EXENATIDE TREATMENT REDUCES THYROID GLAND VOLUME, BUT HAS NO EFFECT ON THE SIZE OF THYROID NODULES

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Abstract

Context. Exenatide is a Glucagon-like Peptide-1 receptor agonist, which is widely used for type 2 diabetes mellitus (T2DM). Limited and conflicting results are present about the effect of exenatide on the thyroid gland.

Objective. The aim of this study was to evaluate the effect of exenatide treatment on structural and functional features of the thyroid gland in patients with T2DM.

Design. The study was a prospective study, performed between 2015 and 2017. The laboratory values and thyroid ultrasonography features were compared before and after exenatide treatment.

Subjects and Methods. The study included 39 obese diabetic patients. After inclusion to the study exenatide was started and patients were followed up for 6 months. Total thyroid volume, thyroid function tests, serum carcinoembryonic antigen (CEA) and calcitonin levels, the size and appearance of thyroid nodules were compared between baseline and after 6 months of treatment.

Results. Exenatide at a dose of 5 µg bid was started, increased to 10 µg bid after 4 weeks. We found a statistically significant decrease in thyroid volume ($p=0.043$) and serum thyroid stimulating hormone (TSH) levels ($p=0.007$), whereas serum ATPO, ATGI, fT4, fT3, CEA and calcitonin levels did not change with 6 months of exenatide treatment. There were no significant differences in the size and appearance of the thyroid nodules with treatment. The thyroid volume decrease was not correlated with TSH, body mass index and HbA1c reduction.

Conclusion. Exenatide treatment for 6 months decreased serum TSH levels and thyroid volume, but had no effect on thyroid nodules and serum CEA and calcitonin levels.

Keywords: calcitonin, carcinoembryonic antigen, diabetes mellitus, exenatide, thyroid nodule, thyroid volume.

INTRODUCTION

Glucagon-like Peptide-1 (GLP-1) is a gastrointestinal hormone, which has beneficial effects

on blood glucose by increasing glucose dependent insulin secretion, suppressing glucagon release, reducing appetite and delaying gastric emptying (1, 2). Exenatide is a GLP-1 receptor agonist, which was approved for the treatment of type 2 diabetes mellitus (T2DM) (3). Limited data is available about the effect of exenatide on the thyroid gland. GLP-1 receptors have 93% homology between humans and rats (4, 5). Animal studies have shown that GLP-1 receptor agonists liraglutide and exenatide increase serum calcitonin levels, which is a marker for thyroid C-cell diseases (5). With very high doses of administration, liraglutide caused C-cell carcinoma and exenatide increased the risk for C-cell adenomas in rodents (6-8). A study examining the US Food and Drug Administration's database of reported adverse events, found an increased risk for thyroid cancer in patients treated with exenatide (9). Recently, it has been shown that treatment with exenatide for a median time of 3.2 years had no effect on serum calcitonin levels in humans (10). Studies demonstrated that not only C cells and medullary thyroid carcinoma cells, but also the normal thyroid tissue and papillary thyroid carcinoma may express GLP-1 receptors (11, 12). Because of these findings exenatide may have unexpected and several effects on the thyroid gland. To investigate these possible effects, Sencar *et al.* compared the thyroid volume and thyroid stimulating hormone (TSH) levels before and after exenatide treatment and found no difference in thyroid volume, despite a statistically significant decrease in TSH levels (13). To the best of our knowledge, the effect of exenatide on thyroid nodules was not formerly studied.

The aim of this study was to examine the effect of 6 months of exenatide treatment on structural and functional features of the thyroid in obese patients with T2DM.

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MATERIALS AND METHODS

This was a prospective single-center study including obese patients with T2DM, who admitted to our hospital between 2015 and 2017. All patients included in this trial had a body mass index (BMI) over 35 kg/m², and did not use exenatide before the study participation. All patients were using oral antidiabetic drugs, and patients who were using insulin were not included for statistical analysis. Patients who had formerly thyroid surgery, hypothyroidism or hyperthyroidism were excluded from the study. Patients who were taking levothyroxine or statins were also excluded from the study. Patients with Cushing's syndrome, coronary artery diseases, heart failure, cirrhosis and formerly known malignancies were excluded from the study, either. After inclusion in the study, exenatide at a dose of 5 µg twice a day was started and the dose was increased to 10 µg twice daily after 4 weeks. A standard hypocaloric diabetic diet and regular standard aerobic exercise were prescribed under the supervision of a dietician.

At baseline clinical characteristic features, demographic data, the duration of T2DM, comorbidities and concomitant medications were recorded. Weight and height were measured and BMI was calculated as the weight (kg) divided by the square of the height (m), for all patients at the beginning and after 6 months of the treatment. All patients underwent laboratory analysis before the treatment and after 6 months of the study participation. The blood samples were collected between 08:00 and 09:00. The laboratory analysis included creatinine, alanine aminotransferase (ALT), fasting plasma glucose (FGP), post-prandial plasma glucose (PPG), glycated hemoglobin (HbA1c), thyroid function tests [free triiodothyronine (fT3), free thyroxine (fT4) and TSH], calcitonin, carcinoembryonic antigen (CEA) and thyroid antibodies [Anti thyroglobuline (Anti Tg) and Anti-thyroid peroxidase (TPO)]. HbA1c was measured with the high-performance liquid chromatography method and automated direct chemiluminescent immunoassay was used for thyroid function tests, calcitonin, CEA and thyroid antibodies. The normal ranges for TSH, fT3 and fT4 were 0.34-5.6 µIU/mL, 2.5-3.9 pg/mL and 0.61-1.12 ng/dL, respectively. The upper limit of normal calcitonin, CEA, Anti TPO and anti TG were 11.5 pg/mL, 5ng/mL, 34 IU/mL and 115 IU/mL, respectively.

Thyroid ultrasonography (US) was performed at the beginning of the study and after 6 months of exenatide treatment, using high resolution B mode

US images (Hitachi EUB 8500) with a 12-MHz linear array transducer. At US examination the thyroid gland volume was calculated as "volume(mL)=length(cm)×width(cm)×depth (cm)×0.524" for each thyroid lobe separately and the results were summed (14). If nodules were detected on US, the sonographic echogenicity pattern and the three dimensions of nodules were recorded and the largest dimension of the largest nodule was included for analysis. Thyroid fine needle aspiration was performed if indicated. All of the US examinations were performed by the same clinician (D.K.).

The Local Ethics Committee approved the study design and methods and written informed consent was obtained from all individual participants included in the study.

Statistical analyses were performed with SPSS software 15.0 (SPSS, Chicago, IL, USA). Data with normal distribution variables were expressed as mean ± standard deviation (SD), and median (min-max) was used for not normally distributed data. Paired sample t test and the Wilcoxon signed rank test were used to compare values before and after therapy, for normally and not normally distributed data, respectively. Qualitative data were expressed as median (range) and compared with chi-square test. Pearson's correlation analysis was used to establish correlation. Multivariate linear regression analysis was performed to determine the independent predictors of the change in the thyroid volume. p values less than 0.05 were considered as statistically significant.

RESULTS

The study included 55 obese diabetic patients receiving exenatide. Among these patients 8 patients were excluded from the study because of lost to follow-up (n=4) and adverse events (nausea in 2 patients, abdominal pain in one patient and drug - related skin eruption in one patient). Eight patients were excluded because of levothyroxine use. The study included 35 women and 4 men with a mean age of 47.26±7.18 years. All patients were under treatment with oral metformin before the study participation, and 9 patients were using gliclazide with metformin. The dose of metformin or gliclazide did not change throughout the study. The median duration of T2DM was 5 (1-15) years. Among the 39 patients, 11 patients had controlled hypertension.

All of the patients lost weight and reached a reduction in HbA1c levels (Table 1). The laboratory parameters and thyroid ultrasonographic features

are summarized in Table 2. A statistically significant decrease was achieved in TSH ($p:0.007$) and thyroid volume ($p:0.043$), whereas fT3, fT4 and thyroid antibodies did not change with therapy. The multivariate linear regression test showed that BMI, HbA1c or TSH reduction were not correlated with the reduction of the thyroid volume. Correlation analysis showed that the thyroid volume was not correlated with HbA1c, TSH, fT3, fT4 levels and HbA1c.

Before the exenatide therapy was started, 70 thyroid nodules were detected in 24 patients. Of these patients: 10 patients had isoechoic nodules, 7 patients had hypoechoic nodules and 7 patients had mixed echoic nodules. Fine needle aspiration (FNA) was performed in 10 patients and all of the nodules showed benign histopathological findings. No statistically significant difference was detected in the largest nodule diameter before and after exenatide therapy (Table 2). After exenatide treatment 24 patients had a total of 72 thyroid nodules. After treatment, a smaller than 5 mm nodule with mixed echoic pattern was detected in one patient who had no nodule at the beginning of the study. FNA was not required for this nodule. Whereas one millimetric nodule, which was present at the beginning of the study could not be detected after therapy. The sonographic echogenicity

of the thyroid nodules did not change after treatment and none of the patients needed FNA after exenatide treatment. The CEA and calcitonin levels did not change with therapy.

DISCUSSION

In this study we evaluated the effect of exenatide on structural and functional features of the thyroid gland. Our results showed that 6 months of exenatide treatment decreased thyroid volume and TSH levels, whereas nodule size, fT3, fT4, calcitonin and CEA levels did not change over time.

Limited data is available about the relationship between exenatide therapy and the thyroid gland. Animal studies have shown that exenatide increases serum calcitonin levels, and also the risk for C-cell adenomas (5-8). Conflicting and limited results are present in human studies. The odds ratio for thyroid cancer was detected as 4.73 by Elashoff *et al.*, but the subgroups of thyroid cancers were not mentioned in this study (9). However, a recently published study showed no increased risk for thyroid cancer with exenatide treatment (15). The inconsistency between animal and human studies may be due to the higher doses of drugs used in animal studies, and possible species

Table 1. The metabolic and laboratory parameters of the patients before the exenatide therapy and 6 months after exenatide was started

	Before exenatide	6 months after exenatide	P
Weight (kg)	112.83±13.40	103.09±13.85	<0.001
BMI (kg/m ²)	43.92±5.87	40.09±5.58	<0.001
Creatinine (mg/dL)	0.75±0.08	0.72±0.09	0.163
ALT (U/L)	23 (10-100)	19 (11-51)	0.005
Hemoglobin (g/dL)	13.35±1.19	13.15±1.27	0.218
FPG (mg/dL)	147 (100-277)	115 (83-176)	<0.001
PPG (mg/dL)	209 (92-386)	152 (83-217)	<0.001
HbA1c (%)	7.69±1.16	6.68±0.76	<0.001

BMI: Body mass index, ALT: alanine aminotransferase, FPG: fasting plasma glucose, PPG: post-prandial plasma glucose, HbA1c: glycated haemoglobin. Values are expressed as mean±standard deviation for normally distributed data and as median (min-max) for not normally distributed data.

Table 2. The thyroid laboratory parameters and thyroid ultrasonographic features before the exenatide therapy and 6 months after exenatide was started

	Before exenatide	6 months after exenatide	P
TSH (μIU/mL)	2.36±1.17	1.95±0.91	0.007
fT4 (ng/dL)	0.88 (0.65-1.46)	0.90 (0.75-1.03)	0.617
fT3 (pg/dL)	3.36±0.42	3.48±0.32	0.113
Anti TPO (IU/mL)	1.3 (0.1-661.7)	1.5 (0.1-636.8)	0.106
Anti TG (IU/mL)	0.6 (0.1-182.6)	2 (0.1-181)	0.847
CEA (ng/mL)	1.82±1.26	1.58±0.85	0.108
Calcitonin (pg/mL)	2 (2-10.3)	2 (2-10.8)	0.784
Thyroid volume (cm ³)	17.39±15.72	15.72±13.17	0.043
Nodule diameter* (mm)	6.15 (2.5-28)	6 (1-30)	0.648

*The largest diameter of the largest thyroid nodule. TSH: thyroid stimulating hormone, fT3: free triiodothyronine, fT4: free thyroxine, Anti TPO: Anti-thyroid peroxidase, Anti Tg: Anti thyroglobuline. CEA: carcinoembryonic antigen. Values are expressed as mean±standard deviation for normally distributed data and as median (min-max) for not normally distributed data.

specific differences in GLP-1 receptor expression. In our study we found no effect of exenatide on CEA and calcitonin levels, in accordance with the existing literature (10, 13). Moreover, we showed that the size and ultrasonographic features of benign thyroid nodules did not change with 6 months of exenatide treatment. To the best of our knowledge, the effect of exenatide on thyroid nodules was not formerly investigated and our study showed that exenatide treatment for 6 months has no effect on thyroid nodules. We may suggest that exenatide can be used safely in patients with benign thyroid nodules, but studies with extended durations are needed to support this finding. Twenty-four patients (61.5%) had one or more thyroid nodules at the beginning of the study. Thyroid nodules can be detected with ultrasonography in 19%-68% of randomly selected persons, and even higher frequencies are seen in women (16). Most of our patients were female (89.7%), and the detected thyroid nodule incidence in our study population is in accordance with the literature.

Our study showed for the first time in the literature that thyroid volume decreases with exenatide treatment. TSH is the main proliferation factor for the thyroid (17), and the reduction of TSH could be a reason for thyroid volume depletion. But our regression analysis showed no correlation between TSH reduction and thyroid volume decrease. Formerly it has been shown that patients with insulin resistance have larger thyroid glands (18, 19). The treatment with metformin showed also a reduction in thyroid volume (20). All of the patients involved in our study were also using metformin. Because the dose of metformin did not change throughout the study, the decrease of thyroid volume cannot be attributed to the metformin use. The decreases of BMI and HbA1c were also investigated whether they have an effect on the thyroid volume, but statistically significant results were not obtained. It is unknown whether exenatide affects other proliferation factors for the thyroid gland, and such an interaction may be the reason for the thyroid volume depletion. In this study we found a statistically significant decrease in thyroid volume, but the exact mechanism for this change remains unknown.

Administration of exendin to rats reduced TSH levels, and a similar effect was detected by Sencar *et al.* in humans (13,21). Also metformin was found to decrease TSH levels, without an effect on fT3 and fT4 (22). Our results about TSH levels were compatible with the existing literature. The effect of metformin and exenatide on TSH may be related to a common

mechanism of action of both drugs. Former studies evaluated the effect of BMI on TSH levels, but found no interaction (23, 24). Also our study showed no effect of BMI reduction in TSH decrease. The exact mechanisms of exenatide action on thyroid function tests are not known and need to be explained in further studies.

We have to mention some limitations of our study. The study group was rather small, but we included only obese patients with uncontrolled T2DM. Another limitation is that we gave 6 months of exenatide treatment, but a longer duration may have different effects on the thyroid gland. The exact mechanism for thyroid volume depletion could not be detected, and this remains another limitation of our study. However, these limitations do not hinder the results of the study. Our findings need to be supported with further studies including larger study population and longer study duration.

In conclusion, in this study we aimed to examine the effect of exenatide on the thyroid gland. After 6 months of exenatide therapy the thyroid volume and TSH levels decreased, with no change in fT3, fT4, CEA and calcitonin levels. Our study is among the first in the literature demonstrating a decrease in thyroid volume with exenatide treatment. Also the effects of exenatide on benign thyroid nodules were evaluated and no change was detected in nodule size and nodule appearance. This findings need to be supported with longer and larger studies in the future.

Conflict of interest

The authors declare that they have no conflict of interest.

References

1. De Leon DD, Crutchlow MF, Ham JY, Stoffers DA. Role of glucagon-like peptide-1 in the pathogenesis and treatment of diabetes mellitus. *Int J Biochem Cell Biol.* 2006; 38:845–859.
2. Samson SL, Garber A. GLP-1R agonist therapy for diabetes: benefits and potential risks. *Curr Opin Endocrinol Diabetes Obes.* 2013; 20:87–97.
3. Knop FK, Brønden A, Vilsbøll T. Exenatide: pharmacokinetics, clinical use, and future directions, *Expert Opinion on Pharmacotherapy* 2017; 18:555–571.
4. Mayo KE, Miller LJ, Bataille D, Dalle S, Göke B, Thorens B, Drucker DJ. International Union of Pharmacology. XXXV. The glucagon receptor family. *Pharmacol Rev.* 2003; 55:167–194.
5. Bjerre Knudsen L, Madsen LW, Andersen S, Almholt K, de Boer AS, Drucker DJ, Gotfredsen C, Egerod FL, Hegelund AC, Jacobsen H, Jacobsen SD, Moses AC, Mølck AM, Nielsen HS, Nowak J, Solberg H, Thi TD, Zdravkovic M, Moerch U. Glucagon-like peptide-1 receptor agonists activate rodent thyroid C-cells causing calcitonin release and C-cell proliferation. *Endocrinology* 2010; 151:1473–1486.

6. Capen CC, Martin SL. The effects of xenobiotics on the structure and function of thyroid follicular and C-cells. *Toxicol Pathol.* 1989; 17:266–293.
7. Martin-Lacave I, Bernab R, Sampedro C, Conde E, Fernández-Santos JM, San Martín MV, Beato A, Galera-Davidson H. Correlation between gender and spontaneous C-cell tumors in the thyroid gland of the Wistar rat. *Cell Tissue Res.* 1999; 297:451–457.
8. Madsen LW, Knauf JA, Gotfredsen C, Pilling A, Sjögren I, Andersen S, Andersen L, de Boer AS, Manova K, Barlas A, Vundavalli S, Nyborg NC, Knudsen LB, Moelck AM, Fagin JA. GLP-1 receptor agonists and the thyroid: C-cell effects in mice are mediated *via* the GLP-1 receptor and not associated with RET activation. *Endocrinology* 2012; 153:1538–1547.
9. Elashoff M, Matveyenko AV, Gier B, Elashoff R, Butler PC. Pancreatitis, pancreatic, and thyroid cancer with glucagon-like peptide-1-based therapies. *Gastroenterology* 2011; 141:150–156.
10. Bethel MA, Patel RA, Thompson VP, Merrill P, Reed SD, Li Y, Ahmadi S, Katona BG, Gustavson SM, Ohman P, Iqbal N, Gagel RF, Hernandez AF, Buse JB, Holman RR; EXSCEL Study Group. Changes in Serum Calcitonin Concentrations, Incidence of Medullary Thyroid Carcinoma, and Impact of Routine Calcitonin Concentration Monitoring in the EXenatide Study of Cardiovascular Event Lowering (EXSCEL). *Diabetes Care* 2019; 42:1075–1080.
11. Gier B, Butler PC, Lai CK, Kirakossian D, DeNicola MM, Yeh MW. Glucagon like peptide-1 receptor expression in the human thyroid gland. *J Clin Endocrinol Metab.* 2012; 97:121–131.
12. Korner M, Stockli M, Waser B, Reubi JC. GLP-1 receptor expression in human tumors and human normal tissues: potential for *in vivo* targeting. *J Nucl Med.* 2007; 48:736–743.
13. Sencar ME, Sakiz D, Calapkulu M, Hepsen S, Kizilgul M, Ozturk IU, Ucan B, Bayram M, Cagir BB, Akin S, Ozbek M, Cakal E. The Effect of Exenatide on Thyroid-Stimulating Hormone and Thyroid Volume. *Eur Thyroid J.* 2019; 8:307–311.
14. Shabana W, Peeters E, De Maesseneer M. Measuring thyroid gland volume: should we change the correction factor? *AJR Am J Roentgenol.* 2006; 186:234–236.
15. Liang C, Bertoia ML, Ding Y, Clifford CR, Qiao Q, Gagne JJ, Dore DD. Exenatide use and incidence of pancreatic and thyroid cancer: A retrospective cohort study. *Diabetes Obes Metab.* 2018 Nov 25. doi: 10.1111/dom.13597. [Epub ahead of print].
16. Haugen BR, Alexander EK, Bible KC, Doherty GM, Mandel SJ, Nikiforov YE, Pacini F, Randolph GW, Sawka AM, Schlumberger M, Schuff KG, Sherman SI, Sosa JA, Steward DL, Tuttle RM, Wartofsky L. 2015 American Thyroid Association Management Guidelines for Adult Patients with Thyroid Nodules and Differentiated Thyroid Cancer: The American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. *Thyroid.* 2016; 26: 1–133.
17. Kimura T, Van Keymeulen A, Golstein J, Fusco A, Dumont JE, Roger PP. Regulation of thyroid cell proliferation by TSH and other factors: a critical evaluation of *in vitro* models. *Endocr Rev.* 2001; 22:631–656.
18. Rezzonico J, Rezzonico M, Pusiol E, Pitoia F, Niepomniszcze H. Introducing the thyroid gland as another victim of the insulin resistance syndrome. *Thyroid* 2008; 18: 461–464.
19. Rogowicz-Frontczak A, Pilacinski S, Chwialkowska AT, Naskret D, Zozulinska-Ziolkiewicz D. Insulin resistance is associated with larger thyroid volume in adults with type 1 diabetes independently from presence of thyroid autoimmunity. *Scand J Clin Lab Invest.* 2018; 78:287–292.
20. Anil C, Kut A, Atesagaoglu B, Nar A, Bascil Tutuncu N, Gursoy A. Metformin Decreases Thyroid Volume and Nodule Size in Subjects with Insulin Resistance: A Preliminary Study. *Med Princ Pract.* 2016; 25:233–236.
21. Malendowicz LK, Nowak KW. Preproglucagon derived peptides and thyrotropin (TSH) secretion in the rat: robust and sustained lowering of blood TSH levels in exendin-4 injected animals. *Int J Mol Med.* 2002; 10:327–331.
22. Cappelli C, Rotondi M, Pirola I, Agosti B, Formenti A, Zarra E, Valentini U, Leporati P, Chiovato L, Castellano M. Thyreotropin levels in diabetic patients on metformin treatment. *Eur J Endocrinol.* 2012; 167:261–265.
23. MacCuish A, Razvi S, Syed AA. Effect of weight loss after gastric bypass surgery on thyroid function in euthyroid people with morbid obesity. *Clin Obes.* 2012; 2: 25–28.
24. Dall'Asta C, Paganelli M, Morabito A, Vedani P, Barbieri M, Paolisso G, Folli F, Pontiroli AE. Weight loss through gastric banding: effects on TSH and thyroid hormones in obese subjects with normal thyroid function. *Obesity (Silver Spring)* 2010; 18:854–857.