследствие приема ПССП.
МАТЕРИАЛЫ И МЕТОДЫ. Всего обследовано 92 пациента. Разработка метода ВЭЖХ-МС/МС для обнаружения исследуемых ПССП (иПССП) (n=1-глибенкламид, n=1-гликвидон, n=1-гликлазид, n=1-глимепирид, n=1-глипизид, n=1-натеглинид и n=1-репаглинид) в крови осуществлена на группе пациентов с сахарным диабетом 2 типа (n=7), которые получали иПССП, и группе условно здоровых, не получавших никакие лекарственные препараты (n=7). Для валидации метода определение субстанций иПССП осуществлено на группах пациентов с гиперинсулинемической недиабетической гипогликемией (НДГ) неясного генеза (n=11) и с инсулиномой (n=67).
РЕЗУЛЬТАТЫ. При исследовании образцов крови методом ВЭЖХ-МС/МС в группе пациентов с сахарным диабетом 2 типа в 100% случаев подтвержден прием препарата, который получал больной, в группе условно здоровых — иПССП не выявлены. Ложноположительный результат не получен ни у условно здоровых и ни у одного пациента с инсулиномой. У 5 из 11 пациентов группы с гиперинсулинемической НДГ неясного генеза диагностирована АрГ, метод идентифицировал иПССП глибенкламид и гликлазид в образцах крови пациентов. У остальных 6 пациентов этой группы были продолжены обследования и диагностированы другие причины НДГ. Чувствительность метода составила 100% [74%; 100%], специфичность — 100% [95%; 100%].
ЗАКЛЮЧЕНИЕ. Метод ВЭЖХ-МС/МС обладает высокой диагностической точностью в обнаружении и идентификации иПССП (глибенкламид, гликвидон, гликлазид, глимепирид, глипизид, натеглинид и репаглинид) в образцах крови пациентов, получающих данные лекарственные препараты. В настоящее время в силу малой доступности метода данное исследование целесообразно применять у пациентов с гиперинсулинемической гипогликемией при отрицательных результатах методов визуализации инсулиномы первого ряда (компьютерная томография с контрастированием, ультразвуковое исследование и магнитно-резонансная томография органов брюшной полости).

КЛЮЧЕВЫЕ СЛОВА: артифициальная гипогликемия; производные сульфонилмочевины; глиниды; субстанции; высокоэффективная жидкостная хроматография с тандемным масс-спектрометрическим детектированием.

BACKGROUND

Munchausen Syndrome (MS) is a collective term combining artificial (factitious) disorders. An individual with MS intentionally induces or simulates physical/mental signs and symptoms of a certain pathology in order to be perceived by others as sick without any external incentives for such behavior [1].

Artificial hypoglycemic syndrome or artificial hypoglycemia (ArH) is one of the most common artificial disorders [2] and means deliberate use of insulin or oral hypoglycemic drugs with the purpose to reduce blood glucose levels [3]. This condition was described both in individuals without impaired carbohydrate metabolism and in patients with diabetes mellitus (DM) [2, 4, 5]; it is often found in healthcare workers [4], and also in patients having relatives with DM. Another risk factors include comorbid psychiatric disorders and socio-psychological problems [4, 6, 7, 8]. Artificial disorders are often characterized by inadequate changes in the symptoms and the disease course, including changes during treatment, frequent hospitalizations and consultations with various specialty physicians. Unexplained prolonged "remission" of hypoglycemia often occurs, followed by resumption of episodes [9]. It is important to note that patients with MS may forge laboratory tests or other medical documentation [6].

As a rule, patients with ArH deny the use of hypoglycemic drugs, do not recognize this fact even after presenting them with evidence (for example, laboratory test results) [2]. Timely diagnosis of MS is first of all an opportunity to discuss the situation with the patient, to talk about possible irreversible damage to health, including the risk of fatality. In addition, establishing the artificial disorder diagnosis, relieves the need for a variety of diagnostic tools to find the causes of hypoglycemia, which often include invasive and expensive procedures, including surgical interventions. In the management of such patients, a multidisciplinary approach is required with the mandatory participation of a psychiatrist [3, 6, 7, 8].

Diagnosis of ArH is difficult, as laboratory signs are similar to those in other diseases associated with nondiabetic hypoglycemia (NDH), for example, with insulinoma. Only absolute round-the-clock observation of the patient and the exclusion of the use of personal belongings was made it possible to confirm ArH. But such measures should be carried out with the consent of the patient, which is usually not given, in addition, the hospital has to be able to provide a round-the-clock observation. As a result, until recently, in most cases ArH patients in Russia remained with an unspecified diagnosis. At the same time, foreign literature describes cases of successful detection of deliberate use of oral hypoglycemic drugs (OHD) using high-performance liquid chromatography with tandem mass spectrometric detection (HPLC-MS/MS) [10].

PURPOSE OF THE STUDY

Given the relevancy of the problem, the purpose of this study is to optimize the diagnosis of ArH due to the use of OHD by detection of OHD using HPLC-MS/MS.

MATERIALS AND METHODS

Study site and time

National Scientific Center – Endocrinology Research Centre of the Ministry of Health of Russia, 2017–2022.

Study population

Patients examined at the Endocrinology Research Centre of the Ministry of Health of Russia.

Inclusion criteria for all groups: male or female patient aged 18+ years. Exclusion criteria: none.

Study design

Single-center experimental study.

Methods

The study included two phases. At each phase pairs of patient groups were formed.

Phase 1: Development of a HPLC-MS/MS method for the detection of sOHD in the blood of patients with type 2 diabetes mellitus (T2D) treated with sOHD and in the group of apparently healthy subjects.

The flowchart of Phase 1 is shown in Figure 1.

The time for blood sampling in Group 1 (1 hour after taking sOHD) was selected based on the time at which the absorption of any oral drug already begins in the GI tract and the drug enters the bloodstream [11, 12, 13, 14, 15, 16]. The minimum amount of the drug is sufficient for its determination by HPLC-MS/MS, taking into account its high sensitivity – about 10 pg/mL for each component, which makes it possible to detect traces of the drug even a week after administration.

Phase 2: Validation of the HPLC-MS/MS method for detection of sOHD in the group of patients with hyperinsulinemic nondiabetic hypoglycemia (NDH).

The flowchart of Phase 2 is shown in Figure 2.

Laboratory tests

Biochemical and hormonal studies were carried out in the clinical diagnostic laboratory of the Endocrinology Research Centre of the Ministry of Health of Russia. Blood was drawn into vacuum tubes with an inert gel. The obtained samples were centrifuged not later than 15 minutes after sampling using a centrifuge Eppendorf 5810R at a temperature 4 °C at 3000 RPM for 15 minutes and then transferred to further operations. Insulin and C-peptide were determined by enhanced chemiluminescence on COBAS 6000 analyzer (Roche Diagnostics, Switzerland). Blood chemistry was performed using an analyzer Architect plus C 4000 (Abbott Diagnostics, USA).

Detection of sOHD (glibenclamide, glycvidone, gliclazide, glimepiride, glipizide, nateglinide, repaglinide) in blood samples was performed by HPLC-MS/MS. This was done using liquid chromatograph Agilent 1290 Infinity II (Agilent Technologies, Germany) equipped with a four-channel pump, autosampler and column thermostat, and a hybrid three-quadrupole mass spectrometer AB Sciex QTrap 5500 (AB Sciex, Singapore) with linear ion trap mode.

Serum samples were prepared by salting-out assisted liquid-liquid extraction (SALLE). For this purpose, 400 µl of acetonitrile was added to 200 µl of serum, the mixture was stirred in a shaker for 10 minutes and then centrifuged for 1 minute at 14,800 rpm at 5 °C. To the resulting solution 200 µl of 5M ammonium acetate (Fluka, > 99%, Netherlands) was added, stirred for 3 minutes in a shaker and centrifuged for 2 minutes at 14,800 rpm and 5 °C. Then 50 µl of the upper organic layer was collected, placed in a 96-well plate, 50 µl of deionized water was added, and mixed by pipetting. The resulting solution was used for chromatographic-mass spectrometric analysis.

The components were separated chromatographically using the column Accucore PFP 2.1x50 mm, 2.6 µm particle diameter (Thermo Scientific, USA). Acetonitrile (Honeywell, for LC-MS, Germany) and deionized water (MilliQ Advantage A10, Millipore, Germany) were used as eluents. The separation was carried out in gradient mode with a gradient of the organic phase from 5 to 60% from 1st to 6th minute, with an isocratic section from 6th to 7th minute at 60% of acetonitrile

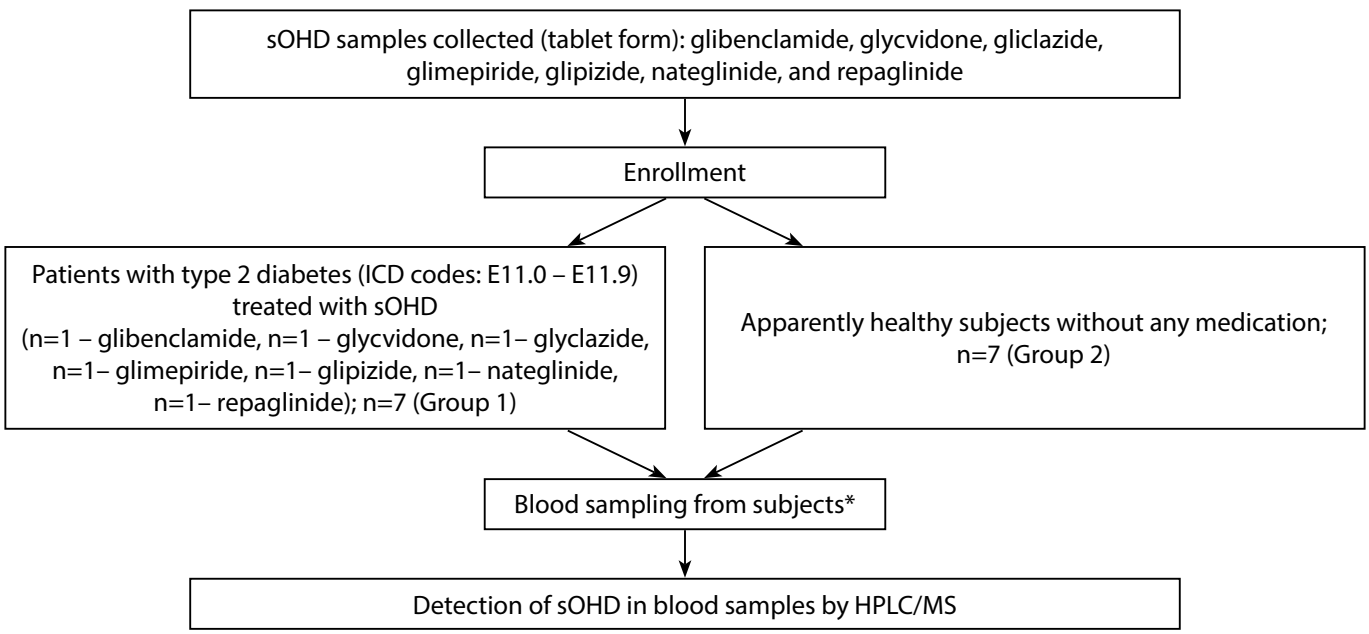

Figure 1. Flowchart of Phase 1

Abbreviations: sOHD – study oral hypoglycemic drugs; HPLC-MS/MS – high-performance liquid chromatography with tandem mass spectrometric detection;

* in group 1, a blood sample was taken in the morning on an empty stomach 1 hour after taking the drug, in group 2 a blood sample was taken in the morning on an empty stomach.

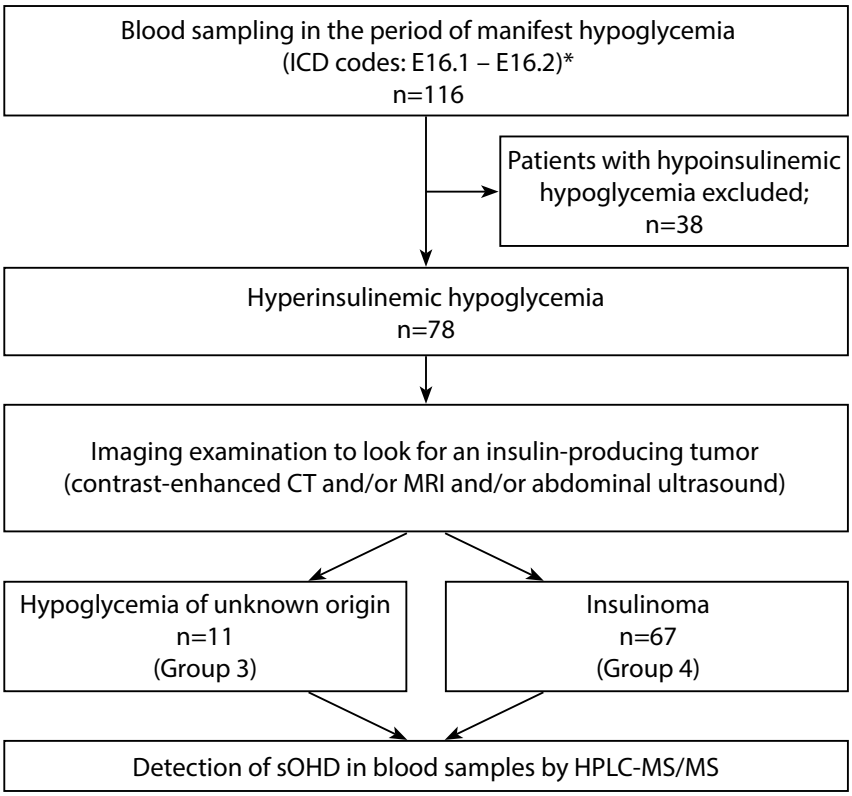

Figure 2. Flowchart of Phase 1

Abbreviations: sOHD – study oral hypoglycemic drugs; HPLC-MS/MS – high-performance liquid chromatography with tandem mass spectrometric detection; CT – computed tomography; MRI – magnetic resonance imaging; U/S – ultrasound study

* Manifest hypoglycemia was defined as venous glucose less than 3 mmol/L; hyperinsulinemic hypoglycemia – C-peptide ≥ 0.6 ng/ml and insulin ≥ 3 μIU/ml; hypoinsulinemic hypoglycemia – C-peptide < 0.6 ng/ml and insulin < 3 μIU/ml [10]. Blood sampling was performed in the period of manifest hypoglycemia during a fasting test or with spontaneous hypoglycemia. Serum samples of patients with hyperinsulinemic hypoglycemia were sent for the detection of sOHD. Without waiting for the results of this analysis, patients underwent a standard imaging examination.

and a return to the initial 5% from 7th to 8th minute, followed by equilibration of the column to 11th minute. The eluent flow was 0,4 mL/min, column temperature – 20 °C, sample injection volume – 20 μL.

Mass spectrometric detection was performed in a multiple reaction monitoring (MRM) mode combined with information-dependent monitoring (IDA), which was implemented on a linear ion trap. An atmospheric pressure chemical ionization (APCI) in positive ion mode was used as ionization source. MRM transitions were selected for each component individually by injecting the reference standard in chromatographic flow directly into the ionization source and sequentially varying the fragmentation parameters. Two MRM transitions were recorded for each component.

The criteria for the identity of the substance from the patient sample with the substance in the control sample are the coincidence of the retention times within 0.2 minutes, the ratio of areas under the chromatographic peaks for two MRM transitions for the substance being determined, as well as the coincidence of the spectra of fragment ions for the sample component and the control.

Analysis management, data acquisition and processing were performed using the software Analyst 1.6.3 (AB Sciex, Canada).

Statistical analysis

Statistical processing of the obtained data was carried out with standard methods for statistical analysis using the software STATISTICA v. 13 (TIBCO Inc., USA). For quantitative variables, the median and interquartile range are presented, in some cases the minimum and maximum values are given. The Mann-Whitney test was used to compare the quantitative data of two independent samples. The threshold for statistical significance in testing statistical hypotheses was 0.05. Bonferroni correction was applied to adjust multiple comparisons. After adjustment, P-values between the calculated value and 0.05 were interpreted as a statistical trend.

Ethical review

The study was approved by the local ethics committee of the Endocrinology Research Centre (Minutes No. 1 of 27.01.2016). All patients signed the informed consent form to participate in the study.

RESULTS

Phase 1. Groups 1 and 2 included 7 consecutive patients. Patient age was 59 [54; 67] years in Group 1, and 45 [41; 51] years in Group 2. Each group includes 4 women and 3 men.

The HPLC-MS/MS study of blood samples of patients from Group 1 confirmed in 100% cases that the patient used an OHD: n=1 – glibenclamide, n=1 – glycvidone, n=1 – glimepiride, n=1 – glipizide, n=1 – nateglinide, and n=1 – repaglinide. The HPLC-MS/MS study of blood samples of patients from Group 2 did not reveal any OHD.

Phase 2. Blood samples were taken from 116 patients in the period of manifest hypoglycemia, of which 38 were diagnosed with hypoinsulinemic hypoglycemia, and these patients were excluded from the study. The remaining 78 patients underwent medical imaging to look for an insulin-producing tumor (contrast-enhanced CT and/or MRI and/or abdominal ultrasound). Hypoglycemia, unspecified, was diagnosed in 11 patients (Group 3), insulinoma was diagnosed in 67 patients (Group 4).

Patient age was 43 [35; 56] years in Group 3, and 51 [34; 62] years in Group 4. Group 3 included 9 women and 2 men, Group 4 included 52 women and 15 men.

No patient in Group 4 had a false positive result. The method identified sOHDs (glibenclamide and gliclazide) in blood samples from 5 of 11 patients in Group 3 (Subgroup 3a). Chromatograms of two of these patients are presented in Figures 3 and 4.

In 6 patients of Group 3 in whose blood samples no sOHD was found (Subgroup 3b), examinations were continued and the following causes of NDH were diagnosed: insulinoma (total n=4; by scintigraphy with SPECT/CT, ^{99m}Tc-Tectrotide (n=3), and endoscopic ultrasound (n=1)), congenital hyperinsulinism due to a mutation in the *ABCC8* gene (n=1), and non-insulin pancreatogenic hypoglycemia

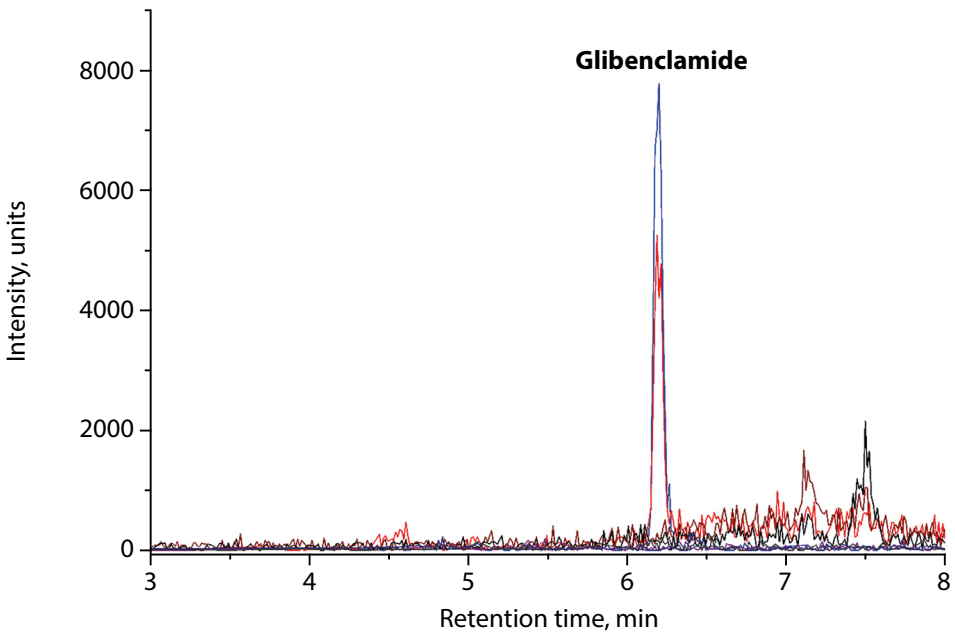

Figure 3. Chromatogram of a blood sample of a patient with artificial hypoglycemia due to glibenclamide [17].

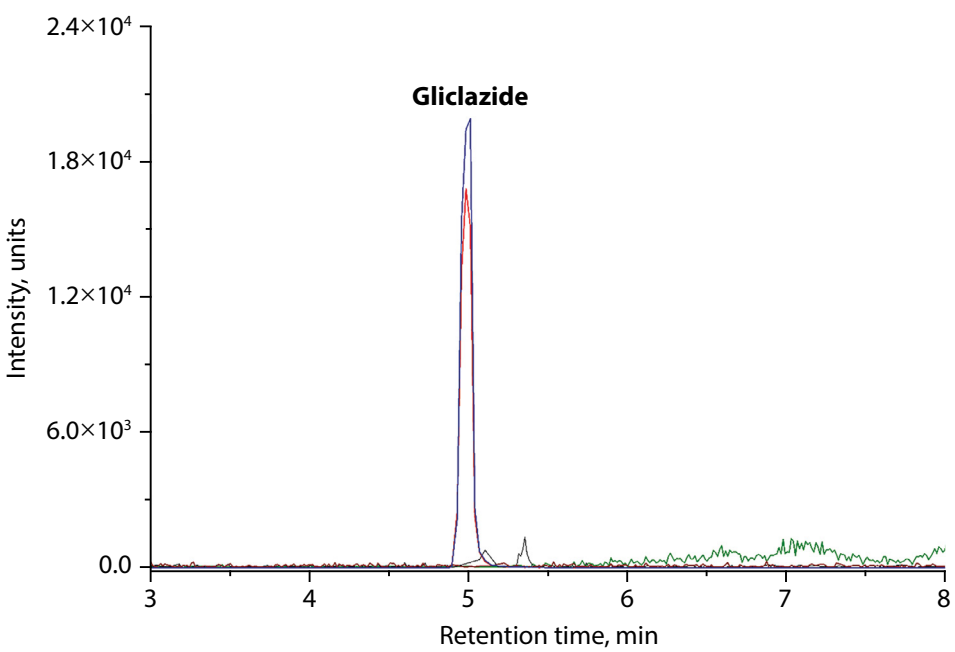

Figure 4. Chromatogram of a blood sample of a patient with artificial hypoglycemia due to gliclazide.

(n=1; confirmed by arterial-stimulated venous blood draw).

Thus, patients in Group 1 (n=7) and Group 3a (n=5) had sOHD detected in the blood, while patients in Group 2 (n=7), Group 4 (n=67) and Group 3b (n=6) had no sOHD detected in the blood. To calculate the performance characteristics of the HPLC-MS/MS method in the detection of sOHD, a cross-tabulation table was compiled in the combined groups: T2D patients receiving sOHD and patients with NDH in whom the diagnosis of insulinoma was excluded by standard methods (n=12); patients with specified causes of NDH, and healthy subjects (n=80) (Table 1). The sensitivity of the method was 100%, 95% CI (74%; 100%), specificity – 100%, 95% CI (95%; 100%).

In order to identify additional serological markers of ArH due to the use of sOHD, which would help to differentiate this condition from laboratory parameters in insulinoma, a comparative analysis of insulin, glucose and C-peptide parameters in the period of manifest hypoglycemia in Group 3a (n=5) and in the combined group of patients with insulinoma (Group 2, n=67) and other causes of non-artificial hyperinsulinemic NDH (Group 3b; n=6). Insulin levels in Group 3a were statistically significantly lower than in Group 2+3b, while glucose and C-peptide levels were not different (Table 2).

A significant increase in insulin in patients No. 2 and No. 3 from Group 3a was noteworthy in the period of manifest hypoglycemia, when compared to other patients (Table 3). It is suggested that insulin levels may be affected by the presence of insulin resistance in the patient. To clarify this hypothesis, HOMA-IR was calculated for all patients of Group 3a after overnight fasting in the period of normoglycemia, which did not exceed the reference value 2.7 (Table 3).

The significant increase in insulin (as well as C-peptide) in patient No. 2 was caused by the end stage of chronic kidney disease due to delayed elimination of hormones (especially C-peptide) [18]. In this regard, a comparative analysis of C-peptide in the period of manifest hypoglycemia was carried out for Group 3a (with patient No. 2 data rejected) and for combined Group 2+3b. It was found that C-peptide level in Group 3a (1.82 [1.18; 2.73]) was lower than in Group 2+3b (p=0.015, U-test).

Table 1. Cross-tabulation of frequencies for calculation of performance characteristics of high-performance liquid chromatography with tandem mass spectrometry in detection of sOHD

Result of HPLC-MS/MS	Groups 1+3a n=12	Groups 2+4+3b n=80
At least one sOHD detected	12 (TP)	0 (FP)
No sOHDs detected	0 (FN)	80 (TN)

Abbreviations: sOHD – study oral hypoglycemic drugs; HPLC-MS/MS – high-performance liquid chromatography with tandem mass spectrometric detection; TP – true positive; FP – false positive; FN – false negative; TN – true negative.

Table 2. Glucose, insulin and C-peptide parameters in the period of manifest hypoglycemia in Group 3a and Group 2+3b (Me [Q1; Q3], (min, max))

	Group 3a, n=5	Group 2+3b, n=73	P*, U-test
Glucose, mmol/L	2.18 [2.13; 2.61] (2.00. 2.72)	2.16 [1.66; 2.41] (0.41. 2.90)	0.289
Insulin, μU/mL	8.99 [6.05; 16.20] (5.91. 21.39)	24.13 [16.00; 35.00] (4.09. 387.30)	0.009
C-peptide, ng/mL	2.30 [1.33; 3.15] (1.03. 23.91)	4.00 [2.79; 5.52] (0.67. 40.00)	0.153

* Threshold R0=0,05:3≈0,017.

Table 3. Results of examination of Group 3a patients in the period of hypoglycemia and normoglycemia

Patient	Manifest hypoglycemia			Normoglycemia (after overnight fasting)
	Glucose, mmol/L	Insulin, μU/mL	C-peptide, ng/mL	HOMA-IR
No. 1	2.72	8.99	2.3	1.833
No. 2	2.18	21.39	23.91	2.321
No. 3	2.13	16.2	3.15	1.855
No. 4	2	5.91	1.03	2.041
No. 5	2.61	6.05	1.33	1.877

Abbreviation: HOMA-IR – Homeostasis Model Assessment of Insulin Resistance

DISCUSSION

The results of phase 1 demonstrated the feasibility of the HPLC-MS/MS method in the detection and identification of OHD substances in clinical practice. In the process of method validation in the group of patients with hyperinsulinemic hypoglycemia during phase 2, we, along with other researches [19, 20, 21, 22], confirmed the high diagnostic accuracy of HPLC-MS/MS in the detection of OHD substances, including in relation to ArH. Thus, the method can be used for diagnosis of deliberate use of drugs of this group and differential diagnosis with other causes of hyperinsulinemic variant of NDH. However, taking into account the relatively rare occurrence of ArH among patients without DM (up to 3.5%) [23], low availability of HPLC-MS/MS in Russia (mainly in specialized centers), routine study in all patients with suspected NDH, as proposed by foreign experts [10], is impractical. Therefore, in our opinion, the detection of OHD substances in patients with hyperinsulinemic hypoglycemia is rational only with negative results of standard insulinoma imaging methods (contrast-enhanced CT, MRI, abdominal and retroperitoneal ultrasound).

Low availability of HPLC-MS/MS in endocrinological hospitals determines the need in other, more accessible, ArH markers. Although our study showed that in ArH, in contrast to other causes of hyperinsulinemic hypoglycemia, insulin levels are significantly lower (8.99 [6.05; 16,20]), taking into account the small number of patients with ArH, these data must be interpreted with caution, further accumulation of data and a study on a larger group of patients are required.

It is important to note that not all patients with ArH had lower insulin levels. In addition to patient No. 2, in whom a significant increase in insulin and C-peptide (21.39 μU/ml and 23.91 ng/ml, respectively) is due to the end stage of chronic kidney disease, a relatively high level of insulin was also noted in patient No. 3 (16.2 μU/ml). After calculating the NOMA index in all patients with ArH, our assumption about the possible effect of insulin resistance on a higher insulin level was rejected – insulin resistance was not detected in any patient in Group 3a. In our opinion, different insulin levels are most likely due to the dose of the drug taken or different sensitivity to OHD. Unlike other researches [22], we did not quantify the content of OHD substances in the blood of patients, since this was not the purpose of our study, but may be useful, for example, to optimize the management of hypoglycemia.

However, with regard to C-peptide level after rejection patient No. 2 data, a statistically significant difference was found between the groups of patients with ArH and other causes of hyperinsulinemic NDH (the median of the C-peptide level in the group of ArH patients was 1.82 [1.18; 2,73]). In perspective, C-peptide can also be considered as an additional indicator in the differential diagnosis of these conditions.

It should be emphasized that we have determined only 7 substances, unlike other researchers [19, 21, 22], who analyzed the presence of more OHDs in the patients' serum. The choice of a limited number of substances is explained by their availability in Russia. As new approved OHDs become available, consideration should be given to their inclusion in our sOHD dashboard. It is also promising to investigate other OHDs (metformin, dapagliflozin, vildagliptin, liraglutide, etc.) that we did not include in the study due to their low likelihood of causing manifest hypoglycemia. But it is possible that such an effect can be achieved, for example, by taking excessively high doses of these drugs.

CONCLUSION

The HPLC-MS/MS method has high diagnostic sensitivity and specificity in the detection and identification of sOHD (glibenclamide, glycvidone, gliclazide, glimepiride, glipizide, nateglinide and repaglinide) in blood samples of patients receiving these drugs. Currently, due to the low availability of the method, it is advisable for use only in patients with hyperinsulinemic hypoglycemia and negative results of first-line insulinoma imaging studies.

Further studies in an expanded group of patients with ArH are required to confirm our data on the possibility of using insulin and C-peptide parameters in the period of manifest hypoglycemia as additional markers of this condition.

A promising area for further research is the inclusion of new hypoglycemic drugs in the sOHD panel as they receive marketing authorizations and are launched in the Russian pharmaceutical market, as well as currently available drugs that are not related to the groups of sulfonylureas or glinides.

FURTHER INFORMATION

Conflict of interest. The authors declare no clear and potential conflicts of interest related to the publication of this paper.

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